# Восточно-Европейский ЖУРНАЛ передовых технологий



### Східно-Європейський ЖУРНАЛ передових технологій

• Technology and equipment of food production

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TECHNOLOGY AND EQUIPMENT OF FOOD PRODUCTIONS

Розроблено технологію кисломолочних десертів з використанням консорціумів штамів лактобактерій — Lb. Acidophilus, S. Thermophilus і біфідобактерій — В. Віfіdum, В. Longum, В. Adolescentis, стійких до дії інгібіторів — шлункового соку, жовчі, фенолу, хлориду натрію, антибіотиків та молочної кислоти.

Для покращення розвитку біфідобактерій, використано біостимулятори. Кількість життездатних клітин біфідобактерій протягом 6 годин ферментації в присутності фруктози зростає з  $1\cdot10^4$  KYO/cm³ до  $8.8\cdot10^9$  KYO/cm³, лактулози — до  $9.9\cdot10^8$  KYO/cm³.

Для отримання стійкої до розшарування структури з глянцевою поверхнею, використали модифікований крохмаль. Динамічна в'язкість дослідних зразків дорівнює  $25 \cdot 10^{-3}$  Па $\cdot$ с, кількість життєздатних клітин біфідобактерій —  $2,5 \cdot 10^{10}$  KVO/см<sup>3</sup>.

Пастеризація при температурі (90±2) °С з витримкою 2 хв гарантує безпечність молочної суміші.

Розроблено рецептуру і технологію десертних ферментованих продуктів з плодово-ягідним збагачувачем. Отримані згустки синбіотичного продукту щільні, консистенція однорідна, ніжна, драглеподібна, в міру в'язка. Смак чистий, приємний, з кольором, присмаком і запахом плодово-ягідного наповнювача.

Після 10 діб зберігання кількість життєздатних клітин біфідобактерій становить 1,5·10<sup>10</sup> КУО/см<sup>3</sup>, після 15–9,5·10<sup>9</sup> КУО/см<sup>3</sup>, що значно перевищує встановлений стандартом необхідний рівень біфідобактерій в кисломолочних продуктах. Оптимальним терміном зберігання десертних продуктів при температурі (3±1) °С без зміни реологічних властивостей є 15 діб.

Використання кисломолочних десертів з біфідогенними властивостями розширює асортимент продуктів, здатних нормалізувати дисбаланс кишкового мікробіоцинозу в організмі людини і стимулювати власну мікрофлору кишечника

Ключові слова: пребіотики, пробіотики, біфідобактерії, лактобактерії, плодово-ягідні збагачувачі, загущувачі, ферментовані кисломолочні десерти

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# SUBSTANTIATION OF THE TECHNOLOGY FOR FERMENTED SOURMILK DESSERTS WITH BIFIDOGENIC PROPERTIES

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### 1. Introduction

While consuming products of milk processing, humans receive at least a third of all nutrients necessary for their normal life. Over the past years there has been a constant increase in the dynamics of consumption of fermented sourmilk products. Their popularity is due to the good taste and medicinal properties, specific consistency, variety of compositions that cater to the requirements of a wide range of consumers of all age groups.

Bifidobacteria that dominate the intestinal microflora of children and adults are a specific factor in protecting the body from the disruption of microbiocynosis of the intestines, the cause of which may be a disease of the digestive system, receival of chemical preparations, antibiotics, etc. Bifidobacteria through their life activities regulate certain quantitative and qualitative composition of normal microflora of the intestine, inhibiting the development of pathogenic and conditionally pathogenic microflora, which is an important factor in protecting the body against various intestinal infections. In this regard, of special importance is the issue of supporting the balance of microbiocinosis in the human gastrointestinal tract, maintaining the qualitative and quantitative composition of intestinal microflora by consuming fermented sour-milk products with bifidogenic properties.

Lactic- and especially bifidobacteria contribute to the processes of enzymatic digestion of food, stimulate intestinal peristalsis and digestion of nutrients. They also take part in the synthesis and absorption of vitamins B, vitamin K, folic and nicotinic acids, better assimilation of vitamin D, calcium salts, increase the immune status of humans. That explains why sour-milk products with bifidogenic properties acquire special significance as a factor in the prevention and treatment of various gastrointestinal diseases.

The most effective way to normalize the imbalance of intestinal microbiocenosis is to use the synbiotics, that is

the complex of probiotics and prebiotics, and the stimulation of own human intestine microflora. A promising direction in the development of dairy industry is the enrichment of products with lactic- and bifidobacteria, as well as the use of biologically valuable products from plant raw materials processing.

Considering that 70 % of people in developed countries experience dysbacterial changes, the issue on creating, maintaining, and restoring the normal intestinal microflora in the organism, should be considered as one of the most pressing ones for human health protection.

That explains the relevance of the work that addresses the expansion of the range of sour-milk products with bifidogenic properties whose consumption normalizes the intestinal microflora of humans, stimulates nutrient absorption, normalizes metabolism, and prolongs life expectancy.

### 2. Literature review and problem statement

The priority in the development of civilized society is human health. The number of people who prefer healthy food products has been steadily on the rise [1]. The normal functioning of human's basic life systems is affected by the entire range of adverse factors. On the one hand, it is the widespread use of pesticides, different food additives, preservatives, colorants, irrational nutrition by most people in the world, on the other hand, a massive uncontrolled application of chemotherapy preparations, including antibiotics. These factors are considered to cause an increase in the frequency of dysbacteriosis and a growth of gastroenterological diseases in people of different age groups.

At present, the products that are created using lactic acid bacteria and bifidobacteria are considered to be the base of healthy eating for humans, which contributes to the prevention of diseases. The positive effect is achieved both through the introduction of live lactobacilli cells directly into the human body and by using these micro-organisms in the composition of ferments in order to obtain sour-milk food products with health-promoting properties. Revitalizing effect is largely predetermined by the biologically valuable properties in the specially selected cultures of lactic acid bacteria and bifidobacteria.

Scientific approaches to the improvement of human body, its active life activity, which are based on the massive use of fermented sour-milk products with probiotic properties, is a new promising area in medicine and nutritiology. The food must provide humans with nutrients and energy, as well as have preventive and therapeutic properties. It is the probiotic products that are most effective for restoring the balance of normal microflora of gastrointestinal tract in case of metabolic disorders after hormonal, antibiotic therapy, chronic diseases, etc.

Fermented sour-milk products are the major suppliers of probiotic microorganisms into the human body. Lactic acid and bifidobacteria belong to the classic probiotics, which are widely used as biologically active components in the manufacture of food products and pharmaceuticals.

At present, special attention is paid to bifidobacteria, using which prevents the development of conditionally pathogenic and pathogenic microorganisms, improving the body's resistance to infectious diseases [2].

There is an increase in the range of products that contain the lactic- and bifidobacteria that are able to normalize work of the gastrointestinal tract of humans. Special attention is given to the development of new technologies to produce functional foods. The use of probiotics and prebiotics is the new promising field in dairy industry, which makes it possible to solve the task on maintaining health and lengthening the duration of human life [3].

Combined use of pro- and prebiotics opens wide opportunities to improve food and biological value of sour-milk fermented products by enriching them with vitamins, mineral and polyphenolic substances, dietary fibers, etc.

A search is underway for the best strains of lactic acid bacteria for industrial use and in order to receive high-quality and safe food products [4]. The selection of strains of lactobacilli is predetermined by the need to obtain compositions with the required acid-forming capability and resistance to adverse conditions of growth and development. Studies are commonly conducted that address the selection of the combined fermented compositions considering the acid-forming properties of lactobacilli, which underlie the technology for fermented sour-milk products with bifidogenic properties [5].

When making probiotic sour-milk products, there are problems related to obtaining and using the ferments whose composition includes bifidobacteria. The low rate of growth, instability at low acidity of environment, contact with the ambient air oxygen prevent the development of bifidobacteria in the production of sour milk products with bifidogenic properties. The use of compositions of lactobacilli with certain properties creates the necessary conditions for the growth and development of bifidobacteria.

Scientists developed the molecular-biological methods for the selection of bacterial cultures that are used for the identification of bacterial strains belonging to the genera *Lactococcus*, *Streptococcus*, *Lactobacillus* and *Enterococcus*. The *S. thermophilus* strains were investigated for the technological and probiotic properties; the most promising cultures were determined for use as ferments in the manufacture of sour-milk products for general and functional purposes [6].

We believe that using modern approaches in order to identify the technological and probiotic properties of the industrially important strains of streptococci, lactococci and bifidobacteria would make it possible to scientifically substantiate the application of bacteria in various biotechnological processes and to select the starting lactic acid bacteria for a variety of fermented sour-milk products.

The global trend in the development of a range of products for healthy nutrition is associated with a decrease in caloric intake, an increase in biological value, the enrichment with functional ingredients that are able to maintain and improve the health of consumers [7].

Therefore, special attention must be given to the production of fermented sour-milk products using the prebiotics – natural substances that stimulate the growth and development of bifidogenic protective microflora, stabilizers, and plant-based fillers. Vegetable or fruit-berry fillers enrich sour-milk products with biologically- and physiologically valuable substances, render them various tasty properties.

Scientists constantly expand components composition and examine conditions for obtaining symbiotic sour-milk products using the consortia of probiotic bacteria and prebiotics of various origin [8].

It should be noted that the use of bifidobacteria in the composition of sour-milk products requires the selection of strains that can flourish under the adverse production and gastrointestinal tract conditions. It is necessary to create new consortiums of microorganisms whose composition would make it possible to maximally implement the physiological, biochemical, and technological potential of applied microorganisms, to enhance the structural-mechanical properties.

To activate the growth of probiotic microorganisms and to simultaneously enrich foods with vitamins, macroand micronutrients, antioxidants, it was proposed to use bio-correctors based on plant raw materials. Bio-protectors stimulate the rate of fermentation, intensify acid formation, growth and development of certain carefully selected consortia of lactic- and bifidobacteria [9].

The created compositions of biologically-active substances promote the optimal development of the selected consortia of probiotics; changing the composition or ratio of the selected probiotics in consortia would lead to a change in activity, the rate of fermentation, the intensity of acid formation.

Designing food for health purposes aimed at people of different age and ethnicity must be addressed based on medical and biological requirements. The age and the condition of health change the needs of the human organism in food, nutrient and energy values [10].

There is an observed extension in the product range of fermented milk drinks with bifidogenic properties for functional purposes, enriched with a wide range of products from the processed dairy and fruit-berry raw materials. Lactulose syrup and sea buckthorn juice are used as bifidogenic stimulants that not only satisfy the physiological needs of the human organism in nutrient requirements, food fibers, and energy, but also perform therapeutic and preventive functions [11].

The data above indicate that the selection of strains of lactic- and bifidobacteria for use in the manufacture of sour milk fermented products and for ensuring optimal conditions for development makes it possible to obtain products with a high concentration of active cells of probiotics.

To obtain fruit and vegetable drinks and desserts, technologists use products made from the processed milk, lactic acid and bifidobacteria concentrates, juices and purees made from fruit and vegetable raw materials, pectolytic enzyme [12].

The proposed technology for fruit and vegetable drinks and desserts contains a large quantity of formulation components that cannot ensure stable conditions for the use of a ferment preparation and its optimal action. It would be more appropriate to use natural thickeners in the form of pectin or alginate, which could simultaneously improve the physiological value of manufactured products.

The structure of nutrition at present does not meet modern principles of rational nutrition. The task to organize and ensure proper nutrition to people, its adequacy and balance, is the most important issue for the joint activities of doctors, technologists, and food-making facilities.

Given this, there is an issue about the techniques to design the composition of the microflora of sour-milk products in order to restore the optimal microflora and improve the state of gastrointestinal tract of humans and to extend the range of fermented milk products with health-promoting properties. The food products to be designed should have antagonistic properties against the competitive, pathogenic, and conditionally pathogenic microflora, resistance to antibiotics, absorb a wide range of nutrients that are formed during food digestion in the human body, have a high rate of growth of probiotic and fermenting cultures. All that would make it possible to provide the required performance of cells of the probiotic strains

in the finished product. When creating probiotic products, it is necessary to select the strains tested for symbioticism so that the probiotic cultures complement each another in terms of biological activity, thereby revealing the synergy in the product. It is advisable to enrich sour-milk products with different strains of bifidobacteria and improve biological value by using prebiotics of various origin.

### 3. The aim and objectives of the study

The aim of this study is to develop a technology for sour-milk dessert fermented products enriched with bifidobacteria, biologically active and physiologically valuable substances of plant origin. This would make it possible to enrich the synbiotic sour-milk products with soluble and non-soluble polysaccharides, polyphenols, vitamins, mineral substances, to stimulate the development of bifidobacteria, etc.

To accomplish the aim, the following tasks have been set:

- to substantiate the formulation of fermentation compositions and to determine the impact of bio-stimulators on the growth and development of bifidobacteria;
- to define changes in the physical and chemical properties of the product in the process of bio-fermentation in the presence of structure-forming agents;
- to devise a technology for dessert fermented products with bifidogenic properties.

## 4. Materials and methods to study sour-milk fermented desserts

Weused the strains of lactobacillus *S. Thermophilus CT-14*, *Lactobacillus acidophilus*, and the most common strains of bifidobacteria that are inherent in the human body, *Bifidobacterium bifidum 791*, *Bifidobacterium longum subsp. longum B 379 M*, *Bifidobacterium adolescentis B-1*.

### 5. Results of studying sour-milk fermented desserts

# 5. 1. Substantiation of the formulation of fermentation compositions and determining the influence of bio-stimulators on the growth and development of bifidobacteria

Complex fermentation compositions based on the consortia of probiotic bacteria from various taxonomic groups are more resistant to adverse conditions and have higher activity compared with fermentation compositions made by using pure monocultures. Criteria for the selection of strains of lactic- and bifidobacteria for the fermentation compositions is the biological activity and technological properties that would make it possible to receive dessert sour-milk fermented products with specific organoleptic, physical-chemical, and rheological properties [13].

We performed the screening of lactic acid bacteria, which were assessed for their capability to ferment lactose, for the level of acid formation and for their proteolytic activity. The nutrient medium used was skimmed milk, sterilized at a temperature of  $(121\pm2)$  °C aged for  $(15\pm5)$  min. (Table 1).

Among the investigated strains of lactobacilli, the high level of milk lactose fermentation is observed when using the cultures *Lactobacillus acidophilus*, *L. delbrueckii ssp. bulgaricus*, *S. Thermophilus*, among which the highest  $\beta$ - galactosidase activity was demonstrated by the strain *S. ther*-

mophilus CT-14. The action of the enzyme  $\beta$ -galactosidase leads to the formation of bifidogenic products of lactose decomposition, which stimulate the development of bifidobacteria and increase activity.

Table 1
Characteristics of the examined strains
of lactobacilli (n=3, P=0.95)

Lactobacillus species	Number of strains	Quantity of lactose used, %	Level of acid accu- mulation, °T	Number of viable cells in a clot, Lg CFU/cm <sup>3</sup>
Lactococcus lactis ssp. lactis	3	17.2±4.7	157.6±2.1	8.9±0.2
Lactococcus lactis ssp. cremoris	3	15.1±6.5	100.8±4.4	8.5±0.2
Lactobacillus casei	3	9.4±6.3	145.7±1.3	8.6±0.2
Lactobacillus plantarum	3	5.9±2.6	127.2±3.2	8.1±0.2
S. thermophilus	3	48.0±5.3	99.8±1.4	8.3±0.2
Lactobacillus acidophilus	3	45.3±6.9	291.9±3.3	8.6±0.2
L. delbrueckii ssp. bulgaricus	3	40.5±7.1	305.0±5.1	8.4±0.2

The active growth and development of lactic acid bacteria requires peptides and amino acids. In terms of the proteolytic activity and the level of accumulation of free amino acids, the most productive were the lactobacilli *L. delbrueckii ssp. bulgaricus* and *Lactobacillus acidophilus*. Protein in milk is hydrolyzed by bacterial proteinase to oligosaccharides, which, under the influence of intracellular peptidase, are hydrolyzed to short-chain peptides and amino acids [14].

The best acid-forming capacity in terms of the accumulation of lactic acid was demonstrated by the milk acid bacteria *L. delbrueckii ssp. bulgaricus* and *Lactobacillus acidophilus*, which produce mostly L(+) – lactic acid, physiologically favorable for the human body. The acidophilic sticks *Lactobacillus acidophilus* are able to produce the antibiotics acidophilus and lactocidine that suppress the development of harmful non-native microflora in the nutrient environment [15].

To create synbiotic systems with functional purpose, we used three strains of bifidobacteria — *Bifidobacterium adolescentis B-1*. *Bifidobacterium bifidum 791*, *Bifidobacterium longum subsp. longum B 379 M*.

Significant impact on the viability of the lactic acid bacteria that come with milk fermented products to the human body is exerted by the digestive system. Therefore, lactic and bifidobacteria were assessed for resistance to inhibitors of development – gastric juice, bile, phenol, sodium chloride, antibiotics, and lactic acid. Duration of growth for the cells of lactobacilli was limited by a concentration of  $1\cdot10^{10}$  CFU/cm<sup>3</sup>.

It was established that all examined strains of lactic and bifidobacteria are resistant to the acidic environment,  $40\,\%$  bile,  $0.3\,\%$  solution of phenol,  $4.0\,\%$  of kitchen salt, penicillin and streptomycin, phage sensitivity of lactobacilli is at the level of  $1.33\,\%$ . All the studied strains of lactic acid bacteria are able to grow in milk, have a high activity to the fermentation of lactose and to the milk protein proteolysis.

We studied the specified strains of bifidobacteria for the technological properties based on such indicators as the activity of milk fermentation, energy of acid formation, active acidity (pH) after fermentation, the number of viable cells in a clot. We applied sterilized skimmed milk with acidity 18 °T, the content of solids is 9.0 %, of the skimmed milk residue – 8.95 %, according to GOST 10163-76. The milk was heated to 40 °C, purified, heated to 65 °C, homogenized at pressure P=15 MPa. The milk, sterilized at  $(121\pm2)$  °C and aged for  $(15\pm5)$  min, was cooled to a temperature of  $(37\pm1)$  °C. The prepared milk was introduced with a ferment of pure cultures of bifidobacteria in the amount of 5.0 %, which contained 1·10<sup>4</sup> CFU/cm³, and then we performed the fermentation at a temperature of  $(37\pm1)$  °C.

Given that the combined use of bifidobacteria strains could give rise to synergies and thus improve the technological properties, we studied the effect of the consortium of the selected strains of bifidobacteria (at ratio 1:1:1) where the content of each strain of bifidobacteria was 1·10<sup>4</sup> CFU/cm<sup>3</sup> (Table 2).

Table 2
Technological properties of the examined strains of bifidobacteria (*n*=3, *P*=0.95)

Bifidobacteria species	Fermentation activity, hours	Active acidity, pH	Acid formation energy during fermentation, °T	Number of viable cells in a clot, Lg CFU/cm <sup>3</sup>
B. bifidum	49±3	4.8±0.2	63±4	8.1±0.2
B. longum	48±5	4.8±0.2	61±2	7.9±0.2
B. adolescentis	49±4	4.7±0.2	64±3	7.8±0.2
Consortium	32±2	4.7±0.1	66±3	8.9±0.1

All the strains of bifidobacteria that we used, as well as the consortium, are very slow at milk fermentation, and form loose clots with separated whey. The obtained clots have low indicators for titrated acidity and pH. This is because at fermentation of lactose by bifidobacteria, together with lactic acid, there builds up to  $30\,\%$  of acetic acid, which has a much higher degree of dissociation, which leads to a decrease in the active acidity of milk. Hydrogen ions, which are formed as a result of the dissociation of lactic and acetic acids, inhibit dissociation and reduce the negative charge of casein micelles. Under the action of lactic and acetic acids, calcium phosphate and organic calcium are cleaved from the caseinate-calcium-phosphate complex, which leads to the destabilization of casein micelles. Insoluble calcium phosphate transforms into soluble calcium lactate. Thus, the data obtained testify to that bifidobacteria are able to grow in the presence of lactose, to accumulate biomass, and to lower the active acidity of milk.

We investigated resistance of the applied consortium of bifidobacteria to adverse conditions of acidity of the stomach and to the conditions for storing finished products. The viability of cells in the bifidobacteria consortium was determined in the presence of HCl over 5.0 hours, and in the presence of lactic acid – over 24 hours. We used as control the sterilized soured milk without a consortium of bifidobacteria.

It was established that compared to control, the number of viable cells of bifidobacteria gradually decreases. During 5-hour storage (pH 3.0), the loss of viable cells of bifidobacteria is  $5.2\,\%$ , at pH  $2.0-9.8\,\%$ . The number of viable cells in the bifidobacteria consortium after 6 hours of storage in the presence of lactic acid begins to gradually decrease. After 24 hours, the loss is, at pH 4.0, 3.4 %, at pH 3.0, 6.2 %. The data obtained indicate that the created consortium of the applied strains of bifidobacteria is effective, and can be used in the manufacture of fermented dessert products.

Therefore, creation of consortia from individual strains of bifidobacteria makes it possible to significantly improve the technological properties of bifidobacteria. When using a consortium of bifidobacteria, the time of clot formation is reduced to 28–32 hours. The number of viable cells increases on average by 3–4 times, indicating a lack of mutual suppression by the used strains of bifidobacteria in the consortium. In this case, the organoleptic indicators for the derived acid milk clots do not change.

It should be noted that the pure cultures of bifidobacteria require anaerobic conditions and, even within the consortium, are weak at acid formation. Thus, the bifidostimulating factors are required, as well as the micro-organisms that are able, in the process of vital activity, to enrich the nutrient environment with nitrogenous and other nutrients available to them.

To implement a clinical effect on the human body, bi-fidofactors should ensure the development of bifidobacteria in the intestines at a level not lower than  $1\cdot 10^6$  CFU/cm³. Therefore, the data obtained testify to the retention of activity of bifidobacteria during the passage through the gastro-intestinal tract and to their ability to get acclimatized in the intestines when consuming sour-milk fermented products.

We used as the stimulant of growth and development of bifidobacteria the prebiotics – fructose and lactulose. The stabilizer and structure-forming agent used are pectin and modified starch. We determined the stimulating action of bifidofactors on the process of milk fermentation based on skimmed sterilized milk.

The prepared milk was introduced with 5.0 % of ferment in the form of a consortium of bifidobacteria at concentration  $1\cdot10^4\,\text{CFU/cm}^3$ . Control was the sterilized skim milk, fermented by the consortium of bifidobacteria in the same amount, without bifidostimulants.

The milk was added with fructose in the amount from 0.1 to 0.5 % in line with TU 9111-011-359-37677-02. The obtained mixture was heated to a temperature of 40 °C, purified, heated to a temperature of 65 °C and homogenized at pressure  $P=(15\pm2)$  MPa. Sterilization of the prepared mixture was conducted at  $(121\pm2)$  °C, aged over  $(15\pm5)$  min, the mixture was then cooled to a temperature of fermentation,  $(37\pm1)$  °C, and the ferment was added to it. Fermentation was carried out to pH 4.6–4.7, that is unto the formation of a clot. The dependence of number of viable cells of bifidobacteria on mass fraction of the introduced fructose is shown in Fig. 1.

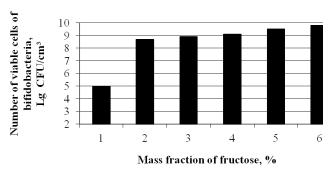


Fig. 1. Dependence of number of viable cells of bifidobacteria in sour-milk clots in the presence of fructose: 1 - control; 2 - 0.1%; 3 - 0.2%; 4 - 0.3%; 5 - 0.4%; 6 - 0.5%

The data obtained suggest that at milk fermentation in the presence of 0.5 % fructose the number of viable cells of bifidobacteria increases to 8.8·10<sup>9</sup> CFU/cm<sup>3</sup>. Fructose is included in the process of lactic acid fermentation as fruc-

tose-6-phosphate and promotes a more rapid accumulation of biomass of bifidobacteria.

Lactulose enters the colon in the unchanged form, where it stimulates the growth and development of native bifidoflora of the intestine, but it is not used in this case as a substrate for the development of pathogenic organisms [16].

The syrup "Laktusan", whose application is allowed in the food industry of Ukraine by the Ministry of Health (P No. 011717/02), was introduced to sterilized skim milk in an amount that corresponds to an increase in the concentration of lactulose in milk from 0.1 to 0.6%. We added the ferment to the prepared mixture at concentration 1·10<sup>4</sup> CFU/cm<sup>3</sup>. The dependence of number of viable cells of bifidobacteria on mass fraction of lactulose in sour-milk clots is shown in Fig. 2.

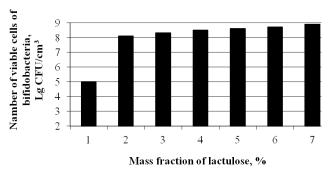


Fig. 2. Dependence of number of viable cells of bifidobacteria in sour-milk clots on mass fraction of lactulose: 1- control; 2-0.1 %; 3-0.2 %; 4-0.3 %; 5-0.4 %; 6-0.5 %; 7-0.6 %

The specified experimental data indicate that the probiotic effect would be achieved at introducing 0.1 % of lactulose. The number of viable cells of bifidobacteria over 6 hours of fermentation increased from  $1\cdot10^4\,\text{CFU/cm}^3$  to  $1.2\cdot10^8\,\text{CFU/cm}^3$ . In the presence of 0.6 % lactulose, it is increased to  $9.9\cdot10^8\,\text{CFU/cm}^3$ .

It is known that along with the probiotic effect on the microflora of the intestine, the prebiotic lactulose has a specific impact on the functioning of the liver and nervous system.

Specialists found that in order to achieve the clinical effect the content of lactulose in sour-milk products must be not less than 0.6 % [17].

To determine the rational technological process parameters of fermentation, we investigated the process of fermenting the skim sterilized milk using a consortium of bifidobacteria in the joint presence of the applied bifidostimulants - fructose and lactulose. The sterilized skim milk was introduced with the preliminary prepared bifidostimulants at a temperature of (55±2) °C. Further processing of the received mixture was performed in the sequence and under the technological modes described above. The prepared mixture was added with 5.0 % of ferment in the form of a consortium of bifidobacteria at concentration 1·10<sup>4</sup> CFU/cm<sup>3</sup>. Control was the sterilized skim milk fermented by a consortium of bifidobacteria in the same quantity, without growth stimulants. The process of fermentation was carried out until the formation of clots (pH 4.6...4.7). In the process of fermentation, we determined a change in the active acidity, titrated acidity, as well as the density of the received clots.

It was established that the duration of the fermentation process to pH 4.6...4.7 and the formation of clots is 6 hours. During this time, active acidity in the presence of the bifidostimulant fructose reaches the level of pH 4.64, lactulose –

pH 4.6, without bifidostimulants (control) – pH 4.7, while the titrated acidity reaches, respectively, 68, 72, and 52 %.

In the samples with bifidostimulants, we observed a lower active acidity and a much higher titrated acidity compared to control.

The increased activity of bifidobacteria can be explained by the presence of bifidostimulants. In the process of fermentation, along with lactic acid, there forms the acetic acid, which is a stronger electrolyte in comparison with lactic acid.

The viscosity of the samples, obtained when using bifidostimulants, remains almost unchanged over the first two hours of the process of fermentation. An increase in viscosity takes place especially fast at the end of the process of fermentation. Over six hours of the process of fermentation by the adapted cultures, the mean value for viscosity of the samples that used fructose reaches  $48\,\mathrm{s}$ , lactulose –  $46\,\mathrm{s}$ , while the viscosity of control sample is  $41\,\mathrm{s}$ .

Thus, the results obtained suggest that the application of bifidostimulants – fructose and lactulose – not only increases the number of viable cells of bifidobacteria, but also greatly increases viscosity of the received clots, which favorably affects the organoleptic properties of the finished product. Therefore, the resulting composition of bifidobacteria with the growth activity stimulants could be used for the creation of synbiotics – a combination of pro- and prebiotics, intended for the manufacture of health promoting products.

Quality of the structure of sour-milk products and its stability depends on the content of dry substances (DS) in milk and is directly connected to its density. Enhancing the content of dry fat-free milk residue (DFMR) in milk helps increase the number of contacts between parts of casein at coagulation per unit volume of the dispersion medium. This increases the intensity of interaction, increases the viscosity of the product, and improves its consistency.

The content of DS in the starting milk was adjusted by adding dry skimmed milk (DSM) with a content of dry substances of 96.0 %, dry fat-free milk residue -95.0 %. An increase in the concentration of DFMR in the nutrient environment increases the number of sulfur-containing amino acids that stimulate the growth and development of bifidobacteria and increase titration. At the same time, there is an increase in the content of caseinate-calcium-phosphate complex (CCPC) in a milk mixture, which forms a buffer system that restrains the growth in acidity with an increase in the biomass.

To obtain a clot of high quality with the required rheological properties, we performed a study to determine the rational DFMR content in milk. We added DSM to sterilized milk with a DS content of 9.0 %, DFMR -8.95 %, protein -3.0 %. The amount of DSM made it possible to increase the content of DFMR by 10, 20, 30, 40, 50 %, and amounted, respectively, to 9.8 %, 10.7 %, 11.6 %, 12.5 %, and 13.4 % DFMR. The amount of protein grew, respectively, by 3.4, 3.7, 4.1, 4.4, 4.8 %. The received mixture was heated to a temperature of 40 °C, purified, heated to 65 °C, homogenized at pressure  $P=(15\pm2)$  MPa. The mixture, sterilized at  $(121\pm2)$  °C, aged for  $(15\pm5)$  min, was cooled to  $(37\pm1)$  °C; then it was fermented by a 5.0 % ferment in the form of a consortium of bifidobacteria at concentration 1·10<sup>4</sup> CFU/cm<sup>3</sup>. Control was the sterilized skim milk fermented by a consortium of bifidobacteria in the same amount, but without adding DSM. The process of fermentation was carried out to pH 4.6.

The duration of clot formation in the control sample containing 8.95 % DFMR is on average 23...24 hours. An increase in the mass share of DFMR in milk shortens the du-

ration of clot formation to 2–3 hours; however, at a content of DFMR exceeding 15 %, the received clots fell apart with the release of whey.

Consequently, our study has shown that it is impossible to ensure the necessary rheological and organoleptic properties, characteristic of milk-based desserts, only by increasing the mass fraction of DFMR in milk. A deterioration in the quality of the dessert fermented products could be prevented using stabilizers, which would eliminate the need to further enhance the content of DFMR in milk.

The quality and properties of sour-milk clots largely depend on the ratio of bifidobacteria and lactobacilli. An increase in the number of bifidobacteria in a fermentation composition leads to a decrease in the viscosity of sour-milk clots, prolongs the process of fermentation and deteriorates the organoleptic properties. Increasing the content of lactobacilli increases the acidity and affects the growth and development of bifidobacteria.

We investigated the influence of joint use of the consortia of bifidobacteria and lactobacilli on the energy of acid formation and the number of viable cells of bifidobacteria in the received clots. Skim milk was heated to a temperature of  $40{\text -}45\,^{\circ}\text{C}$ , normalized for the content of DFMR to the level of  $12.5\,\%$  by using DSM. The use of DSM provides improved consistency of fermented milk products and hinders the process of syneresis of the formed clots. The further technological treatment of the received milk base was carried out under the technological modes described above. The fermentation involved the composition of the consortia of bifidobacteria and lactobacilli, taken in the amount of  $1{\cdot}10^4\,\text{CFU/cm}^3$  and at ratio 2:1.

Results of research into the influence of consortia of the adapted lactic and bifidobacteria, and the combination, on the energy of acid formation and the presence of viable cells of microorganisms in a clot over 6 hours of fermentation are given in Table 3.

Table 3
Technological properties of the examined compositions of microorganisms (n=3, P=0.95)

Applied micro-or-	Active acidity, pH	Energy of acid formation	Number of viable cells in a clot, Lg CFU/cm <sup>3</sup>		
ganisms		during fer- mentation, °T	bifido- bacteria	lactoba- cilli	
Consortium of lacto- bacilli (Lb. acidophi- lus + S. thermophilus) (1:1)	4.5±0.2	73±0.5	I	7.2±0.2	
Consortium of bifido- bacteria (B. bifidum + + B. longum + + B. adolescentis) (1:1:1)	4.7±0.2	66±0.3	8.9±0.2	I	
Composition of the applied consortia of bifidobacteria and lactobacilli (2:1)	4.6±0.2	69±0.5	9.5±0.3	8.0±0.2	

It was established that when using the composition of consortia of lactic and bifidobacteria the energy of acid formation, as compared with the consortium of bifidobacteria, grows. However, compared with the consortium of lactobacilli, it decreases, which is a favorable phenomenon for the growth of bifidobacteria.

An important characteristic of the strains of probiotic bacteria, which are used in the production of functional foods, is the antagonistic effect against pathogenic and conditionally pathogenic microorganisms. Under the *in vitro* conditions we investigated the antagonistic activity of the consortiums of strains of bifidobacteria and lactobacilli, as well as the composition. To determine the antagonistic activity, we used test cultures of the pathogenic and conditionally pathogenic microorganisms *E. coli, Proteus vulgaris, B. subtilis*.

An analysis of antagonistic activity of the experimental samples was carried out using the method of holes. We controlled, in the Petri dishes, the size of zones where growth of pathogenic and conditionally pathogenic test cultures was inhibited at introduction of the created consortia and composition (Table 4).

Table 4 Antagonistic activity of consortia of bifidobacteria and lactobacilli (n=3, P=0.95)

Compositions of micro-organisms	Zone of growth inhibition of test cultures, mm			
	E. coli	P. vulgaris	Bac. subtilis	
Consortium of bifidobacteria (B. bifidum + B. longum + + B. adolescentis)	22.1	18.6	31.3	
Consortium of lactobacilli ( <i>L. ac-idophilus</i> + <i>S. thermophilus</i> )	20.2	17.4	28.6	
Composition of consortia of bi- fidobacteria and lactobacilli (2:1)	26.7	23.3	35.8	

The data obtained show that the antagonistic activity of the consortium of bifidobacteria is larger than that in the consortium of lactobacilli. Combined use of microorganisms increases antagonistic activity. This suggests a possibility of using the resulting composition of bifidobacteria and lactobacilli in the manufacture of milk dessert fermented products for functional purposes.

The use of complex ferments based on the composition of probiotic microorganisms provides an opportunity to obtain a product with a large number of viable cells of bifidobacteria and significant antimicrobial activity.

The results obtained from the joint use of the selected consortia of bifidobacteria and lactobacilli are an innovative approach to creating milk-based dessert fermented products for functional purpose.

# 5. 2. Determining a change in the physical-chemical properties of a product in the process of biofermentation in the presence of structure-forming agents

To provide dessert fermented products with properties characteristic of pastes and puddings, we used hydrocolloids. Application of hydrocolloids makes it possible to receive the required structure, to ensure a certain moisture content, to prevent the lamination of bifidogenic sour-milk products when using fruit-and-berry dressings.

We performed the *in vitro* study to determine the rational concentration of apple pectin as a stabilizer that has prebiotic properties and is a nutrient environment for the growth of own normal microflora of the intestinal tract of humans. In addition, pectin substances are characterized by detoxicating and radioprotection properties. It was established that the presence of pectin increases the activity of proteolytic enzymes and improves the process of digestion [18].

We investigated the influence of pectin on the development of bifidobacteria and the physical-chemical properties of the obtained sour-milk clots.

A batch of pectin in the amount from 0.1 to 0.5 % was mixed in a separate container with 0.% fructose and was added to a small amount of skim milk. At constant stirring, it was heated to  $(90\pm2)$  °C and aged for 5 min. The mixture, cooled to  $(55\pm2)$  °C, was added to milk, normalized for the content of DFMR, heated to a temperature of  $(55\pm2)$  °C and purified. The resulting mixture was heated to  $(65\pm2)$  °C, homogenized at pressure  $P=(15\pm2)$  MPs, pasteurized at  $(90\pm2)$  °C, aged over 5...10 min, and cooled to a temperature of fermentation  $(3\pm1)$  °C.

We introduced to the prepared mixture a ferment in the amount of 5 %, which contains 1·10<sup>4</sup> CFU/cm<sup>3</sup> of the composition of bifidobacteria and lactobacilli, and aged it over 24 hours.

Upon cooling to  $(4\pm2)$  °C, we determined the impact of mass share of pectin on the pH of the fermented milk base, moisture-binding capacity, and viscosity of the received structures. We used a sample without added pectin as control.

It was established that the titrated acidity increases within the first hour in the control and examined samples by 6 and 7 °T, respectively. Over the next two hours it sharply increases and reaches in the control sample 55, in the examined sample  $-60\,^\circ\text{T}$ . Starting from the fourth hour, control samples undergo an intensive growth of titrated acidity, which almost reaches the level of titrated acidity of the examined sample, in which an increase in acidity slows down due to the formation of complex structures between the protein and pectin.

After six hours of fermentation, titrated acidity of the examined sample reaches 72 °T, control – 77 °T. At further aging of the product, titrated acidity of the examined sample reaches  $(80\pm2)$  °T, control –  $(88\pm1)$  °T. Over the fifth hour, the formation of gel occurs in the control sample, but the consistency of clots is not as strong as in the presence of pectin.

Testing the water-retaining capacity of the sour-milk milk clots by using a centrifuge method has shown that in the examined samples with the addition of 0.3 % pectin the water-retaining capacity increases, compared with control, by 4.0 %. The difference in the moisture content can be explained by swelling of pectin, and the emergence of complex structures between milk protein and pectin. The process of structure formation occurs due to electrostatic forces, hydrogen and hydrophobic bonds with the formation of a 3-dimensional spatial grid that retains moisture. The effect of stabilization is achieved also by the formation of additional hydrogen bonds between biopolymers involving the non-dissociated free carboxylic groups.

It should be noted that the examined samples with pectin are better at restoring their structure after mechanical agitation than control, due to the thixotropic properties of a sourmilk product. Pectin, as a hydrocolloid, binds moisture and forms a flexible structure, but its excess content in the product could prolong the duration of cultivation by several hours.

Results of research into the influence of pectin on the process of development of bifidobacteria in the milk base, normalized for DFMR, show that pectin activates the development of bifidobacteria in the process of fermentation. The number of viable cells of bifidobacteria in sour-milk clots increases from  $1\cdot10^4\, \text{CFU/cm}^3$  to  $0.5\cdot10^8\, \text{CFU/cm}^3$ , compared to control, in which the number of bifidobacteria grows only to  $1\cdot10^7\, \text{CFU/cm}^3$ .

In the presence of pectin in the amount from 0.4 to 0.5 %, there forms a non-uniform dense consistency and the taste

of pectin is felt. Therefore, the amount of pectin, which is advisable to use, was limited to  $0.3\,\%$ .

When making sour-milk products with a gelatinous consistency and glossy surface, they use modified starch in line with DSTU 4286:2004, which belongs to the neutral polysaccharides. We performed a study to determine the rational amount of the modified starch to be added, which would make it possible to obtain the structure characteristic of such milk-based desserts as pastes and puddings.

Batches of starch in the amount from 1.0 to 5.0 g were poured over with four times the amount of skimmed milk, heated to 30 °C, stirred and aged over 1 hour for swelling. The resulting mixture, constantly stirring, was heated to (85±2) °C for the complete dissolution of starch, and cooled to a temperature of mixing with the normalized milk, (55±2) °C. We introduced to the mixture, prepared to fermentation, 5 % of the ferment, which contains 1·10<sup>4</sup> CFU/cm<sup>3</sup> of the composition of bifidobacteria and lactobacilli, and aged over 24 hours. Upon cooling to (4±2) °C, we determined water-retaining capacity, acid formation, and viscosity of the derived structures. We used, as control, the fermented milk base without starch. It was established that the best water-retaining properties are observed in the presence of 5.0 % starch. Water-retaining capacity of the fermented milk base, as compared with control, increased by almost 20 %, but the product does not demonstrate a sufficiently homogeneous consistency. Starch affects the acid-forming capacity of the examined samples. Acidity in the control samples reaches almost 88 °T, and with 5.0 % starch it does not exceed 76 °T. Starch, as a neutral hydrocolloid, does not directly affect the fermentation process, but it binds moisture and increases the viscosity that prevents the development of fermentation cultures and slows down the fermentation process.

Dynamic viscosity of the examined samples that use starch, which was determined using the rotational viscosimeter "Reotest-2", increases from  $12\cdot10^{-3}$  to  $25\cdot10^{-3}$  Pa·s. When using 5.0 % of starch, one feels its flavor, therefore, the amount of starch which was limited to 4 %.

The process of homogenization is aimed at fragmentation of casein micelles to submicelles, and milk fat to the balls with a diameter of less than 1.0  $\mu m$  [19]. We selected the optimal regime of homogenization based on an indicator for the settlement of fat fraction after homogenization at temperatures of 55–75 °C and pressure 10–15 MPa. It was established experimentally that the optimal mode of homogenization of milk base, whose efficiency reaches 85 %, is the temperature of 65 °C and a pressure of 15 MPa. Effectiveness of the homogenized milk base with stabilizers at a pasteurization temperature of 65 °C and a pressure of 15 MPa reaches 97–98 %.

Dairy enterprises employ rather strict modes of pasteurization. This is predetermined by the high bacterial contamination of raw materials and the need to receive a product that can be stored for less than 7 days. A pasteurization mode should contribute to the complete destruction of pathogens, maximally destroy the saprophytic microflora, and ensure a minimum change in the main biologically valuable components of the product.

We performed a study into the effectiveness of pasteurization of a synbiotic product and determined the quantitative and qualitative composition of the microflora after pasteurization under the modes of  $(85\pm2)$  °C,  $(90\pm2)$  °C,  $(92\pm2)$  °C. In the examined samples, we determined the

content of colony-forming QMAFAnM, BGKP, and heat-resistant microorganisms (Table 5).

Table 5
Efficiency of heat treatment depending on the mode of pasteurization (*n*=3, *P*=0.95)

Pasteurization mode	Pasteurization effectiveness, %				
Pasteurization mode	QMAFAnM	Heat-resistant	BGKP		
$t = (85\pm 2)  ^{\circ}\text{C},  \tau = 2  \text{min}.$	99.43	99.12	absent		
$t = (90\pm 2) ^{\circ}\text{C},  \tau = 2 \text{min}.$	99.98	99.96	absent		
$t=(92\pm2)$ °C, $\tau=2$ min.	99.99	99.99	absent		

An increase in the temperature of pasteurization from (85±2) °C to (90±2) °C decreases the number of colony-forming QMAFAnM by almost 100 times, heat-resistant microorganisms - by 20 times, BGKP - by 3 times. An increase in the temperature of pasteurization to (92±2) °C reduces the number of all microorganisms by two times. That is, the decisive influence of temperature on the microflora of a synbiotic product occurs at a temperature of pasteurization (90±2) °C. Application of the high-temperature pasteurization at a temperature of (90±2) °C and (92±2) °C with an aging over 2 minutes makes it possible to ensure the safety of the pasteurized milk mixture. Residual microflora under the high-temperature pasteurization of the examined samples are represented by soporific microorganisms that are, from a biochemical point of view, low-active, and do not develop at storing temperatures of (4±2) °C. The effectiveness of pasteurization of synbiotic products at a temperature of (90±2) °C and (92±2) °C, aged over 2 minutes, is almost indistinguishable, so the pasteurization was carried out at a temperature of (90±2) °C, aged over 2 minutes.

We investigated the development of bifidobacteria, as well as the structurally-mechanical properties of a milk base for dessert products, in the presence of the applied bifidostimulants and structure-forming agents. Control samples were the sour-milk clots, received by fermenting the milk, normalized for DFMR and fat, without using bifidostimulants and structure stabilizers. Prepared solutions of stabilizers were mixed at (55±2) °C prior to introducing to the milk base. We added to the milk, normalized for DFMR and fat, a mixture of the prepared bifidostimulants and stabilizers in the preliminary established rational amount. The resulting mixture was purified, heated at stirring to  $(65\pm2)$  °C, homogenized at pressure  $P=(15\pm2)$  MPa, and pasteurized at (90±2) °C over  $\tau$ =2 min. The mixture, cooled to (37±1) °C, was fermented with the adapted composition of bifidobacteria and lactobacilli in the amount of 5 % with a content of microorganisms of 1·10<sup>4</sup> CFU/cm<sup>3</sup> and aged over 6 hours. The end of the fermentation process was defined based on the indicators for titrated and active acidity. In the product, cooled to  $(4\pm2)$  °C, we determined the impact of the applied bifidostimulants and stabilizers on the development of probiotics, titrated and active acidity, and the physical-chemical properties.

The process of gel formation begins at hour three and almost ends at hour five of the process of fermentation. Duration of the lag-phase during fermentation of dessert products with a gel-like structure is 1 hour, indicating the properly defined composition and the number of the used bifidostimulants. The largest increase in titrated and decrease in the active acidity occurs from hour three through hour five of fermentation. Titrated acidity of the examined samples after six hours of

fermentation is 72 °T, control -85 °T, active acidity is, respectively, 4.7 and 4.5. The number of viable cells of bifidobacteria in sour-milk clots after  $6\,h$  of fermentation increased from

 $1.10^4$  CFU/cm<sup>3</sup> to  $2.5\cdot10^{10}$  CFU/cm<sup>3</sup>, compared to control, in which the number of bifidobacteria grows to  $2\cdot10^8$  CFU/cm<sup>3</sup>.

The viscosity of products was determined using the viscosimeter "Reotest-2" (a shear rate gradient Dr=0.3333 s<sup>-1</sup>). It was established that the process of structure formation during fermentation of a milk-based dessert product is almost over when reaching a viscosity of 1.65 ·10<sup>2</sup> Pa·s.

An important component of any product is taste fillers, which not only form the organoleptic properties, but also enrich products with biologically active ingredients vitamins, mineral substances, polyphenols, increase the body's resistance to adverse environmental conditions. Dressings are typically the fruit-berry juices or syrups that are distributed evenly throughout the entire volume of the product. When making sour-milk products for functional purposes, it is advisable to use only the juices of direct extraction from vitamins, polyphenols, minerals, etc.

The complexity of using fruit-berry dressings relates to that the application of additives in the process of fermentation can disrupt the process of fermentation of a milk base, change the color, flavor, and rheological properties of the finished product, which affects the duration of storage of the finished product. It was experimentally proven that the production of dessert fermented products is advisable to carry out using a thermostatic technique, and the fruit-berry dressings should be better introduced after fermentation during agitation.

# 5. 3. Development of a technology for dessert fermented products with bifidogenic properties

Based on the results of our study, we have developed the formulation and technology of production of dessert milk-based fermented products with a fruit-berry dressing (Fig. 3).

Stability of a product to syneresis and the level of its viscosity affect the stability of the structure of sour-milk dessert products and the duration of storage. It should be taken into consideration that fruit-berry fillers have low acidity (pH 2.9–3.6), which can lead to the compaction of the mesh of protein gel, disruption in the structure of dessert food and the occurrence of syneresis. The presence of stabilizers prevent the process of syneresis as a result of the formation of colloidal aggregates between milk proteins and

molecules of hydrocolloids. In addition, to prevent syneresis and maintain the pH of the medium, we added salt of citric acid, three-substituted, in the amount of  $0.12\,\%$ .

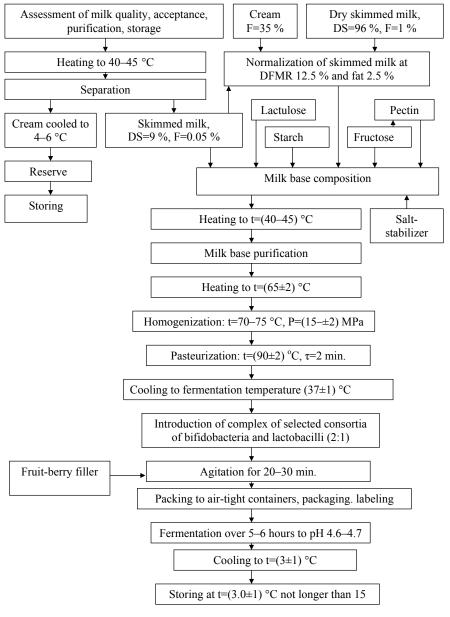


Fig. 3. Technological diagram for making dessert fermented products

The semi-finished fruit-berry juices without pulp, prior to introducing to the fermented product, were exposed to heat treatment at 70–80 °C for 20 min. and cooled to a temperature of (37±1) °C. When using juices with pulp, the semi-finished juice was pressed, homogenized, pasteurized at 80–85 °C for 20 min., cooled to a temperature of (37±1) °C, and used for the production of dessert products. Sour-milk products agree most successfully with raspberry, cherry, cranberry, strawberry, current, apricot juices.

We investigated the physical and chemical properties of the obtained sour-milk dessert products with a strawberry filler and without it (control) immediately after cooling to a temperature of storage of  $(3\pm1)$  °C (Table 7).

The number of viable cells of bifidobacteria in the samples with fruit and berry juices is higher than that in control; syneresis is absent in all samples. The process of clot forma-

tion takes 5–6 hours. The obtained clots of the symbiotic product are dense, their consistency is homogeneous, gentle, gelatinous, viscous to a degree. The taste is pure, pleasant, with the color, taste, and smell of the strawberry filler.

Table 7 Characteristic of dessert products with a fruit-berry filler (n=3, P=0.95)

	Dessert products		
Indicators	Control	Experiment	
Active acidity, units pH	4.5±0.1	4.67±0.1	
Titrated acidity, <sup>o</sup> T	77.5±0.2	75.2±0.2	
Number of viable cells of bifidobacteria, Lg CFU/cm <sup>3</sup>	9.2±2	9.8±2	
Clot formation time, hours	5.0±0.5	5.5±0.5	
Viscosity, η·10 <sup>3</sup> , Pa·s	1.89±0.2	1.93±0.2	
Syneresis, cm <sup>3</sup>	absent	absent	

To influence the state of the microflora in the intestines, the number of probiotics in the composition of sourmilk fermented products should reach a minimum level of  $1\cdot10^7~\rm CFU/cm^3$ . Technological feature of the production of dessert fermented foods is the process of structure formation, which occurs when cooling the finished product to a temperature of 4–6 °C for 6–8 hours.

Our dessert products were kept at a temperature of  $(4\pm2)$  °C for 25 days to determine the optimal shelf life for eating. We controlled the organoleptic, physical-chemical, microbiological and rheological parameters at the time of clot formation, and after 5, 10, 15, 20, and 25 days (Table 8).

Table 8 Characteristic of organoleptic parameters of fermented dessert products depending on the length of storage (n=3, P=0.95)

	Storage duration, days					
Parameters	Freshly- prepared	5	10	15	20	25
D	essert product,	milk-b	ased (c	ontrol)		
Taste and flavor	Pure, sour-m	Pure, sour-milk, without foreign odor and taste				
Color	White with light cream tint, homogeneous throughout the mass					
Consistency and physical appearance	Homogeneous, viscous, gel-like mass, without whey separation, with glossy surface				With negligible release of whey	
Dessert product, milk-based, with a fruit-berry filler						
Taste and flavor	Pure, sour-milk, moderately sweet, with aroma and taste of the filler					
Color	From pale pink to pinkish, uniform throughout the mass					
Consistency and physical appearance	mass, without whey separation, gible				With gible r of w	

It was established that over 15 days of storage an increase in acidity occurs evenly in the control and examined samples made with a fruit-berry filler. Over the next five days, the active acidity dramatically decreases to the level of pH 4.5 on average, and the titrated acidity rises to 86...89 %.

With shelf life is extended up to 25 days, active acidity of control samples is reduced. In the examined samples, an increase in acidity is slowing down, due to the reduction in

the content of free hydrogen ions as a result of water molecules binding to the hydrocolloids. Over 10 days of storage, the number of viable cells of bifidobacteria almost does not change and, in the examined samples, is  $1.5\cdot10^{10}$  CFU/cm³, in control –  $7.5\cdot10^9$  CFU/cm³. In the next 5 day, there begins a gradual die-out of cells of bifidobacteria, but the number of viable cells in the products remains at a high level, in the examined samples –  $9.5\cdot10^9$  CFU/cm³, in control –  $4.2\cdot10^9$  CFU/cm³.

It was established that the structure of control samples of desserts remains unchanged during 15 days, examined samples – 20 days, and then there starts an insignificant release of moisture. In 25 days, syneresis of the examined product is  $1.2\,{\rm cm}^3$ . Probiotic properties of the examined samples and control samples over 20 days of storage are at the level of  $6.3\cdot10^9\,{\rm CFU/cm}^3$ , control –  $0.8\cdot10^9\,{\rm CFU/cm}^3$ . The optimal shelf life of dessert products without a change in the rheological properties was limited for a period of 15 days. The developed production technology of sour-milk desserts with bifidogenic properties has passed industrial testing at the Litinsky dairy factory.

# 6. Discussions of results of research into the development of a technology for the production of fermented sour-milk desserts

In the course of this work, we investigated the strains of lactic acid bacteria that can develop in milk, have a high activity to the fermentation of lactose and milk protein proteolysis. We have determined the strains of lactic acid bacteria resistant to inhibitors of development in the gastric tract, during storage and use.

The complexity of making sour-milk products with bifidogenic properties relates to the search for strains and consortia of lactic acid bacteria. And the need to define the conditions for maximal survival and retention of activity of bifidobacteria depending on a formulation composition of sour-milk milk products and the prebiotics used.

When using the composition of fermenting cultures from the consortia of bifidobacteria ( $B.\ bifidum + B.\ longum + B.\ adolescentis$ ) and lactobacilli ( $L.\ acidophilus + S.\ thermophilus$ ) in the ratio of 2:1, the energy of acid formation in the composition increases, compared to the consortium of bifidobacteria, by 5.3 %. The use of bifidostimulants – fructose, lactulose, and inulin – stimulates the growth and development of bifidobacteria by 8.5 %, 12.2 %, and 15.8 %, respectively.

Increasing a dry fat-free milk residue (DFMR) in the milk base to  $12.5\,\%$  reduces the duration of clot formation by 2-3 hours, as well as stimulates the growth of bifidobacteria. When using a milk base with a DFMR content of  $12.5\,\%$ , growth of viable cells of bifidobacteria, in comparison with control, increases by  $19\,\%$ .

A component composition of the stabilizing system, which contains 0.3 % pectin and 4.0 % starch, contributes to an increase in the number of viable cells of bifidobacteria in dessert products from  $1\cdot10^4$  CFU/cm³ to  $1\cdot10^8$  CFU/cm³ and makes it possible to obtain a homogeneous gel-like structure with a glossy surface, characteristic of pastes and puddings.

We have substantiated the technological parameters for making dessert fermented products: homogenization – t=65 °C, P=15 MPa, pasteurization – t=(90±2) °C,  $\tau$ = =2 min, temperature and duration of storage t=(3±1) °C,

τ=15 days, which ensure obtaining dessert fermented products of high quality with the content of viable cells of bifidobacteria  $1\cdot10^9$  CFU/cm³ and viscosity (1.75± ±0.2)·10³ Pa·s. In the presence of fruit and berry fillers the products acquire a pleasant taste and coloring, enrich with vitamins, polyphenols, mineral substances, soluble and non-soluble food fibers, etc. When stored over 15 days at a temperature of (3±1) °C, the titrated acidity of desserts increases to 82 °T, and viscosity increases slightly due to the formation of new hydrogen bonds and structure compaction.

The research results obtained open the possibility of long-term storage and use of sour-milk desserts with a high activity of bifidobacteria. The work related to the search for possibilities of application of the established consortia with a variety of biologically valuable fillers will continue.

### 7. Conclusions

- 1. Complex ferments based on the consortia pf probiotic bifidobacteria and lactobacilli of various taxonomic groups are more resistant to adverse environmental factors and have a higher activity compared to ferments made using the pure monocultures. Supplements of fructose and lactulose stimulate the growth and development of bifidobacteria.
- 2. The use of pectin and starch as the stabilizing system makes it possible to increase the number of viable cells of bifidobacteria and to obtain the consistency that is characteristic of pastes and puddings.
- 3. The use of fruit-berry dressings promotes the development of bifidobacteria and lactobacilli, accelerates the formation of sour-milk clots. The optimal shelf life of fermented dessert foods at a temperature of (3 $\pm$ 1) °C should not exceed 15 days.

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