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ORIGINAL ARTICLE

Features of heavy metal excretion in dairy cows in agroecosystems around an industrial city and the production of environmentally safe milk

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The strained ecological situation has developed today in agroecosystems around industrial cities in almost all world countries. They are subject to increased anthropogenic pressure due to the use of agrochemicals and polluting emissions from industrial enterprises. Heavy metals Cd and Pb represent the most significant ecological threat to ecosystems. In such an ecological situation, the risk of xenobiotics entering the milk and its contamination by foreign matter significantly increases; they migrate easily in the components of the biosphere through trophic chains entering the organism of farm animals, incl. dairy cows, which negatively affects their health, ecological safety, quality of manufactured milk and human food. In four farms located around the industrial city, scientific and economic experiments were carried out on feeding cows with a specially developed adapted to the actual diets of feeding mineral-vitamin premix "MP-A" and subcutaneous injection of biologically active preparation "BP-9" to enhance the excretion of urinary heavy metals Cd, Pb, Cu, Zn and ensuring the production of high quality environmentally sound milk. In plants grown in farm-feeds that were part of the cows' diet, high levels of Cd, Pb, Cu, and Zn were detected. Getting into the body, toxicants from the gastrointestinal tract are absorbed into the blood, spread around the body, accumulate in organs and tissues, pass into urine and milk. The accumulation of Cd in the blood of test cows in control groups was on average from 77.94 to 101.20 nmol/L, Pb from 4.63 to 8.32 μmol/L. The transfer of Cd from blood to urine was on average 1.7 to 2.0%, Pb-5.4-7.3%. Elimination of heavy metal data from the body through the kidneys with urine is negligible and without the use of additional agents is unlikely. Acute research has shown that the liver, kidneys are "targets" for the action of pollutants. Partially the elements were accumulated in the spleen, muscle, and bone tissue with the milk. There was a chronic form of toxicosis. The applied antidote substances contributed to the exacerbation of heavy metal extermination from the body of animals and the restoration of its homeostasis. In other experimental groups of cows, the transfer of Cd from blood to urine averaged 3.9 to 9.5%, Pb-37.7-103.5%, respectively, in the third-from 7.1 to 12.7% Cd and 70.7-144.1% Pb. The combined effect of the antitoxin premix and the biopreparation in the third experimental group of cows gave a better result of removing toxic elements from the body. Enhanced excretion of pollutants can serve as proof of their high content in the body. The excretion of heavy metals in the urine of cows reflects the level of loading of the body that is caused by the eating of feeds from the excess of investigated elements. The premix and biopreparation developed blocked absorption of the pollutants in the gastrointestinal tract, strengthened the protective effect of the intoxicated organism, and facilitated the elimination of heavy metals with urine for such several migration activities of Pb, Cu, Zn, and Cd; have positively influenced the renal elimination of the excess of essential elements of Cu and Zn, not worsening the health of cows, but rather restoring homeostasis, improving the quality and ecological safety of the produced milk. Further research is aimed at developing more effective anti-nutritional substances.
Keywords: Premix, biopreparations, medicinal plants, cadmium, lead, copper, zinc, contaminated feeds, antidote substances, animal productivity.

Introduction

Agroecosystem is a natural-anthropogenic ecosystem; it is the result of human activity, an artificially formed system of plant and animal groups, has a weakly expressed self-regulation mechanism, predicted productivity is regulated by direct and indirect energy investments, after the termination of critical reduction of which the system degrades (Blancard & Martin, 2014; Gnatiuk, 2018). The degradation of agroecosystems also enhances the anthropogenic impact due to violation of the rules and regulations for the use of agrochemicals: mineral fertilizers and pesticides, and around industrial cities, in addition, emissions from enterprises, which leads to pollution with pollutants and xenobiotics, among which heavy metals are the most dangerous. As a result, negative changes occur in the health status of productive animals, dairy cows, products-milk may contain excess Cd, Pb, Cu, Zn, and other heavy metals and be environmentally hazardous (Song et al., 2004).

Heavy metals are among the priority environmental pollutants, monitoring of which is mandatory in all its components. The term "heavy metals" itself characterizes a broad group of substances; recently it has become widespread. In various scientific and applied scientific works, scientists interpret the meaning of this concept differently (Fersman, 1934-1937; Tsvetkova & Gunko, 2015). Numerous characteristics are used as membership criteria: atomic mass, density, toxicity, the prevalence in the environment, degree of involvement in natural and manufactured cycles. In works that highlight environmental pollution and environmental monitoring problems, heavy metals include more than 40 microelements with an atomic mass of over 40 atomic units. At the same time, conditions play an important role in the classification of heavy metals: their high toxicity to living organisms at relatively low concentrations, the ability to bioaccumulate, and biomagnification (Adriano, 2001; Brygadyrenko & Ivanyshyn, 2015; Tsvetkova & Gunko, 2015).

According to the Fersman classification (1934-1937), metals with a density of more than 8 g/cm^3 should be considered heavy. Formally, many elements correspond to the definition; however, according to scientists engaged in practical activities related to the organization of observations of the state and pollution of the environment, the combination of these elements is far from being equivalent as pollutants. Therefore, in many works (Tsvetkova & Gunko, 2015) there is a narrowing of the scope of the group of heavy metals, following the priority criteria, due to the direction and specifics of scientific research.

The differences in terminology are mainly related to the concentration of metals in the natural environment. On the one hand, the metal concentration can be excessive and even toxic, then this metal is "heavy"; on the other hand, when it is concentrated or deficient, it is referred to as trace elements. Thus, the terms "trace elements" and "heavy metals" are qualitative rather than quantitative categories, tied to extreme variants of the ecological situation (Tsvetkova & Gunko, 2015).

The excretion of heavy metals from the body of animals with feces and urine leads to an increased content of them in organic fertilizers (manure and compost), which also contain a significant amount of heavy metals (Gutyj et al., 2016). As a result of introducing such organic fertilizers into the soil, the concentration of heavy metals such as cadmium, lead, copper, zinc, iron, and manganese increases (Brygadyrenko & Ivanyshyn, 2015).

Studies have collected a large amount of data on the content of bulk and mobile forms of heavy metals in technogenically polluted landscapes in the soil (Bigalke al., 2017; Borah et al., 2018; Gunko et al., 2018). Heavy metals, especially cadmium and lead, accumulate in the soil in significant concentrations, which often exceed the established maximum permissible levels and, under appropriate environmental conditions, migrate to plants that are fed as feed to farm animals, and then enter milk and other products (for example, dry milk, where heavy metals are much more concentrated than in liquid milk (Jolly et al., 2017), thereby worsening its quality and environmental safety (Mamenko et al., 2010; Ren-ju et al., 2015; Tahir et al., 2017).

Pollution of the environment, its ecosystems with heavy metals Cd, Pb, Cu, Zn, negatively affect the body of dairy cows, leadsdairy cows' body, leading to the production of milk of poor quality and safety is an acute problem. Against the background of increased migration of heavy metals Cd, Pb, Ni, Cu from the soil, a decrease in the number of mobile forms of essential mineral elements such as Ca, P, Co is observed, which leads to their low content in the ration feed, as a result: their low content in the blood, the development of osteodystrophy in cows, deformation of the spine and horny covering of the hoof, partial lysis of the last pair of ribs, resorption of the last 2-3 caudal vertebrae (Slivinska et al., 2018). Mixed ligand complexes of zinc, manganese, cobalt, eliminating the deficiency of essential mineral elements in cows' body, improve mineral metabolism, thereby contributing to the growth of milk productivity (Bomko et al., 2018). The use of natural detoxicants of heavy metals, dry apple and dry beet pulp, in pigs' diets helped reduce Cd's accumulation in muscle tissue and animal slaughter products (Dyachenko et al., 2017). Therefore, the study of intoxication of the body of dairy cows with heavy metals, their excretion from the body, and the use of particular antidote substances (premix and biological product) to enhance the excretion of pollutants with feces and urine and in a smaller amount with milk, and thus the production of environmentally friendly milk is relevant (Mamenko & Portjannyk, 2010).

Intakes of Cd and Pb are associated with environmental risks to the body through cumulative toxicity and negative effects on internal organs and systems (Canty et al., 2014; Hashemi, 2018; Roggeman et al., 2014). In animals, the intensity of growth and productivity decreases. The accumulation of the aforementioned heavy metals in environmental components and agroecosystems increases the risk of their intake into cows' body and thereby poses a threat to human and animal health (Fischer et al., 2011; Rezza et al., 2018; Rahimi 2013). In chronic cadmium intoxications (nephrotoxicity, hepatotoxicity, immunotoxicity, osteotoxicity), oxidative stress of liver and kidney cells occurs, damage to them and DNA, which can lead to carcinogenesis and oncological diseases (Liu et al., 2009).

The existing environmental situation negatively affects the efficiency of the livestock industry and, in particular, dairy farming. That is why an in-depth study of the processes of elimination of heavy metals from the body of animals and the development of special antitoxic premixes and biological products that would not only enhance the excretion but also ensure an improvement in the health of cows, increase their productivity and the safety of milk is necessary.

The aim of the research is a quantitative assessment of the elimination of toxic and essential heavy metals from urine and milk of dairy cows under conditions of intoxication of the animal body with Cd, Pb, Cu, Zn against the background of feeding a specially developed mineral-flax-vitamin premix "MP-A" and subcutaneous injection of a biological product " BP-9 "of vegetable origin, while at the same time dyeing the quality and environmental safety of milk.

Materials and Methods

In the farms located around the industrial city, scientific and economic experiments were carried out on dairy cows of Ukrainian black and red-and-white dairy breeds. For the experiments, 126 cows were selected with the silage-hay-concentrate type of feeding, 63 from the silo-silage type, 36 - from the silage-root crops, and 195 from the silage-haylage type of feeding, respectively. The experimental population was divided into three groups: the first control and the second and third research groups. Cows of all groups were fed feed with the content of heavy metals Cd, Pb, Cu, Zn above the established maximum permissible concentrations. Animals of the second experimental group received an additional special antitoxic mineral-vitamin premix "MP-A", and the third-a premix and subcutaneous injection of the biological product "BP-9" includes an extract of nine medicinal plants. The average live weight of cows is 500-545 kg, the average daily milk yield is 14.0-14.8 kg, which per lactation averages 4270-4514 kg of milk. The comparative period was 42 days. The cows selected by the method of analogs in terms of live weight, productivity were in the same conditions of feeding and keeping. The trial period lasted 120 days.

According to the method, mineral-vitamin premix and biologically active preparation "BP-9"were developed according to the method (Portjannyk, 2002). Biological product "BP-9" an extract from 9 medicinal plants. 100 ml of the preparation contains *Schisandra chinensis*-15 ml, Chamomile (pharmacy, peeled) (English Chamomile)-3 ml, *Eleutherococcus senticosus*-15 ml, medicinal sage (*Salvia officinalis*)-17 ml, common barberry (English Berberis)-12 ml, alfalfa (English Alfalfa)-5 ml, kidney tea (*Orthosiphon stamineus*)-15 ml, sea buckthorn (*Hippophae rhamnoides*)-15 ml, *Verbena officinalis*-3 ml. Subcutaneous injection was applied. The dose of administration of the preparation "BP-9"-20 ml/day with the division of this norm into two, 10 ml each in the morning and at intervals of 12 hours in the evening. The frequency of administration of the preparation is 5 times a month, with an interval of administration - once every 6 days. The preparation for injection was used for 42 cows of the third research group according to the silage-silage-concentrate type of feeding 16.8 l (20 ml × 20 (5 times a month × for 4 months of the experiment)=400 ml) silage-haylage-21 cows-8.4 l silage-root crop-12 heads-4.8 l silage-hay-65 heads-26 liters, respectively. A total of 56 liters of the preparation were made and used. Release form-glass bottles with a volume of 250 ml. The biological product was manufactured under sterile conditions in a production laboratory according to the method (Hmel'nic'kij et al., 1994; Sokolov et al., 2002). In the second and third research groups, the rations for feeding dairy cows developed by us were additionally balanced, adapted to the actual daily ration, with a special antitoxic mineral-vitamin Premix "MP-A". The premixes were developed taking into account the synergistic, antagonistic action of macro-, microelements concerning cadmium, lead, copper and zinc, taking into account their concentration per 1 kg of dry matter, which allowed to develop a methodological approach to rationing the content of heavy metals in the diet and the corresponding amount (ratio) essential macro-, microelements, partial deficiency of which was observed in the diets of cows in the control groups. The composition of the mineral-vitamin premix "MP-A" included the following mineral elements: P, Ca, Mg, S, Fe, Mn, Co, J, Se, etc.; vitamins: A, D, E, B2, B3 (niacin, PP), B4, B6, C, H (Biotin) and one of the sulfur-containing amino acids. Feeding the premix based on the introduction of 1% into the diet (Podobed et al., 1996) was: for cows with silage-root crops and silage-hay type of feeding 250 g per head per day, silage-hay-concentrate - 290 g, silage- Hay - 255 g, respectively. Premix "MP-A" was fed for 24 heads of dairy cows of the second and third experimental groups with the silage-root type of feeding 720 kg (250 g per head per day × 24 chapters × 120 days of experience) silage-hay -130 heads -3900 kg; silage-haylage-42 heads -1285.2 kg silage-haylage-concentrate -84 heads -2923.2 kg, respectively. There are a large number of regional firms on the market that, to order, according to the appropriate diet and recipe, can manufacture and sell such a premix to an agricultural enterprise, and this makes it quite convenient to use, which we took into account when conducting experiments. Within the framework of commercial contracts, all experimental farms cooperated with such a company. Therefore the production and supply of the premix along with other products is a standard business activity.

The recipe (formula) of the "MP-A" premix, the "BP-9" preparation, the methodology for the development of the mineral and vitamin premix "MP-A" adapted to the actual daily rations belong to the authors of this publication A. Mamenko and C. Portyannyk (Ukraine).

Biochemical analysis of samples of plant and animal origin: feed, blood, internal organs and tissues, urine, and milk for the content of macro- and microelements, including heavy metals, were carried out by atomic absorption spectrophotometry (spectrophotometer AAS-30) (Praice, 1972). The results obtained were compared with physiological norms for blood (Skukovsky, 1987), for urine (Levchenko et al., 2002). Control over the quality and environmental safety of milk was carried out following GOST 3662-97 (Mamenko et al., 1997), as well as taking into account the requirements of international quality standards (Regulation (EU) No. 853/2004 and No. 1881/2006).

Together with determining the absolute concentrations of metals in biosubstrates, we calculated their relative indicator according to the method (Stus et al., 2010); it is considered the index of renal migration (IRM). We performed this indicator, not in arbitrary units but simply in% about the concentration of metal in the blood as an integral internal environment of the animal organism and a leading source at the beginning of migration. The indicator allows us to reveal more deeply the corresponding links of the mechanism of removal of heavy metals from the body and to carry out a comparative assessment of the elimination qualities of toxicants and microelements when using the antidote substances we have developed - a premix and a biological product.

All manipulations with animals were carried out following the European Convention for the Protection of Vertebrate Animals used for Experimental and Scientific Purposes (Strasbourg, 1986).

The analysis of the data was carried out considering the peculiarities of the results obtained in the study: the size of the sample and the type of distribution of the data, the nature of the variances. For each sample, the mean value of the characteristic in the sample (M) and the standard deviation (SD) were calculated, the estimate is given as M ± SD. Disagreements between mean values were considered statistically significant at $P<0.05$. The calculation was carried out using the STATISTICA software package version 10.0 for the Windows 7 operating system.

Results

Environmental pollution by emissions from industrial enterprises of the city and a gas condensate station (CNG filling station) with the simultaneous use of mineral fertilizers and pesticides led to soil acidification in agricultural systems of farms, which caused increased migration of heavy metals from soil into plants, as a result, into the feed of the main ration of cows of all experimental groups. The feed was found to exceed the maximum permissible concentration for cadmium, lead, copper, and zinc. The content of Cd in the ratio of cows with silage-root crops type of feeding exceeded the established maximum permissible concentrations on average 2.1-3.2 times, Pb-2.4-5.7 times, Cu-1.4-2.3, and Zn-1.2-2.4 times, respectively. The most significant excess of the maximum permissible concentration for Cd and Pb was found in cereal-legume hay (3.2 and 5.7 times), for Cu-in maize mats (2.3 times), Zn-n wheat straw (2.4 times). The concentration of heavy metals in the feed of cows with other types of feeding fluctuated, which is due to the different content of mobile forms of toxicants in the soil and the location of the land where the plants were grown, depending on the distance to the industrial center, highways, CNG filling stations. Therefore, in cows' feed with a silage-hay type of feeding, the highest content of Cd, Pb, Cu, Zn with exceeding the maximum permissible concentrations was found in fodder beets, respectively, in 2.5; 3.4; 3.8; and 4.1 times. It is generally known that plants accumulate heavy metals in the root system to a greater extent, that is, in the part that is in the soil, slightly less heavy metals enter the vegetative system; therefore, of all feeds, it was fodder beets that had the highest level of contamination in all studied elements compared to other feeds. In the feed grown on agricultural land, where cows are fed a silage-haylage-type diet, in addition to exceeding the maximum permissible concentrations for the content of Cd, Pb, Cu, Zn, in comparison with other experiments, high content of zinc in oats and peas was recorded on average in 6.3-6.8 times. Pea bran had the highest cadmium content and the lead among other feeds, and cereal-legume hay was distinguished by the highest content of copper (3.9 times).

Thus, according to the level of contamination of feed rations such as feeding cows, they are arranged as follows (in descending order): Cd contamination of silage-root crops → silage-haylage → silage-hay → silage-haylage-concentrate; Pb silage-haylage-concentrate → silage-root crops → silage-haylage → silage-hay; Cu silage-haylage-concentrate → silage-haylage → silage-hay → silage-root crops; Zn silage-haylage-concentrate → silage-haylage→ silage-hay → silage-root crops.

The average concentrations of heavy metals in blood and urine established by us are the amount of their transition from blood to urine, taking into account, physiological norms are given in Tables 1 and 2.

Animal Feeding Type	Metal	Actual concentration								
		Blood			Urine			Transition, %		
		1 control	2 experiment	3 experiment	1 control	2 experiment	3 experiment	1 control	2 experiment	3 experiment
1	2	3	4	5	6	7	8	9	10	11
Silage-root crops	Cd, (blood-nmol/l); (urine-µmol/l)	98.34 ± 1.03	79.11 ± 2.90 **	49.19 ± 2.41 **	1.90 ± 0.17	3.05 ± 0.22 **	3.48 ± 0.40 **	1.9	3.9	7.1
	Pb, µmol/l	8.32 ± 1.65	3.02 ± 0.99 **	1.98 ± 0.16 **	0.46 ± 0.15	1.27 ± 0.14 **	1.40 ± 0.07 **	5.5	42.1	70.7
	Cu, µmol/l	28.93 ± 1.28	18.04 ± 0.68 **	18.89 ± 0.83 **	0.98 ± 0.19	1.84 ± 0.12 **	1.97 ± 0.17 **	3.4	10.2	10.4
	Zn, µmol/l	29.85 ± 1.11	22.81 ± 1.32 **	21.19 ± 1.45 **	1.05 ± 0.15	2.39 ± 0.40 **	2.61 ± 0.42 **	3.5	10.5	12.3
Silage-hay	Cd, (blood - nmol/l); (urine -	101.20 ± 3.17	54.29 ± 2.64 **	40.72 ± 1.98 **	1.73 ± 0.33	3.57 ± 0.41 *	3.82 ± 0.33 *	1.7	6.6	9.4

	μmol/l)									
	Pb, μmol/l	6.54 ± 0.45	4.01 ± 0.64 **	1.38 ± 0.28 **	0.36 ± 0.10	1.51 ± 0.19 **	1.57 ± 0.16 **	5.5	37.7	113.8
	Cu, μmol/l	30.62 ± 1.24	17.07 ± 0.54 **	16.07 ± 0.98 **	0.91 ± 0.15	2.34 ±0.42 **	2.69 ± 0.37 **	3.0	13.7	16.7
	Zn, μmol/l	17.06 ± 0.40	10.04 ± 0.23 **	9.82 ± 0.24 **	0.90 ± 0.07	3.46 ± 0.28 **	3.51 ± 0.33 **	5.3	34.5	35.7
Silage-haylage	Cd, (blood - nmol/l); (urine - μmol/l)	81.17 ± 0.60	48.19 ± 0.73 **	41.61 ± 1.05 **	1.63 ± 0.30	3.54 ± 0.50 **	3.96 ± 0.24 **	2.0	7.3	9.5
	Pb, μmol/l	5.74 ± 0.32	2.07 ± 0.16 **	1.87 ± 0.09 **	0.31 ± 0.04	1.69 ± 0.19 **	1.76 ± 0.09 **	5.4	81.6	94.1
	Cu, μmol/l	27.62 ± 0.44	16.07 ± 0.37 **	17.13 ± 0.30 **	0.89 ± 0.11	2.63 ± 0.23 *	2.84 ± 0.23 *	3.2	16.4	16.6
	Zn, μmol/l	26.52 ± 0.53	18.53 ± 0.44 **	17.11 ± 0.39 **	1.01 ± 0.13	4.60 ± 0.25 **	4.75 ± 0.25 **	3.8	24.8	27.8
Silage-haylage-concentrat	Cd, (blood - nmol/l); (urine - μmol/l)	77.94 ± 0.99	40.64 ± 0.54 **	32.14 ± 0.55 **	1.40 ± 0.16	3.86 ± 0.30 **	4.07 ± 0.23 **	1.8	9.5	12.7
	Pb, μmol/l	4.63 ± 0.37	1.72 ± 0.17 **	1.27 ± 0.22 **	0.34 ± 0.12	1.78 ± 0.21 **	1.83 ± 0.30 **	7.3	103.5	144.1
	Cu, μmol/l	25.16 ± 0.59	16.03 ± 0.40 **	14.16 ± 0.62 **	0.91 ± 0.15	3.34 ± 0.26 *	3.57 ± 0.35 *	3.6	20.8	25.2
	Zn, μmol/l	28.94 ± 1.60	18.12 ± 0.96 **	16.41 ± 0.65 **	1.11 ± 0.14	4.61 ± 0.64 **	4.82 ± 0.32 **	3.8	25.4	29.4

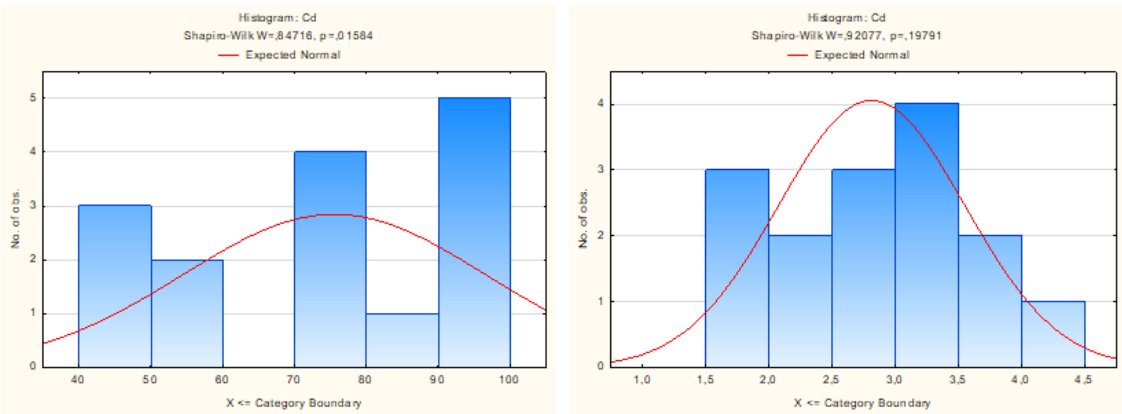
Note: the degree of reliability in comparison with the data of the control group *P<0.05; **P<0.01; n=5.

Table 1. The concentration of metals in biological media and the amount of their transition from blood to urine (M ± SD).

Metal	**Standard concentrations**		
	Blood	**Urine**	**Transition, %**
Cd, (blood - nmol/l); (urine - μmol/l)	20-50	0.89	1.8-4.5
Pb, μmol/l	up to 2	0.22	11
Cu, μmol/l	12.6-18.9	0.79	4.2-6.3
Zn, μmol/l	15.4-23.0	0.86	3.7-5.6

Table 2. Standard concentrations of metals in biological media and the amount of their transition from blood to the urine.

To establish the correspondence of the obtained data to the law of "normal" distribution (Gaussian), we applied the Shapiro-Wilk's W test, which is considered the most powerful, especially in small samples (n<50) of independent groups. The content of cadmium in the blood and urine of cows with silage-root crops type of feeding is not subject to the law of normal distribution (Fig. 1).



a b

Fig. 1. Analysis result of the distribution of the studied parameters (the content of Cd in the blood (nmol/l) (a) and urine (μmol/l) (b) of cows) using Shapiro-Wilk's W test.

The Shapiro-Wilk's W test made it possible to determine the appropriate method for further analysis of the comparison of samples. Considering that the sample size is insignificant, the most effective way to apply the nonparametric analysis of variance is Kruskal-Wallis (or H-test) (Kruskal-Wallis ANOVA) (Fig. 2).

Depend.: Cd	Kruskal-Wallis ANOVA by Ranks; Cd (Spreadsheet1) Independent (grouping) variable: Group Kruskal-Wallis test: H (2, N= 15) =12,50000 p =,0019			
	Code	Valid N	Sum of Ranks	Mean Rank
1	1	5	65,00000	13,00000
2	2	5	40,00000	8,00000
3	3	5	15,00000	3,00000

b

Depend.: Cd	Kruskal-Wallis ANOVA by Ranks; Cd (Spreadsheet1) Independent (grouping) variable: Group Kruskal-Wallis test: H (2, N= 15) =11,18000 p =,0037			
	Code	Valid N	Sum of Ranks	Mean Rank
1	1	5	15,00000	3,00000
2	2	5	43,00000	8,60000
3	3	5	62,00000	12,40000

Fig. 2. The value of the error P for the null hypothesis that the content of Cd in the blood and urine of cows in different test groups does not differ, in our case P<0.05 -the studied groups are statistically significantly different from each other are given.

Fig. 3 shows indicators of descriptive statistics.

Variable	Valid N	% Valid obs.	Mean	Median	Sum	Minimum	Maximum	Lower Quartile	Upper Quartile	Std.Dev.	Standard Error
1 Group	5	33,33333	98,34400	98,76000	491,7200	96,89000	99,51000	97,74000	98,82000	1,029335	0,460332
2 Group	5	33,33333	79,11200	78,71000	395,5600	75,91000	83,62000	77,52000	79,80000	2,904491	1,298928
3 Group	5	33,33333	49,19000	49,26000	245,9500	45,74000	52,01000	48,21000	50,73000	2,407893	1,076843

Descriptive Statistics (Spreadsheet1)

b

Variable	Valid N	% Valid obs.	Mean	Median	Sum	Minimum	Maximum	Lower Quartile	Upper Quartile	Std.Dev.	Standard Error
1 Group	5	33,33333	1,904000	1,820000	9,520000	1,730000	2,150000	1,810000	2,010000	0,171697	0,076785
2 Group	5	33,33333	3,052000	3,150000	15,26000	2,720000	3,250000	2,940000	3,200000	0,219932	0,098356
3 Group	5	33,33333	3,484000	3,510000	17,42000	2,980000	4,040000	3,260000	3,630000	0,398535	0,178230

Descriptive Statistics (Spreadsheet1)

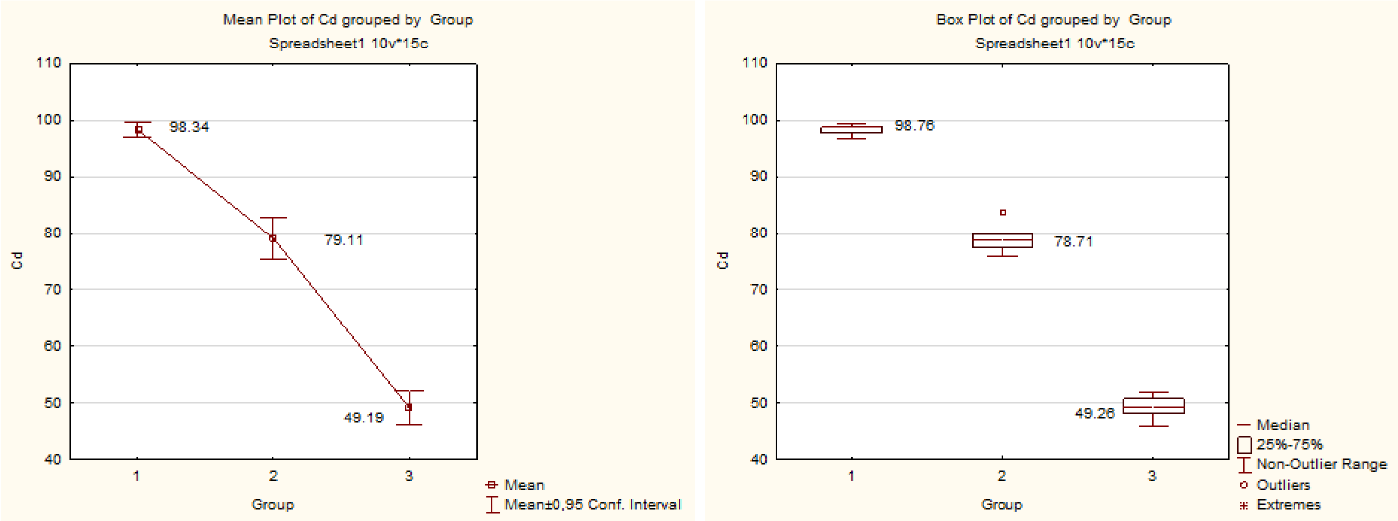
Fig. 3. Descriptive statistics of the studied indicators: arithmetic means M (Mean), standard deviation SD (Std. Dev.), median (Median), lower and upper quartiles (Lower Quartile; Upper Quartile), and other Cd contents in blood and urine of cows of three experimental groups with the silage-root type of feeding.

Analysis of the data obtained indicates that cadmium, according to the average data, is determined in the blood of cows from the control groups with the silage-haylage-concentrate type of feeding at concentrations from 77.94 ± 0.99 nmol/l to 101.20 ± 3.17 nmol/l in cows with the silage-hay type of feeding, and lead-from 4.63 ± 0.37 μmol/l in cows with a silage-haylage-concentrate type of feeding to 8.32 ± 1.65 μmol/l in cows with a silage-root crop ration.

At the same time, the process of accumulation of heavy metals in the body of dairy cows is observed, as a result of feeding feeds with an increased content of pollutants, which is accompanied by a corresponding increase in the content of elements in the blood of the first control groups of animals in all four experiments on Cd and Pb, and in cows with silage-root crops by the type of feeding not only for Cd and Pb but also for Cu and Zn.

In particular, the content of Cd in the blood of dairy cows of the control group with silage-root type of feeding exceeded the established physiological norms by 1.97 times, 2.02 times in animals with silage-hay, 1.62 times-from silage-haylage, and 1.56

times in cows with silage-haylage-concentrate feeding type, by Pb-at 4.16; 3.27; 2.87 and 2.32 times, respectively. Considering such laboratory blood tests, it is possible to confidently say that cows' body is experiencing a toxic shock (Fig. 4 and 5).



a b

Fig. 4. Diagram of mean values (a) and quartile diagram (b) of Cd content in the blood (nmol/l) of cows of three experimental groups with silage-root crops type of feeding.

The concentration of cadmium in the urine of cows of the first control groups ranges from 1.4 µmol/l in animals with a silage-haylage-concentrate type of feeding to 1.9 µmol/l - from a silage-root crop, which exceeds physiological norms, respectively, in animals with a silage-root crop type of feeding at 2.13 times, silage-hay - 1.94, silage-haylage - 1.83 and silage-haylage-concentrate-1.57 times, respectively, for Pb-2.09; 1.64; 1.41 and 1.55. The indicator of the Cd transfer from blood to urine averages from 1.7 to 2.0%, Pb-5.4-7.3%. That is, the elimination of heavy metals from the body with urine is negligible. The likelihood of an increase in the excretion of all heavy metals studied by us in the urine without the use of additional funds is unlikely. Therefore, being distributed by pollutants in the body of cows, it accumulates in various organs and tissues, in particular, the liver, kidneys, as organs "targets", partly in the spleen, muscle, and bone tissue, and is excreted in milk.

A similar situation is with other elements Pb, Cu, Zn. Thus, chronic intoxication of animals is observed, confirmed by the results of an acute experiment. Analysis of muscle tissue, internal organs for the content of Cd, Pb, Cu, Zn, showed that not only milk is impressed by heavy metals but also muscle tissue, especially internal organs directly involved in the neutralization and elimination of heavy metals. The highest accumulation of heavy metals was observed (in descending order): according to Cd-kidneys, muscles, liver, lungs, spleen, heart, and bones.

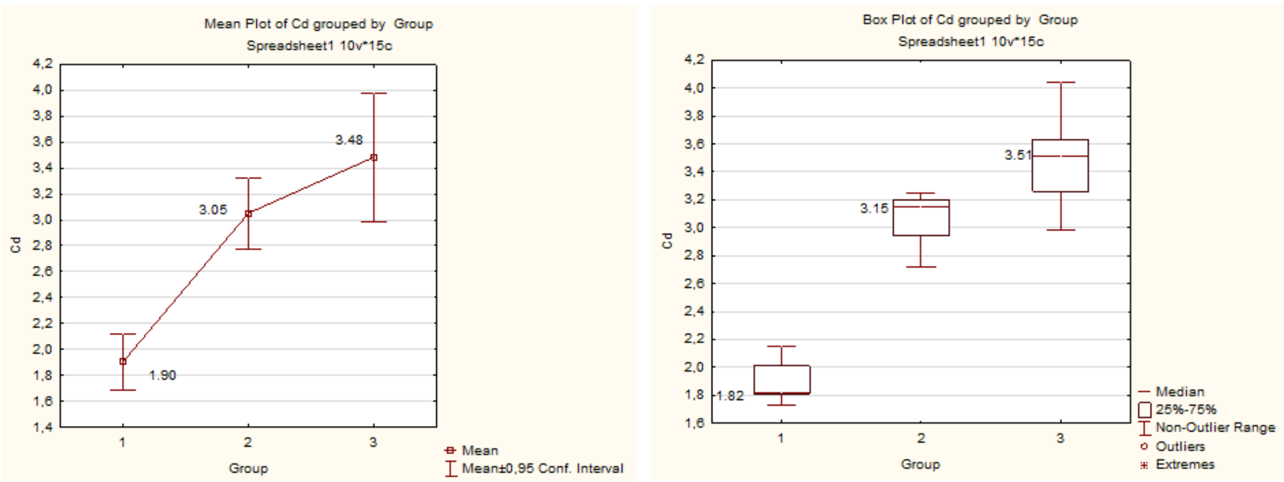
The premixes developed by us are adapted to the actual diets of the cows. During the balancing of rations, the deficit in essential macro-, microelements in the forages of the main diet of cows was investigated in all four experiments. At the same time, the content of heavy metals Cd, Pb, Cu, Zn in some feeds included in the diet exceeded the established maximum permissible concentrations. Based on this, all diets are maximally balanced in terms of macro- and microelement composition, taking into account the mechanism of absorption and the dynamics of movement of heavy metals, especially Cd and Pb, in the body of animals, in the premixes, we included antagonists of the studied ecotoxicants in an amount sufficient to block their absorption by the level of the gastrointestinal tract. The introduction of vitamins into the composition of the premix helped restore the homeostasis of the cows' organism and improve the functioning of organs and systems affected by the oligodynamic action of heavy metals. As a result, we managed to positively affect and reduce the content of heavy metals in the milk of cows in all farms for optimal milk quality and animal productivity. The dynamics of a decrease in the Cd content in milk of dairy cows with silage-haylage-concentrate feeding was 0.053 ± 0.019 mg/kg in the control group, 0.024 ± 0.009 in the second experimental group, and 0.014 ± 0.004 mg/kg in the third experimental group, and Pb 1.794 ± 0.165 mg/kg, 0.331 ± 0.064, and 0.032 ± 0.008 mg/kg, respectively ($p<0.01$) (Table 3).

Animal feeding type	**Mineral elements**	**Cow groups**		
		1 control	**2 experiment**	**3 experiment**
Silage-root	Copper, mg/kg	2.47 ± 0.38	0.31 ± 0.14	0.29 ± 0.09
	Zinc, mg/kg	7.06 ± 0.32	6.01 ± 0.27	4.32 ± 0.84
	Cadmium, mg/kg	0.087 ± 0.008	0.031 ± 0.005	0.018 ± 0.002

	Lead, mg/kg	1.835 ± 0.093	0.614 ± 0.085	0.014 ± 0.003
Silage-hay	Copper, mg/kg	2.54 ± 0.42	0.57 ± 0.26	0.32 ± 0.06
	Zinc, mg/kg	9.93 ± 0.72	6.14 ± 0.56	4.17 ± 0.62
	Cadmium, mg/kg	0.09 ± 0.085	0.031 ± 0.008	0.011 ± 0.003
	Lead, mg/kg	1.641 ± 0.253	0.515 ± 0.064	0.027 ± 0.012
Silage-haylage	Copper, mg/kg	2.36 ± 0.39	0.28 ± 0.09	0.27 ± 0.15
	Zinc, mg/kg	7.93 ± 0.23	4.02 ± 0.16	3.51 ± 0.39
	Cadmium, mg/kg	0.068 ± 0.017	0.017 ± 0.004	0.012 ± 0.002
	Lead, mg/kg	1.734 ± 0.148	0.016 ± 0.004	0.014 ± 0.004
Silage-haylage-concentrate	Copper, mg/kg	2.63 ± 0.42	0.34 ± 0.12	0.35 ± 0.17
	Zinc, mg/kg	8.74 ± 0.40	4.97 ± 0.30	3.87 ± 0.20
	Cadmium, mg/kg	0.053 ± 0.019	0.024 ± 0.009	0.014 ± 0.004
	Lead, mg/kg	1.794 ± 0.165	0.331 ± 0.064	0.032 ± 0.008
Note: the degree of reliability in comparison with the data of the control group is P<0.01; n=5.				

Table 3. Mineral elements and heavy metals in cows' milk at the end of the research period (M ± SD).

At the same time, the maximum permissible concentration of cadmium content according to the DSTU 3662-97 standard is set at 0.03 mg/kg, lead 0.1 mg/kg, and Regulation (EU) No. 1881/2006 at 0.02 mg/kg, copper-0.26-0.35 and zinc-3-5 mg/kg, respectively. Table 3 also shows the total content of mineral elements in the milk of experimental cows, heavy metals in all conducted experiments on different types of animal feeding.



a b

Fig. 5. Diagram of mean values (a) and quartile diagram (b) of Cd content in urine (μmol/l) of cows of three experimental groups with silage-root crops type of feeding.

Thus, acting in a complex premix+preparation, we were able to significantly increase the removal of heavy metals from the body of dairy cows of the second and third research groups in all farms with different types of animal feeding. The index of Cd transfer from blood to urine averaged in the second experimental group from 3.9 to 9.5%, Pb-37.7-103.5%, in the third-from 7.1 to 12.7%, and 70.7-144.1%, respectively. So, the combined action of the premix and the preparation gave a slightly better result in removing elements from the body than the action of the premix itself, since the transition of heavy metals from blood to urine according to standard concentrations (Table 2) is 1.8-4.5% for Cd, and 11 for Pb %. Enhanced renal excretion of cadmium and lead can serve as evidence of a higher level of metals in the body of animals due to their unimpeded intake with feed ration and rapid absorption into the bloodstream in the gastrointestinal tract.

Discussion

Anthropogenic impact on agroecosystems around industrial cities, through the intensification of traditional farming (excessive use of pesticides, mineral fertilizers), unfortunately, is increasing in different countries of the world (Bigalke et al., 2017). The ingress of heavy metals into the soil can lead to the accumulation of concentrations of undesirable agricultural land, endanger fertility, the transfer of such pollutants as cadmium and lead from the soil to plants that are used for animal feed, are included in the diet of any type of feeding, can complicate the production of high acid milk, and hence raw materials for food production. An increased concentration of cadmium and even uranium in the upper fertile soil layer of agricultural land, after applying phosphorus fertilizers, the introduction of sludge from wastewater, was observed (Bigalke et al., 2017). We have investigated more than 216 agricultural land plots in different regions of Switzerland, with mineral fertilizers becoming the dominant source of heavy metal pollution among all, taking into account precipitation from the atmospheric air (Bigalke et al., 2017).

Heavy metals are divided into toxic-lead and cadmium, conventionally non-essential-nickel and essential-copper, zinc, and chromium. Today, with 92 elements found in nature, 81 are found in the human body. At the same time, 15 (Fe, I, Cu, Zn, Co, Cr, Mo, Ni, V, Se, Mn, As, F, Si, Li) are recognized as vital, that is, essential or non-essential washing as nickel. However, they can negatively affect plants, animals, and humans if the concentration of their available forms exceeds the permissible limits. Cd, Pb, Sn, Rb are considered conditionally necessary since, most likely, elements that are not very important for plants and animals and are hazardous to human health, even at relatively low concentrations (Lu et al., 2013). For a long time, interest in geochemical anomalies and endemics of natural origin caused by these anomalies prevailed in biogeochemical studies of trace elements. For example, scientists (Gribovsky, 2000) diagnosed nickel toxicosis enzootic diseases in regions polluted by industrial emissions. It was found that excessive intake of nickel into the body of animals leads to damage to the gastrointestinal tract, liver, heart, blood vessels, brain, retina. Nickel has been insufficiently studied from an ecological and physiological point of view and cadmium, an unidentified range of animal tolerance to the action of this element on their body. More and more studies are devoted to studying the pathogenic effect of this heavy metal, like many others, and primarily as a feed contaminant. However, due to the rapid development of industry and global technogenic pollution, anomalies of elements, mainly heavy metals of industrial origin, began to attract the most significant attention. Already, in many world regions, the environment is becoming more and more chemically "aggressive". In recent decades, the main objects of ecological research have become the territories of industrial cities and the adjacent lands (Patra et al., 2008; Lu et al., 2013), which we have done as well. Many scientists have found that the influence of heavy metals is quite diverse and depends on their concentration in the environment and the degree of need for microorganisms, plants, animals, humans.

Cadmium exhibits toxic effects on several organs and systems, including the cardiovascular, reproductive, excretory (especially kidneys, liver), respiratory, musculoskeletal system (causes osteodystrophy), hematopoiesis (Gutyj et al., 2016). The most hazardous effects include the carcinogenic and mutagenic effects of this element (Gutyj et al., 2016).

There is a large amount of information in the literature on the effect of acute and chronic forms of cadmium toxicosis in humans and experimental animals (Gutyj et al., 2016; Salvatori et al., 2004). The results of many studies indicate significant differences in the metabolic effects of one-time high doses and long-term exposure to low doses of cadmium. Under conditions of intoxication of the animal body with cadmium compounds, anemia, suppression of the immune system's functional state, and other disorders of the processes of hematopoiesis occur (Gutyj et al., 2016).

According to (Neathery & Miller, 1975), most cadmium, lead, mercury accumulate in the kidneys and liver of animals - up to 50%. Our experience, the highest cadmium content - 38%, was found precisely in the kidneys and liver.

It is worth noting that the acute form of cadmium toxicosis, sometimes with a fatal outcome, rarely occurs today, but the syndrome of the chronic form of toxicosis is observed much more often. Scientists note this in their works (Gutyj et al., 2016). Clinical signs of chronic animal poisoning are accompanied by a sharp decrease in feed intake, a decrease in body weight, a slowdown in animal growth, impaired renal function, proteinuria, liver dysfunction, anemia, and an increase in neonatal mortality.

Feeding the cows of the second and third experimental groups specially developed by us antitoxic mineral and vitamin premix "MP-A" and an additional subcutaneous injection of the biological product "BP-9" in the third experimental group changed the situation described above for the better.

Developing these antidote substances, we set ourselves the task of not only achieving an increase in the excretion of dangerous heavy metals, such as cadmium and lead, from the body of dairy cows but also at the same time showing its homeostasis and thereby testing the effectiveness of the premix and preparation in scientific and economic experiments.

Biopreparation "BP-9" is an extract from the corresponding set of medicinal plants with biologically active substances safe for the body, in contrast to the long-known synthetic preparations such as unitiol. For the production of the biological product "BP-9", we selected nine medicinal plants: Chinese magnolia vine, medicinal chamomile (pharmacy, peeled), Eleutherococcus prickly, medicinal sage (pharmacy), common barberry, alfalfa, kidney tea, sea buckthorn, buckthorn.

The overwhelming majority of medicinal substances can penetrate cell membranes through passive transport or active special transport systems. Fat-soluble substances penetrate the membranes through passive diffusion. The transport intensity is directly proportional to the concentration of the substance in the membrane and the fat-water distribution coefficient. Non-polar and polar substances, poorly soluble in lipids, penetrate through water pores (filtration) due to a hydrostatic and osmotic difference on both sides of the membrane.

Each medicinal plant, which was included in the composition of the biological product, has its biological role; we will dwell on some of them in detail - this is Chinese magnolia vine and common barberry.

Chinese magnolia vine extract protects the liver, kidneys, spleen, heart, lungs from the toxic effects of heavy metals; in addition, it reduces the negative carcinogenic processes in the mammary gland associated with the migration of cadmium and lead into the blood and milk, prevents the oxidation of liver lipids, Eleutherococcus extract helps to protect the body animal from hazardous influence of heavy metals and not only Cd and Cu, but also Pb and Zn, which we confirmed in experiments. It activates the intensity of removing pollutants (Cd, Pb, Cu, Zn) from blood, milk, internal organs, and body systems. It normalizes blood pressure, immunostimulating action, resistance to stress, and a stimulating effect on the adrenal glands, which contributed to the restoration of the metabolism of mineral substances in the body simultaneously with an increase in the body's resistance to various diseases, viral origin; the extract of renal tea helped to increase the excretion of cadmium, lead, copper, and zinc, confirmed by experiments, effectively counteracted the development of proteinuria, the restoration of the functioning of the renal tubules and glomeruli of infiltration in the kidneys, prevented degenerative changes in this crucial organ during intoxication. It seemed somewhat more important to us the role in the composition of the developed biological product of common barberry, which in terms of mass ratio took the third place - 12%, which is again explained by active substances, high content of vitamins C and E, carotene, malic and citric acids, which support the normal functioning of the Krebs cycle. According to (Embaby & Afifi, 2016), ascorbic acid is one of the water-soluble antioxidants and has a protective function against Cd-induced histological changes in the liver, kidneys, lungs, and bone marrow cytotoxicity. Fat-soluble vitamin E is one of the most important for intoxicated bodies, an antioxidant factor, protects tissues from oxidative stress, and performs various physiological functions a free radical scavenger, which prevents lipid peroxidation, hepatoprotection of the liver from biochemical disorders, histological changes resulting from exposure to heavy metals (Al-Attar, 2011). Anticarcinogenic and anti-thyroid remedy. Choleretic and diuretic properties strengthen the antidote effect of the preparation "BP-9", about the effect of heavy metals on the body of dairy cows; in addition, it is not difficult to prepare an extract from this plant, since it is prevalent, especially in Ukraine, in fact, like most medicinal plants preparation.

The renal pathway for eliminating heavy metals from the body is a rather complex and dynamic process. An increase in the content of toxic metals in urine in most cases is observed earlier than biochemical and, especially, clinical changes, and in conditions of external intake of pollutants (in the diet with feed) is an informative indicator of the body's load. At the same time, the excretion of elements is a regulating means of the constancy of their content in the body. Our studies have shown that with prolonged chronic intake of heavy metals into the body, their content in biological substrates increases. That is, over time, the pathological effect of heavy metals on cellular metabolism increases, which causes damage to the morpho-functional structure of the kidneys and, if appropriate measures are not taken, then over time, this will lead to pathological conditions and kidney diseases.

If according to the results of our experiment, we place the metals studied by us in a row of elimination activity with urine, according to the value of the transition index, taking into account the action of the premix and the preparation, then we get the following form: the second and third research groups-Pb→Cu→Zn→Cd. The magnitude of the transition is different for metals of different biological activity and characterizes the body as a self-regulating system upon entering which toxic metals are intensively excreted. In the conditions of our experience, we see that we did not succeed in enhancing the excretion of the most dangerous element of cadmium in the urine, in comparison with all other metals, but we succeeded in removing lead, which is no less dangerous toxicant. Most intensively, Cd passed from blood to urine in the experiment on cows of the second and third experimental groups with silage-haylage-concentrate feeding type 9.5 and 12.7%, respectively, further in animals with silage-haylage - 7.3 and 9.5%; silage-hay - 6.6 and 9.4% and silage-root crops - 3.9 and 7.1%, respectively. On average, this indicator ranged from 3.9 to 12.7%, which is a significant result for such an ecological state of agroecosystems with environmentally friendly milk production. Also, a significant transition of Pb from blood to urine occurs in the experiment on cows of the second and third experimental groups with the silage-hay-concentrate feeding type 103.5 and 144.1%, respectively. The result was somewhat more significant at first in animals with silage-hay - 37.7 and 113.8%, and then from the silage-haylage - 81.6 and 94.1%, and, as for Cd, in the last place were animals with the silage-root type of feeding - 42.1 and 70.7%, respectively. On average, the Pb transition ranged from 42.1 to 144.1%, which is typical of agroecosystems, where the level of environmental pollution with Pb is much higher than that of Cd. Among all farms, the cows with the silage-haylage-concentrate type of feeding ate fodder with a large excess of lead content by 7.3 times. In the absolute ratio of Cd and Pb indicators, the transfer of Pb from blood to urine is actually 10 times higher than Cd (3.9% versus 42.1% and 12.7% versus 144.1%). At the same time, there is a remarkably high-quality ecologically safe and biologically complete milk, which corresponded to GOST 3662-97, and in terms of lead content, bacterial contamination and the number of somatic cells, regulations (EU) No. 853/2004 and No. 1881/2006.

In addition, the essentiality of elements such as copper and zinc reduces renal excretion. In cows of the control groups, the transition of Zn from blood to urine averaged 3.5-5.3%, and in animals of the second and third experimental groups, on average, from 10.5 to 35.7% more by an average of 7-30.4%. So, the premix and the preparation perform their protective function, to increase the excretion of this essential element from the body through the kidneys with urine ($P<0.01$) by its excess consumption with the feed of the diet. The biological product used in the third experimental group of cows, with a silage-hay type of ration, did not affect the increase in the transfer of Zn from blood to urine compared to the second research group, where only the premix itself was used since the transition was 34.5% in the second and 35.7% in the third experimental group, the difference is insignificant, only 1.2%. A similar situation occurred for Cu. Its transition from blood to urine averaged 3.0-3.6% in cows of the first control groups and in animals of the second and third experimental groups - 10.2-25.2%. Likewise, but already in cows with a silage-hay type of feeding, the injection of the biological product "BP-9" did not significantly affect the increase in the transition of Cu from blood to urine, since in the second experimental group, the transition was 16.4%, and in the third experimental group - 16.6%, the difference is 0.2%. However, the protective function of the applied antidote substances also acts, protecting the body from excess Cu in the diet, which is confirmed by the results obtained in the second and third experimental groups, where the Cu transition was 16.4 and 16.6%, respectively, compared with the control group-3.2%, more on average by 13%, with a high probability of excretion of the element in the urine ($P<0.01$).

The test cows on the silage-root ration ate the feed with the highest level of Cd contamination. In the experiment, a stable tendency was achieved to increase the excretion of this mineral element in the urine ($P<0.01$), thereby reducing its accumulation in the body of animals, feeding the premix and subcutaneous injection of the biological product ensured the transition of Cd from blood to urine at a low level among all four of our experiments. , which amounted to 3.9% in the second experimental group and 7.1% in the third experimental group, which requires further analysis or the development of new, more effective antidotes for this farm. In general, the result obtained is a significant scientific achievement.

So, having in its composition all the necessary mineral elements: iron, selenium, molybdenum, boron, calcium, chromium, fluorine, sodium, lithium, iodine, silicon, manganese, copper, zinc, nickel, vanadium, bromine, cobalt, Including antagonists of heavy metals cadmium and lead, and among vitamins-A, C, B, PP, E, K, F, with the help of the injection of the biological product "BP-9" we managed to enhance the antitoxic effect of the mineral-vitamin premix "MP A "at the level of tissues, organs, and systems of intoxicated cows. We have described in detail the composition of premixes and biological products, the amount of supplement fed to cows and the dose of preparation administration, the mechanism of antitoxic action, the quality and safety of milk produced by us in previous works (Portjannyk, 2002; Mamenko & Portjannyk, 2010). Included in the "MP-A" premix, selenium performed a vital function: amino acids (selenomethionine and selenocysteine), selenium-forming proteins, and enzymes glutathione peroxidase, which prevents the toxic effects of peroxide radicals on cells, has pronounced antioxidant properties. Glutathione peroxidase destroys not only hydrogen peroxide, but also peroxide compounds formed as a result of the oxidation of unsaturated fatty acids. This process is disrupted by Cd and Pb, which leads to an increase in the amount of under-oxidized products. The key role of selenium and zinc in the prevention of cadmium neurotoxicity has been proven (Branca et al., 2018).

Subcutaneous injection of the biological product "BP-9" developed by us, made from an aqueous extract of 9 medicinal r-saliva, with a daily dose of 20 ml per head per day, in general, in all four experiments carried out, positively influenced the urinary excretion dangerous ecotoxicants Cd and Pb and the maximum preservation in the body of dairy cows of important essential macro- and microelements, like Cu and Zn with different types of animal feeding. Anti-inflammatory, antitumor properties of the preparation, a decrease in the neurotoxicity of cadmium and lead, a hepatoprotective effect increased the excretion of heavy metals in the urine, thereby reducing the content of toxicants in the liver, kidneys, spleen, other internal organs, and tissues, muscle tissue ($P<0.01$). The positive experience of using extracts of medicinal plants has been proven by the studies of domestic and foreign scientists. For example, the leaf extract of the perennial evergreen betel nut (Latin Piper betie) studied by scientists (Milton Prabu et al., 2012) showed an excellent protective effect against cadmium-induced oxidative liver damage in experimental rats. There are also known methods for removing cadmium from the body of experimental animals using betulinic acid, a natural pentacyclic triterpenoid, which can be found in the bark of some plant species, in particular, birch (Betula pubescens), from where it got its name. Betulinic acid has anti-inflammatory and antitumor properties; in the experiment, it increased the excretion of cadmium from the liver and kidneys, the cadmium content in the urine increased, which caused an increase in the excretion of the toxicant (Fan et al., 2018). A positive result in experiments on Cd and Pb intoxication was shown by an aqueous solution of tannic acid, which contains an extract of many plants (Winiarska-Mieczan 2013), and an extract from mangosteen (an evergreen tree from Latin Garcinia Mangostana) relieves lead-induced neurotoxicity (Phyu & Tangpong, 2014). The hepatoprotective properties of royal jelly were studied with cadmium intoxication in rats, which prevented oxidative stress, inflammation, and liver damage (Almeer et al., 2018). Bilberry extract (Vaccinium corymbosum L.) and anthocyanins isolated also have a protective, therapeutic effect against Cd intoxication at the level of metal-chelating complexes (Gong et al., 2014).

Physalis Peruvian or Cape gooseberry (Physalis peruviana, Solanaceae). In experiments, family was used to change the histopathological state of liver and kidney tissues completely, its good hepatorenal protective role was proved lipid peroxidation decreases (Dkhil et al., 2014). In the organs and tissues of cows of the second research group, where the biological product was not used and, primarily, the first control groups, the highest content of the studied heavy metals was recorded, proves the accumulating effect of pollutants. However, the content of elements in the organs and tissues of animals of the second research groups tended to decrease due to the blocking effect of toxicants in the gastrointestinal tract using the mineral-vitamin premix " MP-A " ($P < 0.01$) gradually. The organs of increased contamination of heavy metals were kidneys and liver ($P<0.01$). Ukrainian scientists carried out a lot of research on feeding various feed additives into the diet, which played the role of a kind of adsorbent of heavy metals, which blocked the absorption of elements at the level of the gastrointestinal tract, in particular, the use in feeding dairy cows of the black-and-white breed of the natural zeolite mineral from the Sokyrnytsia deposit, which has absorption, ion-exchange, catalytic and other unique properties in conditions of local pollution of agroecosystems, which positively affects the physiological state, productivity of animals, which increases by 14.6% and the production of environmentally friendly milk in terms of lead content (Kurljak, 2011). Feeding animals as part of the main diet of a pectin preparation made from the waste of the wine industry ensures the removal of mercury from the sheep's body, and its inclusion, together with fodder beets in the diet of fattening pigs, helps to reduce the negative effect of cadmium on the productivity of animals and its transformation into muscle tissue. In order to increase the elimination of lead, copper, zinc, and iron, it is recommended to feed the cows daily for 45 days, 500 ml of decoction of licorice root. Also, the intensity of the transition of heavy metals into livestock products is reduced by the introduction of additives into the diet of animals that adsorb and inhibit their absorption in the gastrointestinal tract into the blood, for example, saponite, glauconites, trapels, complex additives and heterosorbents such as "Belosorb" - a polymer of natural origin, made of cellulose, these substances bind heavy metals, thereby weakening their toxic effect on the body of animals, while positively affecting metabolic processes in the microbial ecosystem of the rumen. It has been experimentally proven that the use of zeolite in the diet of dairy cows helps to reduce lead in milk by 56.8%, cadmium by 22.6%, mercury - by 50%, and at the same time has a positive effect on productivity. The most promising means of preventing the migration of heavy metals in the trophic chain are complexing compounds and enterosorbents, which do not exhibit toxic effects on the body and significantly limit the absorption of heavy metals in the gastrointestinal tract (Papaioannou et al., 2005; Savchenko & Savchuk 2013).

We have tested a methodology for developing particular antitoxic mineral and vitamin premixes, such as "MP-A", adapted to the actual feeding ration, introducing them into the daily ration for feeding dairy cows. Milk, since many enterprises for the production of various feed additives are represented on the market. Sometimes even agricultural enterprises themselves have their workshops for the production of feed additives necessary for them. The premix's antitoxic effect consists of the introduction into its composition in the appropriate ratio of mineral elements, which act as antagonists of heavy metals Cd and Pb, displacing them in metabolism. The vitamin group aims to maximize the normalization of the physiological functions of animals intoxicated with incorporated heavy metals (especially Cd and Pb), and the added sulfur-containing amino acid acts as a chelating complex with heavy metals. The antitoxic effect of the premix on sending the excretion of heavy metals, especially Cd and Pb with urine and their excretion from the body of animals, was better manifested in cows of the second and third experimental groups with the silage-silage-concentrate type of feeding. Good results were also obtained in cows of the experimental groups with the silage-haylage and silage-hay type of feeding. The weakest effect of the premix on the transfer of heavy metals from blood to urine and, accordingly, excretion from the body was observed in research groups of cows with the silage-root type of feeding, which requires additional analysis. In all four experiments, feeding the premix to cows helped to improve the quality and environmental safety of the milk produced, which mehe requirements of DSTU 3662-97 "Whole cow's milk.

Requirements for purchases", and the content of lead, microorganisms and somatic cells of the standard (regulation (EU) No. 853/2004 and No. 1881/2006) of the countries of the European Union; an increase in the milk productivity of animals ($P<0.01$), which averaged 17-22 kg per day in cows of the second and third experimental groups compared to the control group-14 kg ($P<0.01$).

Conclusions

The excretion of heavy metals from the urine of dairy cows reflects their load on the animal body caused by the consumption of feed that exceeds the maximum permissible concentrations of the studied elements. The health of dairy cows with a particular antitoxic mineral and vitamin premix and the protective effect of the injection of a biologically active preparation in agroecosystems, which are located near an industrial city, enhanced the elimination of heavy metals in the urine of intoxicated animals by such a range of migration activity of Pb, Cu, Zn, and Cd. The antidote effect of the premix and the biological product had a positive effect on the renal excretion of excess essential elements Cu and Zn from the body without sacrificing cows' health, but on the contrary restoring its homeostasis, improving the quality and environmental safety of the milk produced.

Further research is aimed at the development of antidote substances that contributed to the increased excretion of hazardous heavy metals such as Cd and Pb from the body of animals in the urine compared with the essential elements Cu and Zn while maximizing the body's needs for essential elements following physiological norms.

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