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## BIO-FERMENTED MILK PRODUCT BASED ON *L.ACIDOPHILUS* FORTIFIED BY *SANGUISORBA OFFICINALIS* EXTRACT

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*The development of new functional food products containing probiotics and prebiotics of plant origin has actual scientific and practical values. The presented study purpose is the creation of fermented milk products based on*

*L.acidophilus with the introduction of an extract of the Sanguisorba officinalis or great burnet as a prebiotic. Extracts from the great burnet rhizomes and roots have an antimicrobial activity, specifically against the bacteria Escherichia coli, Pseudomonas aeruginosa, and Staphylococcus aureus with pronounced bactericidal effect on various microbes of the dysenteric and paratyphoid groups, a detrimental effect on Trichomonas, fungi of the genus Candida and Giardia. Physicochemical and microbiological indicators during ripening and storage of the obtained fermented milk products based on L.acidophilus with great burnet extracts are considered. Titratable acidity, water-holding capacity, the number of lactobacilli, viscosity, and organoleptic values of fermented milk products are analyzed. It is established that the product with great burnet dry extract at a concentration of  $10^{-3}$  g/cm<sup>3</sup> has maximum scores for organoleptic, physicochemical, and microbiological indicators. The developed bio-fermented milk product enriched by antioxidants from extracts of biologically active compounds of great burnet will meet the national and international consumers' demands with recommendations for therapeutic and preventive nutrition and a healthy lifestyle.*

**Keywords:** *Lactobacillus acidophilus*, prebiotics, extract of *Sanguisorba officinalis*, functional food products, dairy products.

### БИОКИСЛОМОЛОЧНЫЙ ПРОДУКТ НА ОСНОВЕ *L.ACIDOPHILUS*, ОБОГАЩЕННЫЙ ЭКСТРАКТОМ *SANGUISORBA OFFICINALIS*

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*Разработка новых продуктов функционального питания, содержащих пробиотики и пребиотики растительного происхождения, имеет актуальное научное и практическое значение. Целью представленного исследования является создание кисломолочных продуктов на основе *L.acidophilus* с введением в качестве пребиотика экстракта *Sanguisorba officinalis* или кровохлебки. Экстракты из корневищ и корней кровохлебки обладают противомикробным действием, в частности в отношении бактерий *Escherichia coli*, *Pseudomonas aeruginosa* и *Staphylococcus aureus* с выраженным бактерицидным действием на различные микробы дизентерийной и паратифозной групп, губительно действуют на *Trichomonas*, грибы рода *Candida* and *Giardia*. Рассмотрены физико-химические и микробиологические показатели при созревании и хранении полученных кисломолочных продуктов на основе *L.acidophilus* с экстрактами кровохлебки. Анализированы титруемая кислотность, влагоудерживающая способность, количество лактобактерий, вязкость и органолептические показатели кисломолочных продуктов. Установлено, что продукт с экстрактом кровохлебки в концентрации  $10^{-3}$  г/см<sup>3</sup> имеет максимальные баллы по органолептическим, физико-химическим и микробиологическим показателям. Разработанный биокисломолочный продукт, обогащенный антиоксидантами из экстрактов биологически активных соединений кровохлебки, будет отвечать запросам отечественных и зарубежных потребителей с рекомендациями по лечебно-профилактическому питанию и здоровому образу жизни.*

**Ключевые слова:** *Lactobacillus acidophilus*, пребиотики, экстракт *Sanguisorba officinalis*, продукты функционального питания, молочные продукты.

### SANGUISORBA OFFICINALIS СЫҒЫНДЫСЫМЕН БАЙЫТЫЛҒАН *L. ACIDOPHILUS* НЕГІЗІНДЕГІ БИОСҮТҚЫШҚЫЛДЫ ӨНІМ

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*Құрамында пробиотиктер мен өсімдік тектес пребиотиктер бар функционалды тағамдардың жаңа түрлерін әзірлеу өзекті ғылыми және практикалық маңызға ие. Ұсынылған зерттеудің мақсаты - *Sanguisorba officinalis* немесе қандышөптің сығындысын пребиотик ретінде *L. acidophilus* негізіндегі ашытылған сүт өнімдерін өндіруде қолдану болып табылады. Қандышөптің тамырларынан алынған*

сығындылар микробқа қарсы әсерге ие, атап айтқанда *Escherichia coli*, *Pseudomonas aeruginosa* және *Staphylococcus aureus* бактерияларына дизентериялық және паратифоздық топтағы әртүрлі микробтарға айқын бактерицидтік әсер етеді, *Trichomonas*, *Candida* және *Giardia* тұқымдас саңырауқұлақтарға зиянды әсер етеді. Мақалада қандышөп сығындысы қосылған дайын *L.acidophilus* негізінде ашытылған биосүтқышқылды өнімдердің сақтау кезіндегі физика-химиялық және микробиологиялық көрсеткіштері қарастырылады. Дайын биосүтқышқылды өнімдердің қышқылдығы, ылғал ұстау қабілеті, лактобактериялар саны, тұтқырлығы және органолептикалық көрсеткіштері талданады. Қандышөп сығындысының  $10^{-3}$  г/см<sup>3</sup> концентрациясы қосылған өнім органолептикалық, физика-химиялық және микробиологиялық көрсеткіштері бойынша максималды көрсеткішке ие болды. Қандышөп биологиялық белсенді қосылыстарының сығындылары антиоксиданттарымен байытылған биосүтқышқылды өнім емдік-профилактикалық тамақтану үшін ұсынылады және салауатты өмір салтын ұстану бойынша отандық және шетелдік тұтынушылардың сұраныстарына жауап береді.

**Негізгі сөздер:** *Lactobacillus acidophilus*, пребиотиктер, *Sanguisorba officinalis* сығындысы, функционалды тағамдар, сүт өнімдері.

### Introduction

Nowadays, one of the directions in the production of beneficial health nutrition is a fortification of traditional food products containing probiotic microorganisms (*Bifidobacterium* and *Lactobacillus*) with functional ingredients - prebiotics.

Analyzing the bio-products of the Republic of Kazakhstan dairy enterprises, it becomes apparent that the majority of offered products are conventional or standard yogurts. Nonetheless, the availability of fermented milk products enriched with probiotics, particularly *Bifidobacterium*, remains relatively limited in the market. For example, “Food Master” company (Almaty) offers a product known as “Bio-C immun” yogurt, which contains *L.Rhamnosus Immunalis*. Only “Natzhe dairy factory” (Karaganda) produces two types of fermented milk drinks with the addition of the microorganisms *L.acidophilous* “Acidophilus” and “Narine-L” designed specifically for children. In this regard, it becomes an actual need to expand the range of dairy products, as well as the development of innovative bio-fermented functional food products containing probiotics and prebiotics microorganisms along with natural antioxidants.

*Lactobacillus acidophilus* is part of the normal microflora of the human large intestine and in the course of its life secretes enzymes (proteases,  $\beta$ -galactosidases, lactate dehydrogenases) that contribute to the active digestion of proteins, carbohydrates, fats, the assimilation of essential amino acids, trace elements, and also produces a number of vitamins [1].

*L.acidophilus* provides the accumulation of free amino acids, such as valine, glycine, and histidine, which also stimulate the development of the human intestinal microflora [2].

*L. acidophilus* has immunomodulatory properties, exhibits high anti-inflammatory and antitumor activity, and also assists in reducing

cholesterolemia by blocking the hydroxymethylglutarate-CoA reductase enzyme that limits the rate of cholesterol synthesis [3]. The antagonistic properties of *L.acidophilus* bacillus are due to the antibiotics produced by it - acidophilus and lactocidin, as well as lactic acid [4].

Based on the above mentioned, the inclusion of *L.acidophilus* in food formulations can enhance their functional properties. The use of such products is necessary after prolonged use of antibiotics, as they can prevent the development of concomitant diseases that occur as a result of inhibition of the intestinal microflora [5].

As prebiotics, plant extracts are often used, for example, extracts of licorice root, burdock, alfalfa, etc. [6].

*Sanguisorba officinalis*, commonly known as great burnet is used to treat diseases of the gastrointestinal tract with gastritis, colitis, etc. [7].

Extracts from the rhizomes and roots of great burnet have an antimicrobial activity, specifically, against the bacteria *Escherichia coli*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. A pronounced bactericidal effect on various microbes of the dysenteric and paratyphoid groups, a detrimental effect on *Trichomonas*, fungi of the genus *Candida* and *Giardia* were noted [8].

The polyphenolic complex of the great burnet rhizomes and roots includes hydrolysable tannins of the pyrogallol group - up to 23%, ellagic and gallic acids, pyrogallol, catechin and gallocatechin. Mainly, these parts of the great burnet consist of starch (until 30%), essential oil (1.8%), saponins: sanguisorbin and pyoterin - up to 4%, dye compounds, calcium oxalate salts, flavonoids derivatives of quercetin and kaempferol, hyperin, 3,7-diramnoside kaempferol, 3-galactosido-7-glucoside catechins, chromones. Roots and rhizomes also contain steroids:  $\beta$ -sitosterol,  $\beta$ -D-glucoside of  $\beta$ -sitosterol, stigmasterol; carotenoids, vitamin C; macro- and

microelements. Also, in the composition of the great burnet carbohydrates (until 7%), organic acids (until 1%), carotenoids, ascorbic acid (until 0.9%) are found in great amounts. The quantitative content of various groups of biologically active substances in the great burnet rhizomes and roots is following: polysaccharides (5.18±0.24%), saponins (3.71±0.17%), hydroxycinnamic acids 1.91±0.08 %), tannins in terms of gallic acid (14.52±0.51%) [7, 9].

Plant extracts have a therapeutic effect, which is due to the complex of biologically active substances that they contain [10].

Biologically active substances can stimulate the growth of fermented milk microorganisms by intensification of their activity. Extracts of them possess antioxidant properties and often contain simple sugars, oligosaccharides, and polysaccharides that serve as a nutrient source for microorganisms in functional food products. The resulting product can serve as a source of useful substances or antioxidants that are essential for the human body, such as vitamins, amino acids, and more [11].

In this regard, the development of new functional food products containing probiotics and prebiotics of plant origin has actual scientific and practical values. The presented study purpose is the creation of the fermented milk products based on *L.acidophilus* with the introduction as a prebiotic of dry extract of the *Sanguisorba officinalis* or great burnet.

The novelty of the work is the application of plant extract from domestic raw material - *Sanguisorba officinalis* L. with prebiotic and antioxidant properties obtained by microwave extraction at low concentrations in bio-fermented milk product based on *L.acidophilus*.

#### Materials and research methods

To obtain a dry extract, raw great burnet root is utilized (Producer “Zerde-Fito” LLP, Shymkent). It is an applied microwave extraction approach [12].

A pure culture of *L.acidophilus* n.v. Ep 317/402 is used as a starter for the production of fermented milk products.

For the preparation of fermented milk products based on *L. acidophilus* with a dry extract of great burnet, *L. acidophilus* bacillus is added as a starter culture at a rate of 2.5% to prepared milk with a fat content of 2.5% at a temperature of 37°C. Subsequently, an aqueous extract of great burnet is introduced into the mixture at concentrations of 10<sup>-3</sup> g/cm<sup>3</sup> and 10<sup>-5</sup> g/cm<sup>3</sup>.

The process of milk fermentation is carried out within 7 hours until a titratable acidity of 75-80°T is reached. Then the product is cooled to a temperature of 10 °C and left for storage. According to the above mentioned method, the following samples are obtained: Acidophilic product 1 - fermented milk product with great burnet extract at a concentration of 10<sup>-5</sup> g/cm<sup>3</sup>; Acidophilic product 2 – fermented milk product with great burnet extract at a concentration of 10<sup>-3</sup> g/cm<sup>3</sup>; Acidophilic product 3 (control) is a fermented milk product without any addition of great burnet extract.

Titratable acidity is expressed in Turner degrees (°T) [13].

The viscosity of the products is determined by viscosimeter VZ-246 [14].

The water-holding capacity is evaluated by centrifuge [15].

Organoleptic assessment of functional fermented milk products is determined by assessment of consistency, taste, smell, and color.

Statistical data processing is carried out by using «Excel 7.0» (MS Office, USA), «Statistica 6.0» (StatSoft, USA) softwares. Values are expressed as means ± standard deviations from three independent experiments (n = 3) at 90% confidence level (P = 0.90)

#### Results and discussion

As a prebiotic, in the obtaining of the functional fermented milk products, a dry extract of great burnet is used. The physicochemical and microbiological indicators of fermented milk products based on *L.acidophilus* with great burnet extract during ripening are shown in Table 1.

Table 1 – Physicochemical and microbiological indicators during ripening

Name	0 h		24 h		
	A, °T	lg CFU/cm <sup>3</sup>	A, °T	lg CFU/ cm <sup>3</sup>	Clot characteristic
					WHC, %
Acidophilic product 1	18,80±0,04	6,00±0,01	94,40±0,08	8,13±0,21	25
Acidophilic product 2	18,80±0,04	6,00±0,01	95,30±0,12	8,45±0,22	20
Acidophilic product 3 (control sample)	18,80±0,04	6,00±0,01	97,20±0,11	7,60±0,21	30

A - acidity, °T;

Ig CFU colony forming unit, is a number of cells in 1 cm<sup>3</sup> of the product;

WHC - water-holding capacity, %

By 24 hours, as the control product and prebiotic products mature, the acidity increases equally. The addition of great burnet extract at a concentration of 10<sup>-3</sup> g/cm<sup>3</sup> makes it possible to grow the content of *L.acidophilic* bacillus in Acidophilic product 2 by 11% higher than in the Ac-

dophilic product 3 (control sample). The introduction of a prebiotic into the product leads to an increase in the density of the formed clot, a 23% growth in viscosity (as shown in Fig. 1), and a 1.5-fold increase in water-holding capacity.

The viscosity of fermented milk products based on *L.acidophilus* with great burnet extract (the relative error of the method does not exceed 10.90%, n=3, P=0.95) is shown in Figure 1.

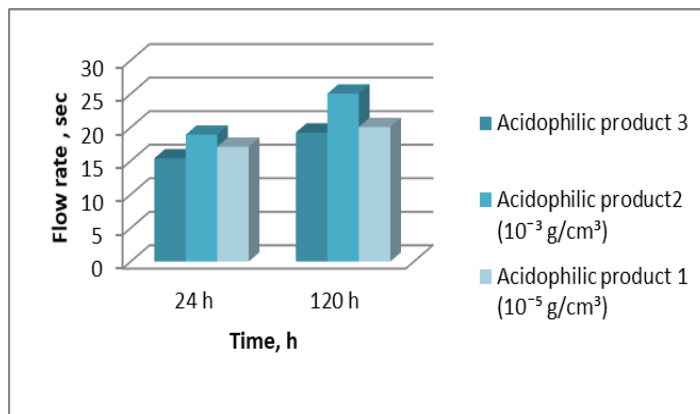


Figure 1 – Viscosity of fermented milk products

All prepared fermented milk products have a milky-white color, milky taste, sour-milk smell, and uniform texture. In Acidophilic products 1 and 2, a slightly sweet taste and a thicker texture are observed compared to the control.

The physicochemical and microbiological indicators of the obtained fermented milk products based on *L.acidophilus* with great burnet extract during the storage period are presented in Table 2.

Table 2 – Physicochemical and microbiological indicators during storage

Name	1 day (24 h)			5 days (120 h)		
	A, °T	Ig CFU/ cm <sup>3</sup>	WHC, %	A, °T	Ig CFU/ cm <sup>3</sup>	WHC, %
Acidophilic product 1	94,40±0,08	8,13±0,21	25	136,00±0,05	8,50±0,22	20
Acidophilic product 2	95,30±0,12	8,45±0,22	20	135,80±0,08	8,72±0,18	20
Acidophilic product 3 (control sample)	97,20±0,11	7,60±0,21	30	137,30±0,07	8,12±0,18	20

A - acidity, °T;

Ig CFU colony forming unit, is a number of cells in 1 cm<sup>3</sup> of the product;

WHC - water-holding capacity, %

As observed in Table 2, by 5 days of storage, the acidity in all products increases by almost 1.5 times. The amount of Lactobacillus in the Acidophilic product 2 during storage is 7.4% higher compared to the Acidophilic product 3 (control sample). All products maintain the same water-holding capacity. Acidophilic product 1 and Acidophilic product 2 have a higher viscosity compared to Acidophilic product 3 (control sample).

During storage, the products have a milky-white color, a milky taste, refreshing, without foreign tastes and odors; more pronounced sour-milk smell. Organoleptic indicators of the obtained fermented milk products based on *L.acidophilus* with great burnet extract are shown in Figure 2. According to the organoleptic indicators, fermented milk product with great burnet extract at a concentration of 10<sup>-3</sup> g/cm<sup>3</sup> has the maximum score for all controlled parameters, compared with other products. It has a softer and more delicate taste and a delicate smell.

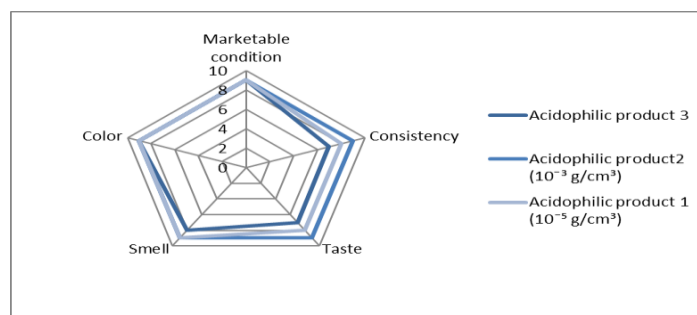


Figure 2 – Organoleptic indicators of the developed fermented milk products

Thus, fermented milk functional products based on *L.acidophilus* with great burnet dry extracts are developed. These obtained products have an extended shelf life, with a higher *Lactobacillus* count compared to the control product.

### Conclusion

The fermented milk product based on *L.acidophilus* is developed by using great burnet extracts as a prebiotic that has high nutritional and consumer properties. It is shown that a fermented milk product based on *L.acidophilus* with great burnet extract at a concentration of  $10^{-3}$  g/cm<sup>3</sup>, surpasses the control sample in terms of quality: the number of *L.acidophilus* is higher by 11.0%, the viscosity is higher by 23.0%, respectively. It has a more delicate and soft taste. Bio-fermented milk products enriched by antioxidants from extracts of biologically active compounds of great burnet are recommended for people intolerant to lactose, taking advantage of the presence of probiotic bacteria, a health-promoting bioactive ingredient. Thus, consumption of developed dairy products can meet the national and international consumers' demands in order to achieve better health status of the population, as well as in therapeutic and preventive nutrition.

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## САРЫ СУ КОНЦЕНТРАТЫМЕН БАЙЫТЫЛҒАН СҮТ ЖӘНЕ ЕТ ӨНІМДЕРІНІҢ ТЕХНОЛОГИЯСЫ

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Бұл мақалада екінші реттік сүт шикізатты пайдалана отырып, ешкі сүті мен ешкі еті негізінде ет және сүт өнімдерін өндірудің жетілдірілген технологиясы туралы мәліметтер ұсынылған. Зерттеулер мақсаты - сарысу ақуыз концентратымен байытылған жұмсақ ірімшіктің және калориясы төмен шұжық өнімдерінің технологиясы мен компоненттік құрамын әзірлеу болып табылады. Жобаның ғылыми жаңалығы Павлодар өңірінің жергілікті тұқымды ешкілердің сүті мен еттің химиялық құрамы мен технологиялық қасиеттерін және Заанен және Горький тұқымды әкелінген ешкілердің сүтін зерттеу бойынша эксперименттік зерттеулер жүргізу болып табылады. Алынған ақуыз ұйытынды қажетті тығыздығы мен тұтқырлығын қамтамасыз ететін ферменттік препарат және оның концентрациясы да негізделеді және таңдалады. Ет өнімін сарысуы бар ақуыздармен байы тудың жаңа әдісі жасалды, ол жергілікті ешкі етінен жасалған шұжық өнімдерінің жаңа түрінің кесіндісінде сыртқы түрі мен көрінісін қалыптастырады. Жұмыстың практикалық маңыздылығы ет және сүт өнімдерінің жаңа түрлерін, ешкі сүті мен ешкі етінен, сүт сарысуын пайдалана отырып, тағамдық және биологиялық құндылығын арттыратын ет және сүт өнімдерінің жаңа түрлерін өндірудің технологиялық процесін әзірлеу және оңтайландыру болып табылады. Ғылыми зерттеу жобаны орындау барысында ешкі сүтінен жасалған жетілусіз табиғи мәйекті жұмсақ ірімшіктерді және ешкі еті негізінде жасалған құс еті қосылған шұжық өнімін өндіру технологияларын әзірлеу бойынша тәжірибелік зерттеулер жүргізілді. Ешкі сүті мен еті негізінде жасалған жетілусіз жұмсақ ірімшік және шұжық өнімінің рецептуралық құрамын әзірлеу үшін екінші реттік сүт шикізатты, толтырғыштарды және тағамдық қоспаларды іріктеу және негіздеу бойынша теориялық және тәжірибелік зерттеулер жүргізілді. «Биотехнология» кафедрасының зертханалық жағдайында сары су концентратымен байытылған жетілусіз жұмсақ ірімшіктің және шұжық өнімінің жаңа түрлерінің рецептуралары апробацияланып, сақтау қабілетілігі зерттеулер нәтижесінде анықталды. Сонымен қатар, дайын өнімдерінің тәжірибелік үлгілерінің сапалық (органолептикалық, физика – химиялық) көрсеткіштері анықталды.

Негізгі сөздер: ешкі сүті, ешкі еті, екінші реттік сүт шикізат, сары су, сары сулы концентраттар, табиғи мәйекті жұмсақ ірімшік, шұжық өнімі.