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Survival of lactic acid bacteria when using the developed yogurt from the milk of small cattle under in-vitro conditions

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Abstract

Functional nutrition is the use in everyday life of products of natural or artificial origin, which have a regulatory effect on the physiological functions, biochemical reactions of a person through the normalization of his microecological status. Cow remains a common raw material for yoghurts. In this article, the milk of small ruminants is considered as a raw material. The functionality of yogurt, shelf life, survival of bacteria in in vitro conditions are being studied. The purpose of the article is to identify the number of live microorganisms in terms of storage from - days, and in vitro conditions. Therefore, we chose the second method in order to study the viability of probiotics in the developed yogurts from sheep and goat milk, by modeling the gastrointestinal tract in vitro in comparison with the information available in the scientific literature. The studies were carried out in two stages: the stage of activation and preparation of strains, the second stage of modeling the gastrointestinal tract and checking at each stage of digestion. In this study, there were 8 samples, all samples showed a constant number of viable cells during 14 days of storage, even at 21 days there were viable bacteria. The number of viable cells was maintained at 107 and 106 CFU/mL until the 21st day of storage at 4 °C. The difference between BB counts may be related to its sensitivity to oxygen. Based on the experimental data obtained, it can be concluded that the samples showed a good initial concentration of probiotics - above 1010 CFU/g, after activation, as well as after simulating passage through the gastrointestinal tract with an average decrease of 106 and 107 CFU/g, which proves the therapeutic ability yoghurts from sheep's milk obtained according to the claimed method.

Keywords: milk of small cattle; acidity; viability; bacteria.

Practical Application: Determination of lactic acid bacteria survival in terms of shelf life.

1 Introduction

Fermented foods enrich the diet and allow nutrients to be stored and delivered to the human body in a complex blend of taste, aroma and texture. Conventional fermentation improves the overall content or availability of therapeutic potentials that have a profound impact directly on the health of the consumer. These microorganisms alter the biochemical constituents of raw materials, thereby improving the taste, digestibility, aroma, nutritional value in some fermented foods. Numerous reviews of biological, chemical and nutritional components of fermented foods from countries such as Asia, Africa and America have been published (Coskun & Dirican, 2019). The consumer is primarily motivated based on the organoleptic properties of food. Fermenting microorganisms use certain organic compounds contained in food substrates to produce special flavors and aromas as by-products. These by-products, including organic acids, esters and carbonyls, are compounds with intense aroma and are often produced by yeast and LAB (Kostov et al., 2020).

Cultures that enhance the characteristic flavors and texture of fermented dairy products are considered additives. Accessory crops may be homofermentative, such as the genera Pediococcus, Enterococcus and Streptococcus; otherwise, auxiliary crops are heterofermentative, releasing lactic acid, acetic acid, and CO2 as fermentative products (Macori & Cotter, 2018).

Heterofermenters include Leuconostoc spp., Lb. brevis and Lb. Fermentum and other Lactobacillus species are facultative heterofermenters as they produce CO_2 and other by-products from certain substrates (Jeantet & Jan, 2021).

The development of a new probiotic yogurt with a mixture of cow's and sheep's milk and the assessment of the physicochemical, textural and sensory parameters of these products was carried out by a group of researchers. The authors claim good taste, smell and texture, but many people suffer from cow protein intolerance and this yogurt does not solve this problem (Vianna et al., 2017).

A study was made of the effect of honey on the quality characteristics of goat yogurt containing the probiotic Lactobacillus acidophilus on the technological, physico-chemical and sensory characteristics of goat yogurt during 28 days of refrigerated storage. Four formulations of goat yogurt were prepared, each varying in added amount of bee honey [(0%, 5%, 10% and 15%], but all were inoculated with L. acidophilus La-05 probiotic (0.1 g/L goat's milk).The inclusion of bee honey positively

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affected several characteristics, namely color, syneresis, viscosity, sensory acceptability, of the developed yogurt. All formulations of yogurt present an amount of L. acidophilus La-05 above 6.0 log CFU/g to 28 days of storage, but the presence of honey increased the amount (about 1 log CFU/g) of L. acidophilus La-05 and yoghurt starter bacteria up to 21 days of storage.The results of this study showed the successful inclusion of both the probiotic L. acidophilus La-05 and honey, produced by a local Brazilian bee as ingredients of a novel goat milk product with satisfactory nutritional and sensory quality and added market value due to its potential functional properties (Machado et al., 2017).

Dairy products, because they naturally contain probiotic microorganisms, are generally a good source of their viability during storage. This is because dairy derivatives promote the survival of probiotics in the gastrointestinal tract through fat globules that can protect viable cells from the highly acidic conditions of the stomach and intestinal bile salts. In addition, this is a great opportunity to offer probiotics and increases the chances of consumption by the public, because they are sold on supermarket shelves and are not considered as medicine. Accordingly, for the preservation and delivery of probiotics to the human body, yogurt must be fatty. To be beneficial to humans, probiotics must survive passage through the gastrointestinal tract, the acidic conditions of the stomach, and be able to reach the large intestine in sufficient quantities to allow colonization and reproduction. By adhering to intestinal epithelial cells, probiotics can improve the microbiota and digestive process, protect against pathogens, and generate potential anti-carcinogenic properties. However, most probiotics cannot survive in large amounts due to low gastric pH, which limits their effectiveness in most functional foods. Resistance to gastric acid and tolerance to bile salts are the two main properties of microorganisms that should be considered as probiotics, allowing them to survive in the acidic conditions of the stomach and the presence of bile salts in the small intestine during passage through the gastrointestinal tract (Verruck et al., 2020; Dantas et al., 2022; Wang et al., 2022).

Probiotics should be present in food at 108-9 CFU/g in the daily product recommendation before ingestion to ensure that a sufficient therapeutic minimum of 6-7 CFU/g^{-1} can reach the colon (Okpara, 2022).

The main difficulty that industries face when adding probiotic bacteria to functional foods is to maintain the viability of these cultures. In-vitro digestion models are widely used to study by modeling gastrointestinal conditions, structural changes, digestibility and release of compounds present in food (Caillard & Lapointe, 2017).

Justifying the benefits of probiotics requires evidence of survival in the human digestive tract and evidence of exposure in humans.

Thus, several researchers have already assessed the resistance of microorganisms in gastrointestinal fluids by modeling the digestive process using in-vitro tests (Günter et al., 2022; Jin et al., 2020)

It is known that exposure to oxygen and a decrease in pH reduces the concentration of probiotics, as well as the presence of sucrose, as a potential inhibitor, some products of the initial metabolism of lactic acid (diacetyl, acetaldehyde, acid lactic acid) can also be associated with the loss of viability of probiotic bacteria (Prestes et al., 2021; Haji et al., 2022).

The low acidity of the stomach is the first barrier against microorganisms, many ingested bacteria are killed, however, acid-fast bacteria such as Lactobacillus sp., Bifidobacterium sp. and Streptococcus sp. able to survive. The contact time of the microorganism at low pH is a critical factor for cell viability. Slower digestion at the stomach stage will have a lower concentration of probiotics. In addition, the pH of the stomach changes throughout the day depending on the food eaten, and it may be more effective to consume probiotics with foods and drinks with a higher pH.

In this context, milk and some of its derivatives may protect microorganisms by helping to maintain viability during digestion when exposed to low pH conditions (below 2.0), which is related to this protection due to the presence of fat globules and milk proteins, mainly casein (Łętocha et al., 2022).

Thus, probiotic cultures may be more viable if they are associated with acid-tolerant bacteria and serve as protective cultures for them. A probiotic strain must meet certain selection criteria in order to be able to have a positive effect. It is important to take into account some aspects of safety, such as origin (healthy human gastrointestinal tract) and non-pathogenicity.

Functional aspects include acid tolerance and human gastric acid; bile tolerance; adherence to epithelial surfaces and persistence in the human gastrointestinal tract; immunostimulation; antagonistic activity against pathogens; antimutagenic and anticarcinogenic properties.

Some technological aspects are good sensory properties; phage resistance; viability during processing; stability in product and during storage (Raza et al., 2022; Yetiman & Ortakci, 2023).

An urgent area of research is the study of the positive impact on the human body of lactic acid strains that can normalize the functioning of the microflora of the gastrointestinal tract and regulate the health of the human body.

Currently, scientists are increasingly considering lactic propionic acid bacteria as probiotics. This paper discusses technological aspects when using strains of Streptococcus thermophillus, Lactobacillus acidophilus, Bifidobacterium bifidum, Propionibacterium shermanii used for the production of yogurt (Vianna et al., 2017).

Some strains of Streptococcus thermophilus are able to synthesize exopolysaccharides (EPS), which act as natural biothickeners that improve the texture of fermented foods. The production of EPS on site during milk fermentation offers clear benefits for the food industry. Streptococcus thermophilus strains that produce EPS can reduce syneresis and improve product texture and viscosity. Thus, these types of cultures are commonly used as substitutes for commercial stabilizers in yoghurt production. Moreover, studies have shown that EPS may have beneficial effects on human health, including cholesterol lowering, prebiotic effects, immunomodulatory and antitumor activity (Al-Emran et al., 2022).

2 Methods and materials

2.1 The rate of nutrient medium acidification and biomass accumulation with given quality parameters

Tubes containing 0.1% sterile peptone water and yoghurt samples were homogenized by vortexing. After homogenization, the samples were sequentially diluted to a decimal degree in 0.1% sterile peptone water, poured onto plates with ST agar (aerobiosis) at 30 ºC/48 h for *Str.thermophilus* (*ST*), in MRS agar with bile (aerobiosis) at 37 °C/72 hours for L. Acidophilus (LAC) and in LP-MRS (lithium propionate-MRS) agar (anaerobiosis) at 37 °C/72 hours for Bifidobacterium bifidum (BB).

2.2 Organoleptic characteristics

The evaluation of sensory attributes was carried out using the methodology of the acceptance test with the participation of 39 untrained experts on a 10-point balanced scale. Each member of the commission received 20 g of yogurt samples. The samples were codified and presented to the examiner in a randomized manner, as suggested by the scientific and technical activity.

The data were analyzed by ANOVA using the Benferroni test to assess differences between means. A P value < 0.05 was considered significant.

Preclinical studies for the humane treatment of laboratory animals are shifting to more cost-effective and substitute methods such as in-vitro simulations of the gastrointestinal tract. Therefore, we chose the second method in order to study the viability of probiotics in the developed yogurts from sheep and goat milk, by modeling the gastrointestinal tract in vitro in comparison with the information available in the scientific literature.

Sheep milk was taken from four breeds of peasant farms in Kazakhstan. Goat milk was obtained from one farm, 3 breeds of goats were considered.

Determination of the survival of bifidobacteria and lactobacilli under In-vitro conditions with simulation of the gastrointestinal tract.

2.3 Analysis progress

Sample preparation

The samples were prepared as indicated in the scientific study materials (Silva et al., 2021). Probiotic cultures (Bifidibacterium and Lactobacterium) were activated prior to use as recommended by the manufacturers. The activation method used was in MRS culture medium (broth) using 1 g probiotic for every 100 mL of broth, followed by incubation at 37 °C in an incubator for 15 hours. At the end of the incubation, to activate the cultures along with the broth, it was centrifuged at a turnover of 4500 × g for 15 min in a centrifuge at 4 °C, washed twice in NaCl solution (0.85% w/v). Viable probiotic cell count.

Viable probiotic cells were counted in two stages as described in (Sheth et al., 2022):

1. After preparation for use (activated);
2. After modeling the GIT, count at each stage of passage through the gastrointestinal tract of the in vitro experiment (esophagus/stomach, duodenum, and ileum).

For analysis, samples of 1.0 g were transferred in appropriate dilutions (in triplicate) into disposable Petri dishes. For Lactobacillus, the deep seeding method on MRS agar was used, and 0.5% dicloxacillin at 0.01% w/v, 1.0% 11.0% w/v lithium chloride was added to determine the concentration of Bifidobacterium and 0.5% solution of 10.0% w/v cysteine chloride in broth with the same inoculation. After inoculation, Petri dishes were incubated upside down in an anaerobic vessel at 37 °C for 72 hours (Rakotonirina et al., 2022).

Modeling to evaluate the survival of commercial probiotics under gastrointestinal conditions was performed as described (Haji et al., 2022), with some modifications (Figure 1). The assay was performed in a water bath maintained at 37 °C to mimic human body temperature, and mechanical agitation was used to mimic peristaltic bowel movements at a similar intensity.

Survival was assessed sequentially in media simulating different parts of the gastrointestinal tract (esophagus/stomach, duodenum, and ileum). 9 mL of peptone water was added to 1 g of the sample.

An acidic solution (0.1 mol L^{-1} HCl) and an alkaline solution (0.1 mol L^{-1} $NaHCO_3$) were pre-prepared and sterilized to control the pH of the samples throughout the experiment. Initially, the pH was adjusted to 7 to simulate acidity in the mouth, then after 3-4 minutes they moved on to the next step.

At the esophageal stage, 25 mg/mL of enzyme pre-prepared in HCl was used. This solution was added in equal doses throughout the gastric phase. At the end of this step, 3 samples were taken for plating to analyze the number of surviving probiotic cells in the simulated passage through the stomach.

In the duodenal step, the enzymes were taken and the pH was adjusted to 5.0 by adding bile enzymes and incubated for 20 minutes at 45 °C with stirring.

At the end, the next 3 samples are removed for analysis to determine the number of surviving probiotic cells.

In the ileum step, the pH is increased to 6.5 using a 0.1 mol/L NaHCO3 solution. At the end of this step, the samples are also analyzed to count the probiotic cells that survived the simulation.

The data obtained were entered into Microsoft Office Excel spreadsheets and triplicates were averaged for each microbiological analysis and with standard deviation representation (Ospanov et al., 2022; Mazzantini et al., 2022).

The purpose of simulation of the GI tract in in-vitro conditions is to determine the survival and concentration of the used bacterial species in the stomach, duodenum and ileum in the presence of enzymes and pH changes, as well as the adherence and physiological effect of the samples.

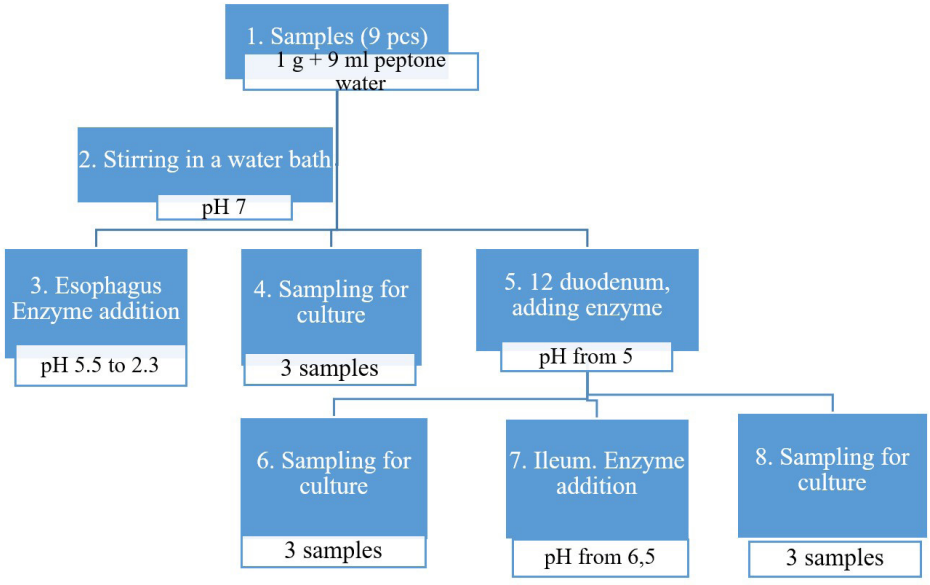

Figure 1. Stages of modeling the gastrointestinal tract (esophagus/stomach, duodenum and ileum). The scheme is shown for the analysis of 1 type of yogurt, the rest were repeated in the same direction.

Starter cultures used to make yogurt

1. Manufactured in Denmark, consisting of: Streptococcus thermophillus, Lactobacillus bulgaricus, Bifidobacterium bifidum;
2. Production Russia VIVO, consisting of: Streptococcus thermophillus, Lactobacillus bulgaricus, Bifidobacterium bifidum, Lactobacillus acidophilus.
3. Production Chr. Hansen (Denmark), consisting of: Streptococcus thermophillus, Lactobacillus acidophilus, Bifidobacterium bifidum.

3 Results

The rationale for the recipe, the rationale for combining milk for the production of live yoghurts is given in (Ospanov et al., 2022). The process of fermentation with selected cultures of live yoghurts from sheep, goat and cow milk was studied (Table 1).

Based on the results of the studies presented in Table 1, it can be concluded that the indicator of the total duration of milk fermentation corresponds to the regulatory documentation for milk for fermented milk products and ranges from 3 to 5 hours. The number of somatic cells is within the normal range (up to 400 thousand [cm]^3), which indicates the safety of the milk supplied for processing.

Table 2 also analyzes the values found for Lactobacillus sp. and Bifidobacterium sp. in every sample.

The samples showed concentrations corresponding to the values indicated on the label, and amounted to 9.9 and 10.7 CFU g^{-1}, respectively. The standard deviation is determined by a triple repetition at each point of the experiment. The results are expressed in CFU g^{-1}.

However, studies show that for foods or probiotic supplements to offer benefits, they must reach the ileum of the small intestine at a minimum concentration of 6 CFU/g. However, the concentration of viable strains is expected to decrease by about 2 CFU/g as it passes through the gastrointestinal tract, so it is recommended that the food or supplement have a concentration of at least 8 CFU g^{-1} (Mazzantini et al., 2022; Liu et al., 2022).

Sensory differences between the 3 samples (sheep yogurt supplemented with starter combinations (#1, #2, #3) were significant ($P < 0.05$) as storage time increased.

On day 1 of refrigeration, group members' most preferred fermented dairy products were No. 1 and No. 2 samples, which did not differ ($P < 0.05$) from each other in Experiment 1. These results are similar to those obtained after 21 days storage.

After 7 days of refrigerated storage, the sensory scores of the experiment showed significant differences ($P < 0.05$) among the selected combinations of all combinations.

Similar results were obtained when stored for 21 days. The most preferred combinations #1 and #3 retained their positions in order of preference at days 14 and 21 of storage, although they did not differ significantly ($P < 0.05$).

It was noted that after 7 days of storage #2 had the lowest sensory scores and was significantly different ($P < 0.05$) from #3 in the experiment.

Table 3 and Table 4 can also analyze the values found for Lactobacillus sp. and Bifidobacterium sp. in every sample.

The regulation of commercial release of probiotic foods and supplements establishes that the manufacturer is responsible for demonstrating to the inspection agency that the product provides the consumer with probiotic benefits, that is, that the concentration and viability of the probiotics are sufficient to provide

Table 1. Table of fermentation of live yogurt with starter cultures.

Product name	Temperature, °C	Fermentation time, hour	Acidity, °T
culture number 1			
Yogurt from cow's milk	40	4 hours	78
Yogurt from sheep milk	40	3 hours	75
Yogurt from goat milk	40	4 hours 15 minutes	82
culture number 2			
Yogurt from cow's milk	40	3 hours 20 minutes	82
Yogurt from sheep milk	40	2 hours 25 minutes	78
Yogurt from goat milk	40	3hours 30 minutes	84
culture number 3			
Yogurt from cow's milk	40	3 hours 30 minutes	83
Yogurt from sheep milk	40	2 hours 32 minutes	80
Yogurt from goat milk	40	3 hours 35 minutes	84

Table 2. Probiotic viability of microorganisms before and after modeling of the gastrointestinal tract.

Samples	Gastrointestinal stimulation			
	Initial	Esophagus	12 duodenum	The Ileum
Lactobacillus sp.				
Edilbay	9.8 ± 0.2	8.7 ± 0.1	8.0 ± 0.1	8.1 ± 0.1
Meat merino	10.5 ± 0.1	9.4 ± 0.2	9.3 ± 0.1	9.0 ± 0.1
Kazakh fine - wool	9.9 ± 0.1	7.1 ± 0.1	7.1 ± 0.3	7.0 ± 0.1
South Kazakh Merino	10.7 ± 0.2	9.4 ± 0.1	8.3 ± 0.1	8.6 ± 0.1
Bifidobacterium sp.				
Edilbay	10.5 ± 0.2	9.2 ± 0.2	7.3 ± 0.1	7.2 ± 0.1
Meat merino	7.7 ± 0.1	3.3 ± 0.1	3.6 ± 0.1	3.5 ± 0.1
Kazakh fine - wool	5.6 ± 0.2	5.4 ± 0.1	5.2 ± 0.1	5.1 ± 0.2
South Kazakh Merino	8.0 ± 0.1	6.7 ± 0.2	6.9 ± 0.1	6.9 ± 0.2

Table 3. Probiotic viability of commercial microorganisms before and after simulation of the gastrointestinal tract. The analysis was performed on 1 g of the sample.

№ n\o	Sheep breeds whose milk was used to make yogurt	Simulation of the gastrointestinal tract			
		Primary	Esophagus	12 duodenal intestine	ileum
1	*When making probiotics Lactobacillus sp. in yogurt*				
1.1	Ordabasy	10^{10}	10^{8}	10^{7}	10^{6}
1.2	Meat merino	10^{10}	10^{9}	10^{8}	10^{7}
1.3	Kazakh fine-fleece	10^{10}	10^{7}	10^{7}	10^{7}
1.4	South Kazakh merino	10^{10}	10^{9}	10^{8}	10^{8}
2	*When introducing probiotics Bifidobacterium sp. in yogurt*				
2.1	Ordabasy	10^{10}	10^{9}	10^{7}	10^{7}
2.2	Meat merino	10^{10}	10^{7}	10^{7}	10^{6}
2.3	Kazakh fine-fleece	10^{10}	10^{8}	10^{7}	10^{7}
2.4	South Kazakh merino	10^{10}	10^{7}	10^{7}	10^{6}

Triple repeat and standard deviation (± 0.1). The results are expressed in CFU g^{-1}.

Table 4. Probiotic viability of commercial microorganisms in formulated yogurt.

№ n\o	Yogurt from the milk of the Zaanenskaya goat breed, when applied:	Simulation of the gastrointestinal tract, CFU / g			
		Primary	Esophagus	12 duodenal intestine	ileum
1	- probiotics Propionibacterium	10^{10}	10^{9}	10^{8}	10^{7}
2	- probiotics Lactobacillus acidophilus	10^{10}	10^{8}	10^{7}	10^{7}
3	- probiotics Bifidibacterium bifidum	10^{10}	10^{8}	10^{7}	10^{6}
4	- consortium of the above probiotics in a ratio of 0.5:1:1, respectively	10^{10}	10^{9}	10^{8}	10^{7}

such benefits. Therefore, it is not possible to determine the ideal concentration of probiotics in a product prior to consumption.

Based on Table 2, we can conclude that all samples showed a good concentration. Sample #1 with the probiotic Propionibacterium had an initial concentration of 1010 CFU/g, after simulation of the gastrointestinal tract, it showed a concentration of 107 CFU/g, which is considered sufficient for colonization of the intestine. Sample #2 with probiotic Lactobacillus acidophilus showed an initial concentration of 1010 CFU/g, after the in vitro test a viability of 107 CFU/g was found. In sample #3, the initial concentration was 1010 CFU/g, after the test it showed a decrease close to 106 CFU/g, which is considered the norm, which carries a probiotic advantage.

However, studies show that for foods or probiotic supplements to offer such benefits, they must reach the ileum of the small intestine at a minimum concentration of 6 CFU/g. However, the concentration of viable strains is expected to decrease by about 2 CFU/g as it passes through the gastrointestinal tract, so it is recommended that the food or supplement have a concentration of at least 8 CFU g^{-1} (Prestes et al., 2021; Haji et al., 2022).

Tables 1-2 presents the results of the probiotic viability of the analyzed products, showing the initial concentration and survival during passage through the simulated model of the gastrointestinal tract. During the passage through the stomach, the number of viable cells and the survival of probiotic microorganisms are reduced due to the extreme pH of gastric acid, which limits the effectiveness of probiotic microorganisms (Prestes et al., 2021; Haji et al., 2022).

The samples showed a good initial concentration, above 108 CFU g^{-1}, after activation, as well as after simulated passage through the gastrointestinal tract with an average decrease of 104 CFU g^{-1}, as expected.

The sample (Kazakh Fine Wool) with Lactobacillus and Bifidobacterium was at an initial concentration of 109 CFU g^{-1}, after GI tract simulation it showed a concentration of 107 CFU g^{-1}, an amount considered sufficient for intestinal colonization.

Based on the experimental data obtained, it can be concluded that the samples showed a good initial concentration of probiotics - above 1010 CFU/g, after activation, as well as after simulating passage through the gastrointestinal tract with an average decrease of 106 and 107 CFU/g, which proves the therapeutic ability yoghurts from sheep's milk obtained according to the claimed method.

In the literature, other authors have also reported that the activity of the starter for yogurt and probiotic bacteria caused certain changes in foods that affect sensory characteristics, and that fermented foods cultured with bifidobacteria differed from yogurts made using traditional starters.

Differences in order of preference between samples may be related to other organic acids produced by Bulgaricus or Thermophilus. For example, producing more acetic acid from days 7 to 14 of storage may cause sensory differences that the group member may perceive negatively.

Propionic acid bacteria are Gram-positive, facultative anaerobes. During fermentation, they form a specific substance. It is propionic acid, hence the name of the microorganism. The peculiarity of propionic acid bacteria is that they increase the shelf life of products, enrich them with living cells of probiotic microorganisms and B12 vitamins, and protect milk and whey fermentation products from spoilage. Bifidobacteria live in the large intestine. The peculiarity of bifidobacteria is that they protect against the penetration of microbes and toxins into the internal environment of the body, synthesize amino acids and proteins, and enhance the absorption of calcium, iron, and vitamin D ions.

Yoghurts based on goat's milk, in comparison with similar yogurts from cow's milk, will differ in quality characteristics, high functional properties and attractiveness for consumers of all age groups.

The results regarding sensory preferences can probably be explained by the behavior of microorganisms, similar to those described in previous works. The increase and decrease in the sensory samples observed after 7 days in experiment 1 and after 14 days of storage may be due to acid formation.

4 Conclusion

An analysis of the market for the production of goat and sheep milk in the Republic of Kazakhstan shows an increased interest in both milk and processed products. The increased and stable growth of interest of the scientific community in the topic of small cattle milk and processed products is also justified.

The range of goat and sheep milk products is expanding every year. In the world, yoghurts, butter, and fermented products are also produced from the milk of small ruminants. The change in the quality indicators of the listed products from combined raw materials is also being studied. Works are given that consider combined raw materials, camel milk-sheep, cow-sheep, cow-goat, goat-sheep, etc.

The direction in the development of products from the milk of small ruminants is also the use of secondary raw materials, the analysis of production modes, syneresis, quality indicators. The issue of lossless delivery of a group of bifidobacteria through the human gastrointestinal tract is being studied, both in vitro and in vivo. Many works are devoted to the enrichment of the composition with probiotics and prebiotics, synbiotics and metabiotics.

In this regard, most of the works are devoted to the direction of development of the technology of functional yoghurts. The composition and modes are also studied.

In this study, there were 8 samples, all samples showed a constant number of viable cells during 21 days of storage.

The number of viable cells was maintained at 107 and 106 CFU/mL until the 28th day of storage at 4 °C. The difference between BB counts may be related to its sensitivity to oxygen. While stirring the milk in the experiment, more oxygen could be added.

References

Al-Emran, H. M., Moon, J. F., Miah, M. L., Meghla, N. S., Reuben, R. C., Uddin, M. J., Ibnat, H., Sarkar, S. L., Roy, P. C., Rahman, M. S., Alam, A. S. M. R. U., Islam, O. K., & Jahid, I. K. (2022). Genomic analysis and in vivo efficacy of Pediococcus acidilactici as a potential probiotic to prevent hyperglycemia, hypercholesterolemia and gastrointestinal infections. *Scientific Reports*, 12(1), 20429. http://dx.doi.org/10.1038/s41598-022-24791-5. PMid:36443433.

Caillard, R., & Lapointe, N. (2017). In vitro gastric survival of commercially available probiotic strains and oral dosage forms. *International Journal of Pharmaceutics*, 519(1-2), 125-127. http://dx.doi.org/10.1016/j.ijpharm.2017.01.019. PMid:28093323.

Coskun, F., & Dirican, L. K. (2019). Effects of pine honey on the physicochemical, microbiological and sensory properties of probiotic yoghurt. *Food Science and Technology*, 39(Suppl. 2), 616-625. http://dx.doi.org/10.1590/fst.24818.

Dantas, D. L. S., Viera, V. B., Soares, J. K. B., Santos, K. M. O., Egito, A. S., Figueirêdo, R. M. F., Lima, M. S., Machado, N. A. F., Souza, M. F. V., Conceição, M. L., Queiroga, R. C. R. E., & Oliveira, M. E. G. (2022). Pilosocereus gounellei (xique-xique) flour: improving the nutritional, bioactive, and technological properties of probiotic goat-milk yogurt. *LWT*, 158, 113165. http://dx.doi.org/10.1016/j.lwt.2022.113165.

Günter, E. A., Martynov, V. V., Ananchenko, B. A., Martinson, E. A., & Litvinets, S. G. (2022). The gel strength and swelling in the gastrointestinal environment of pectin/κ-carrageenan gel particles based on pectins with different degrees of methylesterification. *Materials Today. Communications*, 33, 104986. http://dx.doi.org/10.1016/j.mtcomm.2022.104986.

Haji, F., Cheon, J., Baek, J., Wang, Q., & Tam, K. C. (2022). Application of Pickering emulsions in probiotic encapsulation-a review. *Current Research in Food Science*, 5, 1603-1615. http://dx.doi.org/10.1016/j.crfs.2022.09.013. PMid:36161224.

Jeantet, R., & Jan, G. (2021). Improving the drying of *Propionibacterium freudenreichii* starter cultures. *Applied Microbiology and Biotechnology*, 105(9), 3485-3494. http://dx.doi.org/10.1007/s00253-021-11273-3. PMid:33885925.

Jin, H. S., Fei, Y. S., Yan, C. K., Kuan, C. H., & Wei, S. W. Y. (2020). Effect of gums coating materials on the survival of microencapsulated probiotics under simulated gastrointestinal conditions. *Materials Today: Proceedings*, 29, 16-19. http://dx.doi.org/10.1016/j.matpr.2020.05.685.

Kostov, G., Denkova-Kostova, R., Denkova, Z., Nenov, N., Shopska, V., Dzhivoderova-Zarcheva, M., Teneva, D., Goranov, B., Petelkov, I., Bouarab, L., Demarigny, Y., Oulahal, N., Husson, F., Waché, Y., & Degraeve, P. (2020). Biopreservation of emulsified food and cosmetic products by synergistic action of probiotics and plant extracts: a Franco-Bulgarian perspective. *Food Science And Applied Biotechnology*, 3(2), 167-184. http://dx.doi.org/10.30721/fsab2020.v3.i2.93.

Łętocha, A., Miastkowska, M., & Sikora, E. (2022). Preparation and characteristics of alginate microparticles for food, pharmaceutical and cosmetic applications. *Polymers*, 14(18), 3834. http://dx.doi.org/10.3390/polym14183834. PMid:36145992.

Liu, J., Li, X., Zhang, X., Wang, R., Han, S., Qin, P., Xing, X. H., & Zhang, C. (2022). Gut lumen-targeted oral delivery system for bioactive agents to regulate gut microbiome. *Journal of Future Foods*, 2(4), 307-325. http://dx.doi.org/10.1016/j.jfutfo.2022.08.003.

Machado, T. A. D. G., Oliveira, M. E. G., Campos, M. I. F., Assis, P. O. A., Souza, E. L., Madruga, M. S., Pacheco, M. T. B., Pintado, M. M. E., & Queiroga, R. C. R. E. (2017). Impact of honey on quality characteristics of goat yogurt containing probiotic Lactobacillus acidophilus. *LWT*, 80, 221-229. http://dx.doi.org/10.1016/j.lwt.2017.02.013.

Macori, G., & Cotter, P. D. (2018). Novel insights into the microbiology of fermented dairy foods. *Current Opinion in Biotechnology*, 49, 172-178. http://dx.doi.org/10.1016/j.copbio.2017.09.002. PMid:28964915.

Mazzantini, D., Calvigioni, M., Celandroni, F., Lupetti, A., & Ghelardi, E. (2022). In vitro assessment of probiotic attributes for strains contained in commercial formulations. *Scientific Reports*, 12(1), 21640. http://dx.doi.org/10.1038/s41598-022-25688-z. PMid:36517529.

Okpara, M. (2022). Microbial enzymes and their applications in food industry: a mini-review. *Advances in Enzyme Research*, 10(1), 23-47. http://dx.doi.org/10.4236/aer.2022.101002.

Ospanov, A., Velyamov, S., Makeeva, R., Tlevlessova, D., & Tastanova, R. (2022). Development of probiotic yogurt from small cattle milk. *Eastern-European Journal of Enterprise Technologies*, 4(11 (118)), 53-59. http://dx.doi.org/10.15587/1729-4061.2022.261574.

Prestes, A. A., Verruck, S., Vargas, M. O., Canella, M. H. M., Silva, C. C., Barros, E. L. S., Dantas, A., Oliveira, L. V. A., Maran, B. M., Matos, M., Helm, C. V., & Prudencio, E. S. (2021). Influence of guabiroba pulp (campomanesia xanthocarpa o. berg) added to fermented milk on probiotic survival under in vitro simulated gastrointestinal conditions. *Food Research International*, 141, 110135. http://dx.doi.org/10.1016/j.foodres.2021.110135. PMid:33642002.

Rakotonirina, A., Galperine, T., & Allémann, E. (2022). Fecal microbiota transplantation: a review on current formulations in Clostridioides difficile infection and future outlooks. *Expert Opinion on Biological Therapy*, 22(7), 929-944. http://dx.doi.org/10.1080/14712598.2022.2095901. PMid:35763604.

Raza, H., Ameer, K., Zaaboul, F., Shoaib, M., Zhao, C.-C., Ali, B., Shahzad, A. T., Abid, M., Ren, X., & Zhang, L. (2022). Physicochemical, rheological, & sensory characteristics of yogurt fortified with ball-milled roasted chickpea powder (Cicer arietinum L.). *Food Science and Technology*, 42, e61020. http://dx.doi.org/10.1590/fst.61020.

Sheth, U., Sankaranarayanan, A., & Srinivasan, R. (2022). Recent advancements in the production of probiotic fermented beverages. In J. Singh & A. Vyas (Eds.), *Advances in dairy microbial products* (pp. 247-270). Duxford: Woodhead Publishing. http://dx.doi.org/10.1016/B978-0-323-85793-2.00009-6.

Silva, M. N., Tagliapietra, B. L., Flores, V., & Richards, N. S. P. S. (2021). In vitro test to evaluate survival in the gastrointestinal tract of commercial probiotics. *Current Research in Food Science*, 4, 320-325. http://dx.doi.org/10.1016/j.crfs.2021.04.006. PMid:34095855.

Verruck, S., Barretta, C., Miotto, M., Canella, M. H. M., Liz, G. R., Maran, B. M., Garcia, S. G., Silveira, S. M., Vieira, C. R. W., Cruz, A. G., & Prudencio, E. S. (2020). Evaluation of the interaction between microencapsulated Bifidobacterium BB-12 added in goat's milk Frozen Yogurt and Escherichia coli in the large intestine. *Food Research International*, 127, 108690. http://dx.doi.org/10.1016/j.foodres.2019.108690. PMid:31882106.

Vianna, F. S., Canto, A. C. V. C. S., Costa-Lima, B. R. C., Salim, A. P. A. A., Costa, M. P., Balthazar, C. F., Oliveira, B. R., Rachid, R. P., Franco, R. M., Conte-Junior, C. A., & Silva, A. C. O. (2017). Development of new probiotic yoghurt with a mixture of cow and sheep milk: effects on physicochemical, textural and sensory analysis. *Small Ruminant Research*, 149, 154-162. http://dx.doi.org/10.1016/j.smallrumres.2017.02.013.

Wang, X., Gao, S., Yun, S., Zhang, M., Peng, L., Li, Y., & Zhou, Y. (2022). Microencapsulating alginate-based polymers for probiotics delivery systems and their application. *Pharmaceuticals*, 15(5), 644. http://dx.doi.org/10.3390/ph15050644. PMid:35631470.

Yetiman, A. E., & Ortakci, F. (2023). Genomic, probiotic, and metabolic potentials of Liquorilactobacillus nagelii AGA58, a novel bacteriocinogenic motile strain isolated from lactic acid-fermented shalgam. *Journal of Bioscience and Bioengineering*, 135(1), 34-43. http://dx.doi.org/10.1016/j.jbiosc.2022.10.008. PMid:36384719.