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PROBIOTICS FOR DIETARY DAIRY PRODUCT

The present invention relates to compositions containing probiotic microorganisms and methods of making and using probiotic associations containing these microorganisms. More particularly, the invention relates to an association of probiotic lactic acid microorganisms including newly identified strains of microorganisms and their use in dietary products. These microorganisms are cultured to produce a protective shell that enhances self-stability of compositions and products of the invention. The isolated microorganisms include new strains of Lactobacillus delbrueckii subsp. bulgaricus, Lactobacillus helveticus, Lactobacillus delbrueckii subsp. lactis, and Streptococcus thermophilus.

Claims

1. A method of producing a dietary product comprising:
   a) mixing spring water form Mountain Stara Planina in Northern Bulgaria with pasteurized milk in a ratio of about 1:2 (v/v) to allow microorganisms in the spring water to multiply and to produce a starter material wherein the microorganisms in the spring water comprise strains: Lactobacillus delbrueckii subsp. bulgaricus DWT1 registered in CCM under no. 7992, Lactobacillus helveticus DWT2 registered in CCM under no. 7993, Lactobacillus delbrueckii subsp. lactis DWT3 registered in CCM under no. 7994, Streptococcus thermophilus DWT4 registered in CCM under no. 7992, Streptococcus thermophilus DWT5 registered in CCM under no. 7993, Streptococcus thermophilus DWT6 registered in CCM under no. 7994, Streptococcus thermophilus DWT7 registered in CCM under no. 7995, Streptococcus thermophilus DWT8 registered in CCM under no. 7996.
   b) mixing the starter material of step a) with pasteurized milk in ratio of about 3:10 (g/L) to allow fermentation;
   and
   c) lyophilizing the fermentation product to produce a dietary product.

2. The method of claim 1 further comprising combining the dietary product with nutritional supplement.

3. The method of claim 2, wherein the dietary product is shelf stabile for at least 12 months.

4. The method of claim 2, wherein the dietary product is shelf stabile for at least 22 months.

5. A method of producing a starter material, the method comprising mixing spring water from Mountain Stara Planina in Northern Bulgaria. with pasteurized milk in a ratio of about 1:2 (v/v) to allow microorganisms in the spring water to multiply and to produce a biomass for use as starter material; wherein the microorganisms in the spring water comprise strains: Lactobacillus delbrueckii subsp. bulgaricus DWT1 registered in CCM under no. 7992, Lactobacillus helveticus DWT2 registered in

CCM under no. 7993, Lactobacillus delbrueckii subsp. lactis DWT3 registered in CCM under no. 7994, Streptococcus thermophilus DWT4 registered in CCM under no. 7992, Streptococcus thermophilus DWT5 registered in CCM under no. 7993, Streptococcus thermophilus DWT6 registered in CCM under no. 7994, Streptococcus thermophilus DWT7 registered in CCM under no. 7995, Streptococcus thermophilus DWT8 registered in CCM under no. 7996.

6. A method of producing a dietary product comprising:

a) mixing a starter material obtained from using the method of claim 5 with pasteurized milk in ratio of about 3:10 (g/L) to allow fermentation; and

b) lyophilizing the fermentation product of step a) to produce a dietary product.

7. The method of claim 6, wherein the dietary product after lyophilizing contains alive cells of probiotic association of alive cells with concentration from $8.0x10^7$ cfu/g to $1.2x10^8$ cfu/g.

wherein the concentration of alive cells of the strains Streptococcus thermophilus ranges from $4,6x10^7$ cfu/g to $5,9x10^7$ cfu/g, including the strain Streptococcus thermophilus DWT4, CCM reg. No. 7992, strain Streptococcus thermophilus DWT5, CCM reg. No. 7993, strain Streptococcus thermophilus DWT6, CCM reg. No. 7994, strain Streptococcus thermophilus DWT7, CCM reg. No. 7995, strain Streptococcus thermophilus DWT8, CCM reg. No. 7996;

wherein the concentration of alive cells of strain Lactobacillus delbrueckii subsp. lactis DWT1, CCM reg. No. 7994 ranges from $6.1\ x10^6$ cfu/g to $7.8x10^6$ cfu/g,

wherein the concentration of alive cells of strain Lactobacillus delbrueckii subsp. bulgaricus DWT1, CCM reg. No. 7992 ranges from $1.4x10^7$ cfu/g to $2.9x10^7$ cfu/g and the concentration of alive cells of the strain Lactobacillus helveticus DWT2, CCM reg. No. 7993 ranges from $1.4x10^7$ cfu/g to $2.8x10^7$ cfu/g.

8. The method of claim 6, wherein the dietary product after rehydratation and in the extreme conditions of the gastro-intestinal tract contains a concentration no less than $1.1x10^7$ cfu/g of alive cells of the strain selected from Lactobacillus delbrueckii subsp. bulgaricus DWT1 registered in CCM under no. 7992, Lactobacillus helveticus DWT2 registered in CCM under no. 7993, Lactobacillus delbrueckii subsp. lactis DWT3 registered in CCM under no. 7994, Streptococcus thermophilus DWT4 registered in CCM under no. 7992, Streptococcus thermophilus DWT5 registered in CCM under no. 7993, Streptococcus thermophilus DWT6 registered in CCM under no. 7994, Streptococcus thermophilus DWT7 registered in CCM under no. 7995, Streptococcus thermophilus DWT8 registered in CCM under no. 7996.

9. A method of producing a dry starter culture comprising:

a) mixing about 3 gram of a starter material obtained from using the method of claim 5 to every 10 litres of pasteurized milk to allow fermentation and produce a liquid culture;

b) mixing the liquid culture from step a) with pasteurized milk in a ratio of about 1:60 (v/v) to make a mixture and to allow fermentation;

c) neutralizing the mixture of step b), if necessary, to optimize pH for the fermentation; and

d) lyophilize the fermentation product of step b) or c) to produce the dry starter culture.

10. A method of producing a dietary product comprising:

a) mixing a dry starter culture obtained from using the method of claim 9 with pasteurized milk in a ratio of about 3:5 (g/L) to allow fermentation; and

b) lyophilizing the fermentation product of step a) to produce a dietary product.

11. The method of claim 2, wherein the nutritional supplement in the dietary product is selected from the group consisting of coffee, cocoa, blueberry extract, papaya extract, oils, honey, dry plant extracts, fibres, enzymes, and any combinations thereof.