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## BIOLOGICAL SAFETY OF COWS' MILK UNDER THE CONDITIONS OF TECHNOGENIC AGRICULTURAL ECOSPHERE WHEN USING BIOLOGICALLY ACTIVE SUBSTANCES

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Article history: Received 12 November 2018 Received in revised form 12 February 2019 Accepted 14 February 2019 Available online 15 February 2019 <i>Keywords</i> : technogenic agricultural pollution; diary produce; chemical substances; biologically active substances; biological safety; Heavy metal.	These days environmental pollutions are occurred all over in the world. Unfavourable environmental background significantly affects animals' health and productivity. At the modern stage of development of dairy cattle breeding, quality of milk and new methods of agricultural production meeting latest sanitary and hygienic requirements have become very important. The article presents the data on the research on influence of a complex of biologically active substances on ecological safety of milk of the cows kept in the territories contaminated with Ni, Pb and Cd. It is stated that the usage of a complex of biologically active substances under the conditions of technogenic agricultural ecosystem significantly reduces excess iron status in cows' blood and milk and increases content of Zinc (Zn) and copper (Cu), so that it restores balance of essential microelements. The complex shows high adhesive properties regarding Nickel (Ni), Lead (Pb) and Cadmium (Cd), that are potentially dangerous elements. Content of Pb in cows' blood by the 14th and 28th days of research is proved to reduce by 1,5 times, and content of Cd – by 3 times, as compared with the original status. At the same time, content of Pb in cows' milk reduced by 9 times, and content of Cd – by 3 times. That fact proves the necessity to use a complex of biologically active substances, in order to provide biological safety of cows' milk.

## 1. INTRODUCTION

The issue of production of ecologically safe animal products is closely connected with continuously growing man-made contamination of environment with toxicants [1, 2, 6, 8]. The Ural area, where concentration of industry is 4-5 times higher than average level in the Russian Federation,



is considered to be the one of the most ecologically unsafe regions [8]. The growth of emissions into the atmosphere results in increased content of heavy metals salts, harmful chemical substances and their compounds in environmental compartments. Contamination of biosphere with ecotoxicants significantly affects level of animals' productivity, as well as biological value of animal products, including milk. Lately proportion of milk samples and dairy products not meeting hygienic standards according to health criteria has increased and been proved to be the highest among all the groups of food products. Because of unfavourable ecological situations in a number of the areas in Russia, development and usage of the latest biologically active complexes that make it possible to improve animals' autarcesis, reduce negative effect of technogenic factors, improve the quality and provide ecological safety of animal products, is becoming especially important and practically significant [3,4,5,7,9,10,11,12]. In this respect, the medications based on mineral elements, vitamins and various groups of organic acids are of special interest.

#### 2. AIM AND METHODS OF RESEARCH

According to the above-mentioned, the aim of the work was research on the effect of new combination of biologically active substances on biological safety of milk of the cows kept under the conditions of technogenic agricultural ecosphere. Scientific and production experiments were done at an agricultural enterprise with methods of animal welfare and breed and age distribution of animals typical for the Ural area, average level of productivity, and with high content of Ni and Cd in its environmental compartments. The objects of the research were blood and milk of black-and-white cows selected according to analogue principle, with body weight of 500–550 kg, two lactations. According to analogue principle, the cows were divided into two groups, 20 cows each. Feeding and welfare were similar.

In the course of experiment, the cows from the experimental group were given a complex of biologically active substances (mineral elements, vitamins – pyridoxin, vitamin C, cholecalciferol in micelle solution, carboxylic and amino acids) in the form of water slurry (1 litre per each cow a day), together with common feed stuff. A complex of biologically active substances was added to cows' diet for 28 days, starting with the first day of the second phase of lactation. Blood for the research was taken before the experiment, and then on the 7<sup>th</sup>, 14<sup>th</sup> and 28<sup>th</sup> day from the moment of the usage of the medication. Content of mineral elements in cows' milk was tested at the same time. Concentration of chemical elements (Fe, Cu, Zn, and Ni) in cows' blood and milk was measured by the method of atomic absorption spectrophotometry (GOST 26929–94 "Primary produce and food products"). Content of Pb and Cd, as potentially dangerous substances, was measured according to GOST 26932–86 "Primary produce and food products. Methods of Pb test» and GOST 26933–86 "Primary produce and food products. Methods of Cd test".

# 3. RESULTS OF RESEARCH

According to the results of the research (Table 1), it shows that when using a complex of biologically active substances there was a tendency towards reduced concentration of Fe in blood serum of the cows from the experimental group. Thus, on the 14<sup>th</sup> day of research, concentration of Fe in their blood was 17.85 mcmole/l (micromoles per litre) that was 12.99 % lower, as compared to the original level and 11.65 % lower, as compared to the cows from the control group. By the 28<sup>th</sup>

day that parameter was 1.6 times lower than the control one. It is worth to note that on the 7<sup>th</sup> day from the moment of the usage of a complex of biologically active substances content of Fe in cows' blood reduced by 10.46%. Probably, it was caused by an ability of elements in the complex to complex free ferrous ions with proteins, that improves bod defense reaction against possible toxic effect of free ferrous ions. In the blood of the cows from the control group content of Fe changed insignificantly and did not have any verified differences as compared with the original data during the whole course of research. It is worth to note that concentration of ions of the metal in blood of the animals from the experimental group was within phisiologically normal state, whereas in the control group on the 7<sup>th</sup> day of research it was 4.7 % more than the limit; on the 14<sup>th</sup> day – 1.16% more; on the 28<sup>th</sup> day – 1.47 times more (P<0.001).

<b>F1</b>	Standard	Group	Baseline value	Days of research		
Element				7	14	28
Fe	16.1-19.7	control	19.87±0.71	20.62±0.85	19.93±0.96	29.00±1.12
		experimental	20.17±0.64	18.26±0.90	17.85±0.77	18.10±1.20***
Cu	14.1-15.7	control	12.69±0.44	12.58±0.51	12.61±0.39	12.90±0.34
		experimental	12.73±0.52	14.21±0.38	$14.58 \pm 0.46$	14.97±0.30**
Zn	19.9-26.0	control	16.98±0.76	17.79±0.73	14.44±0.95	12.75±0.86
		experimental	17.24±0.68	20.82±0.65	22.05±1.22**	22.94±1.18**
Ni	0.12	control	2.25±0.18	2.45±0.12	2.37±0.17	2.13±0.32
		experimental	2.30±0.21	2.21±0.15	1.32±0.19*	0.49±0.09***
Pb	0.50	control	0.077±0.003	0.093±0.002	$0.096 \pm 0.004$	$0.088 \pm 0.005$
		experimental	0.075±0.002	0.062±0.001	0.048±0.002***	0.047±0.003***
Cd	-	control	0.093±0.003	0.085±0.003	$0.088 \pm 0.002$	0.083±0.003
		experimental	0.095±0.002	0.031±0.001***	0.030±0.001***	-

**Table 1**: Dynamics of mineral elements in cows' blood when using a complex of biologically active<br/>substances, mcmole/l ( $\tilde{O} \pm Sx$ , n = 20)

Note: here and further: \*P<0.05; \*\*P<0.01; \*\*\*P<0.001.

As for content of Cu, the opposite tendency was stated. Thus, in the blood of the cows from the control group, concentration of Cu was within the limits of 12.58–12.90 mcmole/l that was lower than the standards. In the group of the cows given a complex of biologically active substances added to their diet, there was verified increase of concentration of Cu by 16.04% as compared to the control group and by 17.59% as compared with baseline value (on the 28<sup>th</sup> day).

Similar tendency was characterized for Zn, that is an element responsible for immune system. Its content in the blood of the cows from the experimental group was within physiologically normal state and proved to increase on the 14<sup>th</sup> (1.53 times) and 28<sup>th</sup> (1.79 times) days of research as compared to the control figure and, respectively, 1.28 and 1.33 times, as compared to the original figure. In the blood of the cows from the control group content of Zn did not reach low limit of physiologically normal state at every stage of research, and its concentration, as compared to baseline value, reduced by 17.59% and by 33.18% on the 14<sup>th</sup> and 28<sup>th</sup> days of research respectively.

The blood of the cows from the control and experimental groups had excess content of Ni that was a few times more than the standards. When using a complex of biologically active substances in the diet of animals from the experimental group, concentration of Ni in blood (by original level of 2.30 mcmole/l) reduced up to 1.32 mcmole/l on the 14<sup>th</sup> day of research and up to 0.49 mcmole/l – on the 28<sup>th</sup> day of research. It is worth to note, that reduction of Ni in cows' blood by the end of the

research period caused increase of content of Zn in blood, because of formation of zink-thionine that is considered to be favourable in the course of correction by a complex of biologically active substances during chronic administration of heavy metal salts into cows' body, as synthesis of zinkthionines is essential for DNA synthesis for liver regeneration. A complex of biologically active substances shows high adhesive properties regarding to Pb and Cd, that are potentially dangerous substances for animal body.

The data in Table 1 show that on the 7<sup>th</sup> day of experiment content of Pb among the cows from the experimental group reduced by 20.97%, as compared to baseline value, and by the  $14^{th}$  and  $28^{th}$  day – by 1.5 times respectively. As compared to the control figures, this property was 1.48 - 1.93 lower. It seems that the effective elimination of Pb from blood was caused by balanced increase of Zn (which is a physiological antagonist of Pb) in cows' body. At the same time content of Pb in the blood of the cows from the control group stayed the same at every stage of research, but it was lower than the allowed standard.

In the blood of the cows from the control and experimental group some insignificant amount of Cd was found. However, concentration of that element changed differently, depending on the usage of biologically active complexes in animals' diet. In the control group of the cows, at every stage of research, concentration of Cd stayed the same and was 0.08 mcmole/l that was 12/5% lower than baseline value. In the blood of the cows from the experimental group content of Cd also stayed the same during the research period, but was 2.67 times lower than the control figures and 3 times lower than baseline value. It is worth to note that on the 28<sup>th</sup> day of research in the blood of the cows from the experimental group no presence of Cd was found, whereas in the control group its concentration was at the same level.

Analysis of the obtained results has proved significant effect of a complex of biologically active substances on mineral composition of cows' milk (Table 2). Thus, by the end of the experiment (the 28<sup>th</sup> day) an effect of a complex of biologically active substances caused reduction of Fe in cows' milk by 1.4 times, as compared to baseline and control value. On the 14<sup>th</sup> and 28<sup>th</sup> days of research content of Cu in the blood of the cows from the experimental group increased by 37,23 and 38,87% respectively, that was 29,64% and 1,5 times higher than control value at those stages of research.

Content of Zn in the milk of the cows from the experimental group was proved to have grown by the end of the experiment by 23.26% as compared to baseline value and by 1.45 times as compared to control value. The control group had the figure of the parameter lower than original concentration of Zn on the 14<sup>th</sup> and 28<sup>th</sup> days of research – by 15.20 and 18.16% respectively. In relation to toxic elements – Ni, Pb and Cd – the usage of a complex of biologically active substances showed high adhesive ability. Verified reduction of content of the above-mentioned elements are registered on the 14<sup>th</sup> and 28<sup>th</sup> days of research. On the 14<sup>th</sup> and 28<sup>th</sup> days of research concentration of Ni in the milk of the cows from the experimental group was respectively 1.75 and 5.38 times lower than original level; Pb – 9 times lower; and Cd – by 3 times. In the milk of the cows from the control group content of Ni, Pb and Cd at every stage of the experiment was at the level of baseline value. Reduction of Fe, Ni, Pb and Cd and increase of content of Cu and Zn improved quality properties of milk – increase of weight content of total protein, casein, fat, lactose and nonfat milk solids (MSNF). Thus, in the milk of the cows from the experimental group on the  $28^{\text{th}}$  day of research content of total protein increased by 2.9 %. In the control group in the course of the experiment the figure of this property reduced by 2.7% - 3.7%.

Element.	Group	Baseline value	Day of research						
mcmole/l			7	14	28				
Fe	control	32.29±1.12	32.72±1.65	31.84±1.48	32.61±1.59				
	experimental	31.98±1.37	29.39±1.30	28.46±1.26	22.78±1.44**				
Cu	control	4.61±0.45	4.76±0.37	4.52±0.41	3.99±0.35				
	experimental	4.27±0.52	5.32±0.29	5.86±0.23*	5.93±0.31**				
Zn	control	32.67±2.15	32.43±1.17	28.96±2.34	27.65±2.29				
	experimental	32.54±3.00	32.87±0.96	36.68±2.10	40.11±2.38*				
Ni	control	0.68±0.03	0.71±0.02	0.65±0.03	0.75±0.04				
	experimental	0.70±0.02	$0.44{\pm}0.03$	0.40±0.03***	0.13±0.02***				
Pb	control	0.10±0.001	$0.09 \pm 0.001$	0.08±0.001	0.09±0.001				
	experimental	0.09±0.001	0.04±0.001***	0.01±0.001***	0.01±0.001***				
Cd	control	0.03±0.002	0.03±0.003	0.03±0.001	0.04±0.002				
	experimental	0.03±0.001	0.01±0.001***	0.01±0.001***	-				

**Table 2**: Dynamics of mineral elements in cows' blood when using a complex of biologically active substances ( $\tilde{O} \pm Sx$ , n = 20)

By the end of the experiment in the experimental group concentration of the main milk protein – casein, that shows nutrition value of milk, - increased by 3.7 % as compared to baseline value. At the same time content of casein in the milk of the cows from the control group reduced insignificantly. The usage of biologically active complexes in the diet of the cows from the experimental group had an effect on content of fat in milk. Its content in cows' milk most significantly increased on the 28<sup>th</sup> day of research (8.9%). Along with increased content of fat in milk there was a tendency towards increased concentration of nonfat milk solids. Thus, content of MSNF in the milk of the cows given a complex of biologically active substances with baseline value of 8.00% by the 14<sup>th</sup> and 28<sup>th</sup> days of research increased by 1.38 and 1.88 %. Weight content of lactose in the milk of the cows from the experimental group during those periods increased by 2.7% and 2,9% respectively, as compared with baseline value.

#### 4. CONCLUSION AND RECOMMENDATIONS

The obtained results and stated tendencies prove the fact that the usage of a complex of biologically active substances under the conditions of technogenic agricultural ecosystem significantly reduces content of potentially dangerous substances (Ni, Pb, Cd) in cows' blood and milk, improves balance of such microelements, such Zn and Cu; increases concentration of total protein, casein, fat, lactose and MSNF. In order to improve sanitary quality and provide biological safety and full value of cows' milk kept under the conditions of technogenic agricultural ecosphere it is recommended to use this complex of biologically active substances in the form of water slurry in the amount of 1 litre per animal a day during the whole lactation period.

## 5. CONFLICT OF INTEREST

The authors confirm that the data presented here have no conflict of interest.



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