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— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*
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(54) **Title:** NEW LACTOBACILLUS PLANTARUM STRAIN IMPARTING HIGH THICKNESS AND/OR HIGH ROPINESS AND/OR HIGH MOUTH THICKNESS TO A DAIRY PRODUCT PRODUCED THEREWITH AND USES THEREOF

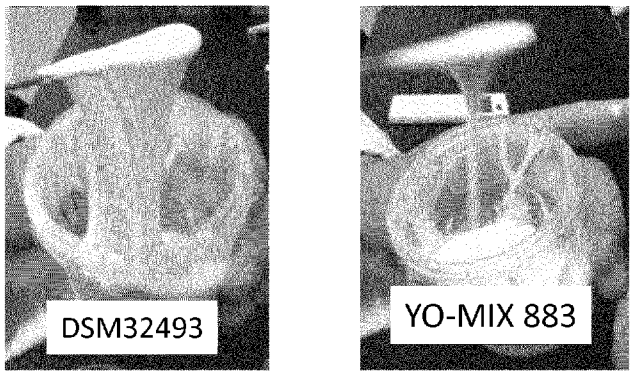

Fig. 6

(57) **Abstract:** The application is directed to a Lactobacillus plantarum strain, having the ability to generate a fermented milk presenting a high thickness and/or a high ropiness and/or a high thickness in mouth. The application also concerns a Lactobacillus plantarum strain, having the ability to generate a fermented milk presenting a high thickness and/or a high ropiness and/or a high thickness in mouth, further being a low post acidification strain at the temperature of fermentation. The application is also about a method to manufacture fermented product, in particular a fermented dairy product, using a L. plantarum strain of the invention or a bacterial composition, composition or kit-of part comprising a L. plantarum strain of the invention.

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NEW LACTOBACILLUS PLANTARUM STRAIN IMPARTING HIGH THICKNESS AND/OR HIGH ROPINESS AND/OR HIGH MOUTH THICKNESS TO A DAIRY PRODUCT PRODUCED THEREWITH AND USES THEREOF

FIELD OF THE INVENTION

The application is directed to a *Lactobacillus plantarum* strain, having the ability to generate a fermented milk presenting a high thickness and/or a high ropiness and/or a high thickness in mouth. The application also concerns a *Lactobacillus plantarum* strain, having the ability to generate a fermented milk presenting a high thickness and/or a high ropiness and/or a high thickness in mouth, further being a low post acidification strain at the temperature of fermentation. The application is also about a method to manufacture fermented product, in particular a fermented dairy product, using a *L. plantarum* strain of the invention or a bacterial composition, composition or kit-of part comprising a *L. plantarum* strain of the invention.

BACKGROUND TO THE INVENTION

Traditional starter cultures for fermented milk have been mainly developed for western countries (Europe, North America). The fermentative pathway is typically carried out by two different bacteria: *Streptococcus thermophilus* (ST) and *Lactobacillus delbruekii* subsp. *bulgaricus*.

Texture is a very important quality parameter for fermented milks and consumers continuously request fermented milk products having new taste and/or new texture. Among important texture descriptors for fermented products, high mouthfeel, long texture and smoothness are well liked by many consumers. There is therefore a need to improve the rheological properties of fermented milks.

Improvement of rheological properties of fermented milks can be obtained by the use of thickening agents, such as alginates. However, there is trend for more natural products perceived as healthy by the consumers, with a minimal of added ingredients (additive-free yoghurts).

WO2011/000879 and WO2011/000883 applications describe cultures comprising respectively *Streptococcus thermophilus* and *Lactobacillus fermentum* strains, and *Streptococcus thermophilus* and *Lactobacillus johnsonii* strains, and their use in the manufacture of low-fat fermented milk products. According to these applications, the replacement of the conventionally used *Lactobacillus delbruekii* subsp. *bulgaricus* by either *Lactobacillus fermentum* or *Lactobacillus johnsonii* - in combination with *Streptococcus thermophilus* strain(s) – would lead to low-fat fermented milk products having improved rheological properties (higher viscosity) and lower post-acidification.

There is still a need for new means to manufacture fermented products with differentiation in texture, in particular high mouth and spoon thickness and high stickiness, while limiting the presence of additives in the fermented products.

SUMMARY OF THE INVENTION

A first aspect of the invention relates to a *Lactobacillus plantarum* strain which – when inoculated into milk - generates a fermented milk presenting a high thickness and/or a high ropiness and/or a high thickness in mouth, in particular presenting one, two or three of the following rheological features, as assayed by test A: a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa; b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa; c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12.

A second aspect of the invention relates to a *Lactobacillus plantarum* strain which – when inoculated into milk - generates a fermented milk presenting a high thickness and/or a high ropiness and/or high thickness in mouth, and further characterized by being a low post acidification strain at the temperature of fermentation.

A third aspect of the invention relates to a bacterial composition comprising or consisting of a *Lactobacillus plantarum* strain of the invention, in particular in combination with at least another lactic acid bacteria.

A fourth aspect of the invention relates to a composition comprising or consisting of *the L. plantarum* strain of the invention or the bacterial composition of the invention and a booster.

A fifth aspect of the invention relates to a kit-of-part comprising or consisting of the *L. plantarum* strain of the invention or the bacterial composition of the invention and a booster.

A sixth aspect of the invention relates to a method for manufacturing a fermented product, comprising inoculating a substrate with the *L. plantarum* strain of the invention, the bacterial composition of the invention, the composition of the invention or the kit-of part of the invention, and fermenting said inoculated substrate, to obtain a fermented product.

A seventh aspect of the invention relates to a fermented product comprising the *L. plantarum* strain of the invention or obtained or obtainable by a method of the invention.

An eight aspect of the invention relates to the use of the *L. plantarum* strain of the invention, the bacterial composition of the invention, the composition of the invention or the kit-of part of the invention, in the manufacture of a fermented dairy product.

DESCRIPTION OF THE DRAWINGS

Figure 1 represents photographs of ropiness assay on *L. plantarum* strains carried out on agar plate. (A) no detectable ropiness; (B) low ropiness; (C) intermediate ropiness; (D) high ropiness.

Figure 2 shows flow curves of fermented milks obtained using 4 different *L. plantarum* strains.

Figure 3 is a schematic representation of the organoleptic properties of fermented milks obtained using 4 different *L. plantarum* strains.

Figure 4 represents photographs showing the appearance of fermented milks obtained using 4 different *L. plantarum* strains.

Figure 5 shows the flow curve of a fermented milk obtained using a *L. plantarum* strain of the invention compared to a flow curve of a fermented milk obtained using a traditional starter culture.

Figure 6 represents photographs showing the long texture descriptor of a fermented milk obtained using a *L. plantarum* strain of the invention as compared to a fermented milk obtained using a traditional starter culture.

Figure 7 shows the acidification profile (variation of pH over time) of milk fermented with two different *L. plantarum* strains of the invention.

Figure 8 shows the container used for fermentation and the stirring rake used for the rheology measurements in test A.

DETAILED DESCRIPTION

The inventors have surprisingly identified a lactic acid bacterium which can be used to obtain a fermented milk with unique texture and rheological properties. This lactic acid bacterium is a *Lactobacillus plantarum* strain (*L. plantarum*), and can be used to provide a fermented milk with high thickness and/or high ropiness and/or high thickness in mouth.

The invention is directed to a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting one, two or three of the following rheological features, as assayed by test A:

a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa;

b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa;

c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12.

For the avoidance of doubt, a *Lactobacillus plantarum* strain is defined herein as described in the literature, in particular in Skerman VBD, McGowan V, and Sneath PH; Int. J. Syst. Bacteriol., 30 (1980), 225-420.

The rheological parameters described herein have been assayed using the following Test A (also described in example 2). The shear stress value measured at shear rate 11.6 s^{-1} as defined herein and the shear stress value measured at shear rate 200 s^{-1} as defined herein are – as calculated by Test A – with a standard deviation of ± 1 Pa, between replicates within the same experiment. The difference value of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} as defined herein is – as calculated by Test A – with a standard deviation of ± 1, between replicates within the same experiment.

Thus, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 11.6 s^{-1} as defined herein and/or a shear stress measured at shear rate 200 s^{-1} as defined herein and/or a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} as defined herein, measured under the following conditions:

Test A

2L UHT milk (3.2% protein, 3.5% fat) added with 6% sucrose and 0.1% of yeast extract powder (LP0021) from Oxoid™ is heat treated at 85°C for 15 min and cooled down to room temperature. A culture of each strain to be tested is inoculated into milk at 1.10^7 CFU/ mL. The inoculated milk is then fermented at 30°C to reach pH 4.5 (typically about 22-24 h). After fermentation, the sample is stirred by a rotator (RZR 2051 control, Heidolph) at 100 rpm for 1.5 min and stored at 4°C overnight, before measurement by rheometer. The used tank and bladder are as disclosed in Fig. 8. The sample is assessed by a rheometer (Anton Paar MCR 302, CC27-SN27450, Austria) using a coaxial cylinder C-CC27-T200/SS and a bob-cup. The rheometer is set to a constant temperature of 10°C during the measurement. Settings are as follows:

- holding time (to rebuild to somewhat original structure): 10 minutes, without any physical stress applied to the sample.
- 25 measuring points over 500 s (one every 20 s)
- rotation step (to measure the shear stress at 11.6 1/s, the shear stress at 200 1/s and the difference of the shear stress at 146 1/s minus the shear stress at 41.1 1/s)

Two steps are designed:

Shear rate: d(gamma)/ dt = [0.1-200] 1/s log and [200-0.1] 1/s log

Each step contained 25 measuring points over 500 s (on every 20 s)

The shear stress measured at shear rate 11.6 s^{-1} represents the thickness of the fermented milk. According to the invention, the claimed *Lactobacillus plantarum* strain has the

ability to generate a fermented milk presenting a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa, as assayed by test A described herein. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 11.6 s^{-1} higher than 32 Pa. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 11.6 s^{-1} higher than 35 Pa. In an embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 11.6 s^{-1} in the range 30-45 Pa. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 11.6 s^{-1} in the range 32-40 Pa. The values or ranges of shear stress measured at shear rate 11.6 s^{-1} as described herein are disclosed individually or in combination with any values or ranges of shear stress measured at shear rate 200 s^{-1} described herein and/or any values or ranges of Δ of the shear stress measured at 146 s^{-1} - the shear stress measured at 41.1 s^{-1} described herein. These values and ranges are assayed by test A described herein.

The shear stress measured at shear rate 200 s^{-1} represents the coating or the mouth thickness (also called the thickness in mouth) of the fermented milk. According to the invention, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa as assayed by test A described herein. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} higher than 65 Pa. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} higher than 70 Pa. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} higher than 75 Pa. In an embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} in the range 60-85 Pa. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} in the range 65-85 Pa. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} in the range 70-82 Pa. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a shear stress measured at shear rate 200 s^{-1} in the range 75-80 Pa. The values or ranges of shear stress measured at shear rate 200 s^{-1} described herein are disclosed individually or in combination with any values or ranges of shear stress measured at shear rate 11.6 s^{-1} as described herein and/or any values or ranges of Δ of the shear stress measured at 146 s^{-1} -

the shear stress measured at 41.1 s^{-1} described herein. These values and ranges are assayed by test A described herein.

The difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} (Δ shear stress 146 s^{-1} - 41.1 s^{-1}) represents the ropiness or stickiness of the fermented milk. According to the invention, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a Δ shear stress 146 s^{-1} - 41.1 s^{-1} higher than 12. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a Δ shear stress 146 s^{-1} - 41.1 s^{-1} higher than 13. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a Δ shear stress 146 s^{-1} - 41.1 s^{-1} higher than 14. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a Δ shear stress 146 s^{-1} - 41.1 s^{-1} higher than 15. In an embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a Δ shear stress 146 s^{-1} - 41.1 s^{-1} in the range 12-18. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a Δ shear stress 146 s^{-1} - 41.1 s^{-1} in the range 13-17. In a particular embodiment, the claimed *Lactobacillus plantarum* strain has the ability to generate a fermented milk presenting a Δ shear stress 146 s^{-1} - 41.1 s^{-1} in the range 14-16. The values or ranges of Δ shear stress 146 s^{-1} - 41.1 s^{-1} described herein are disclosed individually or in combination with any values or ranges of shear stress measured at shear rate 11.6 s^{-1} as described herein and/or any values or ranges of shear stress measured at shear rate 200 s^{-1} described herein. These values and ranges are assayed by test A described herein.

The expression "*one, two or three of the [following] rheological features*" (in the context of the *L. plantarum strain*, the method, the fermented product or the use) means either:

- one rheological feature selected from the group consisting of a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa, b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, or
- two rheological features selected from the following combinations: 1) a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa and b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa, 2) a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, or 3) b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12; or

- three rheological features, *i.e.,* the combination of a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa, b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12.

Thus, in an embodiment, the invention concerns a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa, in particular higher than 32 Pa, in particular higher than 35 Pa, or in the range 30-45 Pa, in particular 32-40 Pa.

In an embodiment, the invention concerns a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa, in particular higher than 65 Pa, in particular higher than 70 Pa, in particular higher than 75 Pa or in the range 60-85 Pa, in particular 65-85 Pa, in particular 70-82 Pa, in particular 75-80 Pa.

In an embodiment, the invention concerns a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, in particular higher than 13, in particular higher than 14, in particular higher than 15, or in the range 12-19, in particular 13-18, in particular 14-17.

In an embodiment, the invention concerns a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa, in particular higher than 32 Pa, in particular higher than 35 Pa, or in the range 30-45 Pa, in particular 32-40 Pa; and b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa, in particular higher than 65 Pa, in particular higher than 70 Pa, in particular higher than 75 Pa or in the range 60-85 Pa, in particular 65-85 Pa, in particular 70-82 Pa, in particular 75-80 Pa. In a particular embodiment, said *Lactobacillus plantarum* strain - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} higher than 32 Pa and b) a shear stress measured at shear rate 200 s^{-1} higher than 70 Pa. In another particular embodiment, said *Lactobacillus plantarum* strain - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} in the range 32-40 Pa and b) a shear stress measured at shear rate 200 s^{-1} in the range 70-82 Pa.

In an embodiment, the invention concerns a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa, in particular higher than 32 Pa, in particular higher than 35 Pa, or in the range 30-45 Pa, in particular 32-40 Pa; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, in particular higher than 13, in particular higher than 14, in particular higher than 15, or in the range 12-19,

in particular 13-18, in particular 14-17. In a particular embodiment, said *Lactobacillus plantarum* strain - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} higher than 32 Pa; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 14. In another particular embodiment, said *Lactobacillus plantarum* strain - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} in the range 32-40 Pa; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} in the range 14-17.

In an embodiment, the invention concerns a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa, in particular higher than 65 Pa, in particular higher than 70 Pa, in particular higher than 75 Pa or in the range 60-85 Pa, in particular 65-85 Pa, in particular 70-82 Pa, in particular 75-80 Pa; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, in particular higher than 13, in particular higher than 14, in particular higher than 15, or in the range 12-19, in particular 13-18, in particular 14-17. In a particular embodiment, said *Lactobacillus plantarum* strain - when inoculated into milk - generates a fermented milk presenting b) a shear stress measured at shear rate 200 s^{-1} higher than 70 Pa; and c) a difference of the shear stress measured at 146 s-1 minus the shear stress measured at 41.1 s-1 higher than 14. In another particular embodiment, said *Lactobacillus plantarum* strain - when inoculated into milk - generates a fermented milk presenting b) a shear stress measured at shear rate 200 s^{-1} in the range 70-82 Pa; and c) a difference of the shear stress measured at 146 s-1 minus the shear stress measured at 41.1 s-1 in the range 14-17.

In an embodiment, the invention concerns a *Lactobacillus plantarum* strain which - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa, in particular higher than 32 Pa, in particular higher than 35 Pa, or in the range 30-45 Pa, in particular 32-40 Pa; b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa, in particular higher than 65 Pa, in particular higher than 70 Pa, in particular higher than 75 Pa or in the range 60-85 Pa, in particular 65-85 Pa, in particular 70-82 Pa, in particular 75-80 Pa; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, in particular higher than 13, in particular higher than 14, in particular higher than 15, or in the range 12-19, in particular 13-18, in particular 14-17. In a particular embodiment, said *Lactobacillus plantarum* strain - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} higher than 32 Pa; b) a shear stress measured at shear rate 200 s^{-1} higher than 70 Pa; and c) a difference of the shear stress measured at 146 s-1 minus the shear stress measured at 41.1 s-1 higher than 14. In another particular embodiment, said *Lactobacillus*

plantarum strain - when inoculated into milk - generates a fermented milk presenting a) a shear stress measured at shear rate 11.6 s^{-1} in the range 32-40 Pa; b) a shear stress measured at shear rate 200 s^{-1} in the range 70-82 Pa; and c) a difference of the shear stress measured at 146 s-1 minus the shear stress measured at 41.1 s-1 in the range 14-17.

Any *Lactobacillus plantarum* strain fulfilling one, two or three of the rheological feature(s) defined herein is part of the invention. The invention has been exemplified with the DSM32493 strain, deposited at the DSMZ. There is no reason to doubt that other *L. plantarum* strains sharing one, two or three of the rheological feature (s) defined herein with the DSM32493 strain exist, with the possibility of using the DSM32493 strain as a positive control.

In a particular embodiment, the invention is directed to the *Lactobacillus plantarum* strain DSM32493 deposited at the DSMZ on April 26th, 2017.

In a particular embodiment, the invention also concerns a variant of DSM32493. By "*variant of DSM32493 strain*", it means a *Lactobacillus plantarum* strain derived from the DSM32493 strain and which generates a fermented milk presenting one, two or three of the rheological features described herein (as assayed by test A), i.e., one, two or three of the following rheological features: a) a shear stress measured at shear rate 11.6 s^{-1} as defined herein and/or b) a shear stress measured at shear rate 200 s^{-1} as defined herein and/or c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1}. In a particular embodiment, the DSM32493 strain variant as defined herein is able to generate a fermented milk presenting, as assayed by test A, a) a shear stress measured at shear rate 11.6 s^{-1} (thickness) higher than 30 Pa; b) a shear stress measured at shear rate 200 s^{-1} (coating/mouth thickness) higher than 60 Pa; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} (ropiness/stickiness) higher than 12. In a particular embodiment, the DSM32493 strain variant as defined herein is able to generate a fermented milk presenting, as assayed by test A, a) a shear stress measured at shear rate 11.6 s^{-1} (thickness) higher than 32 Pa; b) a shear stress measured at shear rate 200 s^{-1} (coating/mouth thickness) higher than 70 Pa; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} (ropiness/stickiness) higher than 14. In a particular embodiment, the DSM32493 strain variant keeps the properties of the DSM32493 strain, with respect to generating a fermented milk presenting the three rheological features, as assayed by test A, a) a shear stress measured at shear rate 11.6 s^{-1} as defined herein, b) a shear stress measured at shear rate 200 s^{-1} as defined herein and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} as defined herein. By "*keep the properties*", it is meant that the values of the rheological features obtained using the DSM32493 variant are at least the values obtained using the DSM32493 strain. The definitions and specific embodiments detailed for the rheological features under the *L.*

plantarum strain characterization apply similarly in the context of any variant of DSM32493 strain, in particular for but not limited to, the minimal values and ranges of the shear stress measured at shear rate 11.6 s^{-1}, the minimal values and ranges of the shear stress measured at shear rate 200 s^{-1}, the minimal values and ranges of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1}, and any individual rheological feature or combination of these rheological features.

A variant of DSM32493 is herein defined as a *Lactobacillus plantarum* strain presenting at least one mutation, such as the addition, deletion, insertion and/or substitution of at least one nucleotide in its genome as compared to the DSM32493 strain. In a particular embodiment, the genome sequence of the variant has an identity of at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, at least 99.1%, at least 99.2%, at least 99.3%, at least 99.4%, at least 99.5%, at least 99.6%, at least 99.7%, at least 99.8%, at least 99.9%, at least 99.92%, at least 99.94%, at least 99.96%, at least 99.98%, or at least 99.99% to the genome sequence of the DSM32493 strain. Such a variant can be for example:

- a natural variant obtained spontaneously from the DSM32493 strain after incubation in a selection medium. A natural variant is thus obtained without any genetic manipulation but only by spontaneous mutation of the strain and selection of the strain in an appropriate medium; an example of protocol used to select particular mutants of the DSM32493 strain is disclosed in example 5; or

- a variant comprising at least one mutation in its genome, said mutation being induced by genetic engineering, for instance by directed mutagenesis or random mutagenesis. Random mutagenesis can be performed with UV radiations or mutagenic compounds such as nitrous acid, ethyl-methanesulfonate, NMethyl- N'-nitro-N-nitrosoguanidine, N-ethyl-N-nitrosourea, acridine orange, proflavine.

The invention is also directed to a *Lactobacillus plantarum* as defined herein (including variant of the DSM32493 strain), which – in addition to provide a fermented milk presenting one, two or three of the following rheological features, as assayed by test A, a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa as defined herein; b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa as defined herein; c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12 as defined herein - is further characterized by being a low post-acidification strain at fermentation temperature.

By "*low post acidification strain at fermentation temperature*", it is meant a *L. plantarum* strain of the invention which - when inoculated into milk - generates a fermented milk, the pH of which – after fermentation - does not fluctuate more than 0.3 unit when the fermented milk

is kept at the fermentation temperature, in particular as assayed by test B (see below). The acidification kinetics is determined by the continuous recording of the pH as a function of time, for example using a Cinac system (CINAC, an automated system for control of lactic starters; Corrieu G, Picque D, Perret B, Quemener P; Process Magazine; 1992: 1068; p.24-27).

The acidification kinetics of milk inoculated with the low post-acidification *L. plantarum* strain of the invention and brought at the fermentation temperature follows a two-phase profile, comprising an initial period of sigmoidal pH decrease down to a pH value between 4.0 and 5.0 (to generate the fermented milk), followed by a second period in which the pH value does not fluctuate more than 0.3 unit (when the fermented milk is kept at the fermentation temperature). To characterize the low post acidifying feature of the *L. plantarum* strain of the invention, the pH is measured once the fermentation step terminates (end of the initial period), the fermented milk is kept at the fermentation temperature, the pH is measured at least 48 hours later, and the evolution of the pH (difference) is measured. In a particular embodiment, the fermentation temperature is 37°C.

In an embodiment, the pH obtained at the end of the initial period is comprised between 4.0 and 5.0. In a particular embodiment, the pH obtained at the end of the initial period is comprised between 4.2 and 4.8. In a particular embodiment, the pH obtained at the end of the initial period is comprised between 4.3 and 4.7. In a particular embodiment, the pH obtained at the end of the initial period is comprised between 4.4 and 4.6.

In a particular embodiment, the low post-acidification strain of the invention is able to generate a fermented milk, the pH of which does not fluctuate more than 0.3 unit, in particular not more than 0.2 unit, in particular not more than 0.1 unit, when kept at the fermentation temperature for a period of 48 hours after the end of the initial period.

In a particular embodiment, the low post acidifying feature characterization is carried out, by assaying Test B described below (see also example 5):

Test B

UHT milk (3.2% protein, 3.5% fat) added with 6% sucrose and 0.1% of yeast extract powder (LP0021) from Oxoid™ is heat treated at 85°C for 15 min and cooled down to room temperature. A culture of each strain to be tested is inoculated into milk at 1.10^7 CFU/ mL and the inoculated milk placed at 37°C (t=0). The inoculated milk is fermented until a pH of 4.5 is reached ($t_{pH\ 4.5}$) and kept at 37°C for at least 48 hours, and the pH of the milk continuously monitored. The delta pH (ΔpH = pH at ($t_{pH4.5}$+48h) – pH 4.5) is used to represent the post-acidification at fermentation temperature.

In a particular embodiment of a low post-acidification strain of the invention is a *L. plantarum* strain having at least one mutation in its ATP-synthase operon (for example point mutation, deletion, insertion, …), such that the strain has a reduced H^+-ATPase activity. The wild-type sequence of the ATP-synthase operon is as set forth in SEQ ID NO:1. The person

skilled in the art knows how to determine whether this operon is mutated and how to measure the H^+-ATPase activity of a bacterium [see for example Jaichumjai et al. 2010; Food Microbiology 27 (2010) 741-748]. In a particular embodiment, a low post-acidification *L. plantarum* strain of the invention as defined herein has at least one mutation in the ATP synthase alpha subunit gene of the ATP-synthase operon (herein referred as "*the ATP synthase alpha subunit gene*"). In a particular embodiment, a low post-acidification *L. plantarum* strain of the invention as defined herein has at least one mutation in the ATP synthase alpha subunit gene of the ATP-synthase operon as defined in SEQ ID NO:2. In a particular embodiment, in combination with the previous embodiment on SEQ ID NO:2 or not, the ATP synthase alpha subunit gene of the low post-acidification *L. plantarum* strain of the invention as defined herein encodes a ATP synthase alpha subunit protein having an aspartic acid residue at position 169. In a particular embodiment, the ATP synthase alpha subunit protein of the low post-acidification *L. plantarum* strain of the invention as defined herein is as defined in SEQ ID NO:5. In a particular embodiment, in combination with the previous embodiment on SEQ ID NO:2 or not, the ATP synthase alpha subunit gene of the low post-acidification *L. plantarum* strain of the invention as defined herein bears the mutation G to A at its position 506 (changing the glycine residue at position 169 by an aspartic acid residue). In a particular embodiment, the ATP synthase alpha subunit gene of the low post-acidification *L. plantarum* strain of the invention as defined herein is as defined in SEQ ID NO:4 (wherein the codon GGT at positions 505-507 is changed to GAT).

In an embodiment, the invention is directed to a low post-acidification variant as defined herein, which is a variant of the *Lactobacillus plantarum* strain DSM32493 deposited at the DSMZ on April 26th, 2017. In a particular embodiment, the invention is directed to a low post-acidification variant of the *Lactobacillus plantarum* strain DSM32493, wherein said variant bears a mutation in the ATP synthase alpha subunit gene (as compared to the DSM32493 strain), in particular bears the mutation G to A at position 506.

The definitions and specific embodiments detailed for the rheological features under the *L. plantarum* strain characterization (including the variant of DSM32493 strain) apply similarly in the context of the low post-acidification variant, in particular for but not limited to, the minimal values and ranges of the shear stress measured at shear rate 11.6 s-1, the minimal values and ranges of the shear stress measured at shear rate 200 s-1, the minimal values and ranges of the shear stress measured at 146 s-1 minus the shear stress measured at 41.1 s-1, and any individual rheological feature or combination of these rheological features.

The invention is also directed to a bacterial composition comprising or consisting of a *L. plantarum* strain of the invention (in particular a low post-acidification *L. plantarum* strain of the invention). In a particular embodiment, the bacterial composition is a pure culture, i.e.,

comprises or consists of a single bacterium strain. In another embodiment, the bacterial composition is a mixed culture, *i.e.,* comprises or consists of the *L. plantarum* strain of the invention and at least one other bacterium strain. Thus, in an embodiment, a bacterial composition of the invention comprises or consists of a *L. plantarum* strain of the invention and at least one lactic acid bacterium, in particular at least another *Lactobacillus plantarum* strain and/or a *Lactobacillus delbrueckii* subsp *bulgaricus* strain. By "*at least*" (in reference to bacterium strain, lactic acid bacterium or another *Lactobacillus plantarum* strain), it is meant 1 or more, and in particular 1, 2, 3, 4 or 5 strains. Thus, in an embodiment, the composition of the invention comprises or consists of - in addition to the *L. plantarum* strain of the invention, 1, 2, 3, 4 or 5 strains, in particular 1, 2, 3, 4 or 5 lactic acid bacteria strains. In a particular embodiment, the bacterial composition of the invention does not contain *Streptococcus thermophilus* strain(s).

In a particular embodiment, the bacterial composition as defined herein, either as a pure or mixed culture as defined above is under frozen, dried, freeze-dried, liquid or solid format, in the form of pellets or frozen pellets, or in a powder or dried powder. In a particular embodiment, the bacterial composition of the invention is in a frozen format or in the form of pellets or frozen pellets, in particular contained into one or more box or sachet. In another embodiment, the bacterial composition as defined herein is under a powder form, such as a dried or freeze-dried powder, in particular contained into one or more box or sachet.

In a particular embodiment, the bacterial composition of the invention, either as a pure culture or mixed culture as defined above, and whatever the format (frozen, dried, freeze-dried, liquid or solid format, in the form of pellets or frozen pellets, or in a powder or dried powder) comprises the *L. plantarum* strain of the invention in a concentration comprised in the range of 10^5 to 10^{12} cfu (colony forming units) per gram of the bacterial composition. In a particular embodiment, the concentration of the *L. plantarum* within the bacterial composition of the invention is in the range of 10^7 to 10^{12} cfu per gram of the bacterial composition, and in particular at least 10^7, at least 10^8, at least 10^9, at least 10^{10} or at least 10^{11} CFU/g of the bacterial composition. In a particular embodiment, when in the form of frozen or dried concentrate, the concentration of the *Lactobacillus plantarum* strain – as pure culture or as a mixed culture - within the bacterial composition is in the range of 10^8 to 10^{12} cfu/g of frozen concentrate or dried concentrate, and more preferably at least 10^8, at least 10^9, at least 10^{10}, at least 10^{11} or at least 10^{12} cfu/g of frozen concentrate or dried concentrate.

The invention is also directed to a composition or a kit-of-part comprising or consisting of a *L. plantarum* strain of the invention (in particular a low post-acidification *L. plantarum* strain of the invention) or a bacterial composition as defined herein (such as pure or mixed culture) and a booster. In a particular embodiment, said booster is a yeast extract or an amino-acid

containing composition. In a particular embodiment, the booster is a yeast extract, such as a yeast extract powder. Any yeast extract (or powder) can be used, for example but not limited to the yeast extract powder LP0021 from Oxoid™.

The expression "*A composition comprising or consisting of a L. plantarum strain of the invention or a bacterial composition of the invention and a booster*" means that the *L. plantarum* or the bacterial composition and the booster are physically mixed together. The booster amount within the composition is such that the booster is inoculated into milk in the range of 0.001% to 2%. In an embodiment, the composition is under frozen format or in the form of frozen pellets. In another embodiment, the composition is under dried or freeze-dried format or in the form of a powder or dried powder. In contrast, the expression "*A kit-of-part comprising or consisting of a L. plantarum strain of the invention or a bacterial composition of the invention and a booster*" means that the *L. plantarum* or the bacterial composition and the booster are physically separated but intended to be used together. Thus, the *L. plantarum* or the bacterial composition and the booster are in different boxes or sachets. In an embodiment, the *L. plantarum* or the bacterial composition and the booster are both under frozen format or in the form of frozen pellets. In another embodiment, the *L. plantarum* or the bacterial composition and the booster are both under dried or freeze-dried format or in the form of a powder or dried powder.

The invention also concerns a method for manufacturing a fermented product, comprising a) inoculating a substrate with a *L. plantarum* strain of the invention (in particular a low post-acidification *L. plantarum* strain of the invention) and b) fermenting said inoculated substrate, to obtain a fermented product. In a particular embodiment, the *L. plantarum* strain of the invention is inoculated as a bacterial composition as defined herein, such as a pure culture or a mixed culture. In an embodiment, the *L. plantarum* strain of the invention is inoculated as a composition as defined herein, i.e., step a) comprises or consists in inoculating the bacterial composition as defined herein. In another embodiment, the *L. plantarum* strain of the invention is inoculated as a kit-of-part as defined herein i.e., step a) comprises or consists in a1) inoculating the *L. plantarum* strain (or the bacterial composition) and a2) inoculating the booster, wherein the *L. plantarum* strain (or the bacterial composition) and the booster are inoculated simultaneously, or wherein the *L. plantarum* strain (or the bacterial composition) is inoculated before the booster or wherein the booster is inoculated before the *L. plantarum* strain (or the bacterial composition). In a particular embodiment, the method of the invention does not comprise inoculation of *Streptococcus thermophilus* strain(s). In a particular and preferred embodiment, the substrate is inoculated in step a) with a pure culture of the *L. plantarum* strain of the invention.

When booster is used, the booster is inoculated into milk in the range of 0.001% to 2%. In a particular embodiment, the booster is inoculated into milk in the range of 0.01 to 1%. In a particular embodiment, the booster is inoculated into milk in the range of 0.05 to 0.5%.

The fermentation time and temperature are parameters which are dependent upon the wanted final fermented product. In an embodiment, the fermentation temperature is comprised between 30 and 45°C, in particular 32 and 42°C, in particular between 35 and 39°C. The time of fermentation is determined according to the final pH desired at the end of the fermentation, and is comprised between 6 and 75 hours, in particular between 12 and 20 hours. Thus, in an embodiment, the fermentation is carried out until a pH comprised between 4.0 and 5.0, in particular between 4.2 and 4.8, in particular between 4.3 and 4.7, in particular between 4.4 and 4.6, is obtained.

Any substrate can be used in the method of the invention. Thus, in a particular embodiment, the substrate is selected in the group consisting of milk, milk of vegetal origin (such as soy milk) or cereal flour.

In a particular embodiment, the substrate used in the method of the invention is milk substrate. Thus, in an embodiment, the invention is directed to a method for manufacturing a fermented milk product comprising a) inoculating a milk substrate with a *L. plantarum* strain of the invention (in particular a low post-acidification *L. plantarum* strain of the invention) or a bacterial composition as defined herein or a composition as defined herein or a kit-of-part as defined herein; and b) fermenting said inoculated milk substrate, to obtain a fermented milk product. By "*milk substrate*", it is meant milk of animal origin. In a particular embodiment, the milk substrate originates from cow, goat, sheep, buffalo, zebra, horse, donkey, or camel, and the like. The milk may be in the native state, a reconstituted milk or a skimmed milk.

In another embodiment, the substrate is of plant origin, and can be obtained from extracts of plant material. In a particular embodiment, the substrate is soy, such as soy milk. In another embodiment, the substrate is cereal flour.

The invention is also directed to a fermented product, which is obtained using a *L. plantarum* strain of the invention (in particular a low post-acidification *L. plantarum* strain of the invention) or a bacterial composition of the invention or a composition of the invention or a kit-of-part of the invention, in particular obtained or obtainable by the method of the invention. Thus, the invention is directed to a fermented product comprising the *L. plantarum* strain of the invention (in particular a low post-acidification *L. plantarum* strain of the invention). In a particular embodiment, the *L. plantarum* of the invention is the only bacterium found in the fermented product of the invention. In a particular embodiment, the *L. plantarum* of the invention is the only acidifying bacterium found in the fermented product of the invention.

In a particular embodiment, the fermented product of the invention is a fermented dairy product, in particular a fermented dairy food product or a fermented dairy feed product. In a particular embodiment, the fermented dairy food product of the invention is fresh fermented milk.

In a particular embodiment, the fermented product of the invention is a fermented soy product. In a particular embodiment, the fermented product of the invention is a fermented cereal product.

In a particular embodiment, the fermented product of the invention - in particular the fermented dairy food product as defined herein - contains the DSM32493 strain deposited at the DSMZ on April 26th, 2017 or any variant thereof as defined herein. In a particular embodiment, the fermented product of the invention - in particular the fermented dairy food product as defined herein - contains any *L. plantarum* variant of the DSM32493 strain able to generate a fermented milk presenting, as assayed by test A, a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa as defined herein; b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa as defined herein; and c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12 as defined herein. In a particular embodiment, the fermented product of the invention - in particular the fermented dairy food product as defined herein - contains any *L. plantarum* variant of the DSM32493 strain keeping 1, 2 or 3, of the rheological features of the DSM32493 strain. In a particular embodiment, the fermented product of the invention - in particular the fermented dairy food product as defined herein - contains any *L. plantarum* variant of the DSM32493 strain keeping the rheological features of the DSM32493 strain. In a particular embodiment, the fermented product of the invention - in particular the fermented dairy food product as defined herein - contains a low post-acidification variant as defined herein of the *Lactobacillus plantarum* strain DSM32493. In a particular embodiment, the fermented product of the invention - in particular the fermented dairy food product as defined herein - contains a low post-acidification variant of the *Lactobacillus plantarum* strain DSM32493 deposited at the DSMZ on April 26th, 2017, wherein said variant bears a mutation in the ATP synthase alpha subunit gene (as compared to the DSM32493 strain), in particular bears the mutation G to A at its position 506, in particular wherein its ATP synthase alpha subunit gene is as defined in SEQ ID NO:4.

The definitions and specific embodiments detailed for the rheological features under the *L. plantarum* strain characterization apply similarly in the context of the fermented dairy product of the invention, in particular for but not limited to, the minimal values and ranges of the shear stress measured at shear rate 11.6 s^{-1}, the minimal values and ranges of the shear stress measured at shear rate 200 s^{-1}, the minimal values and ranges of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1}, and any individual rheological feature

or combination of these rheological features. Similarly, the definitions and specific embodiments detailed for the *L. plantarum* strain characterization apply similarly in the context of the fermented dairy product of the invention, in particular for but not limited to, the DSM32493 variant, the low post-acidification variant and the mutation of the ATP synthase alpha subunit gene, in particular the mutation G to A at its position 506, in particular the ATP synthase alpha subunit gene as defined in SEQ ID NO:4.

The invention is also directed to the use of the *L. plantarum* strain of the invention (in particular a low post-acidification *L. plantarum* strain of the invention) or the bacterial composition of the invention or the composition or the kit-of part of the invention, in the manufacture of a fermented dairy product.

In a particular embodiment, the *L. plantarum* strain is the DSM32493 strain deposited at the DSMZ on April 26th, 2017 or any variant thereof as defined herein, in particular a low post-acidification variant of the *Lactobacillus plantarum* strain DSM32493 as defined herein, in particular a low post-acidification variant of the *Lactobacillus plantarum* strain DSM32493 bearing a mutation in the ATP synthase alpha subunit gene. The definitions and specific embodiments detailed for the *L. plantarum* strain characterization apply similarly in the context of the use of the invention, in particular for but not limited to, the DSM32493 variant, the low post-acidification *L. plantarum* and the mutation of the ATP synthase alpha subunit gene, in particular the mutation G to A at its position 506, in particular the ATP synthase alpha subunit gene as defined in SEQ ID NO:4.

SEQUENCES

SEQ ID NO:1: *L. plantarum* ATP-synthase operon

GTGGGTGATCCAGTTCCTACAGTCAAATTCCTTGGACTGACGTTTAATATCGCGAATGAC
ATCTCAGTAATTGTGACTTGTCTGATTGTTTTCTTGTTTGTTTTTTTACTTTCGCGACAT
TTAACAATGAAGCCCAAGGGTGGACAAAATGTGCTGGAGTGGCTCATCGAGTTCACGAAT
GGCATTGTCAAAGGGTCGATCAAGGGTAACGAAGCGTCTAACTTCGGTTTGTACGCATTT
ACATTGTTTCTCTTTATCTTCATCGCTAACCAACTTGGATTGTTCATTCACGTTCAGGTC
GGGCAGTATACGTATCTGAAGAGTCCAACCGCCGATCCGATTGTGACTTTGACGTTATCG
TTTATGACCGTTGCACTTGCACATGCTGCGGGTGTTCGTAAGAAAGGTATGGGTGGTTAT
TTGAAAGAATACACACAACCTTTTGCTGTTTTCTCGGTTGTTAACGTCTTTGAACAATTT
ACCGATTTCCTAACTTTAGGTCTTCGGCTGTTCGGGAACATCTTTGCTGGTGAAATGTTA
CTAACGAAGGTTGCTGATTTGGCAAAGAGCAACGGTTGGTTGAGCTATGTTTACTCATTT
CCAATTGAACTCTTATGGCAAGGTTTCTCAGTGTTTATCGGGAGCATTCAAGCGTTCGTG
TTCGTAACCTTGACTTCAGTTTATATTTCTCAGAAGGTTAACGACGAGGAATAATTTCTA
GTTTTTTAATTTTAAGGAGGATACACAGATTATGGGAGCAATTGCTGCAGGTATTGCTAT

GTGTGGTGCCGCTATAGGTGCTGGTATTGGTAACGGTTTGGTTATTTCTAAGATGCTTGA
AGGGATGGCCCGTCAACCAGAATTATCTGGTCAATTACGGACTAACATGTTCATCGGTGT
TGGGTTGGTCGAATCAATGCCTATAATTTCCTTCGTTGTTGCTTTGATGGTTATGAACAA
GTAATCATTGGTCAACGAGTTCATTTTAATGAAAATGAAAGAAGGAGGTGTCATTAGATG
CTCTCGCATTTAATTATCGGTGCATCCGGTCTCTACCTTGGTGATATGTTGTTTATCGGG
ATTAGCTTTATTGTTTTGATGGCATTGATCTCTGTTGTTGCTTGGAAGCCCATCACAAAA
ATGATGGCTGATCGAGCCGACAAGATTGCGAACGACATTGATTCAGCACAAAAGTCTCGG
CAAGAAGCGAGTGACTTAGCTGATCAACGGCGTGATGCGCTATCACACTCTCGCGCTGAA
GCGAGTGAAATTGTCGCTGACGCGAAAAAGAGTGGCGAAAAGCAACGGTCAAGTATCATT
GCCGATGCGCAAAACGAAGCAACGCAGTATAAACAAAATGCGCGTAAGGATATTGAACAG
GAGCGTCAAGATGCCTTGAAGAACGTCCAATCAGACGTCGCTGACATTTCGATTGCGATT
GCTACGAAGATTATTAAGAAGCAATTGGATCCGGAAGGCCAACAGGCATTAATTAATTCG
TATATTGAAGGGTTGGGAAAGCATGAGTCTTGATAATCTTACAATTGCAAGTCGTTATTC
AAAGGCACTCTTTGAACTTGCAGTTGAAAAAGATCAGACCGAAGCATTCCTGGCCGAGTT
AAAGCAATTACGGCAAGTCTTTGTCGACAACCCGCAATTGGCAGAGGTCCTCTCAGGATC
ATTGCTTCCGGTTGATCAAAAACAGACAACGTTGTCAACTTTGACTGACCACGCTTCAGA
ATACATTAAAAACTTTATTCAAATGTTGTATGATTACGGCCGCATGTCGAACTTAGTTGG
CATTGTTGACGCGTTTGAAGCACGTTTCGATGAGAGTCGCAAAATAGTGCATGCCGAAGT
AACGTCTGCGGTCAAGTTGTCAGATGAGCAAGCTGATGCAATCGCAAAGGCATTCGCCAA
ACGTGTTGGGGCCAATCAGGTTGTTTTGTCACGTAAAGTCGATGAAGCAATCATTGGCGG
TGTAATTGTGAAGTCAAATAATCAAACGTTTGATGGTAGCGTTGCGTTACAACTAACGAA
TTTAAGACGAGCACTCATCAACAATTAGTTTACGAAGAGGTGAAACTTTTATGAGCATTA
AATCTGAAGAAATCAGTGCTCTAATCAAACAACAATTAGAAAGTTATCAAACTGAGCTCT
CAGTTGCTGAAACCGGTACTGTCACCTACGTTGGTGATGGGATCGCCCGTGCTCACGGAC
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AAAACCTCGAAAGCAACGATGTTGGTATCGTTGTTTTAGGGGATTTTGATGGTATTCGTG
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TTGGCCGGGTCGTTAACCCATTAGGTCAACCAGTTGACGGTTCAGGTGAGATTAAGACCA
CGAATACGCGGCCAATCGAACATAAAGCTCCTGGTATTATGCAACGGCAATCAGTTAGCG
AACCACTTCAAACTGGGATCAAGGCCATTGATGCCTTAGTTCCAATTGGTCGGGGCCAAC
GTGAATTGATTATCGGTGACCGTAAGACTGGGAAGACGTCCGTTGCCATTGATGCCATTT
TGAACCAAAAGGACCAAGACATGATTTGTGTCTACGTTGCAATCGGTCAAAAGGACTCAA
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TAACTGCCGGACCTGCTGAACCAGCGCCATTACTGTACTTAGCTCCTTATGCTGGGGCAG
CGATGGGTGAAGAATTTATGATGAACGGCAAGCACGTTTTGATCGTCTATGATGACCTTT
CAAAGCAAGCAACGGCTTACCGTGAACTTTCCTTGATCCTCCGTCGTCCTCCAGGTCGTG
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TGAGCGATGAATTGGGTGGCGGTTCAATGACGGCCTTACCAATTATCGAAACGCAAGCTG
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TGGATAGTGATTCATTCTATTCAGGTGTGCGGCCAGCGATTGATGCCGGGGCCTCTGTTT
CCCGGGTTGGTGGGGATGCGCAAATTAAAGCGATGAAGTCCGTTGCCGGGACCTTGCGTC

TTGACTTGGCTTCTTATCGTGAATTGGAATCCTTCTCACAATTCGGTTCTGACTTGGATG
CTGCAACCCAAGCGAAATTAAATCGTGGGCAACGGATCGTTGAAGTCTTAAAACAACCTG
TTCATTCACCATTGAAGGTCGAAGAACAAGTAATGATTTTATATGCTTTGACCAACGGTT
ATTTGGATAAAGTGGCAGTTGATGATATTGCCCGTTACCAAAGTGAATTGTTTGAATTTA
TTCATGCTAGTCATCAGGACCTCTTTGATACGATTTTGGCAACCAAGAAGTTACCAGAAG
CTGATAAGATGAATGGGGCCTTAGATGCGTTTGCAGAACAATTCCAGCCAACCGCTGCCG
CTGCGAAGTAGTTATGGCTGAAAAGGATGGTGAGTAGTGCATGGCAGAATCATTAATGGA
TGTCAAGCGCCGAATTGACTCAACAAAGAAGACTCATCAAATTACGTCGGCAATGCAAAT
GGTCTCAACTTCAAAATTGAACCAGATTCAAAAGCATACCAGCACGTATCAGGTGTACGC
TTCTAAAGTTGAAAGCATCGTTTCACATCTTGCCAAAGCTCATCTGATGTCAGCAAGTGC
CGGTGTTGCTAACAGTAATTCGAACACGATTTCAGTTAGTGAATTGCTCGCGCAACGCCC
CGTTAAAAAGACTGGTTTATTGGTGATCACTTCGGACCGTGGCCTCGTTGGTAGTTACAA
CAGTAACGTGTTGAAACAGACTAACGATTTCATGCGGACGCACAAGGTTGATGCCGATAA
CGCAGTCGTTTTGGCGGTTGGTGGCACTGGTGCGGATTTCTATAAAAAGAACGGGTTAAA
CGTGGCTTATGAGTACCGCGGCGTCTCTGATGTCCCAACTTTTAAAGAGGTTCGTGAAAT
CGTTAAGACAGTCACATCAATGTACCACAACGAAGTCTTTGATGAACTTTACGTCTTCTA
CAACCACTTTATTAATCGGCTCTCTTCTGGTTTTCGGGCCGTTAAGATGTTACCGATCTC
CGAAGAGACCTTTGAACAAAGTGAGTCAGATAATCGTAAAGCCAAGGATAGCCGGGTAGA
TGTCGGTCCCGAGTATGAAATGGAACCGTCAGAAGAAGCCATTTTGTCGGCCGTGTTGCC
ACAATATGCTGAAAGCTTGGTTTATGGTGCAATCTTGGATGCCAAGACTGCTGAACATGC
TTCGTCGTCAACCGCGATGAAGGCTGCATCAGATAACGCTGGCGATTTAATCGATAAATT
AAATCTGAAATATAACCGTGCGCGTCAAGCTGCTATTACCACTGAAATCACTGAAATCAC
TGGTGGTTTGGTTGCGCAAGAATAACGAAGTGGGAGGAATTAACGACTAATGAGTACAGG
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ACCCGATATTAATAACGCCTTGATCATTCAGAAGGACAACGATGACACTTTAACGGTGGA
AGTATCGTTGGAATTAGGTGATGGGGTTGTTCGGACCGTCGCGATGGATGGTACGGATGG
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AGAGACGTTAGGCCGGGTCTTCAACGTTTTAGGGGAAACCATTGATGGTGGTCCAGAATT
CGGTCCAGACGCAGAACGTAACCCGATTCATCGGGATGCGCCTAAATATGATGAATTAAC
GACCAGTACTGAAGTATTGGAAACTGGAATTAAAGTTATTGACCTCTTAGCACCTTATGT
TCGTGGTGGTAAGATTGGGTTGTTCGGTGGTGCCGGTGTTGGTAAAACTGTTTTAATCCA
GGAATTAATTCATAACATTGCCCAAGAACATAACGGGATTTCCGTGTTTACCGGTGTTGG
TGAACGGACGCGTGAAGGGAATGACCTTTACTTCGAAATGAAGGCTTCCGGCGTTTTGAA
GAATACCGCCATGGTTTATGGTCAAATGAACGAACCACCTGGTGCCCGGATGCGGGTGGC
CTTGACCGGTTTGACGATTGCGGAATACTTCCGTGATGTTCAAGGTCAAGACGTGTTGTT
ATTCATCGACAATATCTTCCGGTTCACGCAAGCTGGTTCTGAAGTTTCCGCCTTACTTGG
TCGGATTCCTTCAGCCGTTGGTTACCAACCAACCTTAGCCACTGAAATGGGTCAATTACA
AGAACGGATCACTTCTACCAAGAAGGGGTCAGTTACTTCGATTCAAGCCGTTTATGTACC
TGCCGATGATTATACCGACCCGGCACCTGCAACGACTTTCGCCCATTTGGATGCGACGAC
CAACTTGGAACGTTCTTTGACGGAACAAGGGATCTACCCAGCCGTTGACCCATTAGCTTC
TTCTTCAATCGCTCTGGACCCATCAATCGTGGGCGAAGAACATTATCAAGTTGCAACGGA

AGTTCAACGGGTCTTGCAACGTTATCGTGAATTGCAAGATATTATCTCGATTTTAGGGAT
GGATGAATTATCTGACGAAGAAAAGACAACTGTTGCGCGTGCACGGCGGATTCAATTCTT
CTTGTCACAAAACTTCTTCGTTGCCGAAAACTTTACGGGCCAACCTGGTTCGTATGTGCC
AATCAACGATACCATCAAGGGCTTCAAAGAAATTCTTGAAGGTAAATATGATGACCTACC
AGAAGACGCATTCCGTCAAGTTGGTAAGATCGACGACGTGGTCGAAAAAGCGAAATCGAT
GGTAACTGATTAGGAGGGGTTTACATGGCTGACAATGCAAAATCATTAACCGTTAGCATC
GTAACTCCAGACGGTCAGGTCTATGAGAATAAGACGCCAATGTTGATCGTGCGAACGATT
GACGGCGAACTCGGAATTTTGCCGAACCATATTCCTGTGATTGCATCGCTTGCAATCGAT
GAGGTTCGGATCAAGCAACTTGAAAGTGATCAGGAAGATGACGAAATTGCCGTTAATGGT
GGTTTTGTTGAGTTCAGTAATAATACGGCAACGATTGTTGCCGATAGTGCTGAACGTCAG
AATGACATTGACGTTGCTCGAGCTGAAAATGCACGGAAACGCGCTGAAACACGGATTCAA
AATGCCCAACAAAAGCACGATGATGCTGAGTTGGCGCGGGCCCAAGTCGCTTTGCGGCGT
GCCATGAACCGTTTGAATGTTGCTCGGCATTAA

SEQ ID NO:2: ATP synthase alpha subunit gene of the DSM32493 strain

ATGAGCATTAAATCTGAAGAAATCAGTGCTCTAATCAAACAACAATTAGAAAGTTATCAA
ACTGAGCTCTCAGTTGCTGAAACCGGTACTGTCACCTACGTTGGTGATGGGATCGCCCGT
GCTCACGGACTCGACAACGCCTTACAAGGTGAATTACTCGAATTCAGTAACGGAGTTTAC
GGGATGGTACAAAACCTCGAAAGCAACGATGTTGGTATCGTTGTTTTAGGGGATTTTGAT
GGTATTCGTGAAGGCGATACTGTTAAGCGGACTGGCCGCATCATGGAAGTTCCAGTCGGT
GACGCCATGATTGGCCGGGTCGTTAACCCATTAGGTCAACCAGTTGACGGTTCAGGTGAG
ATTAAGACCACGAATACGCGGCCAATCGAACATAAAGCTCCTGGTATTATGCAACGGCAA
TCAGTTAGCGAACCACTTCAAACTGGGATCAAGGCCATTGATGCCTTAGTTCCAATTGGT
CGGGGCCAACGTGAATTGATTATCGGTGACCGTAAGACTGGGAAGACGTCCGTTGCCATT
GATGCCATTTTGAACCAAAAGGACCAAGACATGATTTGTGTCTACGTTGCAATCGGTCAA
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ACAATCGTTGTAACTGCCGGACCTGCTGAACCAGCGCCATTACTGTACTTAGCTCCTTAT
GCTGGGGCAGCGATGGGTGAAGAATTTATGATGAACGGCAAGCACGTTTTGATCGTCTAT
GATGACCTTTCAAAGCAAGCAACGGCTTACCGTGAACTTTCCTTGATCCTCCGTCGTCCT
CCAGGTCGTGAAGCTTATCCTGGGGATGTCTTCTACTTGCACTCACGGTTACTCGAACGG
GCTGCCAAGTTGAGCGATGAATTGGGTGGCGGTTCAATGACGGCCTTACCAATTATCGAA
ACGCAAGCTGGGGATATTTCGGCTTATATTCCAACTAACGTTATTTCAATCACCGATGGG
CAAATCTTCTTGGATAGTGATTCATTCTATTCAGGTGTGCGGCCAGCGATTGATGCCGGG
GCCTCTGTTTCCCGGGTTGGTGGGGATGCGCAAATTAAAGCGATGAAGTCCGTTGCCGGG
ACCTTGCGTCTTGACTTGGCTTCTTATCGTGAATTGGAATCCTTCTCACAATTCGGTTCT
GACTTGGATGCTGCAACCCAAGCGAAATTAAATCGTGGGCAACGGATCGTTGAAGTCTTA
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ACCAACGGTTATTTGGATAAAGTGGCAGTTGATGATATTGCCCGTTACCAAAGTGAATTG
TTTGAATTTATTCATGCTAGTCATCAGGACCTCTTTGATACGATTTTGGCAACCAAGAAG
TTACCAGAAGCTGATAAGATGAATGGGGCCTTAGATGCGTTTGCAGAACAATTCCAGCCA
ACCGCTGCCGCTGCGAAGTAG

SEQ ID NO:3: ATP synthase alpha subunit protein of the DSM32493 strain

MSIKSEEISALIKQQLESYQTELSVAETGTVTYVGDGIARAHGLDNALQGELLEFSNGVY
GMVQNLESNDVGIVVLGDFDGIREGDTVKRTGRIMEVPVGDAMIGRVVNPLGQPVDGSGE
IKTTNTRPIEHKAPGIMQRQSVSEPLQTGIKAIDALVPIGRGQRELIIGDRKTGKTSVAI
DAILNQKDQDMICVYVAIGQKDSTVRAQVETLKKLGAMDYTIVVTAGPAEPAPLLYLAPY
AGAAMGEEFMMNGKHVLIVYDDLSKQATAYRELSLILRRPPGREAYPGDVFYLHSRLLER
AAKLSDELGGGSMTALPIIETQAGDISAYIPTNVISITDGQIFLDSDSFYSGVRPAIDAG
ASVSRVGGDAQIKAMKSVAGTLRLDLASYRELESFSQFGSDLDAATQAKLNRGQRIVEVL
KQPVHSPLKVEEQVMILYALTNGYLDKVAVDDIARYQSELFEFIHASHQDLFDTILATKK
LPEADKMNGALDAFAEQFQPTAAAAK

SEQ ID NO:4: ATP synthase alpha subunit gene of a low post-acidification mutant of DSM32493 strain

ATGAGCATTAAATCTGAAGAAATCAGTGCTCTAATCAAACAACAATTAGAAAGTTATCAA
ACTGAGCTCTCAGTTGCTGAAACCGGTACTGTCACCTACGTTGGTGATGGGATCGCCCGT
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GGGATGGTACAAAACCTCGAAAGCAACGATGTTGGTATCGTTGTTTTAGGGGATTTTGAT
GGTATTCGTGAAGGCGATACTGTTAAGCGGACTGGCCGCATCATGGAAGTTCCAGTCGGT
GACGCCATGATTGGCCGGGTCGTTAACCCATTAGGTCAACCAGTTGACGGTTCAGGTGAG
ATTAAGACCACGAATACGCGGCCAATCGAACATAAAGCTCCTGGTATTATGCAACGGCAA
TCAGTTAGCGAACCACTTCAAACTGGGATCAAGGCCATTGATGCCTTAGTTCCAATTGGT
CGGGGCCAACGTGAATTGATTATCGATGACCGTAAGACTGGGAAGACGTCCGTTGCCATT
GATGCCATTTTGAACCAAAAGGACCAAGACATGATTTGTGTCTACGTTGCAATCGGTCAA
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GATGACCTTTCAAAGCAAGCAACGGCTTACCGTGAACTTTCCTTGATCCTCCGTCGTCCT
CCAGGTCGTGAAGCTTATCCTGGGGATGTCTTCTACTTGCACTCACGGTTACTCGAACGG
GCTGCCAAGTTGAGCGATGAATTGGGTGGCGGTTCAATGACGGCCTTACCAATTATCGAA
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CAAATCTTCTTGGATAGTGATTCATTCTATTCAGGTGTGCGGCCAGCGATTGATGCCGGG
GCCTCTGTTTCCCGGGTTGGTGGGGATGCGCAAATTAAAGCGATGAAGTCCGTTGCCGGG
ACCTTGCGTCTTGACTTGGCTTCTTATCGTGAATTGGAATCCTTCTCACAATTCGGTTCT
GACTTGGATGCTGCAACCCAAGCGAAATTAAATCGTGGGCAACGGATCGTTGAAGTCTTA
AAACAACCTGTTCATTCACCATTGAAGGTCGAAGAACAAGTAATGATTTTATATGCTTTG
ACCAACGGTTATTTGGATAAAGTGGCAGTTGATGATATTGCCCGTTACCAAAGTGAATTG
TTTGAATTTATTCATGCTAGTCATCAGGACCTCTTTGATACGATTTTGGCAACCAAGAAG
TTACCAGAAGCTGATAAGATGAATGGGGCCTTAGATGCGTTTGCAGAACAATTCCAGCCA
ACCGCTGCCGCTGCGAAGTAG

SEQ ID NO:5: ATP synthase alpha subunit protein of a low post-acidification mutant of DSM32493 strain

MSIKSEEISALIKQQLESYQTELSVAETGTVTYVGDGIARAHGLDNALQGELLEFSNGVY
GMVQNLESNDVGIVVLGDFDGIREGDTVKRTGRIMEVPVGDAMIGRVVNPLGQPVDGSGE
IKTTNTRPIEHKAPGIMQRQSVSEPLQTGIKAIDALVPIGRGQRELIIDDRKTGKTSVAI
DAILNQKDQDMICVYVAIGQKDSTVRAQVETLKKLGAMDYTIVVTAGPAEPAPLLYLAPY
AGAAMGEEFMMNGKHVLIVYDDLSKQATAYRELSLILRRPPGREAYPGDVFYLHSRLLER
AAKLSDELGGGSMTALPIIETQAGDISAYIPTNVISITDGQIFLDSDSFYSGVRPAIDAG
ASVSRVGGDAQIKAMKSVAGTLRLDLASYRELESFSQFGSDLDAATQAKLNRGQRIVEVL
KQPVHSPLKVEEQVMILYALTNGYLDKVAVDDIARYQSELFEFIHASHQDLFDTILATKK
LPEADKMNGALDAFAEQFQPTAAAAK

SEQ ID NO:6: forward primer

Primer-F: TCAACCAGTTGACGGTTCAG

SEQ ID NO:7: reverse primer

Primer-R: TTTGGTTCAAAATGGCATCA

DEPOSIT and EXPERT SOLUTION

The following deposit was made according to the Budapest treaty on the international recognition of the deposit of microorganisms for the purposes of patent procedure.

- *Lactobacillus plantarum* strain (DGCC12411) deposited under accession number DSM32493 on April 26th, 2017, at the DSMZ [Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH, Inhoffenstrasse 7B, D-38124 Braunschweig - Germany].

It is requested that the biological material shall be made available only by the issue of a sample to an expert nominated by the requester. In respect to those designations in which a European Patent is sought, a sample of the deposited microorganism will be made available until the publication of the mention of the grant of the European patent or until the date on which application has been refused or withdrawn or is deemed to be withdrawn, only by the issue of such a sample to an expert nominated by the person requesting the sample, and approved either i) by the Applicant and/or ii) by the European Patent Office, whichever applies (Rule 32 EPC).

Various preferred features and embodiments of the present invention will now be described by way of non-limiting examples.

EXAMPLES

Example 1: selection of a *Lactobacillus plantarum* with high ropiness on MRS

The following method was used for screening *Lactobacillus plantarum* strains of the DuPont collection. Thus, 214 *Lactobacillus plantarum* isolated from various sources were screened as follows:

- a single colony of each *L. plantarum* was inoculated into MRS agar plate, and incubated at 30°C for 2 days;
- the ropiness was manually scored based on the strength of filament formed when touch the colony by an inoculation loop, according to the following classification: no detectable ropiness (no filament formed), low ropiness, intermediate ropiness or high ropiness (see respectively Figure 1A to 1D).

From the 214 tested strains, the following classification was obtained:

- 1 strain was classified with a high ropiness
- 2 strains were classified with intermediate ropiness
- 17 strains were classified with low ropiness
- 194 strains were classified with no detectable ropiness

One *L. plantarum* strain representative of each class was then assayed for rheological and sensory analyses.

Example 2: rheological properties of milk fermented with DSM32493 and comparison with 3 other *L. plantarum* strains

4 *L. plantarum* strains selected from example 1 were used to ferment milk and the rheological properties of the obtained fermented milks were determined.

Strains: The following strains were used:

- the DSM32493 strain, showing a high ropiness in example 1;
- the LP12111 strain showing an intermediate ropiness in example 1;
- the LP12428 strain showing a low ropiness in example 1;
- the *L. plantarum* 115 strain (also known as DGCC4715, deposited as DSM22266 in patent EP2245943B1) showing no detectable ropiness in example 1.

The following **Test A** was carried out:

Milk: 2L UHT milk (3.2% protein, 3.5% fat) added with 6% sucrose and 0.1% of yeast extract powder (LP0021) from Oxoid™ was heat treated at 85°C for 15 min and cooled down to room temperature.

Fermentation: For each strain, a frozen culture was inoculated into milk at 1.10^7 CFU/ mL. The inoculated milk was then fermented at 30°C to reach pH 4.5 (typically about 22-24 h).

Rheological measurements: After fermentation, the samples were stirred by a rotator (RZR 2051 control, Heidolph) at 100 rpm for 1.5 min and stored at 4°C for overnight, before measurement by rheometer. The used tank and bladder are as disclosed in Fig. 8. The samples were assessed by a rheometer (Anton Paar MCR 302, CC27-SN27450, Austria) using a coaxial cylinder C-CC27-T200/SS and a bob-cup. The rheometer was set to a constant temperature of 10°C during the measurement. Settings were as follows:

- holding time (to rebuild to somewhat original structure): 10 minutes, without any physical stress applied to the sample.
- 25 measuring points over 500 s (one every 20 s)
- rotation step (to measure the shear stress at 11.6 1/s, the shear stress at 200 1/s and the difference of the shear stress at 146 1/s minus the shear stress at 41.1 1/s)

Two steps were designed:

Shear rate: d(gamma)/ dt = [0.1-200] 1/s log and [200-0.1] 1/s log

Each step contained 25 measuring points over 500 s (on every 20 s)

The flow curves of fermented milks obtained using either the DSM32493 strain, the LP12111 strain, the LP12428 strain or the Lp115 strain were obtained (Figure 2). As show in Figure 2, as compared to the LP12111, LP12428 and Lp115 strains which share close flow curves, the flow curve of the DSM32493 strain is atypical due to its significantly higher thickness and ropiness.

For each fermented milk, the shear stress at 11.6 1/s, the shear stress at 200 1/s and the difference between the shear stress at 146 1/s and the shear stress at 41.1 1/s were then calculated (Table 1). The shear stress at 11.6 1/s was correlated to sensory thickness, the shear stress at 200 1/s was correlated to mouth-thickness and the difference between the shear stress at 146 1/s and the shear stress at 41.1 1/s was correlated to ropiness.

Strain	Thickness (shear rate[1/s] 11.6)	Mouth thickness (shear rate[1/s] 200)	Ropiness (Δ146-41.1)
DSM32493	36.6	77	15.5
LP12111	19.7	38.1	8.7
LP12428	16.9	32.6	7.3
Lp115	16.8	33.5	8

Table 1: shear stress at 11.6 1/s, shear stress at 200 1/s and difference between the shear stress at 146 1/s and the shear stress at 41.1 1/s of fermented milks obtained with either the DSM324931 strain, the LP12111 strain, the LP12428 strain or the Lp115 strain.

As detailed in Table 1, the fermented milk obtained using DSM32493 shows high values of thickness, mouth thickness and ropiness, almost double those obtained using LP12111, LP12428 and Lp115 strains.

Thus, these results show that a fermented milk having high values of thickness, mouth thickness and ropiness (as defined herein) can be obtained using a *L. plantarum* of the invention (in particular the DSM32493 strain).

Interestingly and as an advantage of the invention, these results also show that a fermented milk having high values of thickness, mouth thickness and ropiness (as defined herein) can be obtained using a pure culture of *L. plantarum* of the invention (in particular a pure culture of the DSM32493 strain). This is in contrast with fermented milk obtained with compositions described in the literature which comprise several strains (of which at least a *Streptococcus thermophilus* strain).

Example 3: sensory analysis of milk fermented with DSM32493 and comparison with 3 other *L. plantarum* strains

An expert panel comprised of five culture/dairy application specialists evaluated the sensory properties of each fermented milk obtained using the DSM32493 strain, the LP12111 strain, the LP12428 strain or the Lp115 strain. Seven flavour and texture descriptors were evaluated and reported on a spider chart (Figure 3). Whereas the fermented milks obtained using LP12111, LP12428 and Lp115 strains show comparable charts, the fermented milk obtained using the DSM32493 strain shows significant differences in several of these descriptors. Thus, fermented milks obtained using one of LP12111, LP12428 and Lp115 strains show grainy texture (so a non-shining appearance), low ropiness and smoothness, and low to medium thickness in mouth and spoon. In contrast, fermented milk obtained using the DSM32493 strain shows very high thickness in mouth and spoon, a very long texture, high ropiness and smoothness and has a shining appearance. Appearance of fermented milks

obtained using either the DSM32493 strain, the LP12111 strain, the LP12428 strain or the Lp115 strain is shown in Figure 4.

Example 4: rheological properties of milk fermented with DSM32493 strain and comparison with a traditional starter

The rheological properties of a fermented milk obtained using DSM32493 was then compared with the ones of a milk fermented using a commercial yogurt starter culture with strong texturing property (Danisco; YO-MIX® 883) consisting of a combination of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp *bulgaricus* strains. Test A as described in example 2 was carried out, with inoculation of either the DSM32493 strain or the YO-MIX® 883 culture, and the flow curve of the fermented milks were obtained (Figure 5). For each fermented milk, the shear stress at 11.6 1/s, the shear stress at 200 1/s and the difference between the shear stress at 146 1/s and the shear stress at 41.1 1/s were then calculated (Table 2).

Culture	Thickness (shear rate[1/s] 11.6)	Mouth thickness (shear rate[1/s] 200)	Ropiness (Δ146-41.1)
DSM32493	36.6	77	15.5
YO-MIX® 883	29.5	63.4	15.5

Table 2: shear stress at 11.1 1/s, shear stress at 200 1/s and difference between the shear stress at 146 1/s and the shear stress at 41.1 1/s of fermented milks obtained with either the DSM324931 strain or the YO-MIX® 883 culture.

As show in Figure 5 and detailed in Table 2, the DSM32493 strain alone is sufficient to manufacture a fermented milk with a thickness and ropiness as good as the ones obtained using a traditional culture of *S. thermophilus* and *L. bulgaricus*. Moreover, this comparative example confirms the uniqueness of the DSM32493 strain to give a fermented milk with a high mouth-thickness.

This high mouth-thickness value provided by the DSM32493 strain is confirmed by the long texture clearly visible on the left photograph of Figure 6 as compared to the right photograph [obtained using YO-MIX® 883].

These results confirm that the DSM32493 is industrially interesting to obtain fermented milk presenting a high thickness, a high ropiness and a high mouth-thickness.

Example 5: selection of a low post-acidification *L. plantarum* variant of the DSM32493 strain

The DSM32493 strain was streaked on MRS agar plate and anaerobically incubated at 30°C for 2 days. A single colony was picked up and grew in 10mL MRS broth 30°C overnight. 200µL of fresh culture (O.D = 0.8) was inoculated into 10mL of 0.5x MRS containing 700µg/mL of neomycin, and then incubated at 30°C for 2 days. A serial dilution of the fresh culture was prepared and plated on 0.5x MRS agar plate with 700µg/mL of neomycin and incubated at 30°C for 2 days. Single colonies were picked up, and each inoculated into both of 0.5x MRS (pH 6.3) and 0.5x MRS (pH adjusted to pH 4.5) in 96-well plates and incubated at 30°C for 2 days. Colonies which grew in 0.5x MRS (pH 6.3) but not 0.5x MRS (pH 4.5) were selected. The acidification curve of each selected colony in MRS at 30°C was monitored using iCinac for 4 days. Colonies with significant higher end pH (and considered as low post acidification mutants at fermentation temperature) were selected and their DNA sequenced.

One mutant (Lp12733) selected by the protocol described above was shown to contain the mutation G to A in position 506 of the ATP synthase alpha subunit gene of the ATP-synthase operon, resulting in the substitution of the glycine residue at position 169 by an aspartic acid residue. Presence of this mutation can be checked by PCR using the primers as defined in SEQ ID NOs: 6 and 7, and then DNA sequencing using the primer as defined in SEQ ID NO:6.

The DSM32493 strain and its low post acidification mutant Lp12733 were used to ferment milk and the pH was recorded over time, using Test B described herein. Cinac curves were obtained and are shown in Figure 7.

Test B:

Milk: UHT milk (3.2% protein, 3.5% fat) added with 6% sucrose and 0.1% of yeast extract powder (LP0021) from Oxoid™ was heat treated at 85°C for 15 min and cooled down to room temperature.

Fermentation: For each strain, a freeze-dried culture was inoculated into milk at 1.10^7 CFU/ mL and the inoculated milk placed at 37°C (t=0). The inoculated milk was fermented until a pH of 4.5 ($t_{pH4.5}$) is reached and kept at 37°C for at least 48 hours, and the pH of the milk continuously monitored.

Post-acidification measurements: The delta pH (ΔpH = pH at ($t_{pH4.5}$+48h) – pH 4.5) was used to represent the post-acidification at fermentation temperature.

As shown in Figure 7, the pH of fermented milk obtained using the DSM32493 strain at $t_{pH4.5}$+48h hours is 4.1 whereas the pH of the fermented milk obtained using the Lp12733 strain (low post acidification mutant of the DSM32493 strain) is 4.5 (i.e. the pH of the fermented milk is steady at pH 4.5). Thus, the ΔpH of the DSM32493 strain is about 0.4, whereas the ΔpH of the Lp12733 strain is 0.

These results confirm that the Lp12733 strain (low post acidification mutant of the DSM32493) is industrially interesting to obtain fermented milk not only presenting a high thickness, a high ropiness and a high mouth-thickness, but also having a low post-acidification at fermentation temperature.

PCT

0-1	**Form PCT/RO/134 Indications Relating to Deposited Microorganism(s) or Other Biological Material (PCT Rule 13bis)**	
0-1-1	Prepared Using	CMS Online Filing Version CMS 1.15 MT/FOP 20020701/0.20.5.20
0-2	**International Application No.**	
0-3	**Applicant's or agent's file reference**	NB41149-WO-PCT2

1	**The indications made below relate to the deposited microorganism(s) or other biological material referred to in the description on:**	
1-1	**page**	22
1-2	**line**	22-24
1-3	**Identification of deposit**	
1-3-1	Name of depositary institution	DSMZ Leibniz-Institut DSMZ - Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH (DSMZ)
1-3-2	Address of depositary institution	Inhoffenstr. 7B 38124 Braunschweig, Germany
1-3-3	Date of deposit	26 April 2017 (26.04.2017)
1-3-4	Accession Number	DSMZ32493
1-4	**Additional Indications**	
1-5	**Designated States for Which Indications are Made**	All designations
1-6	**Separate Furnishing of Indications** These indications will be submitted to the International Bureau later	Form PCT/RO/134

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0-4	**This form was received with the international application:** (yes or no)	yes
0-4-1	Authorized officer	Kuiper-Cristina, Nathalie

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CLAIMS

1. A *Lactobacillus plantarum* strain which – when inoculated into milk - generates a fermented milk presenting one, two or three of the following rheological features, as assayed by test A:

a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa;

b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa;

c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12.

2. The *Lactobacillus plantarum* strain according to claim 1, which is the strain DSM32493 deposited at the DSMZ on April 26th, 2017, or a variant of the DSM32493 strain generating a fermented milk presenting one, two or three of the following rheological features, as assayed by test A: a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa; b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa; c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, or a variant of the DSM32493 strain keeping 1, 2 or 3, of the rheological features of the DSM32493 strain.

3. The *Lactobacillus plantarum* strain according to claim 1 or 2 which is further characterized by being a low post acidification strain at the temperature of fermentation.

4. The *Lactobacillus plantarum* strain according to claim 3, which is mutated within the ATP-synthase operon.

5. The *Lactobacillus plantarum* strain according to claim 4, which bears a mutation in the ATP synthase alpha subunit gene of the ATP-synthase operon, in particular which bears the mutation G to A at its position 506.

6. The *Lactobacillus plantarum* strain according to any one of claims 3 to 5, which is a variant of the strain DSM32493 deposited to DSMZ on April 26th, 2017.

7. A bacterial composition comprising or consisting of the strain according to any one of claims 1 to 6, in particular in combination with at least another lactic acid bacteria, in particular with a *Lactobacillus delbrueckii* subsp *bulgaricus* strain or with another *Lactobacillus plantarum* strain.

8. A composition or kit-of part comprising or consisting of the *L. plantarum* strain according to any one of claims 1 to 6 or the bacterial composition according to claim 7, and a booster, such as a yeast extract or an amino-acid containing composition.

9. A method for manufacturing a fermented product, comprising a) inoculating a substrate with the *L. plantarum* strain according to any one of claims 1 to 6 or the bacterial composition according to claim 7 or the composition or kit-of part according to claim 8, and b) fermenting said inoculated substrate, to obtain a fermented product.

10. The method for manufacturing a fermented product according to claim 9, wherein said *L. plantarum* strain is inoculated together with a booster, such as a yeast extract or an amino acid-containing composition.

11. The method according to claim 9 or 10, wherein said substrate is milk and said fermented product is a fermented dairy product.

12. A fermented product comprising the *L. plantarum* strain according to any one of claims 1 to 6 or obtained or obtainable by a method according to any one of claims 9 to 11.

13. The fermented product according to claim 12, which is a fermented dairy product, such as a fresh fermented milk.

14. The fermented product according to any one of claims 12 to 13 which contains the *Lactobacillus plantarum* strain DSM32493 deposited at the DSMZ on April 26th, 2017, or a variant of the DSM32493 strain generating a fermented milk presenting one, two or three of the following rheological features, as assayed by test A: a) a shear stress measured at shear rate 11.6 s^{-1} higher than 30 Pa; b) a shear stress measured at shear rate 200 s^{-1} higher than 60 Pa; c) a difference of the shear stress measured at 146 s^{-1} minus the shear stress measured at 41.1 s^{-1} higher than 12, or a variant of the DSM32493 strain keeping 1, 2 or 3, of the rheological features of the DSM32493 strain

15. Use of the *L. plantarum* strain according to any one of claims 1 to 6 or a bacterial composition according to claim 7 or a composition or kit-of part according to claim 8, in the manufacture of a fermented dairy product.

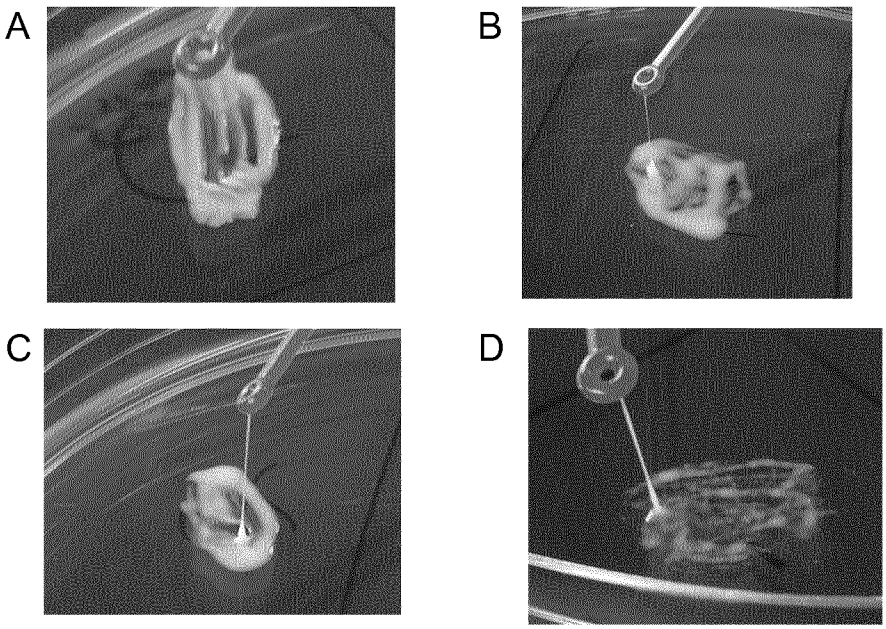
A
B
C
D

Fig. 1

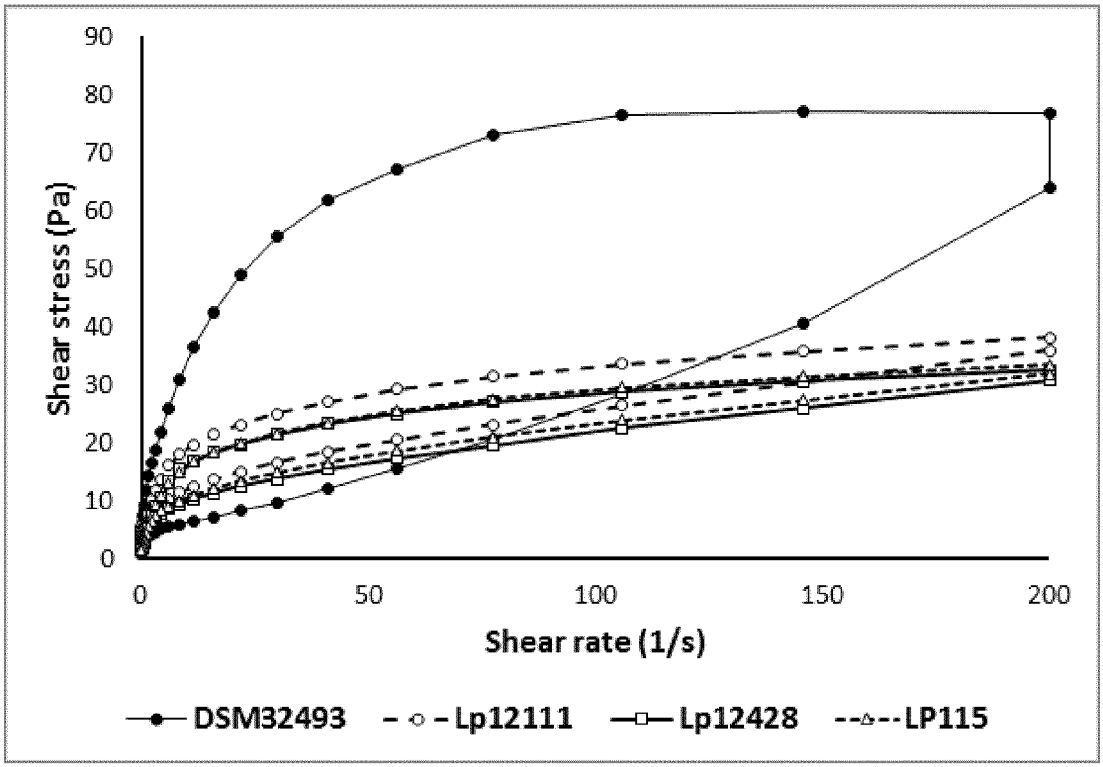
Shear stress (Pa)
0
10
20
30
40
50
60
70
80
90
0
50
100
150
200
Shear rate (1/s)
DSM32493
Lp12111
Lp12428
LP115

Fig. 2

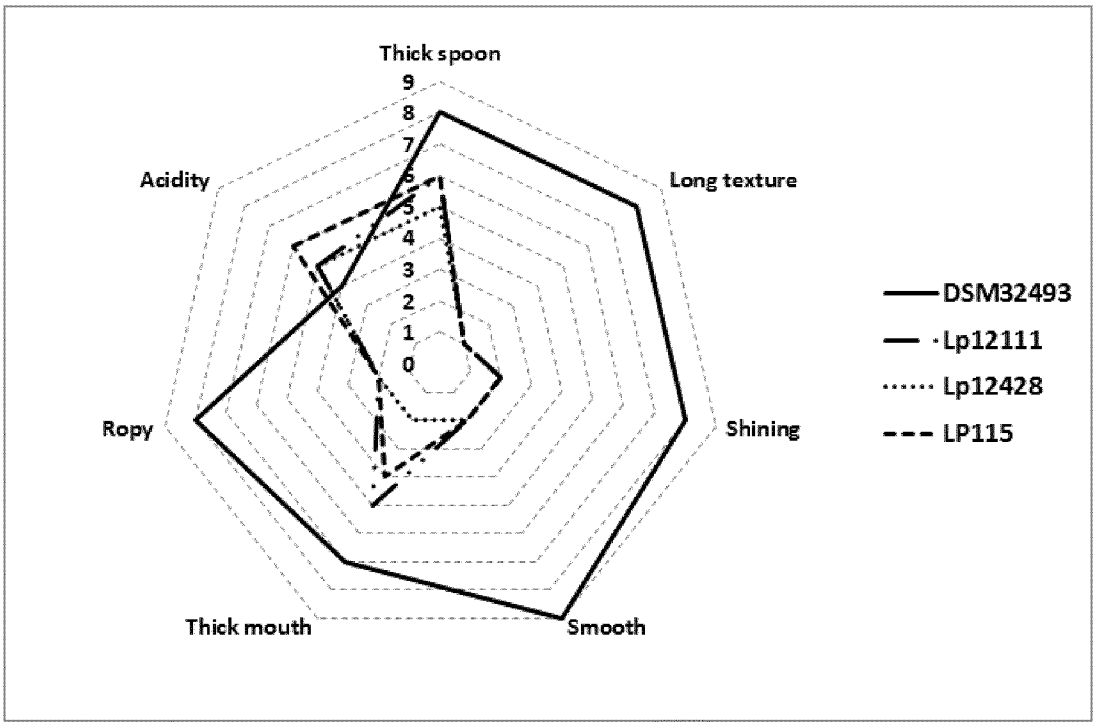
Thick spoon
9
8
7
6
5
4
3
2
1
0
Long texture
Shining
Smooth
Thick mouth
Ropy
Acidity
DSM32493
Lp12111
Lp12428
LP115

Fig. 3

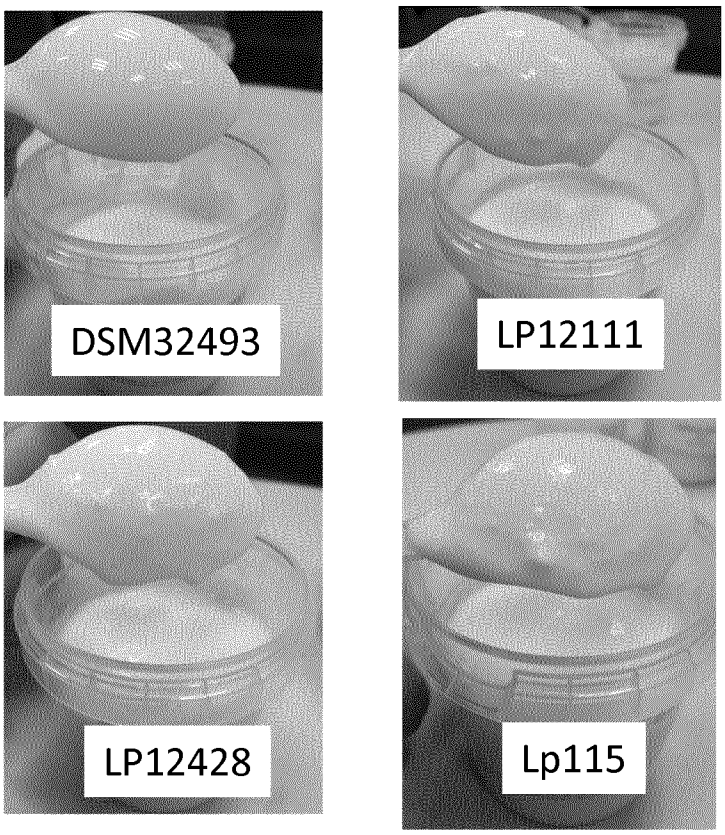
DSM32493
LP12111
LP12428
Lp115

Fig. 4

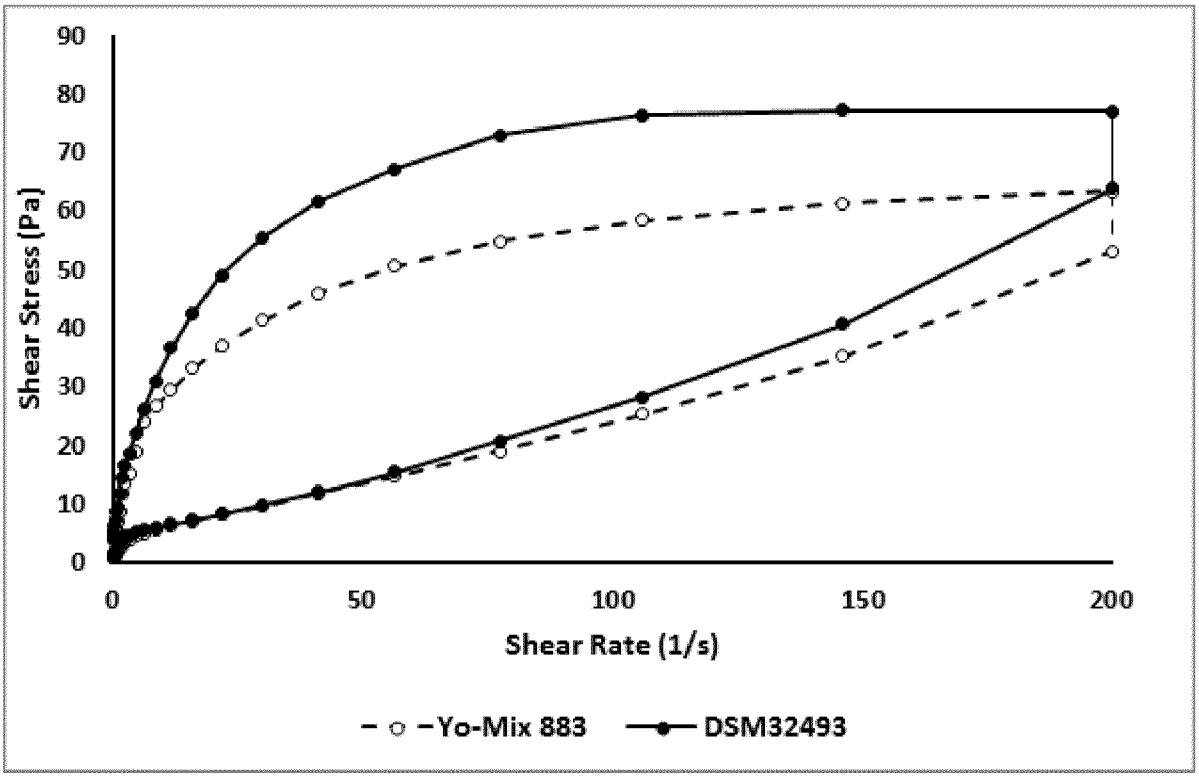
Shear Stress (Pa)
90
80
70
60
50
40
30
20
10
0
0
50
100
150
200
Shear Rate (1/s)
Yo-Mix 883
DSM32493

Fig.5

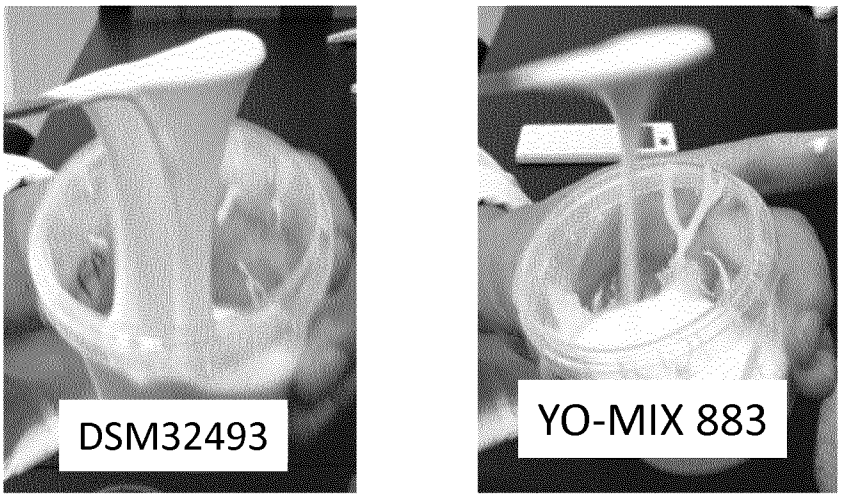
DSM32493
YO-MIX 883

Fig. 6

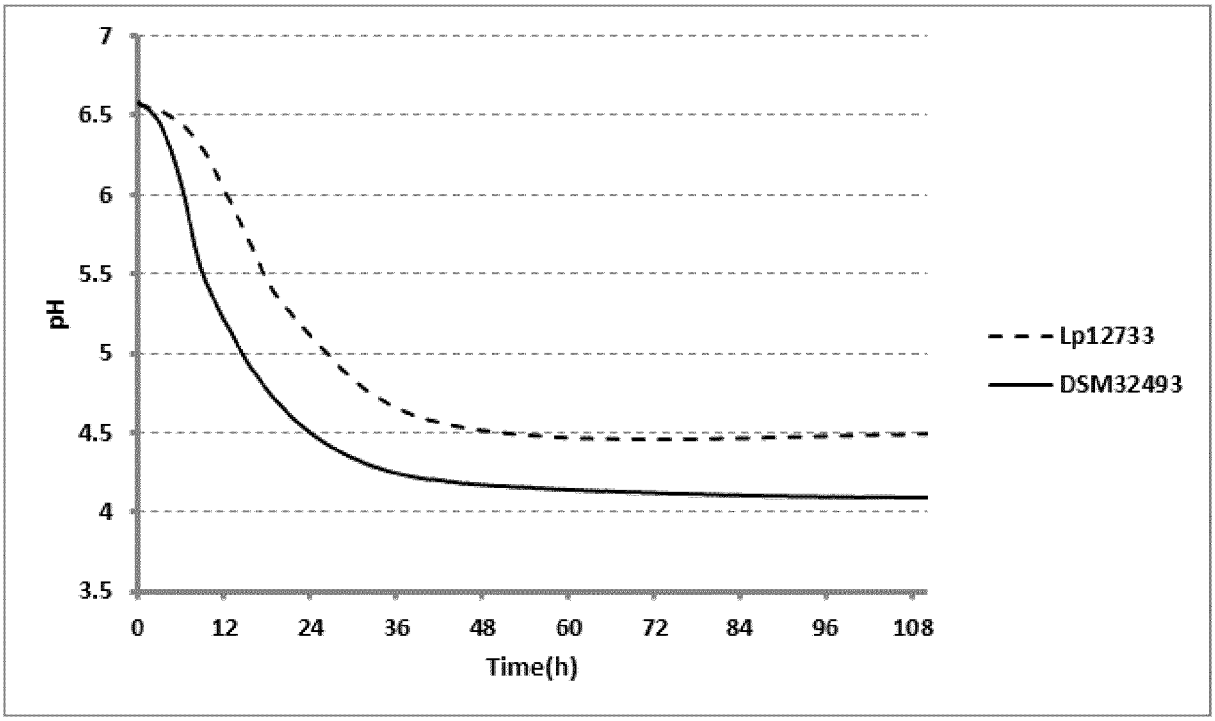
7
6.5
6
5.5
5
4.5
4
3.5
pH
0
12
24
36
48
60
72
84
96
108
Time(h)
Lp12733
DSM32493

Fig. 7

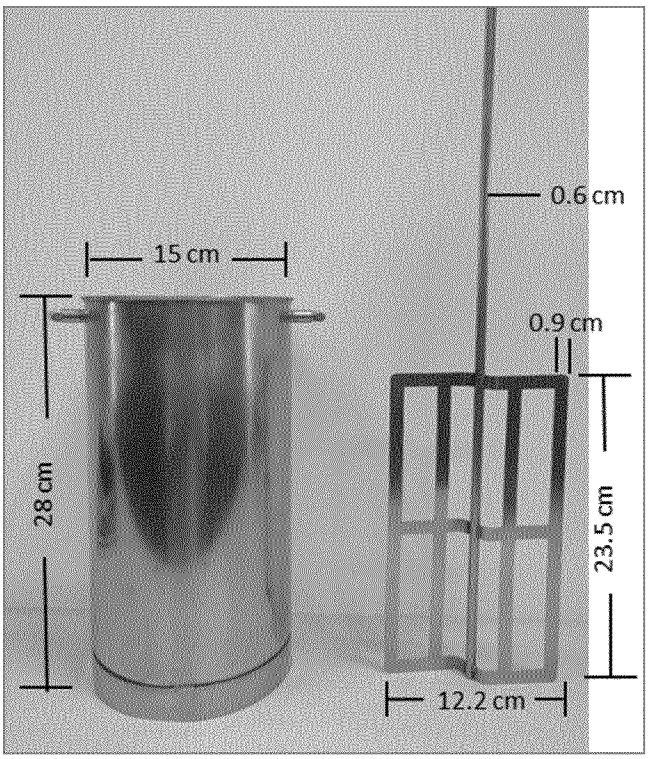
0.6 cm
15 cm
0.9 cm
28 cm
23.5 cm
12.2 cm

Fig. 8

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2018/076229

A. CLASSIFICATION OF SUBJECT MATTER
INV. C12R1/25 A23C9/123 A23L33/135
ADD.

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)
C12R A23C A23L

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)

EPO-Internal, FSTA, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	JI WANG ET AL: "Characterization of an exopolysaccharide produced by Lactobacillus plantarum YW11 isolated from Tibet Kefir", CARBOHYDRATE POLYMERS., vol. 125, 9 March 2015 (2015-03-09), pages 16-25, XP055538664, GB ISSN: 0144-8617, DOI: 10.1016/j.carbpol.2015.03.003	1,2,6-15
Y	abstract page 16, column 1, paragraph 1 - page 17, column 1, paragraph 3 page 18, column 1, paragraph 5 page 20, column 1, paragraph 2 - column 2 figures 1,5	3-5

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[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

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"A" document defining the general state of the art which is not considered to be of particular relevance

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"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
15 January 2019	21/01/2019

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Schlegel, Birgit

3

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C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X Y	US 2014/322273 A1 (AI LIANZHONG [CN] ET AL) 30 October 2014 (2014-10-30) paragraphs [0004], [0006] - [0027], [0099] - [0102]; claims 1-11	1,2,6-15 3-5
X Y	CN 104 498 402 A (UNIV BEIJING TECH & BUSINESS) 8 April 2015 (2015-04-08) paragraphs [0002] - [0006], [0029]; claims 1-9	1,2,6-15 3-5
X Y	CN 105 104 533 A (UNIV NANCHANG) 2 December 2015 (2015-12-02) paragraphs [0003] - [0010], [0040]; claims 1-3; table 1	1,2,6-15 3-5
X Y	CN 101 748 083 A (JILIN ACADEMY OF AGRICULTURAL) 23 June 2010 (2010-06-23) paragraphs [0002] - [0010], [0021], [0022], [0099] - [0129]; claims 1-10	1,2,6-15 3-5
A	WO 2013/160270 A1 (CHR HANSEN AS [DK]) 31 October 2013 (2013-10-31) page 2, line 17 - line 24; claims 1-10; example 3 page 3, line 13 - page 4, line 2 page 6, line 1 - line 23	1-15
A	US 2017/042172 A1 (MALJAARS CORNELIA ELIZABETH PAULINA [NL] ET AL) 16 February 2017 (2017-02-16) paragraphs [0014], [0015], [0080] - [0094]; claims 1-16; examples 1-6; tables 1,2	1-15
Y	BIHONG JIA ET AL: "Screening of Lactobacillus plantarum LPM21 with F1F0-ATPase ^¦^beta;-subunit Mutation Used as Probiotics Adjunct in Sichuan Pickle", FOOD SCIENCE AND TECHNOLOGY RESEARCH, vol. 19, no. 6, 31 December 2013 (2013-12-31), pages 1045-1050, XP055539257, CH ISSN: 1344-6606, DOI: 10.3136/fstr.19.1045 abstract page 1047, column 1, paragraph 3 - page 1049	3-5

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/EP2018/076229

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