Evaluation of *Lactibacillus rhamnosus* LGG and *Bifidobacterium* BB-12 growth in mixed plant-based beverages in industrial pilot production

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Summary
Plant-based milk substitutes are the most widespread plant-based alternatives today. Their biological value can be increased by fermentation and the activity of lactic acid bacteria. In this study, effect of temperature (1–8 °C, 20 °C and 37 °C) and short-term storage (up to 18 days) on *Lactibacillus rhamnosus* LGG (Chr. Hansen, Hoersholm, Denmark) and *Bifidobacterium* BB-12 (Chr. Hansen) growth and on the quality characteristics (titratable acidity, pH, dry matter content) of model almond-coconut-strawberry probiotic beverage was evaluated. Higher storage temperatures resulted in a significant ($p < 0.05$) increase in titratable acidity and viable cell counts to values above 10^9 CFU·ml^{-1}, while pH and dry matter content decreased significantly ($p < 0.05$). On the contrary, viable cell counts of *Lb. rhamnosus* and *Bifidobacterium* sp. varied slightly around average values of 5.1×10^6 CFU·ml^{-1} and 8.1×10^6 CFU·ml^{-1}, respectively, during refrigerated storage. In conclusion, the results showed that both bacterial strains have sufficient potential for industrial production of plant-based beverages, the production of which will contribute to the sustainable development of food production and ensure food security.

Keywords
fermentation; storage; temperature; almond-coconut-strawberry probiotic beverage; *Lactobacillus rhamnosus*; *Bifidobacterium*

Probiotics, along with prebiotics, phytonutrients and lipids, are among the segments of food products receiving increasing attention. Such products are mostly found on the market in the form of dairy products such as yoghurts, fermented milks or cheeses [1–3], while plant-based probiotic products are very rare. However, the popularity of plant-based products is increasing, primarily due to the growing percentage of people with lactose intolerance or allergic to milk protein [1, 2, 4–6]. Plant-based milk substitutes are the most widespread plant-based alternatives today. They are used for the production of plant-based yoghurts, kefir, cheese, butter, ice cream or cooking cream [5, 7].

The biological value of plant-based beverages can be enhanced through fermentation and the activity of lactic acid bacteria. Thus, for nutritional reasons, manufacturers began to test the lactic acid fermentation of plant substrates with the aim of developing fermented beverages from fruits, vegetables and other plant bases. During fermentation, the product is acidified (pH decreases), certain nutrients are degraded and lactic acid accumulates, which may be a challenge for the survival of probiotic bacteria.[1, 4, 5, 8] However, some studies showed that probiotic bacteria are able to survive under these adverse conditions and maintain themselves live in numbers higher than the minimum recommended doses, which range from 10^6 CFU·ml^{-1} to 10^7 CFU·ml^{-1}. Fermentation has a number of advantages, the most important of which is its conversation effect, i. e. ensuring the product's microbiological stability. However, in the case of beverages based on cereals, fruits or vegetables, it has a major impact on their nutritional and sensory quality [1, 6–8]. Fermentation produces organoleptically active compounds that cause changes in smell, taste, colour and/or consistency. Organic acids such as lactic acid or acetic acid are formed, giving the product an acidic and, above all, very specific taste [7]. Substances such

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as diacetyl, carbon dioxide or ethanol can also be formed. In addition to the sour taste, there is also a change in the sweetness of the product, mainly due to hydrolysis of oligosaccharides into shorter carbohydrates with higher sweetness [1].

Bacteria from the genera *Lactobacillus*, *Bifidobacterium* and *Streptococcus*, *Pediococcus* have been previously tested for the production of probiotic plant-based milk alternatives [1, 5, 6, 8, 9]. However, in the case of plant matrices, the correct selection of probiotic bacteria is extremely important, maintaining their viability in a non-dairy base is more challenging. This is because most probiotic strains have been isolated from dairy products and plant products may not be a suitable environment for them. Further problems may be caused by the nutrient content, anti-nutritional compounds, unfavourable pH and lack of buffering activity. However, scientific studies proved that many plant matrices can support the growth and reproduction of probiotic microorganisms [5, 6, 8, 10, 11].

In general, probiotics are thought to have a number of beneficial effects on host health. Probiotic bacteria are a natural part of the gut microbiome [2]. The microflora is believed to play an important role in human health, influencing the maturation of the immune system, metabolic responses, barrier function, regulation of the energy system and even brain behaviour through the brain-gut axis [6, 9]. A large variety of probiotic bacterial strains have been studied for their potential health benefits. *Lactobacillus rhamnosus* GG (LGG) and *Bifidocaterium animalis* ssp. *lactis* BB-12 are the best documented strains among them [12].

The potential of lactic acid bacteria is of great interest for plant-based fermentation [10]. This work sought to describe the growth dynamics of the probiotic strains *Lb. rhamnosus* LGG (Chr. Hansen, Hoersholm, Denmark) and *Bifidobacterium* BB-12 (Chr. Hansen) in plant-based substrates in industrial production, i. e. to determine the optimal conditions for their growth, maintaining of their viability and good sensory properties of the monitored beverages. Further, the aim was to assess the effect of temperature on the levels of probiotic bacteria, pH, titration acidity and solids content.

MATERIALS AND METHODS

Preparation of plant-based beverage

The plant-based beverage, which was produced on the line as a trial production at McCarter (Bratislava, Slovakia), was made form almonds, coconut cream, coconut water, rice flour, strawberry purée, lecithin, natural flavours and vitamin D. Ingredients were sterilized at 140 °C for 10 s. The product composition and processing conditions were designed with the aim of obtaining maximum shelf-life and not to complete the fermentation in the shortest possible time. The sterile plant base was aseptically inoculated with the combination of probiotic bacteria *Lb. rhamnosus*, Nu-Trish LGG DA and *Bifidobacterium* BB-12 (both Chr. Hansen). The inoculum was prepared for both dried and frozen forms by dissolving the preparation in 9 $g \cdot l^{-1}$ NaCl and dosage was adjusted to reach a concentration of 10^6 $CFU \cdot ml^{-1}$ of the product at a temperature of 5–10 °C. The resulting beverage was aseptically packaged in 200 ml polyethylene terephthalate (PET) bottles. The process was carried out without any further incubation, as it was the final consumer packaging. The products were stored and monitored at three temperatures, specifically, 1–8 °C (the commonly used temperature range in the retail chain), 20 °C and 37 °C using constant climate chambers HPP 108 (Memmert, Büchenbach, Germany). Only samples stored at a temperature of 1–8 °C were monitored up to 18 days, as storage at other temperatures led to deterioration of sensory properties and changes in physico-chemical parameters. Therefore, monitoring of samples stored at 20 °C and 37 °C was completed after 10 days. Sensory quality was evaluated by qualified sensory assessors during development, production and storage. In the case of the industrial test, acceptance of the sour taste and content of probiotic bacteria were monitored to obtain the maximum shelf-life of the product.

pH and dry matter content

A pH meter Accumet AB250 (Thermo Fischer Scientific, Waltham, Massachusetts, USA) was used to determine pH values in 25 ml of sample, following the procedure of AOAC method No. 981.21 [13]. Gravimetric dry matter content was determined using a moisture analyser MB90/MB120 (Ohaus, Nänikon, Switzerland). Briefly, 2 g of ground sample was spread on aluminium foil and dried at a maximum temperature of 105 °C to the constant weight.

Titratable acidity

Titratable acidity was determined by the titration method according to the AOAC method No. 942.15 [13]. Briefly, 5 g of sample was diluted by 100 ml of distilled wated and titrated with 0.1 $mol \cdot l^{-1}$ NaOH to a pink-coloured end point using phenolphthalein as an indicator. Results were expressed as concentration of citric acid [13].

Microbiological analysis

Counts of *Lb. rhamnosus* LGG were determined on De Man, Rogosa and Sharpe (MRS) agar (Biokar Diagnostics, Allonne, France) with the addition of 50 mg·kg^{-1} of vancomycin (Thermo Fisher Scientific). Diluted samples were inoculated using the pouring method and subsequently incubated for 72 h at 37 °C under anaerobic conditions created using AnaeroGen (Oxoid, Basingstoke, United Kingdom). After the incubation period, dishes with 15 to 150 colonies were evaluated. Counts of *Bifidobacterium* BB12 were determined on MRS agar with the addition of 25 mg·kg^{-1} of mupirocin (Dr Kulich Pharma, Hradec Králové, Czech Republic) and 500 mg·kg^{-1} of cysteine hydrochloride (VWR, Leuven, Belgium). The same procedure as for *Lb. rhamnosus* LGG was performed for *Bifidobacterium* BB-12. The resulting number of colony forming units (N) was calculated according to Eq. 1 [10]:

$$N = \frac{\sum c}{V(n_1 + 0.1n_2)} \times d \quad (1)$$

where Σc is sum of all colonies counted on Petri dishes; V is volume of the diluted sample used to inoculate the agar plate (1 cm^3), n_1 is number of dishes from the first dilution used for calculation, n_2 is number of dishes from the second dilution used for calculation, d is dilution factor (first dilution selected for calculation).

Statistical analysis

All measurements were performed in triplicate and results were expressed as mean ± standard deviation. Statistically significant differences between samples stored at different temperatures were evaluated by analysis of variance with Tukey's honesty significance test (ANOVA-Tukey HSD) at the level of significance of $p < 0.05$.

RESULTS AND DISCUSSION

Effects of cold short-term storage

The almond-coconut-strawberry probiotic beverage prepared by inoculation with *Lb. rhamnosus* LGG and *Bifidobacterium* BB-12 was monitored for 18 days at 1–8 °C, 20 °C and 37 °C. Acidity and pH are an important indicator that affects the quality of fermented beverages and are used primarily to estimate consumption quality. As shown in Tab. 1, storage caused significant differences ($p < 0.05$) in the pH values of fermented beverages between day 1 and day 18. The pH values decreased from 5.700 ± 0.010 to 5.180 ± 0.010, while the titratable acidity remaining stable within 10 days of refrigerated storage, followed by its significant increase in the final period of storage. The gradual decrease in pH may be attributed to the persistent metabolic activity of probiotic bacteria during cold storage [14, 15].

The dry matter content increased slightly to 818 ± 2 g·l^{-1} at the beginning of storage but decreased significantly to 808 ± 1 g·l^{-1} at the end of the storage period. This phenomenon could be attributed to degradation of carbohydrates and proteins consumed by bacteria as an energy source. However, decrease in dry matter content was lower compared to previous studies, probably due to low carbohydrate content in coconut and almond milk [16–18]. In line with our expectations, a strong negative correlation ($r = –0.9147$) was observed between pH and titratable acidity, while dry matter content was positively correlated with pH ($r = 0.8622$). The observed changes in pH and titratable acidity were slightly lower than those reported for fermented and non-fermented coconut and hemp milk [14] or soya, oat and coconut milk [10]. These differences could be related to the quality of the raw materials used in the production of fermented beverages, as the content of

Tab. 1. Parameters of almond-coconut-strawberry probiotic beverage inoculated with a combination of *Lactobacillus rhamnosus* LGG and *Bifidobacterium* BB12 stored during 18 days at 1–8 °C.

Storage time	pH	Dry matter [g·l^{-1}]	Titratable acidity [mg·l^{-1}]
Day 1	5.700 ± 0.010	818 ± 2	560 ± 10
Day 4	5.660 ± 0.010 *	826 ± 2 *	560 ± 20
Day 6	5.630 ± 0.020	824 ± 2	560 ± 10
Day 10	5.570 ± 0.010 *	807 ± 1 *	560 ± 10
Day 14	5.400 ± 0.020 *	799 ± 2 *	700 ± 20 *
Day 18	5.180 ± 0.010 *	808 ± 1 *	840 ± 10 *

* – statistically significant differences ($p < 0.05$) between consecutive storage days in the same column.

proteins, lipids and carbohydrates has significant impact on the development and activity of lactic acid bacteria [14].

Probiotic bacteria are living microorganisms, activity of which can easily be lost during processing and storage [19]. The most important parameter determining the quality of fermented plant-based beverages are the total bacterial counts. The efficacy of the added probiotic strains depends on the food matrix, fermentation conditions or inoculum level, and their viability must be maintained throughout storage of the product [20–23]. Probiotic drinks must contain a minimum of 10^6–10^7 CFU·ml^{-1} viable particular microflora at the end of storage [23]. The recommended daily dose of probiotics is 10^8–10^9 CFU, which corresponds to the consumption of 100 g of the fermented product containing 10^6–10^7 CFU·ml^{-1} [14]. As shown in Fig. 1, the concentrations of both bacterial strains varied slightly during the short-term storage. During the first 6 days, higher viable cell counts were observed for *Bifidobacterium* BB-12 (from 6.8×10^6 CFU·ml^{-1} to 7.4×10^6 CFU·ml^{-1}) compared to *Lb. rhamnosus* LGG (from 2.8×10^6 CFU·ml^{-1} to 4.1×10^6 CFU·ml^{-1}). The reverse trend was observed in the later stages of storage. The observed differences between viability of the two strains during storage were in agreement with previous studies [10, 20]. However, values of viable cell counts for both strains were lower compared to the study of Masiá et al. [10], who reported *Lb. rhamnosus* LGG and *Bifidobacterium* BB-12 counts in fermented coconut beverages to range from 5.7×10^7 CFU·g^{-1} to 6.2×10^8 CFU·g^{-1} and from 1.8×10^7 CFU·g^{-1} to 3.2×10^8 CFU·g^{-1}, respectively. These differences could be related to the different fermentation conditions and inoculum levels, as mentioned above. Overall, total counts of the probiotic lactic acid bacteria increased from 9.6×10^6 CFU·ml^{-1} to 1.4×10^7 CFU·ml^{-1}, which fulfilled the criteria of recommended concentrations for probiotics. Based on this result, it could be concluded that almond-coconut-strawberry probiotic beverage stored for 14 days at 1–8 °C could be beneficial for human consumption.

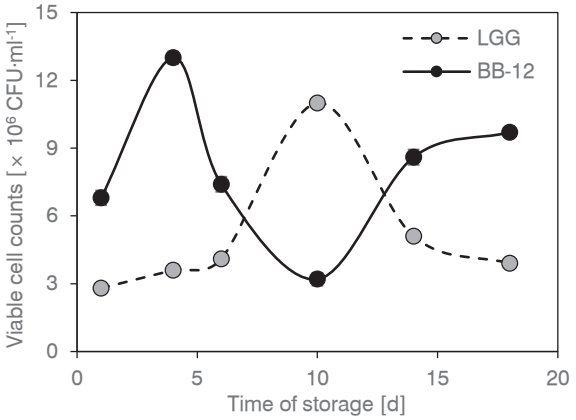

Fig. 1. Comparison of viable cell counts of *Lactobacillus rhamnosus* LGG and *Bifidobacterium* BB-12 in model almond-coconut-strawberry probiotic beverage during storage at 1–8 °C.

LGG – *Lactobacillus rhamnosus* LGG, BB-12 – *Bifidobacterium* BB-12.

Effects of storage temperature

Although fermentation processes are intended to increase food stability, intrinsic and extrinsic factors (composition of ingredients, processing or storage conditions) influence the quality characteristics and survival of probiotics in fermented foods. Storage time and temperature are main factors affecting the bacterial survival. In general, higher temperatures significantly reduce the viability of microorganisms, while lower temperatures have been reported to be better for the survival of certain probiotic bacteria [24].

The results for the effect of storage temperature on the analytical characteristics and counts of *Lb. rhamnosus* LGG and *Bifidobacterium* BB-12 are presented in Tab. 2 and Tab. 3. Compared to the temperature of 1–8 °C, storage for 10 days at 20 °C and 37 °C resulted in a sharp decrease in pH (approximately pH 3.5, $p < 0.05$) and a statistically significant ($p < 0.05$) increase in titratable acidity (up to 5 740–7 420 mg·l^{-1}). Dry matter content decreased significantly ($p < 0.05$) during storage at all temperatures. This phenomenon, as mentioned above, is related to degradation of carbohydrates used by the bacteria as an energy source. Despite the relatively low carbohydrate content of almond and coconut "milk", the decrease in dry matter content may be associated also with the breakdown of carbohydrates present in strawberry purée. In addition, reduced pH and increased titratable acidity at higher temperatures had a negative effect on sensory properties of the final products, as the intensity of sour taste became unpleasant.

Total counts of probiotic lactic acid bacteria increased significantly with the increasing temperature. At the end of the observation period (10 days), they reached values of 2.9×10^9 CFU·ml^{-1} and 5.1×10^9 CFU·ml^{-1} at 20 °C and 37 °C, respectively. Our results agree with those of a study of Liptáková et al. [22] who monitored growth of *Lb. rhamnosus* LGG in milk at various temperatures. They reported that

Tab. 2. Parameters of almond-coconut-strawberry probiotic beverage inoculated with *Lactobacillus rhamnosus* LGG and *Bifidobacterium* BB-12 during storage at various temperatures.

Temperature	Parameter	Storage time		
		Day 1	Day 6	Day 10
1–8 °C	pH	5.700 ± 0.010^{Aa}	5.630 ± 0.010^{Ab}	5.570 ± 0.020^{Ac}
	Dry matter [g·l^{-1}]	818 ± 2^{Aa}	824 ± 2^{Ab}	807 ± 2^{Ac}
	Titratable acidity [mg·l^{-1}]	560 ± 10^{Aa}	560 ± 10^{Aa}	560 ± 10^{Aa}
20 °C	pH	5.700 ± 0.010^{Aa}	4.120 ± 0.010^{Bb}	3.650 ± 0.010^{Bc}
	Dry matter [g·l^{-1}]	818 ± 2^{Aa}	797 ± 4^{Bb}	802 ± 1^{Bc}
	Titratable acidity [mg·l^{-1}]	560 ± 10^{Aa}	2380 ± 10^{Bb}	5740 ± 20^{Bc}
37 °C	pH	5.700 ± 0.010^{Aa}	3.570 ± 0.020^{BCb}	3.540 ± 0.010^{BCbc}
	Dry matter [g·l^{-1}]	818 ± 2^{Aa}	815 ± 1^{ACb}	801 ± 1^{BCc}
	Titratable acidity [mg·l^{-1}]	560 ± 10^{Aa}	7420 ± 20^{Cb}	7420 ± 20^{Cbc}

Different lowercase superscript letters in the same row indicate statistically significant differences ($p < 0.05$) between the consecutive storage days. Different uppercase superscript letters in the same column indicate statistically significant differences ($p < 0.05$) between the storage temperatures for each parameter.

Tab. 3. Comparison of viable cell counts of *Lactobacillus rhamnosus* LGG and *Bifidobacterium* BB-12 in model almond-coconut-strawberry probiotic beverage during storage at various temperatures.

Temperature	Bacterial strain	Viable cell counts [CFU·ml^{-1}]		
		Day 1	Day 6	Day 10
1–8 °C	*Labtobacillus rhamnosus* LGG	2.8×10^6	4.1×10^6	1.1×10^7
	Bifidobacterium BB-12	6.8×10^6	7.4×10^6	3.2×10^6
	Sum of probiotic bacteria	9.6×10^6	1.2×10^7	1.4×10^7
20 °C	*Labtobacillus rhamnosus* LGG	2.8×10^6	4.0×10^8	1.6×10^9
	Bifidobacterium BB-12	6.8×10^6	4.7×10^8	1.3×10^9
	Sum of probiotic bacteria	9.6×10^6	8.7×10^8	2.9×10^9
37 °C	*Labtobacillus rhamnosus* LGG	2.8×10^6	3.1×10^9	2.5×10^9
	Bifidobacterium BB-12	6.8×10^6	1.7×10^9	2.6×10^9
	Sum of probiotic bacteria	9.6×10^6	4.8×10^9	5.1×10^9

after 10 days at 6 °C, the growth of the bacterial strain was still in the exponential phase and the counts did not exceed 10^7 CFU·ml^{-1}, while stationary phase (3.2×10^7 CFU·ml^{-1}) was reached after 9 days at 8 °C. Our results are partially in agreement also with study of Fowoyo et al. [25], who reported stable counts of viable *Limosilactobacillus fermentum* in fermented milk with baobab fruit pulp stored for 10 days at 4 °C, while storage at room temperature resulted in a significant decrease in viable counts. Contrary to our results, de la Bastida et al. [26] reported stable counts of *Limosilactobacillus mucosae* during cold storage of soya-based beverages, while the viability of *Bifidobacterium pseudocatenulatum* decreased continuously, reaching uncountable levels on day 28. Similarly, Montero-Zamora et al. [27] described a gradual loss of viability of *Lb. rhamnosus* LGG in a whey-based beverage containing Costa Rican guava fruit pulp. The gradual increase in viable cell counts at higher temperatures in our study may be related to the differences in the technology of the model beverage. However, higher storage temperatures resulting in decreased pH, increased titratable acidity and high viable cell counts after 10 days of storage, made the studied almond-coconut-strawberry probiotic beverage unsatisfactory from a sensory point of view.

CONCLUSIONS

This study showed that the entire process of the plant-based beverage production can be controlled by changing the temperature, as storage temperature significantly affected viability of bacterial

strains and physico-chemical characteristics of the final product. Viability of *Lb. rhamnosus* LGG and *Bifidobacterium* BB-12 in model beverages stored at refrigeration temperature varied around average values of 5.1×10^6 CFU·ml^{-1} and 8.1×10^6 CFU·ml^{-1}, respectively. Concentration of both strains significantly increased in beverages stored at higher temperatures (20 °C and 37 °C). Higher storage temperature negatively affected the overall acceptability of the final product from a sensory point of view, the intensity of sour taste became unpleasant. Samples stored at higher temperatures were characterized by significantly higher titratable acidity, lower pH and lower dry matter content. Future research will be focused on studying other matrices and bacterial strains with regard to the quality of plant-based beverages, as fermented plant-based alternative beverages represent a promising segment with potential growth.

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REFERENCES

1. Küçükgöz, K. – Trząskowska, M.: Nondairy probiotic products: functional foods that require more attention. Nutrients, *14*, 2022, article 753. DOI: 10.3390/nu14040753.
2. Sharma, R. – Mokhtari, S. – Jafari, S. M. – Sharma, S.: Barley-based probiotic food mixture: Health effects and future prospects. Critical Reviews in Food Science and Nutrition, *62*, 2022, pp. 7961–7975. DOI: 10.1080/10408398.2021.1921692.
3. Dicks, L. – Botes, M.: Probiotic lactic acid bacteria in the gastro-intestinal tract: health benefits, safety and mode of action. Beneficial Microbes, *1*, 2010, pp. 11–29. DOI: 10.3920/BM2009.0012.
4. Aydar, E. F. – Tutuncu, S. – Ozcelik, B.: Plant-based milk substitutes: Bioactive compounds, conventional and novel processes, bioavailability studies, and health effects. Journal of Functional Foods, *70*, 2020, article 103975. DOI: 10.1016/j.jff.2020.103975.
5. Panghal, A. – Janghu, S. – Virkar, K. – Gat, Y. – Kumar, V. – Chhikara, N.: Potential non-dairy probiotic products – A healthy approach. Food Bioscience, *21*, 2018, pp. 80–89. DOI: 10.1016/j.fbio.2017.12.003.
6. Rasika, D. M. D. – Vidanarachchi, J. K. – Luiz, S. F. – Azeredo, D. R. P. – Cruz, A. G. – Ranadheera, C. S.: Probiotic delivery through non-dairy plant-based food matrices. Agriculture, *11*, 2021, article 599. DOI: 10.3390/agriculture11070599.
7. Jeske, S. – Zannini, E. – Arendt, E. K.: Past, present and future: The strength of plant-based dairy substitutes based on gluten-free raw materials. Food Research International, *110*, 2018, pp. 42–51. DOI: 10.1016/j.foodres.2017.03.045.
8. Rasika, D. M. D. – Vidanarachchi, J. K. – Rocha, R. S. – Balthazar, C. F. – Cruz, A. G. – Sant'Ana, A. S. – Ranadheera, C. S.: Plant-based milk substitutes as emerging probiotic carriers. Current Opinion in Food Science, *38*, 2021, pp. 8–20. DOI: 10.1016/j.cofs.2020.10.025.
9. Vinderola, G. – Burns, P. – Reinheimer, J.: Probiotics in nondairy products. In: Mariotti, F. (Ed.): Vegetarian and plant-based diets in health and disease prevention. London : Academic Press, 2017, pp. 809–836. ISBN: 9780128039687.
10. Masiá, C. – Geppel, A. – Jensen, P. E. – Buldo, P.: Effect of *Lactobacillus rhamnosus* on physicochemical properties of fermented plant-based raw materials. Foods, *10*, 2021, article 573. DOI: 10.3390/foods10030573.
11. Dahiya, D. – Nigam, P. S. – Dahiya, D. – Nigam, P. S.: Nutrition and health through the use of probiotic strains in fermentation to produce non-dairy functional beverage products supporting gut microbiota. Foods, *11*, 2022, article 2760. DOI: 10.3390/foods11182760.
12. Flach, J. – van der Waal, M. M. – Kardinaal, A. F. M. – Schloesser, J. – Ruijschop, R. M. A. J. – Claassen, E.: Probiotic research priorities for the healthy adult population: A review on the health benefits of *Lactobacillus rhamnosus* GG and *Bifidocaterium animalis* subspecies lactis BB-12. Cogent Food and Agriculture, *4*, 2018, article 1452839. DOI: 10.1080/23311932.2018.1452839.
13. Horwitz, W. – Latimer, G. (Ed.): Official methods of analysis of AOAC International. 19th edition. Gaithersburg : AOAC International, 2012. ISBN: 9780935584837.
14. Szparaga, A. – Tabor, S. – Kocira, S. – Czerwińska, E. – Kuboń, M. – Płóciennik, B. – Findura, P.: Survivability of probiotic bacteria in model systems of non-fermented and fermented coconut and hemp milks. Sustainability, *11*, 2019, article 6093. DOI: 10.3390/su11216093.
15. Bartkiene, E. – Zokaityte, E. – Lele, V. – Sakiene, V. – Zavistanaviciute, P. – Klupsaite, D. – Bendoraitiene, J. – Navikaite-Snipaitiene, V. – Ruzauskas, M.: Technology and characterisation of whole hemp seed beverages prepared from ultrasonicated and fermented whole seed paste. International Journal of Food Science and Technology, *55*, 2019, pp. 406–419. DOI: 10.1111/ijfs.14285.
16. Chawafambira, A. – Jombo, T. Z. – Mkungunugwa, T.: Effect of *Lacticaseibacillus rhamnosus* yoba fermentation on physicochemical properties, amino acids, and antioxidant activity of cowpea-peanut milk. Journal of Food Quality, *2022*, 2022, article 3192061. DOI: https://doi.org/10.1155/2022/3192061.
17. Bernat, N. – Cháfer, M. – Chiralt, A. – González-Martínez, C.: Development of a non-dairy probiotic fermented product based on almond milk and inulin.

Food Science and Technology International, *21*, 2015, pp. 440–453. DOI: 10.1177/1082013214543705.
18. Ziarno, M. – Derewiaka, D. – Dytrych, M. – Stawińska, E. – Zaręba, D.: Effects of fat content on selected qualitative parameters of fermented coconut "milk" beverage. Journal of Food and Nutrition Research, *59*, 2020, pp. 155–162. ISSN: 1336-8672. <https://www.vup.sk/en/download.php?bulID=2062>
19. Shudong, P. – Guo, C. – Wu, S. – Gui, H. – Suo, H. – Duan, Z.: Bioactivity and metabolomics changes of plant-based drink fermented by *Bacillus coagulans* VHProbi G08. LWT – Food Science and Technology, *156*, 2022, article 113030. DOI: 10.1016/j.lwt.2021.113030.
20. Łopusiewicz, Ł. – Drozłowska, E. – Trocer, P. – Kostek, M. – Kwiatkowski, P. – Bartkowiak, A.: The development of novel probiotic fermented plant milk alternative from flaxseed oil cake using *Lactobacillus rhamnosus* GG acting as a preservative agent against pathogenic bacteria during short-term refrigerated storage. Emirates Journal of Food and Agriculture, *33*, 2021, pp. 266–276. DOI: 10.9755/ejfa.2021.v33.i4.2679.
21. Valík, Ľ. – Medveďová, A. – Čižniar, M. – Liptáková, D.: Evaluation of temperature effect on growth rate of *Lactobacillus rhamnosus* GG in milk using secondary models. Chemical Papers, *67*, 2013, pp. 737–742. DOI: 10.2478/s11696-013-0365-1.
22. Liptáková, D. – Valík, Ľ. – Medveďová, A.: Characterization of the growth of *Lactobacillus rhamnosus* GG in milk at suboptimal temperatures. Journal of Food and Nutrition Research, *47*, 2008, pp. 60–67. ISSN: 1336-8672. <https://vup.sk/download.php?bulID=65>
23. Mantzourani, I. – Terpou, A. – Bekatorou, A. – Mallouchos, A. – Alexopoulos, A. – Kimbaris, A. – Bezirtzoglou, E. – Plessas, S.: Functional pomegranate beverage production by fermentation with a novel synbiotic *L. paracasei* biocatalyst. Food Chemistry, *308*, 2020, article 125658. DOI: 10.1016/j.foodchem.2019.125658.
24. Cabello-Olmo, M. – Oneca, M. – Torre, P. – Díaz, J. V. – Encio, I. J. – Barajas, M. – Araña, M.: Influence of storage temperature and packaging on bacteria and yeast viability in a plant-based fermented food. Foods, *9*, 2020, article 302. DOI: 10.3390/foods9030302.
25. Fowoyo, P. T. – Ogunbanwo, S. T. – Popoola, O. O. – Adeniji, P. O.: Development of non-dairy symbiotic fruit beverage using *Adansonia digatata* (baobab) fruit pulp as prebiotic. Fermentation, *8*, 2022, article 673. DOI: 10.3390/fermentation8120673.
26. De la Bastida, A. R. – Peirotén, Á. – Langa, S. – Curiel, J. A. – Arqués, J. L. – Landte, J. M.: Effect of storage and heat treatment on the levels of bioactive flavonoids produced in fermented soy beverages. LWT – Food Science and Technology, *154*, 2022, article 112872. DOI: 10.1016/j.lwt.2021.112872.
27. Montero-Zamora, J. – Cortés-Muñoz, M. – Esquivel, P. – Mora-Villalobos, J.-A. – Velázquez, C.: Growth conditions and survival kinetics during storage of *Lactobacillus rhamnosus* GG for the design of a sustainable probiotic whey-based beverage containing Costa Rican guava fruit pulp. Journal of Food Science, *85*, 2020, pp. 3478–3486. DOI: 10.1111/1750-3841.15430.

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