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NATURE OF THE

COMPOSITION

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ANALYSIS OF THE

SUBSTANCES OF SOUR-

PLANT-BASED FILLERS

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MILK DESSERT WITH

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The nature and interaction of the constituent substances that make up the sour-milk dessert with plant-based fillers have been studied by the method of IR spectroscopy. This method is used to study the diverse nature of substances. The spectral range applied was in the range of 500-4,000 s⁻¹.

It was found that the intensity of functional groups absorption in the range of $2,500-3,500 \text{ s}^{-1}$ is due to the valence vibrations of NH-, CH and S-H-groups, indicating the presence of free organic acids, aromatic substances. In addition, in the spectra of sour-milk dessert with plant-based fillers, an absorption intensity in the range of $1,470-1,335 \text{ s}^{-1}$ is observed, which indicates the presence of soluble pectin. Proteins characteristics in the samples are observed at absorption in the range of $3,300-3,500 \text{ cm}^{-1}$, which is due to the valence vibrations of the N-H bond in the -NH₂ groups.

The use of fruits in the form of a freeze-dried powder together with milk protein concentrate in the technology of sour-milk desserts helps reduce the content of free moisture, hence a stable structure.

Sour-milk dessert with plant-based fillers is a system consisting of particles of different dispersion, which will affect its physical and chemical properties. In particular, there is a slight coarsening of whey proteins and redistribution between particles in the range of 1-10 nm and 1-100 nm.

The use of plant-based fillers in the form of a freezedried powder in the technology of sour-milk desserts would not only improve its physical and chemical properties but also could make it possible to enrich the product with minerals.

The mineral composition of the sour-milk dessert is marked by the calcium content (122 mg/100 g), potassium (97 mg/100 g), phosphorus (82 mg/100 g), sodium (50 mg/100 g), and sulfur, iron

Keywords: IR-spectra, dispersion, mineral substances, sour-milk product, functional groups, physical properties

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1. Introduction

Sour-milk products are characterized by a viscous (pseudoplastic), coagulation structure. It is due to a combination of factors, such as total dry matter, protein, lipids, dispersion of fat globules and casein micelles, the degree of hydration and aggregation, species composition and fermentation culture properties, heat treatment regimes, etc. [1].

The most important processes that occur during the production of sour-milk products are casein coagulation, that is, the transition of the colloidal system of milk from the free-dispersed state (sol) to the bound-dispersed (gel). The consistency of sour-milk products, as well as the maintenance of the structure during storage, depends on their proper course [2].

In the process of microflora fermentation, proteins are partially broken down into peptones and other simple substances, lactic acid is formed from lactose, and vitamins, enzymes, aromatic compounds, and others are accumulated [2].

More than 95 % of casein is contained in the form of micelles. These are almost spherical particles with a size of 20 to

300 nm. The properties of casein micelles are determined by the properties of monomeric primary α sl-, α s2-, β - and κ -caseins. Their molar mass is 19,000–24,000 g/mol. All caseins contain phosphate residues esterified with the hydroxyl group of serine, due to the presence of phosphoserilic groups caseins are characterized by a high ability to bind calcium and magnesium. At low values of active acidity of milk, their surface has a negative charge, which leads to association [3, 4].

The main whey proteins, in contrast to case in, show a fairly wide range of molar mass (14,000–150,000 g/mol) and are more finely dispersed, the particle size is 15–30 nm [2].

Carbohydrates in dairy products are sucrose and lactose. Lactose is the main nutrient for yeast microorganisms. Under the influence of lactic acid bacteria, 25 % of lactose is broken down to form lactic acid. Lactic acid removes calcium from caseinates, forms soluble salts of calcium lactic acid and calcium phosphate.

Lipids that are part of sour-milk products are represented in the form of fat globules with a size of $1.0-2.0 \mu m$, which prevents destabilization of the fat phase and the stratification process of the milk-fat mixture [5].

Therefore, the study of the components of sour-milk dessert to form a stable structure during storage is relevant.

2. Literature review and problem statement

Sour-milk desserts have good consumer properties, high nutritional, and biological value. Their production involves a wide range of flavorings, fillers, flavors, stabilizers that regulate the processes of structure formation, prevent the deposition of filler particles, and denaturation of proteins during the heat treatment of mixtures [6].

In [7], the IR spectra of dry films of model samples of gelatin gems with the addition of these components were studied. As a result of studying the spectra, the samples were arranged in a series with increasing strength of the formed structure. It is determined that the model system containing gelatin, sodium caseinate, and sunflower seed kernel concentrate has a stronger structure. However, the influence of the dispersed composition of the model samples on the strength of the structure has not been studied.

The state of moisture in milk-protein mixtures with rice and wheat extrudates before thawing and after defrosting was studied [8]. Infrared spectroscopy has shown that grain extrusion improves the moisture-binding properties. The effect is observed when comparing the relative intensities of the water and proteolytic proton bands of sour-milk cheese, which decrease after defrosting, which are characterized by an increase in the content of strongly bound moisture. For mixtures of sour-milk cheese with wheat, the opposite pattern is realized – during defrosting, the amount of both strongly and weakly bound moisture increases significantly, which indicates the filling of the voids of the crystal lattice. However, the authors did not study the effect of extrudates on the mineral composition of milk-protein mixtures.

Litchi juice fermentation in the production of sour-milk desserts was studied. IR spectroscopy has shown that S-type bonds increase during milk fermentation. The viscosity of the sample also increased significantly. Thus, the use of litchi juice in the technology of fermented milk products can accelerate the fermentation process [9]. However, the authors did not study the effect of litchi juice on the mineral composition of a sour-milk dessert.

In [10], the methods of accelerating the process of cheese ripening and the formation of aromatic compounds are considered. Infrared technologies were used to assess the biochemical transformations that occur during the maturation process. These methods are proposed to be used to monitor the maturation process or predict various signs of cheese quality. But the effect of the cheese ripening process on its dispersed composition is not considered.

It has been proven in [11] that infrared spectroscopy of milk can be used to predict the detailed composition of milk fat. Besides, polymorphisms have been identified that have a significant effect on the composition of milk fat. However, the dispersed composition of milk components has not been studied.

Studies of the particle size distribution of yogurt samples with Jerusalem artichoke powder have shown that the introduction of Jerusalem artichoke powder in the amount of 1 to 5 % is optimal. In the samples, there is an active increase in the particle size of Jerusalem artichoke powder in yogurt, there is a process of gradual swelling of the powder [12]. However, the authors did not study the mineral composition of yogurt with the addition of Jerusalem artichokes. The effect of sea buckthorn powder on the properties of sour cream, which is the basis of whipped sweet dishes, has been studied [13]. Studies of the microstructure of the sample showed that there are uniformly distributed small balloons and fat in the microstructure. Fine plant-based powder strengthens the microstructure of sour cream, as it promotes its thickening, associated with the formation of complexes of polysaccharides (pectins, hemicelluloses, pentosans) with casein micelles of sour cream and uniform distribution of fat globules. But the nature of the components that form complexes in sour cream has not been studied.

The influence of the particle size of wheat germ on the physical-chemical properties of dairy dessert has been studied in [14]. With an increase in the content of wheat germ and particle size, a dense consistency is formed, the separation of moisture decreases. But, at the same time, there is a rough consistency, which loses its elasticity.

Our review of evidence-based information has shown that the study of the phase components of sour-milk dessert requires further research.

3. The aim and objectives of the study

The study aims to analyze the interaction of the components of sour-milk dessert with plant fillers. This will make it possible to improve the technology of fermented milk products.

To achieve the aim, the following tasks were set:

to determine the nature of the constituent substances
by the results from acquiring the IR spectra of the product;
to examine the analysis of variance of the product;

- to determine the mineral composition of fermented milk products with plant-based fillers.

4. The study materials and methods

A sour-milk dessert with plant-based fillings was chosen for the study, the recipe of which is given in [15]. It consists of skim milk, whey protein concentrate, apple and banana powder, gelatin. The technological process of production was carried out according to the technological scheme given in [16].

To determine the nature of the constituent substances that are part of the sour-milk dessert with plant-based fillers, the method of IR spectroscopy was used. The method is typically applied to study substances of various natures. The research was performed using an IR spectrometer with a spectral range of 500-4,000 cm⁻¹ [17].

The main regions (Table 1) of IR spectroscopy include the area of valence oscillations of simple bonds (OH, N–H, C–H, S–H with a spectral range of 4,000–2,500 cm⁻¹). It is also an area of valence oscillations of multiple bonds (C=C, C=O, C=N, C=C, C=N with a spectral range of 2,500–1,500 cm⁻¹). And there can be a region of valence oscillations of simple bonds (C–C, C–N, C–O) and deformation oscillations of simple bonds (C–H, O–H, N–H) with a spectral range of 1,500–500 cm⁻¹ [17].

Analysis of variance was conducted using a Zetasizer NANO S laser particle size analyzer (Malvern Instruments, UK). The measuring range of the device is from 0.3 nm to 10,000 nm [18].

Valence oscillations of groups, cm ⁻¹				
OH	NH	СН	S–H	C=O
3,6452,500	3,5003,300	3,3502,850	2,6002,550	1,7501,720
Valence oscillations of groups, cm ⁻¹				
С-О-	СООН	S=S	C=N	CH ₃
1,3001,000	1,7501,700	550450	1,2301,030	1,4701,355

Main regions of IR spectroscopy

We determined the mineral component of sour- milk dessert with plant-based fillers at the X-ray diffractometer "D8 ADVANCE" ("Bruker") under CuKa radiation (λ =1,542 nm) in the range of diffraction angles from 20 to 130 degrees. The principle of diffractometer operation is based on the X-rays diffraction from the atomic planes of the crystal lattice of the test substance. X-ray diffraction follows Wolf-Bragg's law. Structurally, diffractometers consist of an X-ray source with anodes made of copper, cobalt, chromium, molybdenum, iron, tungsten, titanium or silver, goniometer, detection units and control systems, data collection, and processing. Diffractometers are built on an optical scheme in which a flat sample is located in the center of the goniometer. Diffractometers are made in the form of a single module, inside which there are all the components, the control computer is located outside the diffractometer housing. The registration of the diffraction pattern is carried out by rotating the detection unit, X-ray source, and goniometer axes with the required angular velocities [19].

The obtained measurement results and graphical representation of experimental data were treated using standard statistical software Microsoft Excel 2010. The accuracy of the obtained results was ensured by repeating the experiments three to five times.

5. Results of studying the component composition of the sour-milk dessert

5.1. Determining the nature of the composition substances based on the results of product IR-spectra

Analyzing the IR-spectra of a sour-milk product without filler (Fig. 1) and a sourmilk dessert with plant-based fillers (Fig. 2), it can be seen which atomic groups of macromolecules are involved in molecular interaction. Fig. 1, 2 show the IR- spectra of product samples from which moisture was removed (sample 1) and calcined product at a temperature of 1,000 °C (sample 2).

The spectra show the presence of absorption bands in the range of 3,600-3,200 cm⁻¹ and 1,646 cm⁻¹, which is typical of both studied samples of fermented milk product with and without the addition of fillers. The existing bands correspond to the valence and deformation oscillations of the hydroxyl groups. They also indicate the presence of coordination-bound moisture in the samples of hydroxyl groups. It should be noted that in the samples before calcination (1), the intensity of these bands is significantly higher compared

to calcined samples. This may indicate the presence of hydroxyl groups in such samples in C–OH fragments.

In sample 1 of the sour-milk product without the addition of fillers, there are absorption bands in the range of $2,850-3,000 \text{ cm}^{-1}$, which correspond to the valence and deformation oscillations of the CH bonds in the resulting sample. These bands are absent in the spectrum of the calcined sample (2), which indicates the removal of the organic component.

Characteristic for sour-milk dessert with plant-based fillings is that in sample 1 there are absorption bands in the range of 1,310-1,450 cm⁻¹. They also correspond to the valence and deformation oscillations of the CH bonds in the resulting sample. These bands are absent in the spectrum of the calcined sample (2), which indicates the removal of the organic component. The spectrum of sample 1 also has an absorption band at 1,743 cm⁻¹, which can be attributed to the valence fluctuations of the C–O bond in aliphatic compounds. The specified band is missing for the calcined sample. And, for it, the absorption band at 1,092 cm⁻¹ increases significantly, which can be attributed to the valence P–O oscillations.

Sour-milk desserts contain 86–89 % moisture, including 83–86 % free, and 3–5 % bound. Therefore, the use of fruit in the form of freeze-dried powder together with milk protein concentrate in the technology of sour-milk desserts is appropriate. This combination will help reduce the content of free moisture, stable structure, which could prolong the shelf-life of dairy products.



Fig. 1. IR-spectra of the sour-milk product (1) and the sour-milk product residue after calcination at 1,000 °C (2)



Fig. 2. IR spectra: 1 – of the sour-milk product with plant-based fillers; 2 –of the sour-milk product residue after calcination at 1,000 $^\circ C$

5.2. Examining the product analysis of variance

The structure and consistency of sour-milk desserts are affected by the dispersion of protein particles. Fig. 3 shows the dispersed composition of the fermented milk product with plant-based fillers. Based on its results, it is possible to conclude the structural characteristics and origin of the product, some physical properties, which will be important at the stages of control of the technological process.



Fig. 3. Dispersion composition of the sour-milk product with plant-based fillers in the range: 1 - 6,000...7,000 nm; 2 - 1...10 nm; 3 - 1...100 nm

During the study of the sour-milk product with and without the addition of fillers, particles of different sizes were found. In particular, there is a slight aggregation of whey proteins and redistribution between particles in the range of 1–10 nm and 1–100 nm. It can be assumed that this is due to the aggregation of whey proteins [20].

It was also found that the heavy fraction of fruit filler particles in the form of freeze-drying powder, which is part of the product, undergoes sedimentation, and there is a gradual swelling. As a result, this leads to an increase in the size of the product components to an average of 6,000–7,000 nm. For the fermented milk product without the addition of plant-based fillers, such a fraction was absent.

5. 3. Determining the mineral composition of the sour-milk product with plant-based fillers

To characterize the total mineral content, the concept of "ash" is used, which means the residue after burning and processing. Therefore, in the first stage, moisture was removed from the dairy product using a rotary evaporator. The sample was dried at a temperature of 50–70 °C. After that, to determine the content of the mineral component, the original sample was calcined at a temperature of 1,000 °C for 2 hours (Fig. 4).

It was determined that the ash content in the fermented milk product without fillers is 0.62 g/100 g, and for fermented milk dessert with plant-based fillers –

0.7 g/100 g of product. The use of plant-based fillers in the form of freeze-dried powder in the technology of sour-milk desserts will not only improve its physical and chemical properties but also enrich the product with minerals. Fig. 5 shows the elemental composition in mg in terms of 100 g per finished product.



Fig. 4. X-ray-fluorescent spectrum of the sour-milk product residue after calcination at 1,000 °C

Accordingly, a sour-milk dessert with plant-based fillers contains a significant amount of minerals. The largest share is calcium (122 mg/100 g), potassium (97 mg/100 g), phosphorus (82 mg/100 g), sodium (50 mg/100 g). These elements are physiologically important for the normal development of the human body. It should be noted that the content of toxic elements in a sour-milk dessert was not detected.

About 80 % of a person's daily need for minerals is provided by dairy products. Also, dairy products contain such important macronutrients as calcium, potassium, sodium, magnesium, which are involved in building a number of vitamins, enzymes, and hormones.



Fig. 5. Elemental composition of sour-milk products

6. Discussion of results from determining the interaction of composition components of a sour-milk dessert with plant-based fillers

It has been established that some groups of different molecules give absorption bands with approximately the same wavelength (Fig. 1, 2). The intensity of absorption of functional groups in the region of 2,500–3,500 cm⁻¹ is due to the valence vibrations of NH–, CH– and S–H-groups, which indicates the presence of free organic acids and aromatic substances. These compounds can be released as a result of lactic acid fermentation in the production of sour-milk desserts. Proteins' characteristics in the samples are observed in the absorption band in the range of 3,300–3,500 cm⁻¹, which is due to the valence fluctuations of the N–H bond in the –NH₂ groups. In turn, they are capable of structuring and gelation. Such absorption bands are typical for both samples with and without the addition of plant fillers.

For a sour-milk dessert with plant-based fillers (Fig. 2), the absorption intensity in the range of 1,470-1,335 cm⁻¹ is observed on the spectra, which indicates the presence of soluble pectin.

The content of groups in the molecule that donate protons, such as R-OH; R_2NH ; R_3CH , and protons are taken, for example, C=O; C-SI; -SO₂, will give a sweet taste to the product [21].

Thus, the strength of the structure, moisture state, fermentation process, maturation, and composition of dairy products are examined by IR-spectroscopy. The location of the absorption bands of IR-spectra and their intensity determine the nature of atomic groups that are part of the macromolecules of the product [7-11].

It is proved that the use of plant ingredients affects the dispersion and physical-chemical parameters of dairy products [12–14].

This paper also reveals the effect of fillers on the size of the components of the product. Sour-milk dessert with plant-based fillings is a system consisting of particles of different dispersion, which will affect its physical and chemical properties (Fig. 3). There is a slight aggregation of whey proteins and redistribution between particles in the range of 1–10 nm and 1–100 nm. This is due to components such as whey protein, amino acids, carbohydrates, vitamins, and minerals.

The use of plant-based fillers in the form of freeze-dried powder in the technology of sour-milk desserts will not only improve its physical and chemical properties but also enrich the product with minerals.

The mineral composition of sour-milk dessert with plant-based fillers is marked by the content of calcium, phosphorus, potassium, sodium, sulfur, and iron (Fig. 5). These elements are physiologically important for the normal development of the human body. The content of toxic elements in a sour-milk dessert was not detected.

The presence of more than 80 % moisture in the food leads to limitations and complications of this

study. Such methods require additional treatment of the samples, namely the evaporation of moisture. Although there are some complications, the results of the research can be used to improve the technology of dairy products. These studies suggest that the use of functional and technological ingredients will help reduce the content of free moisture and the stable structure of fermented milk products.

7. Conclusions

1. The nature of the constituent substances was determined based on the results from acquiring the IR-spectra of the product. The spectra show the available absorption bands in the range of 3,600-3,200 cm⁻¹ and 1,646 cm⁻¹, which correspond to the valence and strain oscillations of the hydroxyl groups. They also indicate the presence of coordination-bound moisture in the samples of hydroxyl groups. It was found that the intensity of absorption of functional groups in the region of 2,500-3,500 cm⁻¹ is due to the valence vibrations of NH-, CH- and S-H-groups, which indicates the presence of free organic acids and aromatic substances. The absorption intensity is also observed in the spectra in the range of 1,470-1,335 cm⁻¹, which indicates the presence of soluble pectin. Proteins' characteristics in the samples are observed in the absorption band in the range of 3,300-3,500 cm⁻¹, which is due to the valence fluctuations of the N–H bond in the –NH₂ groups.

2. The dispersion analysis results of the product revealed a slight aggregation of whey proteins and redistribution between particles in the range of 1–10 nm and 1–100 nm.

3. Sour-milk dessert with plant-based fillers contains a significant amount of minerals. The largest share is calcium (122 mg/100 g), potassium (97 mg/100 g), phosphorus (82 mg/100 g), sodium (50 mg/100 g).

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