

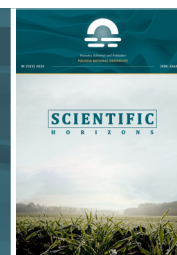
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Accumulation of cadmium and lead in the organs and tissues of dairy cows depending on breed and diet

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Abstract. Agroecosystem monitoring in a time of full-scale war has shown a high level of anthropogenic impact and an increase in the contamination of the environment with heavy metals, including toxic Cd and Pb. This study aimed to analyse the content of cadmium and lead in the kidneys, liver, lungs, heart, spleen, bone and muscle tissues of dairy cows depending on breed and diet under the conditions of environmentally safe cow's milk production technology. Experiments were conducted on dairy cows of different breeds in farms located within the Forest-Steppe soil and climatic zone of Ukraine, with control slaughter performed at the end of the experiment. The highest Cd content was found in the kidneys of cows of the Ukrainian Black-and-White Dairy breed on a silage-hay diet with the values 2.968 ± 0.193 mg/kg and on a silage-root crop diet with the values 3.241 ± 0.318 mg/kg, and the highest Pb content was found on the silage-hay diet with the values 4.387 ± 0.092 mg/kg and silage-root crop diet with the values ± 0.546 mg/kg. Analysis of muscle tissue for Cd content showed the highest concentration in Ukrainian Black-and-White Dairy cows fed a silage-hay diet, amounting to 0.125 ± 0.026 mg/kg, and a silage-root crop diet, amounting to 0.140 ± 0.006 mg/kg. The Pb concentration in muscle tissue was also highest in Ukrainian Black-and-White Dairy cows, reaching 1.472 ± 0.065 mg/kg under the silage-hay diet and 1.691 ± 0.092 mg/kg under the silage-root crop diet. The highest concentrations of Cd and Pb in the kidneys, liver, lungs, spleen, heart, bones and muscle tissue were identified in Ukrainian Black and-White Dairy cows compared with animals of the Ukrainian Red-and-White Dairy breed, which was associated more with the higher intake of pollutants from the basal diets and feeding type than with breed characteristics. The obtained data may be used in the development of recommendations for selecting diet types and feeding technologies for dairy cows in order to reduce the accumulation of heavy metals in internal organs and tissues, as well as to improve the environmental safety of cows' milk and dairy products

Keywords: ecotoxics; feed; feeding type; heavy metals; productive animals; agroecosystems

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INTRODUCTION

The production of environmentally safe livestock products under conditions of technogenic environmental contamination is one of the key challenges facing the modern agricultural sector. High anthropogenic loads of ecotoxicants on agrobiogeocenoses cause changes in the natural state of soils, water bodies, flora, fauna and microorganisms. Similar processes are also characteristic of territories affected by military activity, where the use of ammunition and military equipment results in additional metal input into the environment, according to A.J. Barker *et al.* (2021). Heavy metals, especially the elements of cadmium and lead, have a particularly dangerous characteristic due to their highly bioaccumulative features and tendency to accumulate in the body of livestock, thereby affecting their physiological status and the safety of products from farm animals. Cattle, in this case, are of great importance as a source of milk and meat, as well as indicators of the environmental situation of agricultural land. In addition, the pollution of livestock with heavy metals is aggravated by the consequences of Russia's military aggression against Ukraine, which has been ongoing since 2022, due to the release of various toxic compounds, including cadmium, lead, mercury, copper, and zinc, during the explosion of landmines and explosives on agricultural land. It has been proven by M. Solokha *et al.* (2023) that military actions, through field and remote sensing observation, lead to significant changes in the physicochemical parameters of soils and accumulation of toxic elements in the environment, which leads to their penetration into the feed base and further to their accumulation in the organisms of animals. Therefore, researching the regularities of cadmium and lead accumulation in the organs and tissues of productive dairy cows is particularly relevant for assessing the environmental safety of livestock products and selecting appropriate mitigation strategies.

The accumulation of heavy metals, in particular cadmium and lead, in the soil results in their transfer into plant products, resulting in the risk of contamination of the food chain and harmful effects on human health. According to V.O. Velychko (2023) and S. Portiannyk *et al.* (2024), under conditions of increased anthropogenic pressure on the environment and farm animals, monitoring of heavy metals within the "soil – plant (feed) – animal products (milk, meat, etc.) – human" system remains a priority area of scientific research. Cadmium and lead entering the bodies of cows through feed are readily absorbed in the gastrointestinal tract into the bloodstream, distributed throughout the organism and excreted with milk and faeces; however, part of these metals accumulates in internal organs and tissues, where they may persist for long periods. An important area of research for Ukrainian scientists is the assessment of the effects of different diet types and the sorbent saponite, various protein-vitamin-mineral supple-

ments in feed mixtures, mineralvitamin premixes and biologically active preparations on the level of cadmium accumulation in the muscle tissue of bulls, pigs and dairy cows. I.M. Savchuk *et al.* (2021, 2023) substantiated the feasibility of using silage-concentrate, silage-concentrate-root crop and silageconcentrate-haylage diet types for fattening young cattle and determined their effects on cadmium and lead accumulation in animal products. The use of silage-concentrate-root crop and silage-concentrate-haylage diets in bull feeding reduced cadmium accumulation in muscle tissue by 34.1%-66.7% and its concentration in the liver by 15.3%-46.6% compared with silage-concentrate diets.

Researchers from Malaysia, Z.F. Anual *et al.* (2023), collected 378 samples of edible animal by-products from markets to determine heavy metal content. The highest lead concentration was detected in cattle lungs (0.11 ± 0.20 mg/kg), as well as in the spleen (0.09 ± 0.14 mg/kg) and rumen (0.09 ± 0.12 mg/kg). With regard to cadmium, the highest concentrations were identified in cattle liver (0.13 ± 0.12 mg/kg), pig liver (0.08 ± 0.05 mg/kg) and chicken liver (0.03 ± 0.02 mg/kg). M.H. Emami *et al.* (2023) determined the content of lead and cadmium in muscles, lungs, heart, liver, spleen and other organs and tissues. The study showed that the accumulation of the heavy metals cadmium and lead in cattle tissues was lower than in goats or lambs, which was attributed to differences in animal size, feeding regimes and housing conditions. The muscle tissue of cows kept outdoors accumulated more lead than that of cows housed indoors.

R.A. Samy *et al.* (2025) examined 150 samples of buffalo muscle, liver and kidney tissues in order to determine heavy metal content and assess risks to consumer health. Cadmium and lead were found in all tissues at varying concentrations, with the highest levels being in the liver and kidneys and the lowest in muscle. A. Mushtaq *et al.* (2024), having analysed 180 samples of cattle muscle tissue, liver and kidneys obtained from slaughterhouses in Pakistan for six heavy metals, found that the liver, compared with muscle tissue and kidneys, was the primary organ for the bioaccumulation of chromium, lead and mercury. Investigating the problem of heavy metal nephrotoxicity in cattle and its treatment, I. Tahir & K.A. Alkheraije (2023) concluded that lead and cadmium exhibit high nephrotoxicity, while these pollutants cause acute kidney diseases in cattle, accumulate in other parts of the body and penetrate the liver and muscles, although with lower intensity compared with the kidneys.

At the same time, despite the considerable number of studies conducted in different countries worldwide, the issue of heavy metal accumulation in livestock products under Ukrainian conditions, particularly with regard to the specific features of anthropogenic pressure and the consequences of military activities, remains

insufficiently investigated. Of particular relevance is the study of the influence of breed and feeding type on the level of cadmium and lead accumulation in the tissues of lactating cows. This study aimed to analyse the accumulation of the toxic heavy metals cadmium and lead in the internal organs and tissues of lactating cows depending on breed and feeding type under conditions of applying new technological approaches to the production of environmentally safe cow's milk on cattle farms.

MATERIALS AND METHODS

The study was conducted in four farms located in the Lubny District of Poltava Region within the Forest-Steppe zone of Ukraine. The study involved 420 dairy cows of the Ukrainian BlackandWhite Dairy and Ukrainian Red-and-White Dairy breeds. All animal manipulations were performed in accordance with the

Law of Ukraine No. 249 (2012) and the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (1986). The laboratory complied with the requirements of ISO/IEC 17025:2005 (2006) concerning the general requirements for the competence of testing and calibration laboratories.

The agricultural lands where the animals were kept and where feed components for the basal diets were cultivated were located near environmentally hazardous anthropogenic facilities and differed in the level of technogenic pressure, including the Kyiv-Kharkiv-Dovzhanskyi highway, oil pipelines, gas pipelines and gas compressor stations. The cows were fed four types of diets: silage-root crop, silage-hay, silage-haylage and silage-haylage-concentrate. The distribution of cows according to feeding type and location is presented in Table 1.

Table 1. Classification of the feeding types according to location

Feeding type	Industrial facility	Distance from the facility, km	Number of animals, head
Silage-root crop	Automobile gas-filling compressor station	3	130
Silage-hay	Gas condensate station	10	800
Silage-haylage	Industrial centre	18	250
Silage-haylage-concentrate	Kyiv-Kharkiv-Dovzhanskyi highway	30	500

Source: developed by the author

The study was carried out in the autumn–spring periods of cow keeping (2017-2018) in two stages, namely comparative (42 days) and experimental (120 days). The cows were divided into three groups: control group I (n = 12) and experimental groups II and III (n = 12 and n = 12, respectively). The animals were selected by the analogue method according to live weight and productivity, and were in the third lactation period, which corresponds to peak milk productivity. The average live weight of the cows at the time of the study ranged from

500 to 545 kg. The average daily milk yield was 14.0-14.8 kg. The breed composition of cows, depending on the type of feeding, is given in Table 2.

The animals of all experimental groups were fed by the developed basic diet balanced on feed units, digestible protein and other nutrients, although in all samples of feeding materials there was an excess of the concentrations of cadmium, lead, copper and zinc above the maximum permissible level. The composition of each feeding type is presented in Table 3.

Table 2. Breed composition of cows depending on the type of feeding

Breed	Feeding type	Number of animals in each group, head			
		I	II	III	Total
BlackandWhite	Silage-root crop	12	12	12	36
	Silage-hay	65	65	65	195
Red-and-White	Silage-haylage	21	21	21	63
	Silage-haylage-concentrate	42	42	42	126

Source: developed by the author

Table 3. Diet composition and cadmium and lead content depending on feeding type

Feeding type	Designation	Composition	Heavy metal content, mg/kg	
			Cd	Pb
Silage-root crop	BD1	grass-legume hay	0.96	28.59
		wheat straw	0.71	13.62
		maize silage	0.69	13.51
		lucerne haylage	0.75	16.78

Table 3. Continued

Feeding type	Designation	Composition	Heavy metal content, mg/kg	
			Cd	Pb
Silage-root crop	BD1	fodder beet	0.64	12.22
		ground maize grain	0.96	20.51
Silage-hay	BD2	lucerne hay	0.57	11.22
		wheat straw	0.51	12.5
		maize silage	0.39	9.13
		lucerne haylage	0.45	9.54
		fodder beet	0.75	17.21
		ground barley grain	0.54	11.6
Silage-haylage	BD2	lucerne hay	0.63	16.51
		grass-legume hay	0.66	18.5
		maize silage	0.48	12.5
		lucerne haylage	0.54	15.5
		ground oat grain	0.75	23.12
		ground pea grain	0.78	23.51
Silagehaylageconcentrate	BD2	lucerne hay	0.69	16.52
		grass-legume hay	0.75	36.51
		maize silage	0.42	10.54
		lucerne haylage	0.48	14.23
		ground maize grain	0.54	18.51
		ground pea grain	0.63	20.53

Note: maximum permissible concentrations in feed: Cd – 0.3 mg/kg; Pb – 5.0 mg/kg

Source: developed by the author

In the second experimental group, the basal diet was supplemented with the mineral-vitamin premix “MP-A” to correct macro- and microelement nutrition and reduce the toxic heavy metal burden on the organism. The premix contained mineral elements including iron, manganese, cobalt, selenium, sulphur, magnesium, calcium, phosphorus and iodine, as well as the amino acid methionine and vitamins A, D, E, B2, B3, B4, B6, C and H. In the third experimental group, in addition to feeding the basal diet supplemented with the premix, intramuscular injections of the phytobiological preparation “BP-9”, based on extracts of medicinal plants, were administered. The

preparation contained five-flavour fruit (*Schizandra chinensis* (Turcz.) Baill.), chamomile (*Chamomilla recutita*), Siberian ginseng (*Eleutherococcus senticosus* (*Acanthopanax senticosus*)), common sage (*Salvia officinalis*), common barberry (*Berberis vulgaris*), lucerne (*Medicago sativa*), Java tea (*Orthosiphon stamineus*), sea buckthorn (*Hippophae rhamnoides*) and common vervain (*Verbena officinalis*). The role of the biological preparation was to enhance the antidotal effect of the premix and influence target organ cells in order to intensify the elimination of ecotoxins from the organism. The feeding scheme for each group of cows is presented in Table 4.

Table 4. Scheme of experimental groups in each experiment

Feeding type	Group		
	I	II	III
Silage-root crop	BD1	BD1 + premix “MP-A1”	BD1 + premix “MP-A1” + preparation “BP-9”
Silage-hay	BD2	BD2 + premix “MP-A2”	BD2 + premix “MP-A2” + preparation “BP-9”
Silage-haylage	BD3	BD3 + premix “MP-A3”	BD3 + premix “MP-A3” + preparation “BP-9”
Silagehaylageconcentrate	BD4	BD4 + premix “MP-A4”	BD4 + premix “MP-A4” + preparation “BP-9”

Note: BD – basal diet; BP – biologically active preparation; MP – mineral premix

Source: developed by the author

At the last stage of the study, an acute experiment was conducted, involving the slaughter of animals from each group (n = 3) and collecting samples of biologically relevant tissue in the internal organs to conduct veterinary and sanitary analysis of it. Sampling of muscle tissue, internal organs (kidneys, liver,

lungs, heart and spleen), and bone tissue was carried out on the basis of the methodical recommendations of E.I. Chygrinov *et al.* (1998) and O.M. Yakubchak *et al.* (2005). Representative samples were prepared by collecting three-point samples weighing 100 g each from different parts of every organ, followed by

their combination and homogenisation to obtain an integrated sample weighing 250-300 g. The collected samples were placed in sterile sealed containers, stored at a temperature of +2°C to +8°C and transported to the laboratory together with the accompanying documentation (sampling report). Statistical analysis was performed on the basis of the calculated mean value of the parameter in the sample (M) with standard deviation (SD). The results are presented as $M \pm SD$. Differences between mean values were considered statistically significant at $p < 0.05$. The level of

significance was assessed using one-way ANOVA. Calculations were performed using STATISTICA software version 10.0.

RESULTS AND DISCUSSION

During the examination of the internal organs after control slaughter, no pathological changes or abnormalities in shape or colour were detected. Table 5 presents the content of the ecotoxins cadmium and lead in the internal organs and tissues of animals from control group I, depending on breed type.

Table 5. Cadmium and lead content in the organisms of cows from group I (mg/kg fresh weight)

Organ	Breed			
	Black-and-White		Red-and-White	
Kidneys	2.968-3.241	4.387-4.522	2.305-2.634	3.452-3.981
Liver	0.701-0.752	1.731-1.865	0.611-0.683	1.607-1.694
Lungs	0.451-0.513	2.465-2.763	0.331-0.393	1.812-2.228
Spleen	Cd 0.462-0.486	Pb 1.480-1.503	Cd 0.361-0.391	Pb 1.029-1.279
Heart	0.431-0.459	1.091-1.146	0.337-0.395	0.840-0.957
Bones	0.411-0.423	1.512-1.685	0.392-0.407	1.234-1.480
Muscles	0.125-0.140	1.472-1.691	0.094-0.115	1.083-1.264

Source: developed by the author

The data presented in the table demonstrate that the principal target organs for cadmium and lead accumulation in both breeds were the kidneys and liver. Accumulation of heavy metals was also recorded in the lungs, spleen, heart, as well as in bone and muscle tissues. The highest Cd concentration was identified in the kidneys of Black-and-White cows fed silage-hay diets, amounting to 2.968 ± 0.193 mg/kg, and silage-root crop diets, amounting to 3.241 ± 0.318 mg/kg, compared with Red-and-White cows, in which the pollutant concentration was recorded at 2.305 ± 0.033 mg/kg in animals receiving the silage-haylage-concentrate diet

and 2.634 ± 0.093 mg/kg in those fed the silage-haylage diet.

Table 6 presents descriptive statistics of cadmium content in the kidneys. A similar pattern was observed for the other dangerous ecotoxin – Pb. The maximal concentration of this ecotoxin in kidneys was detected in Ukrainian Black-and-White Dairy cows fed silage-hay (4.387 ± 0.092 mg/kg) and silage-root crop (4.522 ± 0.546 mg/kg) diets, while it was lower in Ukrainian Red-and-White Dairy cows, which were fed silage-haylage-concentrate diet (3.452 ± 0.017 mg/kg) or silage-haylage diet (3.981 ± 0.274 mg/kg).

Table 6. Statistics of cadmium concentration in the kidneys of cows from group I, mg/kg

Breed	Feeding type	Mean	Median	Minimum	Maximum	Lower Quartile	Upper Quartile	Std. Dev	Standard Error
BlackandWhite	Silage-root crop	3.241	3.322	2.891	3.511	2.891	3.511	0.318	0.183
	Silage-hay	2.968	2.858	2.855	3.191	2.855	3.191	0.193	0.112
	Silage-haylage	2.634	2.619	2.549	2.734	2.549	2.734	0.093	0.054
Red-and-White	Silagehaylageconcentrate	2.305	2.318	2.268	2.329	2.268	2.329	0.033	0.019

Source: developed by the author

According to the quartile diagram presented in Figure 1, the degree of variation can be assessed. The graph reflects the dispersion of lower and upper values, while the median does not differ substantially from the arithmetic mean. Among the maximal concentration values, the highest one was noted in the

kidneys of the animals fed a silage-root crop diet (3.511 mg/kg), while the lowest one occurred under a silage-haylage-concentrate diet (2.329 mg/kg). The minimum of cadmium values, 2.268 mg/kg, was identified among animals receiving a silagehaylageconcentrate diet.

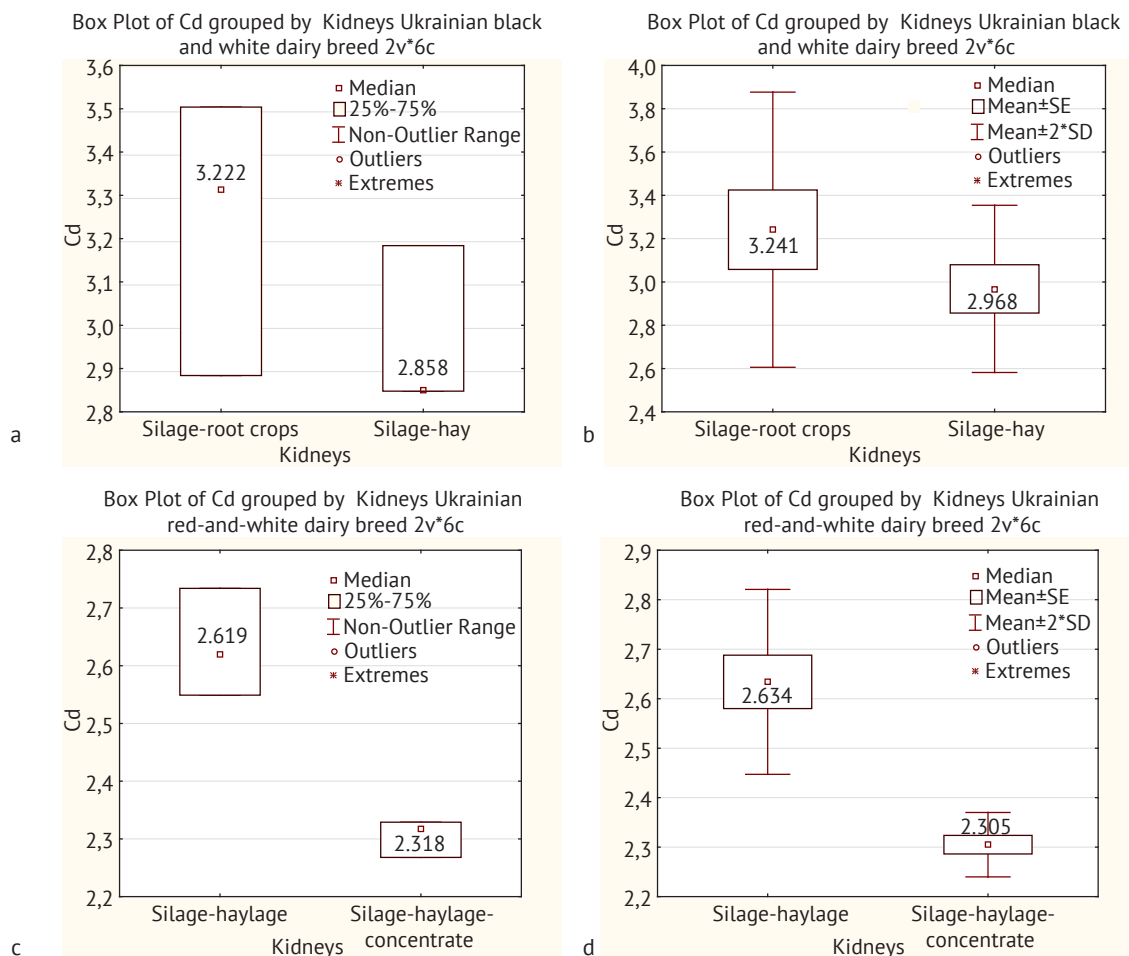


Figure 1. Diagrams of mean values and quartile plots of Cd content in the kidneys (mg/kg) of cows from the first control group

Note: a – Black-and-White cows fed a silage-root crop diet; b – Black-and-White cows fed a silage-hay diet; c – Red-and-White cows fed a silage-haylage diet; d – Red-and-White cows fed a silage-haylage-concentrate diet

Source: developed by the author

Analysis of muscle tissue for cadmium content showed that the highest concentration of the pollutant was found in Ukrainian Black-and-White Dairy cows fed a silage-hay diet (0.125 ± 0.026 mg/kg) and a silage-root crop diet (0.140 ± 0.006 mg/kg), compared with Ukrainian Red-and-White Dairy cows receiving silage-haylage-concentrate diets (0.094 ± 0.008 mg/kg) and silage-haylage diets (0.115 ± 0.020 mg/kg). The highest lead concentration in muscle tissue was also detected in Black-and-White cows: 1.472 ± 0.065 mg/kg under the silage-hay diet and 1.691 ± 0.092 mg/kg under the silage-root crop diet. In Ukrainian Red-and-White Dairy cows, the corresponding values were lower,

amounting to 1.083 ± 0.008 mg/kg under the silagehaylageconcentrate diet and 1.264 ± 0.067 mg/kg under the silage-haylage diet.

The concentration of heavy metals, particularly cadmium and lead, in the liver, kidneys, muscle tissue, bones, spleen, heart and lungs of animals is one of the key indicators of pollutant intake into the organism via feed. The increased values of these indices indicate that there is a risk of consumption of feed based on plants grown in ecotoxicant-polluted soils. Table 7 shows the cadmium and lead content in different types of diets for Ukrainian Black-and-White and RedandWhite Dairy cows per 1 kg of dry matter.

Table 7. Content of toxic heavy metals in diets of different feeding types for cows (mg per 1 kg of dry matter)

Breed	Feeding type	Cadmium content, mg/kg	Lead content, mg/kg
Black-and-White	Silage-root crop	2.177-2.833	49.567-58.370
	Silage-hay		
Red-and-White	Silage-haylage	1.098-1.271	31.728-34.746
	Silage-haylage-concentrate		

Source: developed by the author

Cadmium and lead levels were higher in the basal diets of the silage-root crop and silage-hay feeding types (2.177-2.833 mg/kg and 49.567-58.370 mg/kg, respectively) of the Ukrainian Black-and-White Dairy breed compared to the silage-haylage and silage-haylage-concentrate ones (1.098-1.271 mg/kg and 31.728-34.746 mg/kg, respectively) of the Ukrainian Red-and-White Dairy breed, in the case of 1 kg dry matter of the feed mixture. The premix and biological preparation used in the experiment contributed to a reduction in Cd and Pb levels in the kidneys, liver, spleen, lung and

muscle tissues, heart and bones. In the experimental Ukrainian Black-and-White Dairy cows receiving the silage-root crop diet, the greatest reduction in cadmium content was observed in the kidneys, averaging a 2.5-fold decrease in the second experimental group and a 3.6-fold decrease in the third experimental group compared with the first control group. Cd content in the kidneys decreased from 3.241 ± 0.318 mg/kg in the first control group to 1.294 ± 0.033 mg/kg in the second experimental group and 0.903 ± 0.011 mg/kg in the third experimental group ($p < 0.001$) (Table 8).

Table 8. Reduction of cadmium content in the kidneys, mg/kg

Breed	Feeding type	I	II	III
BlackandWhite	Silage-root crop	3.241 ± 0.318	1.294 ± 0.033	0.903 ± 0.011
	Silage-hay	2.968 ± 0.193	0.884 ± 0.015	0.701 ± 0.013
Red-and-White	Silage-haylage	2.634 ± 0.093	0.831 ± 0.061	0.695 ± 0.026
	Silagehaylageconcentrate	2.305 ± 0.033	0.721 ± 0.027	0.563 ± 0.056

Source: developed by the author

In the second experimental group, over the 120-day experimental period, it was not possible to reduce the Cd content to the permissible limit using the mineral-vitamin premix "MP-A" alone. In the other samples of the internal organs and tissues studied, among the specimens taken from animals of both breeds, the amounts of cadmium in the second and third experimental groups were within the normal values. The administration of the premix had a positive effect, reducing Cd content in muscle tissue in the second and third experimental groups by 66.4%-69.2% compared with the first control group, which in the long term supports the application of technological approaches for the production of environmentally safe beef. A reduction in Cd content was also observed in the liver, spleen, lungs, heart and bone tissue, confirming the beneficial effect of the "MP-A" premix and the "BP-9" biological preparation on the animals' organism. No statistically significant differences in the reduction of cadmium content in lung, heart and muscle tissues were identified between cows of the second and third experimental groups of the Ukrainian BlackandWhite and Red-and-White Dairy breeds; however, the third experimental group demonstrated more favourable values ($p < 0.001$). Therefore, in order to alleviate the intoxication of the organs (the most severe Cd intoxication occurred in the kidneys and liver), a subcutaneous injection of the biological preparation was needed.

The mineral-vitamin premix and the biological preparation demonstrated a stronger antitoxic effect in Ukrainian Red-and-White Dairy cows fed silage-haylage and silage-haylage-concentrate diets. The Cd content in the second and third experimental groups was reduced to the greatest extent. The second and third experimental groups of both breeds complied with

maximum permissible concentrations of the pollutant ($p < 0.001$). The reduction in cadmium levels in muscle tissue was most pronounced, compared with the second and third experimental groups, in Ukrainian Black-and-White Dairy cows fed silage-root crop and silage-hay diets. Thus, the decrease was 4.65.5 times in the group of cows that received a silage-haylage diet, 4.1-5.5 times in the group of cows that received a silage-haylage-concentrate diet, and 3.0-3.3-fold in the group of animals that received a silage-root crop diet, and 4.0-4.5-fold in the group of animals that received a silage-hay diet.

In this regard, a significant decrease in the amount of Cd content in the kidney samples is also noted: 3.2-3.8-fold in Ukrainian Red-and-White Dairy cows fed a silage-haylage diet, 3.2-4.1-fold in Ukrainian Red-and-White Dairy cows fed a silage-haylage-concentrate diet and 2.5-3.6-fold in Ukrainian Black-and-White Dairy cows fed a silage-root crop diet and 3.4-4.2-fold in Ukrainian Black-and-White Dairy cows fed a silage-hay diet. No statistically significant differences were found in cadmium content in the muscle tissue, liver, spleen, lungs, heart, and bones of experimental cows between the second and third experimental groups fed silage-haylage and silagehaylage-concentrate diets. The results of the experiment indicate the appropriateness of applying technological approaches involving the administration of the antitoxic premix "MP-A" and injection of the biological preparation "BP-9" in dairy cows fed silage-haylage and silagehaylage-concentrate diets, regardless of breed, as confirmed by the reduction in toxic heavy metal content in the organism. The intensity of Pb removal from the cows' organism was also affected by the pollutant concentration in the diet of experimental animals:

the maximum amount of lead in feed was registered in the diet of Ukrainian Black-and-White Dairy cows fed the silage-root crop and silage-hay diet, and the

minimum was in the silage-haylage and silage-haylage-concentrate diet of Ukrainian Red-and-White Dairy cows (Table 9).

Table 9. Reduction of lead content in the kidneys, mg/kg

Breed	Feeding type	I	II	III
BlackandWhite	Silage-root crop	4.522 ± 0.564	0.931 ± 0.019	0.851 ± 0.010
	Silage-hay	4.387 ± 0.092	0.804 ± 0.010	0.735 ± 0.037
Red-and-White	Silage-haylage	3.981 ± 0.274	0.772 ± 0.025	0.639 ± 0.055
	Silagehaylageconcentrate	3.452 ± 0.017	0.785 ± 0.032	0.721 ± 0.014

Source: developed by the author

The level of lead in the analysed samples of internal organs and tissues was significantly affected by the feeding regime; it provided the most significant removal of this contaminant in Ukrainian Red-and-White Dairy cows on a silage-haylage diet (66%-86.4% of the average) and 63.1%-88% in the case of the silage-haylage-concentrate diet ($p < 0.001$), which is of great significance. None of the experimental groups reached the permissible limits of the contaminant's level using only the mineral-vitamin premix, though the inclusion of this compound in the "MP-A" feed premix supplied to the cows of both breeds without the introduction of the phytobiological preparation resulted in a consistent decrease trend of Pb levels in their bodies, regardless of the feeding regime ($p < 0.001$). Using the herbal extract preparation caused stimulation of circulatory, nervous and endocrine system activity, intensification of blood supply to the lungs and removal of Pb from the lungs by 71.6%-80.7% in Ukrainian Black-and-White Dairy cows fed a silage-root crop diet, by 70.7%-83.3% in those fed a silage-hay diet, by 67.7%-86.4% in Ukrainian Red-and-White Dairy cows fed a silage-haylage diet, and by 63.1%-88% in those receiving a silagehaylageconcentrate diet as compared with the first control groups ($p < 0.001$). The type of feeding had a greater effect on the level of ecotoxins in the internal organs and tissues of cows than the breed of cattle, even in the conditions of using modern technological approaches for obtaining environmentally safe cattle products. The obtained results are consistent with the findings of Y. Akar *et al.* (2018) and I. Tahir & K.A. Alkheraije (2023), who reported that the kidneys and liver are target organs for heavy metal toxicity. Organs such as the lungs, spleen, heart, bone and muscle tissue are capable of accumulating significant amounts of ecotoxins, particularly highly hazardous ones such as cadmium and lead. This requires further study, in particular, on different breeds of cattle used for milk production.

Researchers from Mongolia, B. Bataa *et al.* (2021), reported maximum lead concentrations in cattle liver at 0.349 mg/kg and in kidneys at 1.08 mg/kg, with minimum levels of 0.2 mg/kg and 0.25 mg/kg, respectively. It follows that this level is much lower than in the presented study, when it was 4.387-4.522 mg/kg

and 3.452-3.981 mg/kg, respectively. As for the accumulation of these elements in the internal organs, the authors explained that this occurs due to environmental factors, in which animals are located in areas with an anthropogenic environmental impact. The established regularities of cadmium and lead accumulation in the organs of cattle are in line with the concept of biogeochemical migration of heavy metals in the "soil-plant-animal" system, where the level of their bioavailability and degree of accumulation depends on the physicochemical properties of the soils and feed (Alloway, 2013). According to M.J. Canty *et al.* (2014), 15% of soil in Ireland exceeded the EU norm (1 mg/kg for cadmium) for Cd in soil. Cd concentrations in kidneys ($n = 393$) ranged from 0.040 to 8.630 mg/kg of fresh weight, whereas As, Hg, and Pb concentrations were comparatively low.

Under the conditions of the present study, Cd concentrations in the kidneys ($n = 3$) of cows from the first control groups of the Ukrainian Black-and-White and Red-and-White Dairy breeds ranged from 2.305 to 3.241 mg/kg fresh weight. By comparison, Irish researchers reported an upper cadmium concentration limit that was almost 2.7 times higher (8.630 mg/kg versus 3.241 mg/kg), which was attributed to a higher level of soil contamination and, consequently, of feed contamination in diets. Lead concentrations reported by those authors were relatively low, whereas in the present experiment, Pb concentrations in the kidneys ($n = 3$) of cows from the first control groups of both breeds ranged from 3.452 to 4.522 mg/kg fresh weight. According to M.J. Canty *et al.* (2014), as for the weighted proportion of animals with a high level of cadmium concentration in the kidneys (≥ 1 mg/kg), according to estimates, this indicator is at the level of 11.25% (95% CI: 8.63%-14.53%). In a conducted experiment, the animals of the first control group in the Ukrainian Black-and-White Dairy breed and in the Ukrainian Red-and-White Dairy breed exceeded this indicator for the content of cadmium in the kidneys (≥ 1 mg/kg). At the same time, in both breeds in the second and third experimental groups, the use of antidotal substances reduced the concentration of cadmium accumulation (< 1 mg/kg) ($p < 0.001$), and the optimal results were

observed in the third experimental groups – 0.563–0.695 mg/kg in Ukrainian Red-and-White Dairy breed cows fed silage-haylage-concentrate and silage-hay diets, respectively, and 0.7010.903 mg/kg in Ukrainian Black-and-White Dairy breed cows fed silage-hay and silage-root crop diets, respectively. Ukrainian Red-and-White Dairy breed cows showed the best results.

According to M.J. Canty *et al.* (2014), the significant predictors of high kidney Cd concentrations were soil Cd, animal age, and the province; breed was not a significant predictor. It was estimated that at a soil Cd level of 1.5 mg/kg, the age threshold required to not exceed 1 mg/kg Cd in kidneys for the majority of the animals would be approximately 3 years in Connacht, >4 years in Ulster, and >5 years in Leinster and Munster. In areas of Ireland with naturally elevated soil Cd levels, kidney cadmium concentrations in cattle may exceed the current EU maximum permissible limit of 1 mg/kg in older animals. The kidneys of most cattle under three years of age would comply with EU requirements. In the present study, the age of the animals was 65 months (over 5 years), which objectively implies prolonged accumulation of cadmium and lead not only in the kidneys but also in other organs and tissues. Given the low efficiency of cellular elimination mechanisms for these pollutants, as well as their ability to persist in depot tissues (particularly the liver, kidneys and intestines), gradual accumulation occurs within the organism. As a result, older animals exhibit elevated concentrations of heavy metals in the liver and kidneys even under relatively low levels of exposure via feed and water (Nriagu *et al.*, 2009).

The highest Cd concentrations in the topsoil layer in Europe have been recorded in Ireland, England, the Western Alps and southern France, the Belgian-German border region, southern Sardinia, eastern Italy, Slovenia, Croatia, Albania and Greece. The elevated levels of Cd in most of these areas are attributed to the intensive application of phosphate fertilisers and sewage sludge in agriculture, except Ireland, southern Sardinia, Poland and the district of Goslar in Germany, where naturally high Cd is recorded (Pan *et al.*, 2010). The Czech researchers J. Drapal *et al.* (2021) analysed cadmium concentration in the muscle, liver and kidneys of cattle of different ages. In cattle older than two years, higher mean cadmium concentrations were observed in the liver (0.10 mg/kg) and kidneys (0.62 mg/kg) compared with cattle under two years of age, where values were 0.06 mg/kg in the liver and 0.24 mg/kg in the kidneys. These values were considerably lower than those obtained in the present research.

Thus, the higher concentrations of cadmium and lead in the internal organs and tissues of cows of different Ukrainian dairy breeds used in the experiment are determined not by breed type, but primarily by the level of pollutant intake through feed of different dietary types; however, a breed-related effect is also

present. The accumulation of ecotoxics in the kidneys, liver, lungs, spleen, heart, and bone and muscle tissues can be counteracted solely by administering specially selected antidotal compounds (such as mineral-vitamin premixes) or adopting environmentally safe feeding systems with an increased share of low-pollutant feeds. In addition, the impact of premixes can be augmented via the cellular-level introduction of biological preparations from medicinal plant extracts aimed at intensifying the protective functions of the organism and promoting the synthesis of specific metal-binding proteins (metallothioneins). In the experiment on the production of environmentally safe cow's milk, the most favourable results were achieved when using silagehaylage and silage-haylage-concentrate diets for Ukrainian Red-and-White Dairy cows. These outcomes may also be influenced by the genetic features that determine the character of metabolism in productive livestock exposed to toxicants from a group of heavy metals. Under the conditions of the conducted study, aimed at producing environmentally safe cow's milk, the most effective feeding systems were the silage-haylage and silage-haylage-concentrate diets used for Ukrainian Red-and-White Dairy cows. Likely, genetically determined metabolic features in productive animals under heavy metal intoxication may have additionally influenced the observed results.

CONCLUSIONS

The internal organs and tissues of dairy cows receiving feed with elevated concentrations of heavy metals (Cd and Pb) accumulate these elements. Cattle of the Ukrainian Black-and-White Dairy breed accumulated the highest levels of cadmium and lead in their kidneys (2.968–3.241 mg/kg and 4.3874.522 mg/kg), the liver (0.701–0.752 mg/kg and 1.731–1.865 mg/kg) compared to animals of the Ukrainian Red-and-White Dairy breed in the kidneys (2.305–2.634 mg/kg and 3.4523.981 mg/kg), liver (0.611–0.683 mg/kg and 1.607–1.694 mg/kg), which is due to a higher intake of pollutants from the feed in the main diets (2.177–2.833 (Cd) mg/kg and 49.56758.370 (Pb) mg/kg of dry matter in the feed, compared with 1.098–1.271 (Cd) mg/kg and 31.728–34.746 (Pb) mg/kg), and the type of feeding, rather than breed-specific characteristics. Dairy cows of the Ukrainian Red-and-White breed fed on the basis of silage-haylage and silagehaylageconcentrate had lower concentrations of cadmium and lead in internal organs, bones, and muscles of the cow. Greater influence on the accumulation of the investigated hazardous ecotoxics in the internal organs and tissues of cows of different breeds may be exerted, first and foremost, by environmental contamination factors affecting atmospheric air, soil, feed and water. Physiological factors are also important, including substantial differences in body weight, milk productivity, stage of lactation and animal age, among others, as well as the influence of

breed-related factors associated with genetic features of metabolism, differences in the intensity of accumulation or elimination of toxicants under varying housing and feeding conditions. These should be subject to a separate investigation for a breed. In future, the correlation and regression analyses of the heavy metal content in the food of feeding diets of different types and in the organs and muscles of dairy cattle should be performed.

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CONFLICT OF INTEREST

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Накопичення кадмію та плумбуму в органах та тканинах дійних корів залежно від породи і раціону

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Анотація. Моніторинг агроєкосистем в умовах повномасштабної війни вказує на великі техногенні навантаження, посилення забруднення довкілля важкими металами, в тому числі токсичними і контамінантними Cd та Pb. Метою досліджень був аналіз вмісту кадмію та плумбуму у нирках, печінці, легенях, серці, селезінці, кістковій і м'язовій тканинах дійних корів в залежності від породи, виду раціону в умовах технології виробництва екологічно безпечного коров'ячого молока. В господарствах Лісостепової ґрунтово-кліматичної зони України було проведено досліді на дійних коровах різних порід з контрольним забоєм в кінці експерименту. Найбільшу концентрацію Cd було встановлено у нирках корів української чорно-рябої молочної породи з силосно-сінним раціоном $2,968 \pm 0,193$ мг/кг та силосно-коренеплодним $3,241 \pm 0,318$ мг/кг, аналогічно по Pb з силосно-сінним типом годівлі $4,387 \pm 0,092$ мг/кг та силосно-коренеплодним $4,522 \pm 0,546$ мг/кг. Аналіз м'язової тканини на вміст Cd показав найбільшу концентрацію у тварин української чорно-рябої молочної породи з силосно-сінним раціоном $0,125 \pm 0,026$ мг/кг та силосно-коренеплодним $0,140 \pm 0,006$ мг/кг. Концентрація Pb у м'язовій тканині теж найбільшою встановлена у корів української чорно-рябої молочної породи $1,472 \pm 0,065$ мг/кг, де згодовувався силосно-сінний раціон і $1,691 \pm 0,092$ мг/кг силосно-коренеплодний. Найбільшу концентрацію Cd та Pb у нирках, печінці, легенях, селезінці, серці, кістках та м'язовій тканині встановлено у корів української чорно-рябої молочної породи в порівнянні з тваринами української червоно-рябої молочної породи, що обумовлене більшим надходженням політантів з кормами основних раціонів, типом годівлі, ніж породними особливостями. Отримані дані можуть бути використані при розробці рекомендацій щодо вибору типів раціонів і технологій годівлі дійних корів з метою зменшення акумуляції важких металів у внутрішніх органах і тканинах, а також для підвищення екологічної безпечності коров'ячого молока та продуктів його переробки

Ключові слова: екотоксиканти; корми; тип годівлі; важкі метали; продуктивні тварини; агроєкосистеми