Title: FERMENTED WHEY PROTEIN-BASED CONCENTRATE, BEVERAGES CONTAINING THE SAME, AND METHODS FOR PRODUCING THEREOF

Abstract: The present invention provides a fermented whey protein-based concentrate comprising, with respect to the total of the weight of the concentrate, 3-10% w/w of total protein, 2-10% w/w of sugar, and 0.001-1.0% w/w of lactic acid bacteria; wherein not less than 60% w/w of the total protein is from whey protein, wherein the lactic acid bacteria is pasteurized. The present invention also provides a beverage containing said fermented whey protein-based concentrate. The invention also provides the methods for the preparation of said fermented whey protein-based concentrate and beverage.
FERMENTED WHEY PROTEIN-BASED CONCENTRATE, BEVERAGES CONTAINING THE SAME, AND METHODS FOR PRODUCING THEREOF

Field of the Invention

The present invention relates to a fermented whey protein-based concentrate, and beverages comprising the fermented whey protein-based concentrate. The present invention also relates to methods for producing the fermented whey protein-based concentrate and the beverages comprising the fermented whey protein-based concentrate.

Technical Background

Whey protein is a high quality powder made from cow's milk. Cow's milk is made up of two proteins - casein (80%) and whey (20%).

It has been widely learnt that whey protein is a high-quality complete protein, with all the essential amino acids, and whey protein is also the richest known source of naturally occurring branched chain amino acids.

Whey proteins have proven data for their excellent nutritional qualities and health benefits. Many drinks supplemented with whey proteins can be seen in market. However, whey proteins impart unpleasant taste, and whey proteins are unstable during heat treatment leading to sedimentation and precipitation in beverages. Because of these drawbacks, many commercially available beverages are only supplemented with little whey proteins. There is not a beverage in which most of the proteins are whey protein. For example, Kosikowski described a method for making of nutritional beverage from acid whey powder comprising blending acid whey powder with frozen, concentrated fruit juices, fresh fruit crystals or flavored powder bases produced acceptable nutrient beverages. Orange, grapefruit, and pineapple juice nutrient beverages showed excellent flavor, texture, and appearance qualities with 4% of powder in the reconstituted product and good qualities at 6% levels. Lemon juice, grape juice, and flavored powder nutrient beverages scored lower because of sedimentation. The acid reaction of a fruit juice and flavored powder base was retained in the nutritional fruit juice. In reconstituted orange nutrient beverage containing 6% whey powder, the maximum pH shift was from 3.75 to pH 3.96. (Kosikowski. (1968), Journal of Dairy Science, 51, 8, 1299-1301. )

US 2008/0187623 A1 introduces a drinking yoghurt comprising casein and whey protein in a casein:whey protein ratio of from 4:96 to 12:88. The present invention relates to a novel drinking yoghurt and a process for manufacturing thereof. While this drinking yoghurt has the same flavour and organoleptic characteristics as traditional drinking yoghurt, it has a different protein structure and a different composition with regard to the content of casein and whey
proteins. In particular, the drinking yoghurt comprises casein and whey protein in a casein:whey protein ratio of from 4:96 to 12:88 (w/w). The drinking yoghurt can be made without the formation of a coagulum after fermentation. In addition, the invention provides a process for the manufacturing of such a drinking yoghurt, comprising adding a quantity of whey product base to a quantity of milk product base to prepare a drinking yoghurt comprising casein and whey proteins in a casein:whey protein ratio of from 4:96 to 12:88 (w/w). However, the bases thereof do not contain lactic acid bacteria.

In addition, in JP2004-121046A, a fermented whey protein beverage was disclosed in which the protein content is about 1.6-5.6% which is originated from fermented whey concentrate. However, it does not use milk powder during fermentation. Also, in RU 22785 1C2, said patent disclosed a diary product which includes fermented whey protein and fermented milk powder although in which most the proteins originated from milk powder (ratio of milk to whey is 1:1 to 9:1).

Lactic acid bacteria is the most commonly used in dairy based and other fermented beverages, and have been studied widely for the generation of flavors and antimicrobial compounds in fermented foods. Lactic acid bacteria can reduce the pH by producing lactic acid and acetic acids, and also produce antimicrobial compounds. Therefore, the lactic acid bacteria bring the fermented products a better shelf life. Proteolytic activity of Lactic acid bacteria can generate small peptides as well as amino acids, that are much easier to be absorbed by the body.

A lot of literatures are available on Dairy fermentation to make yogurt and other beverages and the benefits thereof. Fermented dairy products have a positive image in the market and Whey proteins have been widely studied because of its excellent nutritional qualities and health benefits. For example, Pescuma M. et al. introduce the hydrolysis of whey proteins by *Lactobacillus acidophilus*, *Streptococcus thermophilus* and *Lactobacillus delbrueckii ssp. bulgaricus* grown in a chemically defined medium (Pescuma M., Hebert EM., Mozzi F. Valdez GF. (2007). *JAvyl Microbiol* 103, 5, 1738-1746).

US2009/00683 12A1 relates to a process for producing whey protein-enriched fermented milk of agitated type comprising: (i) adding, to a raw material milk, a reaction product which is obtained by reacting a transglutaminase with a whey protein solution, to obtain a mixture; and (ii) fermenting said mixture by inoculating a lactic acid bacterium. This invention does not use whey protein concentration.

Increasing consumer awareness and demand is noticeable for health and wellness drinks and there is a need for stable, good tasting, nutritional whey protein fermented drink.

**Summary of the Invention**
Accordingly, an object of the present invention is to provide a natural and healthy beverage containing fermented whey protein with excellent flavor, stability and nutrition.

Another object of the present invention is to offer a novel fermented whey protein-based concentrate from which the above-mentioned beverage is obtained by means of dilution.

A further object of the present invention is to provide an economical process for producing said beverage.

Thus, one aspect of the invention relates to a fermented whey protein-based concentrate comprising, with respect to the total of the weight of the concentrate, 3-10% w/w, preferably 4-8% w/w, more preferably 6-7% w/w of protein, and 0.001-1.0% w/w of lactic acid bacterial; wherein not less than 60% w/w, preferably 70% w/w, most preferably 80% w/w of the protein is from whey protein, wherein said lactic acid bacteria is pasteurized after fermentation.

The pH of the fermented whey protein-based concentrate is in the range of 3-5, preferably 3.5-4.5, most preferably 3.8-4.2.

In an optional embodiment, said fermented whey protein-based concentrate further comprises, based on the total weight of the concentrate, 1-20% w/w, preferably 3-10% w/w, most preferably 5-8% w/w of fruit juice or puree.

In another embodiment, said lactic acid bacteria is the mixture of bacteria for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH, wherein the bacteria for yogurt is selected from *Streptococcus thermophilus*, *Lactobacillus delbrueckii subsp. Bulgaricus*, and the mixture thereof, the bacteria for masking the bad taste is *Lactococcus lactis subsp. lactis biovar.diacetylactis*, and the bacteria for lowering the pH is *Lactobacillus acidophilus N'CFM R*, wherein the weight ratio of the bacteria for yogurt to (the bacterium for masking the bad taste + the bacterium for lowering the pH) is 1:1-1:2.

Another aspect of the invention relates to a beverage obtained by diluting said fermented whey protein-based concentrate with water, wherein the total protein concentration is 0.5-4% w/w, preferably 1.0-4.0% w/w, most preferably 1.0-2.0% w/w based on the total weight of the beverage.

The further aspect of the present invention relates to a process for the preparation of said fermented whey protein-based concentrate, comprising:

(i) preparing a concentrated base of whey protein and milk protein by combining the whey protein concentrate, the whole milk powder with water, in which the amount of the total protein content is 3-10% w/w, preferably 4-8% w/w, more preferably 6-7% w/w of the base,
wherein not less than 60% w/w, preferably 70% w/w, most preferably 80% w/w of the protein is from whey protein, the protein content in the whole milk powder is not less than 25% w/w;

(ii) adding sugar into the base in the range of 2-10% w/w, preferably 7-10% w/w of sugar based on the total weight of the base;

(iii) pasteurizing at 63°C/30min to form a pasteurized concentrate base;

(iv) adding a mixture of lactic acid bacteria consisting of consisting of a bacterium for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH, the adding amount of said mixture is 0.001-1.0% w/w, preferably 0.01-1.0% w/w, most preferably about 0.01-0.8% w/w, with respect to the total of the weight of the concentrate;

(v) fermenting at temperature 37-43°C for 6-16 hours;

(vi) pasteurizing the substances obtained from step (v) at 63°C/30min to obtain the fermented whey protein concentrate.

In addition, if necessary, fruit juice or puree including but not limited to mango, grape, apple, pear, peach are added into the concentrate after step (vi). The amount of juice or puree added varies from 1-20% w/w, preferably 3-10% w/w, most preferably 5-8% w/w based on the total weight of the concentrate. Alternatively, the juice or puree may be added into the concentrate before the step (v) of the fermentation.

Furthermore, the present invention also provide a method for producing the beverage containing the fermented whey protein, comprising the steps:

(i) preparing a concentrated base of whey protein and milk protein by combining the whey protein concentrate, the whole milk powder with water, in which the amount of the total protein content is 3-10% w/w, preferably 4-8% w/w, more preferably 6-7% w/w of the base,

wherein not less than 60% w/w, preferably 70% w/w, most preferably 80% w/w of the protein is from whey protein, the protein content in the whole milk powder is not less than 25% w/w;

(ii) adding sugar into the base in the range of 2-10% w/w, preferably 7-10% w/w of sugar based on the total weight of the base;

(iii) pasteurizing at 63°C/30min to form a pasteurized concentrate base;

(iv) adding a mixture of lactic acid bacteria consisting of consisting of a bacterium for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH, the adding amount of said mixture is 0.001-1.0% w/w, preferably 0.01-1.0% w/w, most preferably about 0.01-0.8% w/w, with respect to the total of the weight of the concentrate;

(v) fermenting at temperature 37-43°C for 6-16 hours;

(vi) pasteurizing the substances obtained from step (v) at 63°C/30min to obtain the fermented whey protein concentrate; and
diluting the fermented whey protein-based concentrate obtained from step (vi) with water, to obtain the desired beverage;

wherein the total protein concentration is 0.5-4% w/w based on the total weight of the beverage, comprising step (vii).

According to the present application, the beverage and the method for the production thereof possess the following advantages: (1) fermentation of whey protein in a concentrated form with lactic bacteria resulted in overall good taste and flavor; (2) economical process as the fermented concentrate can be further diluted with water to desired protein content, hence involves less capital expenses requirements, tank space, ease of handling and operations; (3) the method utilizes benefits of high quality whey protein and natural fermentation; all resulting in good tasting, natural, healthy drink with excellent flavor, taste, stability and nutrition.

**Detailed Description of the Preferred Embodiments**

The whey protein concentrate (WPC) used in the present invention has a protein content of not less than 80% w/w. The WPC used in the present invention could be, for example, WPC80 (Fonterra, New Zealand).

In one embodiment, wherein the whey protein concentrate accounts for 50-90% w/w of the total proteins, and the proteins originated from the whole milk powder accounts for 10-50% w/w of the total protein; The Whole Milk Powder used in the present invention could be, for example, WMP (Fonterra, New Zealand). The protein content in the Whole Milk Powder is not less than 25% w/w.

The sugar used in the present invention is commercial sugars, which could be one or more than one sugars selected from the group consisting of Sucrose, fructose, glucose and other chemical sweeteners.

Whey proteins are recognized as being of very high nutritional quality and value. However, whey supplementation quantity is generally limited because whey imparts bad taste as well as whey protein denatures causing sedimentation problem due to heat instability.

The present inventors surprisingly find that if the whey protein concentrate (WPC) is fermented with a lactic acid bacterial mixture consisting of a bacterium for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH, the obtained products possess excellent flavor, taste, stability and nutrition.

In one embodiment, the bacteria for yogurt is selected from *Streptococcus thermophilus, Lactobacillus delbrueckii subsp. Bulgaricus*, and the mixture thereof; the bacteria for masking the bad taste is *Lactococcus lactis subsp. lactis biovar.diacetylactis*, and the bacteria for lowering the pH is *Lactobacillus acidophilus*
NCFM, wherein the ratio of the weight of the bacteria for yogurt to the sum weight of the bacterium for masking the bad taste and the bacterium for lowering the pH is 1:1:1:2.

In another embodiment, the lactic acid bacterial mixture includes *Streptococcus thermophilus, Lactobacillus delbrueckii subsp. Bulgaricus, Lactococcus lactis subsp. lactis biovar.diacetylactis*, and *Lactobacillus acidophilus NCFM*, the fermented beverage has reduced off taste from whey. Concretely, *Streptococcus thermophilus, Lactobacillus delbrueckii subsp. Bulgaricus* belong to conventionally used yoghurt bacteria, *Lactococcus lactis subsp. lactis biovar.diacetylactis* is used to mask the bad taste from Whey, and *Lactobacillus acidophilus NCFM* is for the acidification of the pH as 3.8-4.2.

In another embodiment, the weight ratio of the whey protein concentrate (WPC) to the whole milk powder (WMP) is 1.4-1.0, preferably 1.45-1.0, the most preferably 1.5-1.0.

The amount of the mixture of lactic acid bacteria is 0.001-1.0% w/w, preferably 0.01-0.1%, w/w, most preferably about 0.01-0.8% w/w based on the weight of the concentrate base.

In another embodiment, said fermented whey protein-based concentrate further comprises, based on the total weight of the concentrate, 1-20% w/w, preferably 3-10% w/w, most preferably 5-8% w/w of fruit juice or puree.

Said fruit juice or puree includes, but not limited to, mango, grape, apple, pear, peach and the like. The amount of juice or puree is from 1-20% w/w, preferably 3-10% w/w, most preferably 5-8% w/w based on the total weight of the concentrate. In one embodiment, the juice or puree is added into the fermented whey protein-based concentrate obtained from step (iv). In another embodiment, the juice or puree may be added into the concentrate before the step (v) of the fermentation.

The invention beverage is obtained by diluting the fermented whey protein-based concentrate with water, wherein the total protein concentration is 0.5-4% w/w based on the total weight of the beverage.

In one embodiment, the beverage could be flavored or supplemented with fruit puree, juice to meet additional market and consumers needs.

In another embodiment, the beverage further comprises 0.1-1.0% w/w of, based on the total weight of the beverage, stabilizers and/or 0.1-1.0% w/w of, based on the total weight of the beverage, starch.

The beverage could be prepared by the following steps:

(i) preparing a concentrated base of whey protein and milk protein by combining the whey protein concentrate, the whole milk powder with water, in which the amount of the total protein content is 3-10% w/w, preferably 4-8% w/w, more preferably 6-7% w/w of the base,
wherein not less than 60% w/w, preferably 70% w/w, most preferably 80% w/w of the protein is from whey protein, the protein content in the whole milk powder is not less than 25% w/w;

(ii) adding sugar into the base in the range of 2-10% w/w, preferably 7-10% w/w of sugar based on the total weight of the base;

(iii) pasteurizing at 63°C/30min to form a pasteurized concentrate base;

(iv) adding a mixture of lactic acid bacteria consisting of a bacterium for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH, the adding amount of said mixture is 0.001-1.0% w/w, preferably 0.01-1.0% w/w, most preferably about 0.01-0.8% w/w, with respect to the total of the weight of the concentrate;

(v) fermenting at temperature 37-43°C for 6-16 hours;

(vi) pasteurizing the substances obtained from step (v) at 63°C/30min to obtain the fermented whey protein concentrate; and

(vii) diluting the fermented whey protein-based concentrate obtained from step (vi) with water to obtain the fermented beverage.

Preferably, it further comprises step (viii): the fermented beverage obtained from step (vii) is homogenized, pasteurized and hot / aseptic filled in bottles.

The invention will now be described in further details in the following non-limiting examples.

Examples

Example 1: fermented whey protein concentrate 1

623 g of water at 60°C was used to dissolve 58.0 g of Whey Protein Concentrate, WPC (Fonterra, New Zealand) and 38.52 g of Whole Milk Powder (Fonterra, New Zealand). The ratio between WPC and WMP is 1.5:1.0. The obtained solution was well hydrated under agitation for 15 to 30 minutes. 80 g of sucrose was added and dissolved therein. The obtained solution was filled to bottles, and pasteurized at 63°C for 30 minutes. After pasteurization, the solution was cooled to 40°C for fermentation. Bacterial cultures (Danisco, China) - 0.16 g each of YO MIX 505 (has 2 kinds of bacteria - Streptococcus thermophilus & Lactobacillus delbrueckii subsp. Bulgaricus, CHOOZIT MD88 (Lactococcus lactis subsp. lactis biovar.diacetylactis), NCFM 150 B (Lactobacillus acidophilus NCFM R) were added to the solution and incubated at 40°C for 10 hours. On completion of fermentation, fermented concentrate was cooled and stored at 4°C. The pH of the concentrate is 4.0.

Example 2: fermented whey protein concentrate 2 (different WPC: WMP ratio)
621.52 g of water at 60° C was used to dissolve 58.0 g of Whey Protein Concentrate, WPC (Fonterra, New Zealand) and 40 g of Whole Milk Powder (Fonterra, New Zealand). The ratio between WPC and WMP is 1.45:1.0. The obtained solution was well hydrated under agitation for 15 to 30 minutes. 80 g of sucrose was added and dissolved therein. The obtained solution was filled to bottles, and pasteurized at 63°C for 30 minutes. After pasteurization, the solution was cooled to 40°C for fermentation. Bacterial cultures (Danisco, China) - 0.16 g each of YO MIX 505 (has 2 kinds of bacteria - Streptococcus thermophilus & Lactobacillus delbrueckii subsp. Bulgaricus, CHOOZIT MD88 (Lactococcus lactis subsp. lactis biovar.diacetylactis), NCFM 150 B (Lactobacillus acidophilus NCFM R) were added to the solution, and incubated at 40°C for 10 hours. On completion of fermentation, fermented concentrate was cooled and stored at 4°C. The pH of the concentrate is 4.0

Example 3: fermented whey protein concentrate 3 (different WPC:WMP ratio)

619.52 g of water at 60° C was used to dissolve 58.0 g of Whey Protein Concentrate, WPC (Fonterra, New Zealand) and 42 g of Whole Milk Powder (Fonterra, New Zealand). The ratio between WPC and WMP is 1.40:1.0. The obtained solution was well hydrated under agitation for 15 to 30 minutes. 80 g of sucrose was added and dissolved therein. The obtained solution was filled to bottles, and pasteurized at 63°C for 30 minutes. After pasteurization, the solution was cooled to 40°C for fermentation. Bacterial cultures (Danisco, China) - 0.16 g each of YO MIX 505 (has 2 kinds of bacteria - Streptococcus thermophilus & Lactobacillus delbrueckii subsp. Bulgaricus, CHOOZIT MD88 (Lactococcus lactis subsp. lactis biovar.diacetylactis), NCFM 150 B (Lactobacillus acidophilus NCFM R) were added to the solution, and incubated at 40°C for 10 hours. On completion of fermentation, fermented concentrate was cooled and stored at 4°C. The pH of the concentrate is 4.0

Example 4: fermented whey protein concentrate with mango puree

583 g of water at 60° C was used to dissolve 58.0 g of Whey Protein Concentrate, WPC (Fonterra, New Zealand) and 38.52 g of Whole Milk Powder (Fonterra, New Zealand). The obtained solution was well hydrated under agitation for 15 to 30 minutes. 80g of commercial sugar (swire) and 40 g of Mango puree (Dachuan, China) is added and dissolved therein. The obtained solution was filled manually in bottles, and pasteurized at 63°C for 30 minutes. After pasteurization, solution was cooled to 40°C for fermentation. Bacterial cultures (Danisco, China) - 0.16 g each of YO MIX 505 (has 2 kinds of bacteria - Streptococcus thermophilus & Lactobacillus delbrueckii subsp. Bulgaricus, CHOOZIT MD88 (Lactococcus lactis subsp. lactis biovar.diacetylactis), NCFM 150 B (Lactobacillus acidophilus NCFM R) were added to the solution, and incubated at 40°C for 10 hrs. On
completion of fermentation, the fermented concentrate was cooled and stored at 4°C. pH is 4.0.

Example 5: fermented whey protein concentrate with apple juice

616.2 g of water at 60° C was used to dissolve 58.0 g of Whey Protein Concentrate, WPC (Fonterra, New Zealand) and 38.52 g of Whole Milk Powder (Fonterra, New Zealand). The obtained solution was well hydrated under agitation for 15 to 30 minutes. 80g of commercial sugar (swire) and 6.8 g of concentrates apple juice (HaiSheng) is added and dissolved therein. The obtained solution was filled manually in bottles, and pasteurized at 63°C for 30 minutes. After pasteurization, solution was cooled to 40°C for fermentation. Bacterial cultures (Danisco, China) - 0.16 g each of YO MIX 505 (has 2 kinds of bacteria - Streptococcus thermophilus & Lactobacillus delbrueckii subsp. Bulgaricus, CHOOZIT MD88 (Lactococcus lactis subsp. lactis biovar.diacetylactis), NCFM 150 B (Lactobacillus acidophilus NCFM R) were added to the solution, and incubated at 40°C for 10 hrs. On completion of fermentation, the fermented concentrate was cooled and stored at 4°C. pH is 4.0.

Example 6 - Beverage with fruit (Mango) puree

A beverage was prepared from the obtained concentrate of Example 1 as follows:

500 g of fermented concentrate is taken. 900 g of water at 65°C was used to dissolve dry pre-blend of 18 g Gum (Danisco), 18 g Sugar (swire) and 2 g starch (National starch). Once dissolution is complete, the solution is added to the fermented concentrate. 103 g of Mango Puree (Dachuan, China), 70 g of the commercial sugar, and 2 g of flavor is added. 387 g of water is added and mixed to achieve a final diluted beverage. Beverage is then homogenized, pasteurized, and hot filled in bottles. Final beverage with protein in the range of 1.7% was achieved. pH is 4.0.

Example 7 - Beverage without fruit (Mango) puree

A beverage was prepared from the obtained concentrate of Example 6 as follows:

500 g of fermented concentrate is taken. 900 g of water at 65°C was used to dissolve dry pre blend of 18 g Gum (Danisco), 18 g Sugar and 2 g starch (National starch). Once dissolution is complete solution is added to fermented concentrate. 70 g of commercial sugar (swire) and 2 g of flavor (Symrise) is added. 490 g of water is added and mixed to achieve final diluted beverage. Beverage is then homogenized, pasteurized and hot filled in bottles. Final beverage with protein in the range of 1.7% was achieved. pH is 4.0.
Example 8 - Beverage with fruit (Mango) puree added before fermentation

A beverage was prepared from the obtained concentrate of Example 4 as follows:

500 g of fermented concentrate is taken. 900 g of water at 65°C was used to dissolve dry pre blend of 18 g Gum (Danisco), 18 g Sugar (swire) and 2 g starch (National starch). Once dissolution is complete, the solution is added to fermented concentrate. 70 g of commercial sugar (swire), 78 g of Mango Puree (Dachuan, China) and 2 g of flavor (Symrise) is added. 412 g of water is added and mixed to achieve final diluted beverage. Beverage is then homogenized, pasteurized and hot filled in bottles. Final beverage with protein in the range of 1.7% was achieved. pH is 4.0.

Example 9 (beverage made from the concentrate of Example 2)

500 g of fermented concentrate is taken. 900 g of water at 65°C was used to dissolve dry pre blend of 18 g Gum (Danisco), 18 g Sugar (trade name?) and 2 g starch (National starch). Once dissolution is complete, the solution is added to fermented concentrate. 70 g of commercial sugar (swire), 78 g of Mango Puree (Dachuan, China) and 2 g of flavor (Symrise) is added. 412 g of water is added and mixed to achieve final diluted beverage. Beverage is then homogenized, pasteurized and hot filled in bottles. Final beverage with protein in the range of 1.75% was achieved. pH is 4.0.

Example 10 (beverage made from the concentrate of Example 3)

500 g of fermented concentrate is taken. 900 g of water at 65°C was used to dissolve dry pre blend of 18 g Gum (Danisco), 18 g Sugar (swire) and 2 g starch (National starch). Once dissolution is complete, the solution is added to fermented concentrate. 70 g of commercial sugar (swire), 78 g of Mango Puree (Dachuan, China) and 2 g of flavor (Symrise) is added. 412 g of water is added and mixed to achieve final diluted beverage. Beverage is then homogenized, pasteurized and hot filled in bottles. Final beverage with protein in the range of 1.75% was achieved. pH is 4.0.

Example 11 (beverage made from the concentrate of Example 5)

500 g of fermented concentrate is taken. 900 g of water at 65°C was used to dissolve dry pre blend of 18 g Gum (Danisco), 18 g Sugar (swire) and 2 g starch (National starch). Once dissolution is complete, the solution is added to fermented concentrate. 70 g of commercial sugar (swire), 16.65 g of apple juice (HaiSheng) and 2 g of flavor (Symrise) is added. 412 g of water is added and mixed to achieve final diluted beverage. Beverage is then homogenized, pasteurized and hot filled in bottles. Final beverage with protein in the range of
1.7% was achieved. pH is 4.0.

Example 12 - Beverage with higher protein content

A beverage was prepared from the obtained concentrate of Example 1 as follows:

500 g of fermented concentrate is taken. 675 g of water at 65°C was used to dissolve dry pre blend of 13.5 g Gum (Danisco), 13.5 g Sugar (swire) and 1.5g starch (National starch). Once dissolution is complete, the solution is added to fermented concentrate. 75 g of Mango Puree (Dachuan, China), 40.0 g of commercial sugar (swire) and 1.5 g of flavor (Symrise) is added. 180 g of water is added and mixed to achieve final diluted beverage. Beverage is then homogenized, pasteurized and hot filled in bottles. Final beverage with protein in the range of 2.3% was achieved. pH is 4.0.

Test Example 1: Evaluation of the taste of the invention beverage.

Below format was used for evaluation of various attributes of the product. 50 ml of the samples was served in transparent cups.
You are requested to evaluate the quality of the given **Fermented Whey beverage sample** through organoleptic analysis. Use the appropriate scale to know your attitude by assuming points that best describe your feeling about the sample. An honest expression will help us.

<table>
<thead>
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<th>Sample</th>
<th>Taste</th>
<th>Flavor</th>
<th>Mouth feel</th>
<th>Acidity</th>
<th>Texture</th>
<th>Color</th>
<th>Aftertaste</th>
<th>Off taste</th>
<th>Comments</th>
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</table>

**RATe**

- Very desirable  9
- Desirable  8
- Moderately desirable  7
- Slightly desirable  6
- Neither desirable nor desirable  5
- Slightly undesirable  4
- Moderately undesirable  3
- Undesirable  2
- Very undesirable  1

- Please rinse your mouth before and after tasting each product.  

Signature
Taste test on beverage of Example 6 (with mango puree)

<table>
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<th>Taster number</th>
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<th>Texture</th>
<th>Color</th>
<th>After taste</th>
<th>Off taste</th>
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<td>4</td>
<td>5</td>
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<td>9 ≠ off taste not detected</td>
<td>Can increase mango flavour a bit</td>
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<td>Natural profile but texture can be exceeded</td>
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<td>Better add yoghurt flavor to enhance, a little mild, mango aroma is good</td>
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<td>Sweet acid is not balance, week mango aftertaste, prefer texture</td>
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Average: 6.94, 6.71, 6.65, 6.59, 6.47, 6.35, 6.65, 6.18

Results

A beverage product of Example 6 was tasted, scores fall on acceptable side of the profile.
Taste test on beverage of Example 7 (without mango puree)

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<th>Acidity</th>
<th>Texture</th>
<th>Color</th>
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<th>Off taste</th>
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<td>Little like</td>
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<td>More clear or watery</td>
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<td>Refreshing clear mouthfeel</td>
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<td>None</td>
<td>Very crisp, clean, yogurt like taste</td>
<td>60</td>
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<td>8</td>
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<td>9</td>
<td>0</td>
<td>Product taste very good with clean notes (yogurt) with good mouth feel</td>
<td>61</td>
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</tbody>
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Average: 7.56, 7.67, 7.69, 7.67, 8.11, 8.44, 7.78

Results

A beverage product of Example 7 was and scores fall on acceptable side of the profile.
**Claims**

1. A fermented whey protein-based concentrate comprising, with respect to the total of the weight of the concentrate, 3-10% w/w of total protein, 2-10% w/w of sugar, and 0.001-1.0% w/w of lactic acid bacteria; wherein not less than 60% w/w of the total protein is from whey protein, wherein the lactic acid bacteria is pasteurized.

2. The fermented whey protein-based concentrate of claim 1, wherein the amount of total protein is 4-8% w/w based on the total weight of the concentrate; the amount of sugar is 7-10% w/w based on the total weight of the concentrate; the amount of the lactic acid bacteria is 0.01-1.0% w/w based on the total weight of the concentrate; wherein not less than 70% w/w of the total protein is from whey protein.

3. The fermented whey protein-based concentrate of claim 1, wherein the amount of total protein is 6-7% w/w based on the total weight of the concentrate; the amount of the lactic acid bacteria is 0.01-0.8% w/w based on the total weight of the concentrate; wherein not less than 80% w/w of the protein is from whey protein.

4. The fermented whey protein-based concentrate of any one of claims 1, it further comprises, based on the total weight of the concentrate, 1-20% w/w of fruit juice or puree.

5. The fermented whey protein-based concentrate of claim 4, wherein the amount of fruit juice or puree is 3-10% w/w based on the total weight of the concentrate.

6. The fermented whey protein-based concentrate of claim 5, wherein the amount of fruit juice or puree is 5-8% w/w, based on the total weight of the concentrate.

7. The fermented whey protein-based concentrate of any one of claims 1-6, wherein the pH of the fermented whey protein-based concentrate is in the range of 3-5.

8. The fermented whey protein-based concentrate of claim 7, wherein the pH is in the range of 3.5-4.5.

9. The fermented whey protein-based concentrate of claim 8, wherein the pH is in the range of 3.8-4.2.

10. The fermented whey protein-based concentrate of any one of claims 1-3, wherein the lactic acid bacteria is the mixture of bacteria for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH.

11. The fermented whey protein-based concentrate of claim 10, wherein the bacteria for yogurt is selected from *Streptococcus thermophilus*, *Lactobacillus delbrueckii* subsp. *Bulgaricus*, and the mixture thereof, the bacteria for masking the bad taste is *Lactococcus lactis* subsp. *lactis biovar. diacetylactis*, and the bacteria for lowering the pH is *Lactobacillus acidophilus* NCFM®, wherein the weight ratio of the bacteria for
yogurt to (the bacterium for masking the bad taste + the bacterium for lowering the pH) is 1:1-1:2.

12. A beverage comprising the fermented whey protein-based concentrate of any one of claims 1-11, wherein the total protein concentration is 0.5-4% w/w based on the total weight of the beverage.

13. The beverage of claim 12, wherein the total protein concentration is 1.0-4.0% w/w based on the total weight of the beverage.

14. The beverage of claim 12, wherein the total protein concentration is 1.0-2.0% w/w based on the total weight of the beverage.

15. The beverage of any one of claims 12-14, wherein the beverage further comprises 0.1-1.0% w/w of stabilizers and/or 0.1-1.0% w/w of sugar based on the total weight of the beverage.

16. A process for the preparation of the fermented whey protein-based concentrates of any one of claims 1-11, comprising:

(i) preparing a concentrated base of whey protein and milk protein by combining the whey protein concentrate, the whole milk powder with water, wherein not less than 60% w/w of the total protein is from whey protein, and the proteins originated from the whole milk powder accounts for 10-50% w/w of the total protein, and the amount of total protein is 3-10% w/w with respect to the total of the weight of the concentrate;

(ii) adding 2-10% w/w of sugar into the base obtained from step (i);

(iii) pasteurizing the base obtained from step (ii) at 63°C/30min to form a pasteurized concentrate base;

(iv) adding 0.001-1.0% w/w of a mixture of lactic acid bacteria consisting of a bacterium for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH, into the base obtained from step (iii);

(v) fermenting at temperature 37-43°C for 6-16 hours;

(vi) pasteurizing the substances obtained from step (v) at 63°C/30min to obtain the fermented whey protein concentrate.

17. The process of claim 16, wherein the amount of total protein in step (i) is 4-8% w/w based on the total weight of the concentrate; the amount of sugar in step (ii) is 7-10% w/w based on the total weight of the concentrate; the amount of the lactic acid bacteria in step (iv) is 0.01-1.0% w/w based on the total weight of the concentrate; wherein not less than 70% w/w of the total protein is from whey protein.

18. The process of claim 17, wherein the amount of total protein is 6-7% w/w based on the total weight of the concentrate; the amount of the lactic acid bacteria is 0.01-0.8% w/w based on the total weight of the concentrate; wherein not less than 80% w/w of the protein is from whey protein.
19. The process of any one of claims 16-18, it further comprises step (a), inserted between step (iv) and step (v), or after step (vi): adding, based on the total weight of the concentrate, 1-20% w/w of the fruit juice or puree.

20. The process of claim 19, wherein the amount of fruit juice or puree is 3-10% w/w based on the total weight of the concentrate.

21. The process of claim 20, wherein the amount of fruit juice or puree is 5-8% w/w, based on the total weight of the concentrate.

22. The process of claim 16, wherein the bacteria for yogurt is selected from *Streptococcus thermophilus, Lactobacillus delbrueckii subsp. Bulgaricus*, and the mixture thereof, the bacterium for masking the bad taste is *Lactococcus lactis subsp. lactis biovar.diacetylactis*, and the bacterium for lowering the pH is *Lactobacillus acidophilus NCFM*<sup>R</sup>, wherein the weight ratio of the bacteria for yogurt to (the bacterium for masking the bad taste + the bacterium for lowering the pH) is 1:1-1:2.

23. A method for producing the beverage of any one of claims 12—15, comprising:

(i) preparing a concentrated base of whey protein and milk protein by combining the whey protein concentrate, the whole milk powder with water, wherein not less than 60% w/w of the total protein is from whey protein, and the proteins originated from the whole milk powder accounts for 10-50% w/w of the total protein, and the amount of total protein is 3-10 % w/w with respect to the total of the weight of the concentrate;

(ii) adding 2-10 % w/w of sugar into the base obtained from step (i);

(iii) pasteurizing the base obtained from step (ii) at 63°C/30min to form a pasteurized concentrate base;

(iv) adding 0.001-1.0 % w/w of a mixture of lactic acid bacteria consisting of a bacterium for yogurt, a bacterium for masking the bad taste and a bacterium for lowering the pH, into the base obtained from step (iii);

(v) fermenting at temperature 37-43°C for 6-16 hours;

(vi) pasteurizing the substances obtained from step (v) at 63°C/30min to obtain the fermented whey protein concentrate; and (vii) diluting the fermented whey protein-based concentrate obtained from step (vi) with water to obtain the fermented beverage, so that the total protein concentration is 0.5-4% w/w based on the total weight of the beverage.

24. The method of claim 23, wherein the bacteria for yogurt is selected from *Streptococcus thermophilus, Lactobacillus delbrueckii subsp. Bulgaricus*, and the mixture thereof, the bacterium for masking the bad taste is *Lactococcus lactis subsp. lactis biovar.diacetylactis*, and the bacterium for lowering the pH is *Lactobacillus acidophilus NCFM*<sup>R</sup>, wherein the weight ratio of the bacteria for yogurt to (the bacterium for masking the bad taste + the bacterium for lowering the pH) is 1:1-1:2.
25. The method of claim 23 or 24, wherein the total protein concentration is 1.0-4.0% w/w based on the total weight of the beverage.

26. The method of claim 25, wherein the total protein concentration is 1.0-2.0% w/w based on the total weight of the beverage.

27. The method of claim 24, wherein in step (i), the amount of total protein is 4-8% w/w based on the total weight of the concentrate; in step (ii), the amount of sugar is 7-10% w/w based on the total weight of the concentrate; the amount of the lactic acid bacteria in step (iv) is 0.01-1.0% w/w based on the total weight of the concentrate; wherein not less than 70% w/w of the total protein is from whey protein.

28. The method of claim 27, wherein the amount of total protein is 6-7% w/w based on the total weight of the concentrate; the amount of the lactic acid bacteria is 0.01-0.8% w/w based on the total weight of the concentrate; wherein not less than 80% w/w of the protein is from whey protein.

29. The method of claim 23, it further comprises step (a), inserted between step (iv) and step (v), or after step (vi): adding, based on the total weight of the concentrate, 1-20% w/w of the fruit juice or puree.

30. The method of claim 29, wherein the amount of fruit juice or puree is 3-10% w/w based on the total weight of the concentrate.

31. The method of claim 30, wherein the amount of fruit juice or puree is 5-8% w/w, based on the total weight of the concentrate.

32. The method of claim 23, it further comprises step (viii): the fermented beverage obtained from step (vii) is homogenized, pasteurized and hot / aseptic filled in bottles.
# INTERNATIONAL SEARCH REPORT

**INTERNATIONAL APPLICATION No.**  
PCT/CN2010/071868

## A. CLASSIFICATION OF SUBJECT MATTER

A23C 21/02 (2006.01 i)

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC: A23C

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

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

CNPAT, CNKI, WPI, EPODOC, ISI Web of Knowledge: whey protein, WPC, lactic acid bacteria, sugar, pasteuriz+

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

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<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
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<td>US2009068312A1(AJIN), 12 Mar. 2009(12.03.2009), see whole document.</td>
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* Further documents are listed in the continuation of Box C.  

See patent family annex.

### Special categories of cited documents:

- **A** document defining the general state of the art which is not considered to be of particular relevance
- **E** earlier application or patent but published on or after the international filing date
- **L** document which may throw doubts on priority claim (S) or which is cited to establish the publication date of another citation or other special reason (as specified)
- **O** document referring to an oral disclosure, use, exhibition or other means
- **P** document published prior to the international filing date but later than the priority date claimed

- **T** later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- **X** document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
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### Date of the actual completion of the international search

10 Jan. 2011 (10.01.2011)

### Date of mailing of the international search report

20 Jan. 2011 (20.01.2011)

**Name and mailing address of the ISA/CN**  
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100088  
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WU Li

**Telephone No.** (86-10) 62411046

Form PCT/ISA/210 (second sheet) (July 2009)
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