Probiotic lactic acid cultures in the production of vegetable cream spread

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Abstract. The possibility of creating a vegetable-creamy spread using probiotics and prebiotics have been investigated. The generally accepted and original research methods are used. Probiotic microorganisms suitable for biological souring of milk are analyzed. The most suitable strains of lactic acid cultures for the production of vegetable cream spreads were selected: Streptococcus salivarius subsp. thermophilus (P 20) and Lactobacillus delbrueckii subsp. bulgaricus (D). It is established that milk fermented using selected cultures has the most favourable characteristics. A new recipe and technology of a functional vegetable-creamy spread with the addition of biologically fermented milk and inulin have been developed. The organoleptic and physicochemical properties of the finished product are investigated.

1. Introduction
At the present stage of development of the oil and fat industry, there has been a tendency to produce products that not only satisfy the energy needs of the body but also provide it with the whole spectrum of essential nutrients, helping to prevent nutritionally dependent diseases and preserve health and longevity [1].

This trend has also affected the production of spreads. Spread - emulsion fat product with a total fat weight fraction of not less than 39%, having a plastic consistency, with a fat phase melting point of not more than 36 °C. Spread is made of milk fat, and/or cream and/or butter and natural and/or modified vegetable oils or only natural and/or modified vegetable oils with or without the addition of food additives and other ingredients. The spread contains not more than 2% by weight of oleic acid trans-isomers in fat isolated from the product (in terms of methyl elaidate) (GOST 34178-2017). Currently, the emphasis is being placed on the creation of products with a directionally improved fatty acid composition, with a reduced fat content, and also enriched with various functional ingredients: polyunsaturated fatty acids, macro- and microelements, vitamins, probiotics and prebiotics [2, 3, 4, 5].

Non-pathogenic microorganisms are considered probiotic, capable of restoring the healthy intestinal flora of the human intestine, and at the same time, inhibiting the growth of pathogenic and conditionally pathogenic bacteria. Following the current standard, microorganisms of the Bifidobacterium, Lactobacillus, Propionibacterium genus are considered as probiotics, as well as bacteria of the genus Lactococcus Streptococcus thermophilus species used in associations with them [5, 6].

To create favourable conditions and stimulate the growth of healthy microflora of the large intestine, substances called prebiotics are used. Examples of such substances are oligofructose,
galactooligosaccharides, dietary fibre, pectins and inulin. Inulin is a natural polysaccharide obtained from chicory root by aqueous extraction [7, 8].

2. Background
Reducing the fat phase in the spread necessitates the use of food ingredients that help maintain the natural organoleptic properties: consistency close to butter, as well as milk flavour and aroma.

Inulin, in addition to a recognized prebiotic [7, 8], when dissolved in water, forms gels with a slight texture that can simulate the high-fat content in low-fat products and, therefore, improve consumer properties, bringing them closer to the characteristics of a classic fat product. Besides, inulin favourably affects the ductility and spreadability of the spread. These parameters indicate that inulin is a universal functional ingredient for spreads, allowing you to reduce their fat content without compromising consumer qualities and give them functional properties[2].

As a rule, to receive organoleptic properties similar to butter, cream-milk group fragrances are added to the vegetable-creamy spread recipes, which can give the following flavouring shades: milk, creamy, melted, sour cream. The basis of this group of flavours is synthetic diacetyl. The introduction of biologically fermented milk into the recipe excludes synthetic flavours from the recipe since diacetyl of natural origin is found in fermented milk in an amount sufficient to give a pleasant taste and aroma.

Thus, the use of low-fat spread, inulin and fermented milk in the recipe allow giving the product functional properties and achieve the required technological effects.

This work aimed to develop and study the technology for the production of vegetable-creamy spreads using milk fermented with probiotic lactic acid cultures and inulin as a prebiotic [8, 9].

For achieving the goal, the following tasks were set:
- pick up probiotic lactic acid cultures for effective fermentation of milk [6];
- develop a recipe of vegetable-creamy spread [4];
- to study the quality indicators of the developed vegetable-creamy spread.

When choosing raw materials for the production of vegetable-creamy spreads, the required properties of the finished product were taken into account: organoleptic properties close to sour cream butter, the mass fraction of milk fat in the finished product, the fatty acid composition and the content of trans isomers, as well as the content of solid triglycerides and the melting point [1, 4].

3. Materials and methods
Theoretical studies and part of experimental studies on modelling the ingredient composition of the developed spread was carried out based on the laboratories of the department “Technology of food products from plant raw materials” Kemerovo State University.

Raw materials for the production of spreads were purchased in the Lenta distribution network. For experimental studies, lactic acid cultures purchased at the State Research Institute of Genetics and Breeding of Industrial Microorganisms at the Kurchatov Institute National Research Center were used.

In the process of work, generally accepted and special methods of researching raw materials and finished products were used.

At the initial stage of the experiment, the physicochemical properties and fatty acid composition of the raw materials used in the fat-based spread were studied.

The quality indicators of milk fermented to the consistency of yoghurt in the laboratory were also determined.

Sourdough of lactic acid cultures was prepared in sterile skim milk. Sterilization was carried out at a temperature of 121 °C for 20 minutes. Pure lactic acid cultures of *Streptococcus salivarius subsp. thermophilus* (strain P 20) and *Lactobacillus delbrueckii subsp. bulgaricus* (strain D) were added to milk cooled to 37 ° C and kept in a thermostat at the indicated temperature for 7 hours until a clot forms[5, 10].
The acidity of the fermented milk was determined by the potentiometric method: 10 g of the analyzed sample was placed in a 50 cm$^3$ beaker, 20 cm$^3$ of distilled water was added and thoroughly mixed. The core of the magnetic stirrer was placed in the beaker, and the beaker was mounted on the magnetic stirrer. The mixer motor was turned on, and the electrodes of the potentiometric analyzer were immersed in a beaker with the mixture. The contents of the beaker were titrated with sodium hydroxide solution to an equivalence point of 8.8 pH. When a pH of 8.8 was reached, alkali addition was stopped. The volume of sodium hydroxide solution consumed in the titration of the mixture was measured with a reading of the measurement result up to 0.05 cm$^3$.

**Basic research methods of the finished product**

Measurement of the acidity of the spread was carried out by a method based on the neutralization of free fatty acids, proteins, phosphate and lemon salts with an alkali solution. A weighed portion of the spread was placed into the flask, an alcohol-ether mixture, phenolphthalein was added and titrated with constant stirring.

Acidity was calculated by the formula:

\[ X = \frac{10A}{m} \]  

(1)

where 

A – amount 0.1n. potassium hydroxide solution used for titration;  
m – the sample weight of the test product.

The melting temperature of the fat extracted from the spread was determined by the following method. In essence, the test fat sample was heated in a water bath in a porcelain cup until wholly melted and filtered. A clean, dry, open at both ends capillary tube made of thin glass with an inner diameter of 1–1.2 mm (capillary length 60 mm, thickness 0.2-0.3 mm) was immersed at one end in molten fat so that its height was the capillary was equal to 10 mm. The capillary with fat was kept on ice for 10 minutes.

After that, the capillary was attached to the thermometer (the value of division is 0.1 ° C) with a thin rubber ring so that the column of fat was at the same level as the mercury ball of the thermometer. The thermometer with the capillary was lowered into a glass of water to such a depth that water did not enter the sealed end of the capillary.

With continuous stirring, the water in the beaker was first heated at a speed of approximately 2 ° C per minute, and as it approached the expected melting temperature, no more than 1 ° C per minute. The melting point was the one at which the fat in the capillary began to rise.

4. Results and Discussion

We have analyzed the group of microorganisms that are allowed for the process of fermentation of milk[17]. The characteristics of the compared lactic acid cultures are presented in the Table 1.

After a comparative analysis, Strep to coccus salivarius subs p thermophilus (strain P 20) and Lactobacillus delbrueckiisubs p.bulgaricus (strain D) were selected [5, 10]. These microorganisms have good acid formation, have the lowest risks of malformation of fermented milk, and at the same time, they have a sufficiently high cell death temperature, which positively affects the finished product, allowing pasteurization of the aqueous phase [1, 4].

Organoleptic evaluation of milk fermented using the selected starter culture is given. The samples obtained have clean, sour-milk flavour without extraneous smacks and odours, a homogeneous consistency. Selected strains of microorganisms form a dense clot that holds serum well.

In addition to biologically fermented milk, the following components are included in the water-milk phase: milk powder; salt; sugar; inulin; water; lemon acid.
Table 1. Comparative characteristics of probiotic cultures

| Indicator                          | L. bulgaricus | L. acidophilus | Str. lactis | Str. thermophilus |
|-----------------------------------|---------------|----------------|-------------|-------------------|
| Acid formation limit in milk, °C  | 350           | 300            | 300         | 110-115           |
| Growth temperature, °C            |               |                |             |                   |
| Optimal                           | 40-45         | 37-38          | 40-45       | 40-45             |
| Marginal                          | 22-53         | 20-55          | 20-45       | 50-60             |
| Growth in milk at temperature, °C |               |                |             |                   |
| 10                                | −             | −              | −           | −                 |
| 45                                | +             | +              | +           | +                 |
| 55                                | −             | −              | −           | +                 |
| Cell death temperature, °C        | 75-80         | 70-80          | 70-80       | 70-85             |

In laboratory conditions, samples of a creamy-vegetable spread with the addition of biologically fermented milk were obtained and examined for compliance with the requirements for organoleptic and physicochemical properties. Quality indicators of the finished product are shown in Table 2.

Table 2. Characteristics of the vegetable-creamy spread

| Indicator                          | Measure value                        |
|-----------------------------------|--------------------------------------|
| Taste and smell                   | Sour-milk, without extraneous smells and smells |
| Consistency and appearance at a temperature of 12 ± 2 °C | Homogeneous and plastic. |
| Product colour                    | Light yellow                         |
| Mass fraction of fat,%            | 40.50 ± 0.05                         |
| including milk fat               | 12.5 ± 0.03                          |
| Mass fraction of fat,%            | 30.2 ± 1.0                           |
| including milk fat               | 23.9 ± 0.01                          |
| Moisture and volatiles,%          | 4.2 ± 0.1                            |
| Acidity, °K                       |                                      |

5. Conclusion
Studies of the quality indicators of the developed spread allow us to conclude that the presence of milk fermented with probiotic lactic acid cultures in the aqueous phase of the spread favourably affects the texture of the spread and its organoleptic properties (taste and smell). Inulin was added as a prebiotic, which also positively affected the consistency of the product and organoleptic characteristics. In terms of physical and chemical parameters, the product complies with the requirements of the state standard GOST 34178-2017 Melted spreads and mixtures. General specifications.

The new functional product can be recommended for inclusion in the daily diet of all groups of the healthy population.

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