

# Sustainable Increase of Cow Milk Productivity Using Components of Siberian Forest and Alpha-Amylase Enzyme

Evgeny IVANOV <sup>1,a</sup> Olga IVANOVA <sup>1,b</sup> Vera TERESHCHENKO <sup>1,c</sup> Lyubov EFIMOVA <sup>1,d</sup>

<sup>1</sup> Krasnoyarsk Research Institute of Animal Husbandry - Separate Division of the Federal State Budget Scientific Institution "Federal Research Center "Krasnoyarsk Science Center of the Siberian Branch of the Russian Academy of Sciences", 660049, Krasnoyarsk, RUSSIA

ORCID: <sup>a</sup> 0000-0001-6992-9406; <sup>b</sup> 0000-0002-8740-5267; <sup>c</sup> 0000-0001-5842-0153; <sup>d</sup> 0000-0002-3234-9747

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## Abstract

At the places of harvesting coniferous trees and industrial nuts processing remains, a devastating amount of wastes litters large areas of land, poisons the air and the hydro-environment by the combustion products and toxic compounds released during decomposition. Therefore, the search for solution for the effective use of waste is an actual task. The aim of this research is to investigate the influence of fodder additive made of forest waste resources and the enzymes on milk productivity of cows. The experiment has been held on black-and-white breed in Krasnoyarsk region. The experimental group received, in addition to the main diet, some fodder additives consisted of coniferous flower, crushed pine nut shells, arabinogalactane and amilosubtiline HCl enzyme. The analysis of coniferous flower demonstrated that it contains more copper by 41.6%, zinc-55.6%, magnesium-183.3%, chromium-66.7% than in pine nut shell thus, on the contrary, the content of lignin, cellulose and moisture, was more in the shell of pine nuts-24.2%, 9.7% and 5.0%, respectively. Under the effect of the test additives in cows of the experimental group milk yield was higher than the control group, 9.9% (P<0.05), fat mass fraction - 0.3% (P<0.05), the amount of milk basis of fat content 19.0%, in the blood was higher total protein content 2.7%, glucose - 4.0; albumin - 3.0; - creatinine 6.0; triglycerides by 11.1%; calcium 1.5%; phosphorous - 10.2; iron - on 3.6%, magnesium - 2.6%. Thus, the tested additives had a positive synergistic effect on the body of cows, which affected the increase in the quantity and quality of milk.

**Keywords:** Coniferous flower, Pine nut shell, Arabinogalactane, Amilosubtiline HCl, Milk efficiency, Cow

## Sibirya Orman Ürünleri ve Alfa-Amilaz Enzimi Bileşenlerini Kullanarak İneklerde Süt Veriminin Sürdürülebilir Artışı

### Öz

İğne yapraklı ağaçlar ve endüstriyel fıstık işleme alanlarında geniş arazileri kirleten, yanma ürünleri ile hava ve hidro-ortamı zehirleyen, ayrıştırma sırasında açığa çıkan toksik bileşiklerden oluşan yıkıcı miktarda atık kalır. Bu nedenle, bu atığın etkili kullanımı için çözüm aramak önemli bir sorumluluktur. Araştırmanın amacı, orman atık ürünleri ve enzimlerden yapılan yem katkılarının ineklerin süt verimi üzerindeki etkisidir. Deney Krasnoyarsk bölgesindeki siyah-beyaz ırkta yapıldı. Deney grubuna ana diyet ek olarak iğne yapraklı çiçek, ezilmiş çam fıstığı kabuğu, arabinogalakattan ve amilosubtilin HC1 enziminden oluşan bazı yem katkı maddeleri verildi. İğne yapraklı çiçeğin analizi, çam fıstığı kabuğundan %41.6, çinko - %55.6, magnezyum - %183.3, krom - %66.7 oranında daha fazla bakır içerdiğini gösterdi, aksine lignin, selüloz ve nem içeriği, çam fıstığı kabuğunda sırasıyla - %24.2, %9.7 ve %5.0 daha fazlaydı. Katkı maddelerinin kullanıldığı deney grubundaki ineklerde süt verimi kontrol grubundan daha yüksekti, %9.9 (P<0.05), yağ kütlesi fraksiyonu - %0.3 (P<0.05), yağ içeriğinin süt bazlı miktarı %19.0, kandaki toplam protein içeriği %2.7, glikoz - 4.0; albümin - 3.0; - kreatinin 6.0; trigliseritler %11.1; kalsiyum %1.5; fosfor - 10.2; demir - %3.6, magnezyum -%2.6'ydı. Bu nedenle, söz konusu katkı maddelerinin, inekler üzerinde, süt miktarı ve kalitesinde artışla sonuçlanan pozitif bir sinerjistik etkisi olduğu belirlendi.

**Anahtar sözcükler:** İğne yapraklı çiçek, Çam fıstığı kabuğu, Arabinogalakattan, Amilosubtiline HCl, Süt verimi, İnek

## INTRODUCTION

Highly productive cows have the most intensive metabolism associated with the formation of a large number of nutrients

released from milk. They have increased activity of the cardiovascular, respiratory, endocrine and other systems of the body. The quantity and quality of milk to a large extent depend on the amount of food eaten, its chemical



### Correspondence



+7 923 2133331



e.a.ivanov@bk.ru

composition and the balance of the diet on nutrients. Sometimes it is almost impossible to ensure high productivity of animals only at the expense of feed of own production. Therefore, it is advisable to use feed additives containing various nutrients and biologically active substances that can enrich the diet [1].

Krasnoyarsk territory has one of the largest reserves of forest resources among the regions of Russia [2]. Coniferous wood species - Siberian larch (*Lárix sibirica* L.) and common pine (*Pinus sylvestris*) prevail in terms of logging and wood harvesting on the territory of Siberia [3]. Forest enterprises of the timber industry mainly use stem wood, which is 60-65% of the total biomass of the tree.

Coniferous browses and butt of the tree are the wastes left on the cutting areas without any usage [4]. It negatively affects the environment in places of the felling, and prevents the growth of forests [5], by forming wastes of wood processing that are harmful to the environment associated with the contamination of large tracts of land, poisoning the air and hydrosphere combustion products landfills, toxic compounds released during decomposition, etc [3]. At the same time, from each hectare of logging, coniferous browses can be collected in an amount that is enough to produce 10-12 tons of coniferous flour [6] and a large number of unclaimed butts of the tree that accumulates into waste [7].

Coniferous needle contains carotene, sugar, glucose, fructose, pectin, tannins, cobalt, copper, manganese, zinc, iron, and vitamins B, C, E, and K [6]. Such a rich complex mineral and vitamin nutrition of coniferous flour allows it to show antimicrobial, immunostimulating, antioxidant, anti-inflammatory, hematopoietic action and can be successfully used to stimulate the growth and development of farm animals, increase productivity and improve its quality [8].

The butt of the tree is the most enriched with the polysaccharide arabinogalactane, the content of which reaches 15-20 % by weight [7].

Arabinogalactane is a complex natural water-soluble polysaccharide extracted from larch wood [1,9]. It is larch that makes it possible to obtain arabinogalactane with the most useful properties [10].

The use of this polysaccharide as a feed additive can increase the productivity of farm animals. When exposed to the intestinal micro flora, polysaccharides break down into short-chain fatty acids, which in ruminants are considered the main source of glucose in the blood, which can significantly reduce the energy deficit in the body [11].

A special place in the structure of the natural resources of Siberia is occupied by cedar forests, the main asset of which was the pine nut [12].

Harvesting pine nuts for food purpose has a long traditional

trade of the population in Siberia. For one ton of harvested pine nuts is more than two tons of waste remain [13], a significant amount of which falls on its shell (51-59% of the weight of the nut) [14]. The presence of mineral substances in the nut shell and a high content of carbohydrates characterize it as a source of carbohydrate-mineral complex [15].

In the Krasnoyarsk Territory annual 40.07 million tons of green conifers waste is formed. According to the Federal State Statistics Service management for the Krasnoyarsk Territory, in 2019 the number of cattle in farms of all categories amounted to 368.4 thousand heads, including 141.2 thousand heads of cows.

Considering of the foregoing premises, the use of forest industry waste, arabinogalactane and enzymes in the feeding of dairy cows is relevant.

The aim of the research is to study the effect of a fodder additive from forest resources and an enzyme on the dairy productivity of cows.

## MATERIAL and METHODS

### Ethics Statement

This study was carried out in accordance with the «Rules for the use of experimental animals» (Appendix to the order of the Ministry of Health of the USSR dated 08/12/1977, No. 755). Federal state budget scientific institution "Federal research centre "of the Krasnoyarsk scientific center of the Siberian branch of the Russian Academy of Sciences. (protocol approval number: 7 - 09.12.2019)

The experiment has been held on lactating cows of black-and-white breed in "Breeding Farm Tazhny" Ltd, Sukhobuzimsky area of Krasnoyarsk region.

To conduct the experiment due to analogue concept there were formed two groups of black-and-white breed lactating cows of the second calving 15 animals in each group (Table 1).

The experimental cows were of average fatness (BCS = 3.5), average live weight was 600-620 kg, average yield of milk - 24 kg/day, DIM - from 11 to 100 d in milk. The animals were kept in a typical four-row barn with a year-round

**Table 1.** Experience scheme

Group	Amount of Cattle	Duration (days)	Feed Condition
Control	15	100	Basic Diet (BD)
Experimental	15	100	BD + coniferous flower (50 g/h) + pine nut shell (50 g/h) + arabinogalactane (5 g/h) + amilosubtiline HCl enzyme (5 g/h)

stall system. Each cow was tied (THS), had a separate stall, individual feeder, and drinker.

The animals of the control group received a basic diet contains of haylage of perennial grasses-20 kg, straw-6 kg, barley-2 kg, wheat-2.1 kg, oats-2.5 kg, rapeseed cake-0.8 kg, sunflower cake-1 kg, grain molasses-1.8 kg.

In addition to the main diet, the cows of the experimental group were fed a mixture consisting of coniferous flour, pine nut shells, amilosubtiline hc (hydrochloride) enzyme and arabinogalactane once a day morning. The main diet was given to each cow individually. Experimental additives were daily mixed with concentrated feed (barley, wheat, oats) and given to cows of the experimental group in dry. The TMR system was not used on this farm. Composition and food value of the main diet presented in the *Table 2*.

Observations of the animals in the experimental groups showed that they willingly and without residue consumed concentrated feed containing test additives.

The main diet contained: energetic feed unit (EFU) - 20.3; exchange energy - 203.8 MJ; digestible protein - 2163.3 g; dry matter - 20.46 kg; crude fat - 776.1 g; crude fiber - 4060.7 g; sugar - 1998 g; methionine - 95.5 g; tryptophan - 28.7 g; calcium - 125 g; phosphorus - 76.9 g; magnesium - 42.2 g; carotene - 986.7 mg.

In 1 kg of dry matter of the diet contained 0.99 EFU, carotene - 48.23 mg, digestible protein per 1 EFU - 106.6 g, the sugar-to-protein ratio was 0.92: 1, the calcium-to-phosphorus ratio was 1.4: 1. The dry matter of the diet contained: crude fiber - 19.81%, raw fat - 3.8%, sugar - 9.8%.

Coniferous flour was made from pine branches (*Pinus sylvestris*), which were subjected to extraction with an alcohol-toluene mixture and hot water on an extraction plant, then dried and ground to a state of crumbly flour. Pine nut shells were crushed to a particle size of no more than 4 mm. Preparation of coniferous flour and crushed pine nut shells, as well as their analysis were carried out in a physical and chemical laboratory of wood plants biology in Forest Institute. named after V. N. Sukachev SD RAS.

**Table 2.** Composition and food value of the main diet

Indicator	Feed								Total
	Haylage Perennial Herbs	Straw	Barley	Wheat	Oats	Sunflower Cake	Oilseed Rape	Molasses Rom Rye Grain	
Amount of feed, kg/day	20.0	6.0	2.0	2.1	2.5	1.0	1.8	1.8	37.38
EFU	7.8	2.0	2.4	2.2	2.3	1.0	0.9	1.7	20.3
Exchange energy, mJ	78.0	20.4	23.6	22.5	23.0	10.4	9.0	16.9	203.8
Dry matter, kg	9.0	2.7	1.78	1.79	2.13	0.9	0.72	1.44	20.46
Digestible protein, g	744.0	60.0	222.0	298.2	197.5	324.0	209.6	108.0	2163.3
Crude Fat, g	408.0	60.0	30.0	31.5	100.0	77.0	69.6	0	776.1
Crude fiber, g	2538.0	942.0	60.0	58.8	242.5	129.0	90.4	0	4060.7
Starch, g	94.0	90.0	1120.0	1029.0	800.0	25.0	0	0	3158
Sugar, g	696.0	138.0	30.0	31.5	62.5	62.6	0.0	977.4	1998
Lysine, g	52.0	8.4	10.4	8.2	9.0	13.4	12.6	0	114
Methionine, g	46.0	8.4	4.4	8.6	8.0	15.8	4.3	0	95.5
Tryptophan, g	8.0	1.8	3.6	2.9	2.8	5.2	4.4	0	28.7
Calcium g	74.0	29.4	0.8	1.5	3.8	5.9	3.8	5.8	125
Phosphorus, g	26.0	7.8	6.0	9.0	8.5	12.9	6.3	0.4	76.9
Magnesium, g	16.0	7.8	4.6	2.3	3.0	4.8	3.5	0.2	42.2
Potassium, g	136.0	70.2	10.2	9.7	13.5	9.5	8.9	59.2	317.2
Sulfur, g	16.0	5.4	0	0.8	3.5	5.5	3.6	2.5	37.3
Copper, mg	74.0	30.6	16.6	4.8	12.3	17.2	5.8	8.3	169.6
Zinc, mg	210.0	87.0	62.4	84.0	56.3	40.0	38.8	37.4	615.9
Manganese, mg	386.0	222.6	85.0	86.3	141.3	37.9	35.4	44.3	1038.8
Cobalt, mg	0.6	1.2	0.2	0.0	0.2	0.2	0.2	1.1	3.7
Iodine, mg	1.6	0.6	0	0.2	0.3	0.4	0.3	1.3	4.7
Carotene, mg	810.0	150.0	0	21.4	3.3	2.0	0	0	986.7
Vitamin D ME	2700.0	1080.0	0	0	0	5.0	2.4	0	3787.4
Vitamin E, mg	760.0	210.0	0.0	27.9	32.3	11.0	9.6	5.4	1056.2

Arabinogalactane is produced in JSC "Ametis" (Blagoveshchensk, Amur region). To activate cicatricial digestion, coniferous flour, pine nut shells and arabinogalactane were mixed with amilosubtiline hc (hydrochloride) enzyme produced in PE "Sibbiopharm" (Berdsk, Novosibirsk region).

The animals participating in the experiment were kept in identical conditions in separate stalls using a tie. Milking cows on the farm is carried out in the milk delivery line (a milk line).

Quality determining of composition components (volatile compounds) of samples of coniferous flour and pine nut shells was performed in the Krasnoyarsk Regional Centre of common use FITC KNC SB RAS on the chromat-mass spectrometer "Agilent 5975C-7890A" (USA) using a vapor-phase sampler HeadSpace Sampler G 1888.

Milk productivity was established by conducting daily control milking. The study of chemical indicators of milk and biochemical indicators of blood were carried out at the beginning and at the end of the experiment.

The mass fraction of fat and protein in milk were determined on the milk analyzer "Lactoscan FARM Eco" (Bulgaria).

The suitability of milk for cheese production was determined by the rennet clotting to GOST 32901-2014. The method is based on the ability of raw milk to coagulate under the influence of rennet and microorganisms of raw milk. According to the results of formed clots visual assessment, raw milk was assigned to one of three classes: class I - a clot with a smooth surface, elastic to the touch, without eyes in a longitudinal section, floats in a transparent, non-stretching milk serum; II class - a clot soft to the touch with single eyes (1-10), torn, but not swollen; Grade III - a clot with numerous eyes, spongy, soft to the touch, swollen, floated up.

Blood for biochemical analysis was taken from the tail vein after morning feeding using special needles of vacuum

tubes with a coagulation activator (PUTH, PRC). Blood serum of cows was examined by photometric method on the biochemical and immunoassay blood analyzer "Chem Well 2910 c" (USA).

Setting up and conducting the experiment was carried out by the method of Ovsyannikov [16].

Analysis was done by the method of Plokhinsky [17] using the computer program "Analysis Package for biometric processing of zootechnical data" [18]. Statistical differences between the two groups were calculated with Student's t-test. The level of significance was  $P < 0.05$ ,  $P < 0.01$ ,  $P < 0.001$ .

## RESULTS

Pine nut shell had a rich brown color with a pronounced smell of pine nuts, coniferous flour-yellow had a green color with a smell of pine needles.

In comparison with pine nut shell the coniferous flour had more extractive substances extracted by alcohol-toluene mixture by 23.4%, hot water-by 16.5%, while the amount of lignin, cellulose and moisture, on the contrary, was greater in the shell of pine nuts-by 24.2%; 9.7 and 5.0%, respectively (Table 3).

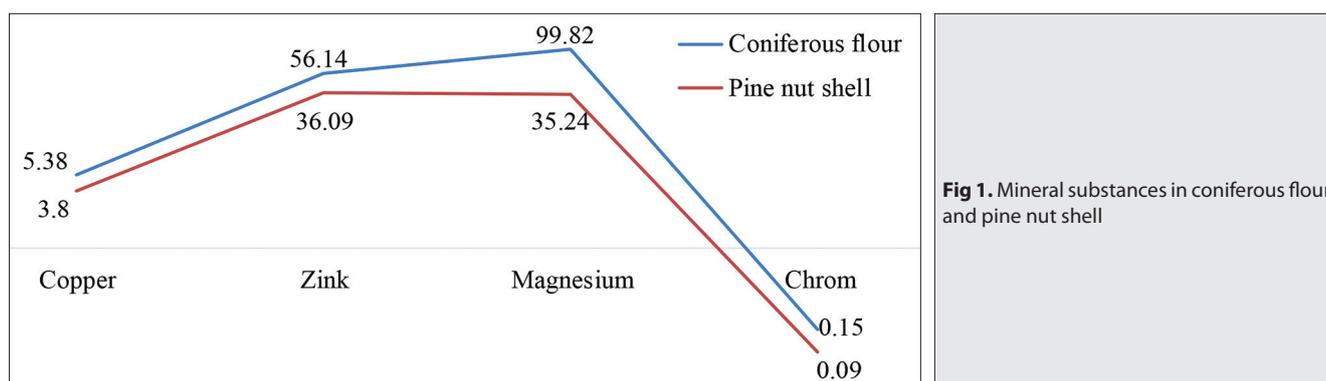
Mineral substances (Fig. 1) in coniferous flour is more than in pine nut shell: level of copper by 41.6%, zinc - 55.6%, magnesium - 183.3%, chromium - 66.7%

Table 4 presents milk efficiency of cows for the initial 100 days of lactation.

The result of feeding tested additives confirms that the animals of the experimental group has higher milk yield than the control group by 9.9% ( $P < 0.05$ ), the mass fraction of fat - by 0.3% ( $P < 0.05$ ), the amount of milk of basic fat content - by 19.0%.

According to the physical and chemical characteristics

<b>Table 3. Physical and chemical characteristics of coniferous flour and pine nut shell</b>		
<b>Characteristic</b>	<b>Coniferous Flour</b>	<b>Pine Nut Shell</b>
<b>Chemical composition</b>		
Monosaccharide, mg/g totally dry weight	0.30	traces
Oligosaccharide, mg/g totally dry weight	8.65	2.37
Starch, mg/g totally dry weight	1.15	0.79
<b>Physical-chemical components</b>		
Moisture %	6.7	11.7
Extractive substances extracted from the sample:		
hot water, %	29.66	4.06
alcohol-toluene mixture, %	20.57	6.26
Polyphenols, g/L	0.19	0.14
Cellulose, %	27.15	36.82
Lignin, %	20.49	44.64



**Fig 1.** Mineral substances in coniferous flour and pine nut shell

Indicator	Group	
	Control	Experimental
Milk yield for 100 days of lactation, kg	2085.36±44.95	2292.65±71.06*
Average daily yield, kg	20.85±0.45	22.93±0.71*
Mass fraction of milk fat, %	3.49±0.07	3.78±0.10*
Mass fraction of protein, %	3.19±0.09	3.19±0.14
FCM (fat corrected milk, 3.4%), kg	2139.51	2546.58
ECM, kg	1936.52	2206.19

\* $P < 0.05$

Indicator	Group	
	Control	Experimental
Mass fraction of milk fat, %	3.49±0.23	3.90±0.32
Mass fraction of protein, %	3.18±0.52	3.42±0.45
Lactose, %	5.15±0.66	5.54±0.56
Mass fraction of MSNF (milk solids non-fat), %	8.35±0.09	8.26±0.16
Salts, %	0.66±0.34	0.28±0.30
Dry component, %	11.10±0.22	11.91±0.27
Water, %	1.14±1.27	1.02±0.03
Density, kg/m <sup>3</sup>	1028.27±0.45	1027.73±0.87
Active acidity (pH)	6.48±0.01	6.41±0.03
Urea, mg/100ml	4.40±1.36	8.24±1.90
Temperature of freeze, °C	0.549±4.87	0.544±1.92

studied at the end of the experiment (Table 5), has been found that the milk of the experimental group cows has higher characteristics in comparison with the control group, such as the mass fraction of fat - by 0.41%, protein - by 0.24%, lactose - 0.39%, dry component - 0.81%, urea - 87.3%.

The results of milk testing for cheese production presented in Table 6.

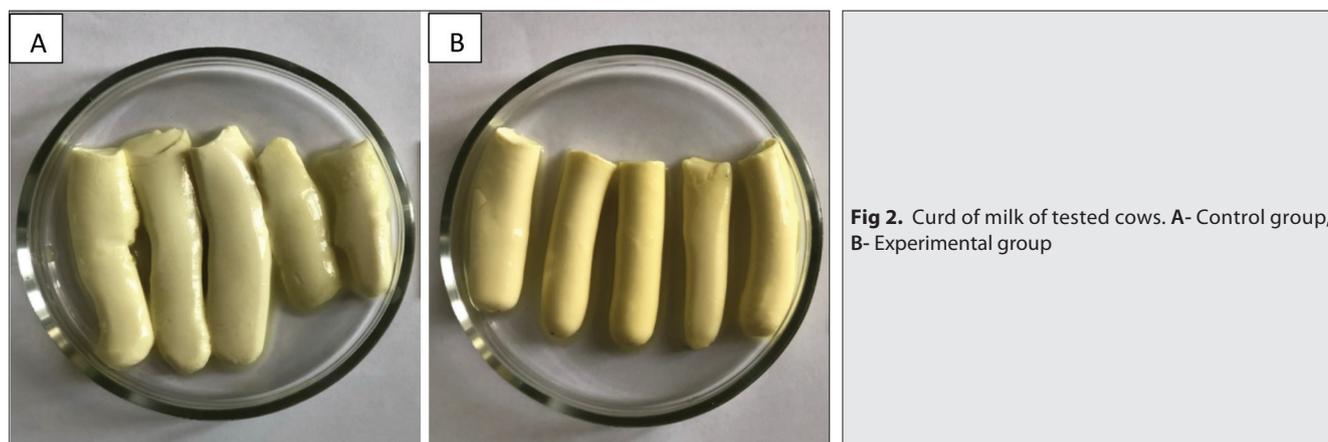
This experience clearly demonstrates that in the experimental group, under the influence of the tested additives, the proportion of cows whose milk corresponded to the first

class of cheese suitability increased and amounted to 100%.

Curd of milk in the experimental group better than in the control group and had a smoother surface, clear shape, density and elasticity (Fig. 2).

Highly productive cows are sensitive to various changes in the feed diet, which is invariably reflected in animal blood composition [19]. As a result of biochemical studies of the blood of cows at the end of the experiment (Table 7), it was found that all the studied blood serum parameters did not exceed the maximum permissible values, but there

Group	At the Beginning			At the End		
	Class Cheese Suitability					
	I	II	III	I	II	III
Control	20	40	40	20	60	20
Experimental	0	80	20	100	0	0



Parameter	Group	
	Control	Experimental
Protein, g/L	83.64±2.27	85.90±3.96
Glucose, mmol/L	3.52±0.23	3.66±0.28
Albumin mmol/L	31.72±1.39	32.66±0.79
Creatinine, mmol/L	94.18±8.58	99.84±3.41
Triglycerids, mmol/L	0.09±0.02	0.10±0.02
Cholesterol, mmol/L	5.22±0.45	5.20±0.68
Amylase, Un/L	13.00±3.48	12.40±2.28
Chlorides, mmol/L	98.00±2.76	93.80±1.39
Calcium, mmol/L	2.67±0.12	2.71±0.15
Phosphorus, mmol/L	2.26±0.19	2.49±0.24
Iron, mmol/L	28.18±0.91	29.18±0.81
Magnezium, mmol/L	1.16±0.02	1.19±0.01

were minor deviations between the groups. In animals of the control group, the level of cholesterol, amylase and chlorides was higher than in the experimental group by 0.4%; 4.8 and 4.5%, respectively. In the experimental group, compared with the control group, the total protein content was higher by 2.7%, glucose-by 4.0%; albumin-by 3.0%; creatinine-by 6.0%; triglycerides-by 11.1%; calcium-by 1.5%; phosphorus-by 10.2%; iron-3.6%, magnesium-2.6%.

The calculation of economic efficiency showed, the cost of used feed additive consisting of coniferous flour, pine nutshell, arabinogalactan, and the enzyme preparation Amilosubtilin G3x for the full period of the experiment per

cows amounted to 845 rubles. The use of these additives in cow feeding allowed to increase milk productivity by 9.9%, reduce the single cost of milk by 7.9%, and increase the level of profitability by 8.4%.

## DISCUSSION

As far as we know, this is the first research of studying the complex influence of coniferous flour, pine nut shells, arabinogalactane and amilosubtiline hc (hydrochloride) enzyme for cow milk efficiency

Previously, we conducted research on feeding cows

coniferous flour and pine nut shells <sup>[20,21]</sup>. The obtained data had a positive effect and allowed to obtain a patent for the invention <sup>[22]</sup>. Studying the effectiveness of each component separately on the body of cows allow us to see the validity of their presence in a diet.

Coniferous flour was fed to animals by researchers <sup>[23]</sup>, but according to their results it had an adverse effect on the animal's body and caused an abortion. However, a team of other scientists <sup>[24]</sup> on the contrary claims that the adding of coniferous flour into a cow's diet did not cause abortions.

More over it contributed to the growth of antioxidant protection <sup>[25]</sup>. Due to the high antibacterial characteristics of coniferous needles, there is a decrease in the content of somatic cells in the milk of cows <sup>[26]</sup>.

Researchers who used pine nut shells in the diet of laboratory animals claim to increase endurance of cows <sup>[27]</sup>. Despite the fact that there are quite few studies about feeding it, we have found evidence of a positive effect of the tested component on the calves <sup>[28]</sup>. The authors claim that adding of pine nut shell into the diet of calves normalizes digestion and metabolism, as well as increases the activity of the body's defenses. This is also confirmed by our research <sup>[29]</sup>, the results show the positive obtained effect.

Using amilosubtiline HCl enzyme, which main part is  $\alpha$ -amylase enzyme, is quite effective in the diet of lactating cows. It is evidenced by the results of researchers <sup>[30,31]</sup>, claiming a tendency of increasing milk productivity while using it. The researchers <sup>[32]</sup> suggest that this enzyme can increase the productivity of animals by increasing the digestion of starch, while the amount of starch does not necessarily increase. However, other researchers have not found this effect determined milk productivity <sup>[33]</sup>, noticing that the absorption of nutrients has increased.

According to researchers <sup>[34,35]</sup> the use of arabinogalactane as a feed additive has a positive effect on the level of natural resistance of the body and on the formation of intestinal microflora. In a more detailed study of the use of arabinogalactane, the authors <sup>[36]</sup> noticed its structure of macromolecules, which consists of 95-99% of arabinogalactane and 1-5% of proteins and has prebiotic characteristics. On its base some prebiotic additives for supporting the growth of bifidobacteria and lactobacilli have been developed. Tests on calves conducted by the authors <sup>[37]</sup> defined that adding of arabinogalactane in the diet helps to improve the efficiency of nutrition and has a positive effect on the hematological status, increasing weight gain. However, the main mechanism of positive influence on newborn calves is its ability to provide a high level of humoral and cellular immunity. In the diet of animals, the presence of arabinogalactane contributed to the strengthening of adaptive and protective functions of the body, and it is also proved that such diets have anti-atherogenic properties <sup>[38]</sup>.

The study on feeding cows coniferous flour (50 g/head/day), pine nut shells (50 g/head/day), arabinogalactane (5 g/head/day) and amilosubtiline HCl enzyme (5 g/head/day) indicate that their complex use had a synergistic effect, resulting in increased milk yield by 9.9% ( $P < 0.05$ ), the mass fraction of fat - by 0.3% ( $P < 0.05$ ), the amount of basic fat content of milk - by 19.0%, as well as improved technological properties of milk and blood biochemical parameters.

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## GRATITUDE

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## STATEMENT OF AUTHOR CONTRIBUTIONS

EI - conducting an experiment, studying the components of a feed additive, examining the blood of animals, processing the data obtained, writing the manuscript of the article; OI - determination of the research topic, control of the analysis of results, statistical processing of research results, interpretation of research results; VT - development and conduct of an experiment, the study of blood and milk of animals, statistical processing of research results; LE - analysis of animal milk.

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