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Accumulation of Cu and Zn in the soils, rough fodder, organs and muscle tissues of cattle in Western Siberia.

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ABSTRACT

The aim of the present study was to determine the Cu and Zn accumulation in the soils, rough fodder, organs and muscle tissue of Hereford and Black-and-White cattle in Western Siberia. The content of copper and zinc in soils and fodder have been studied in two groups of districts of the Novosibirsk Region, distinguished by dominant cattle breeds – Hereford beef cattle and Black-and-White dairy cattle. The concentration of elements in soils and fodder was determined by flame atomic absorption method using "Kvant-2A" spectrometer and counted up to absolute dry matter of samples. The copper and zinc concentration in soils and fodder did not exceed maximum permissible concentrations (MPC). The analysis of organs and muscle tissue with respect to the presence of heavy metals has been conducted by the atomic absorption spectrometry method using Thermo Scientific ICAP-6500 (hair) and Perkin Elmer 360 (other organs and muscle tissue) spectrophotometers. The average population-based level of copper and zinc was determined in hair, liver, kidneys, spleen, heart, lungs and muscle tissue in Black-and-White and Hereford breeds. In the first group of the studied areas, zinc concentration in the fodder was higher than in the second group. The maximum zinc content was detected in the hair and muscles of animals of both breeds, whereas minimum concentration was detected in the spleen of dairy cattle and in the lungs of beef cattle. The highest accumulation of copper was registered in the liver of the animals regardless of their breed, and the lowest one - in muscles of the beef cattle and in the spleen of the dairy cattle. Interbreed differences in terms of zinc content in hair, liver, and spleen as well as the copper content in all organs and muscle tissue were identified. It may be concluded that the satisfactory content of Cu and Zn in soil, fodder and animal muscle tissue in Western Siberia is favorable for obtaining ecologically safe products.
Keywords: accumulation, copper, zinc, soil, fodder,cattle, Hereford breed, Black-and-White breed.

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INTRODUCTION

Copper and zinc are natural components of the environment and essential micronutrients for plants and animals. At the same time, they are dangerous heavy metals that pollute the environment due to anthropogenic activities (García-Vaquero et al., 2011; López-Alonso et al., 2002). Therefore, metals can accumulate in high concentrations in soils and plants, and penetrate further into the organs and muscle tissue of animals, rendering them toxic effects (Chysyma et al., 2003b; Korotkevich et al., 2014). Metal toxicity depends on animal species, as well as a dose and exposure duration. Although contamination of fodder with heavy metals cannot be entirely avoided given the prevalence of these substances in the environment, it is necessary to minimize this contamination. Human being is the final link in the trophic chain. It's therefore important to reduce the direct impact of heavy metals on both the health and productivity of animals, and indirect effects on human health (Miranda et al., 2009). Toxic effects caused by metals have been revealed in animals at relatively low concentrations (Kostial, 1986). One of the earliest effects is a violation of the microelement metabolism (López-Alonso et al., 2002). In these circumstances, the issues of biomonitoring and bioindication of the environment and the trophic chain links become topical to prevent the accumulation of excessive concentrations of heavy metals in animals of different species and humans (Chysyma et al., 2003a). In addition, to ensure high animal productivity and environmental safety of the resulting products, it is necessary to conduct a comprehensive assessment of farm animals interior with regard to content of chemical elements, as well as to determine hematological, biochemical, immunological and other parameters (Miller et al., 2012; Narozhnyh et al., 2012; Petukhova, 2012).

MATERIALS AND METHODS

Soil samples were taken from the 0-20 cm layer from the field where fodder was grown. Further five plots were laid. At least 5 individual soil samples were taken from each plot to prepare the average sample. Besides, samples of hay, straw and compound animal fodder stuff were studied as well. In total, there were selected 84 soil samples and 84 plant samples. Studies were performed in three analytical replicates. Samples of hay and straw stored in ricks and stacks were selected manually around the perimeter of rick and stack at equal distances from each other at a height of 1.0-1.5 m from the ground surface all around from the depth of 0.5-1.0 m. Every spot sample, taken from each stack and rick, weighted 0.3-0.5 kg. Spot samples were combined into unified sample of 2 kg in weight. For this, hay samples were stacked in a thin layer (3-4 cm) on the tape and gently mixed avoiding breaking up plants and the formation of dust. Combined hay sample was used for preparing an average sample for analysis. For this, bundles of hay weighing 100-120 g were selected from no less than 10 different locations throughout the layer area and thickness so that showered parts of the plant were also included into the sample. The obtained average sample with a weight of not less than 1 kg was packed in a dry plastic bag and transported to the laboratory. Samples of compound animal fodder stuff, stored in warehouse in the containers, were sampled with special probes from different layers (upper, middle and lower) as well as in the center. Each sample weighted 0.8-1.0 kg. At that, 5 samples were selected per each 10 ton of fodder tuff. Obtained spot samples were thoroughly mixed to get the common sample, which was laid out in a square on a smooth surface and manually divided diagonally into four triangular sections by means of two short wooden planks. The two of obtained triangular sections were removed, while the remaining two sections were used after a few shuffling for preparing average sample weighting 0.5-1.0 kg. Average sample was placed in a plastic bag and transported to the laboratory.

The total contents of zinc and copper in the soil were determined after decomposition of the samples in an autoclave with a mixture of mineral acids. Labile forms of chemical elements extracted using acetate-ammonium buffer with pH 4.8. The plants were assessed also with regard to the total ash content. We used the atomic absorption spectrometry method making use of “Kvant-2A” photometer. The contents of zinc and copper are presented after recalculating on air-dry specimens.

More than 420 samples of organs and muscle tissue were taken from 31 bull of Hereford breed and 31 bull of Black-and-White breed at the age of 18 months. At the time of slaughter all animals were clinically healthy. Beef cattle were grown in the farming enterprises of Novosibirsk region, namely in Maslyaninsky, Krasnozersky and Novosibirsky areas (zone 1). Animals of Black-and-White breed were selected from Ubinsky district of the Novosibirsk region (zone 2). Samples, 100 g each, were selected from the following organs: muscle tissue, liver, kidneys, heart, and spleen; lungs were taken from the diaphragm, and the hair was cut at the withers. The organs and muscle tissue were frozen and kept at a temperature of -24^{o}C until the study.

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Determination of zinc and copper concentration in the organs and muscle tissue was conducted at the biochemical laboratory of the Siberian Research Institute of Livestock using atomic absorption spectrophotometer Perkin Elmer 360 (USA). Content of these elements in hair was determined by atomic emission spectrometry at the analytical laboratory of Nikolaev Institute of Inorganic Chemistry, Siberian Branch of the Russian Academy of Sciences using spectrometer ICAP-6500 produced by Thermo Scientific (USA).

Sample preparation of the organs and muscle tissue of animals for atomic absorption analysis was carried out in the following order: after washing in soap solution the dishes were washed with tap water, rinsed with redistilled water and then dried. A portion of sample weighing 100 g was crushed to a homogeneous mass, and then dried in an oven at a temperature of 60-70^{o}C degrees for about 12 hours to constant weight. Further, three-gram sample was selected from the obtained dry matter to cinder in a muffle furnace at a temperature of 500-550^{o}C. After 10-15 hours, the mineralization was over and the ash acquired a gray or white color. After cooling the samples at a room temperature, the ash residue was dissolved in 3 ml of 50% hydrochloric acid, and then dried to a dry residue on an electric stove. Then, the residue was transferred into volumetric flask by diluting it to 25 ml of distilled water. Prepared solution was analyzed for elemental composition.

For hair analysis, a hair sample of 100 mg was selected. Selected hair was carefully washed. To clean hair from pollution, hair sample was placed in a flask with distilled water and mixed for one minute with a mixer at a rotation speed of 1000 rpm. Water was changed up to 10 times and procedure was repeated. Then hair was washed in ACS acetone 49-5 within two minutes, then washed 3 times with deonized water, and dried at a room temperature. Then, the sample was dissolved in 2 ml of ACS nitric acid 27-5 and placed in a standard autoclave of a microwave oven MARS-5 ("SEM"). The autoclave was sealed and the dissolution was carried out gradually over 40 minutes raising the temperature to up 180°C.

After the decomposition procedure, the autoclaves were cooled and opened. The resulting solution was quantitatively transferred into a measuring flask. The analysis was subjected to the solutions after 10-fold and 100-fold dilution to the calibration solutions prepared on the basis of multi-element standards MES ("Skat").

STATISTICAL ANALYSIS

The obtained results were analyzed using the STATISTICA 6.0 and Microsoft Excel software. The original data on the zinc and copper contents were tested for normality of distribution using the Shapiro-Wilk test. If the distribution of variables did not conform to normal, thus it was decided to calculate the arithmetic mean, standard deviation, and the standard error of mean using the median, sample size, maximum and minimum values. We used formulas for calculating the mean and the variance values proposed by Hozo et al. (2005):

$$\bar{x} \approx \frac{a+2\mathrm{m}+b}{4} + \frac{a-2\mathrm{n}+b}{4\mathrm{n}} \;;$$

$$S^2 \approx \frac{1}{n-1}\left(a^2+m^2+b^2+\left(\frac{n-3}{2}\right)\frac{(a+m)^2+(m+b)^2}{4} - n\left(\frac{a+2\mathrm{m}+b}{4}+\frac{a-2\mathrm{m}+b}{4\mathrm{n}}\right)^2\right)$$

where *n* – is the sample size; *a* – is the minimum value; *b* – is the maximum value; *m* – is the median. The standard error of mean was calculated based on the obtained arithmetic mean values and the variances. This method is equally effective for both normal and non-normal distributions.

RESEARCH RESULTS AND DISCUSSION

The contents of zinc and copper in animal fodder and soils are presented in Table 1. The concentrations of the investigated elements in soil and fodder were within roughly permissible concentration (RPC) and MPC (Talanov, Khmelevskyi, 1991). The difference between the MPC and gross copper content was much greater (more than 7 times) in the fodder than in the soil. Zinc concentration in the soil and fodder was higher than that of copper, at that, in the soil it was higher by 2 times, while in the fodder – by 7 times. The zinc content in the soil was 2-3 times greater than that in the fodder, whereas the copper concentration in the soil was 7 times higher than in the fodder. In terms of accumulation of zinc and copper in soils and fodder, no differences were found between district groups 1 and 2, with the exception of zinc concentration in the fodder. Thus in the zone 1, the content of zinc in fodder was higher (P<0.001).

Table 1: The total content (mean ± standard deviation) of Cu and Zn in soils and rough fodder in the Novosibirsk Region, ppm

Zone	1		2		RAC	MPL
Indicator	soil	fodder	soil	fodder	soil	fodder
Zink	67.2±19.8	28.9±11.8	68.1±17.5	21.9±11.5	220.0	50.0
Copper	30.8±10.3	4.4±1.1	33.8±8.2	3.8±1.2	130.0	30.0
n	63	63	21	21		

Data on the zinc content in the cattle organs and muscle tissue are presented in Table 2. The level of zinc in the Hereford cattle organs and muscle tissue can be ranked as follows: lungs = heart < spleen < kidneys < liver < muscles < hair in a ratio of 1 : 1 : 1.1 : 1.2 : 2.2 : 2.4 : 7.5, and in Black-and-White breed – spleen < lungs < kidneys = heart < liver < muscles < hair in a ratio of 1 : 1.2 : 1.3 : 1.3 : 2.1 : 3 : 6. This series show that most of the zinc in cattle of both breeds is accumulated in the hair, muscles and liver. In Hereford cattle a minimum level of zinc was observed in the lungs, while in Black-and-White cattle – in the spleen.

Differences between beef and dairy cattle (P<0.001) in the zinc content were revealed in hair, muscles, liver, spleen and heart. Thus, the content of zinc in Hereford cattle as compared to Black-and-White cattle was higher in the hair (by 32%), liver (by 18%), and spleen (by 25.5%). The concentration of this element was higher in dairy cattle as compared to beef cattle in muscles (by 13%) and in heart (by 10.5%). The level of zinc in the kidneys and lungs in animals of the two breeds did not differ.

Table 2 The zinc content in the cattle organs and muscle tissue, ppm

Breed	Hereford beef cattle			Black-and-White dairy cattle		
Organ, tissue	n	Mean±SE*	Range	n	Mean±SE*	Range
Hair	30	143.7±6.4	93.0-220.0	31	97.9±1.4	86.9-121.2
Muscles	31	46.4±1.9	27.5-66.6	31	48.3±1.9	24.6-63.7
Liver	27	42.1±1.1	31.1-51.9	30	34.3±0.9	26.4-45.0
Kidneys	31	23.3±1.0	16.1-35.4	31	20.7±1.2	12.4-35.7
Spleen	31	21.7±0.5	16.3-27.1	30	16.3±0.3	13.6-20.1
Heart	29	20.0±0.6	15.0-26.7	31	20.8±0.5	16.0-27.0
Lungs	31	19.2±0.6	13.8-26.6	28	20.0±0.6	13.7-25.2

*Here in after SE is standard error.

Table 3 shows the level of zinc in the organs and muscle tissue of Hereford and Black-and-White cattle. Series of increase in the level of copper accumulation in beef cattle was as follows: muscles < spleen < lungs < heart < kidneys < hair < liver in the ratio 1 : 1.7 : 1.8 : 2.5 : 3.3 : 9.6 : 26.4, whereas in dairy cattle it was the following: spleen < lungs < muscles < heart < hair < kidneys < liver in the ratio 1 : 1.7 : 2 : 4.7 : 8.6 : 10.2 : 18.7. The maximum content of copper in examined cattle was revealed in the liver, while the minimum content was observed in the muscles of Hereford cattle and the spleen of Black-and-White cattle.

The copper content in the studied breeds of cattle varies widely in all organs (P<0.001). In some cases, the level of copper in the organs differs by more than twice. The concentration of copper in the liver, hair, spleen, and lungs of Hereford cattle was higher than that of Black-and-White cattle by 18-41%. In turn, the level of copper in dairy cattle was higher than that in beef cattle, particularly in the kidneys (by 61%), heart (37%) and muscles (54%).

The zinc content ratio in the chain of soil – fodder – muscle tissue in beef cattle was 1.4 : 0.7 : 1, while in dairy cattle it was 1.4 : 0.5 : 1. The ratio of the Cu level in the chain soil – forage – muscle tissue of beef cattle was different than that for Zn – 29.9 : 4.3 : 1 and for Black-and-White cattle – 19.2 : 2.2 : 1. Since the contents of copper and zinc in the soil and fodders differ slightly, the different distribution in particular of copper may be due to a greater difference of it's interbreed accumulation in muscle tissue. Minor differences in the zinc content ratio are confirmed by the small difference in it's accumulation in the muscle tissue.

Table 3 The copper content in cattle organs and muscle tissue, ppm

Breed	Hereford beef cattle			Black-and-White dairy cattle		
Organ, tissue	n	Mean±SE	Range	n	Mean±SE	Range
Hair	30	27.22±2.47	4.4-53.7	30	16.10±2.10	1.30-43.20
Muscles	31	9.86±0.38	6.20-14.00	31	7.39±0.20	5.10-9.60
Liver	31	3.45±0.17	1.60-5.40	31	8.80±1.20	1.70-26.30
Kidneys	29	2.53±0.20	0.54-4.50	31	4.00±0.18	1.70-5.30
Spleen	31	1.85±0.12	0.75-3.23	30	0.86±0.03	0.66-1.20
Heart	30	1.79±0.08	1.16-2.77	28	1.47±0.06	0.99-2.16
Lungs	28	1.03±0.03	0.80-1.30	31	1.76±0.09	1.10-2.90

The contents of Cu and Zn in the organs and muscle tissue of the cattle of different breeds could be considered as physiological and environmental standards in Western Siberian conditions. The content of these elements was investigated also in the Novosibirsk region in fast-growing meat pigs. The Cu concentration in pig muscle tissue, liver and kidneys was approximately the same as in the examined cattle. However, skeletal muscles and organs of pigs are more enriched with Zn, than those of cattle (by 1.6 – 4 times) (Korotkevich et al., 2009).

Domaradzki et al. (2015) investigated the level of Cu and Zn content in the muscles *longissimus lumborum* and *semitendinosus* from animals of 4 dairy cattle breeds – Polish Red, White-Backed, Polish Black-and-White, and Polish Holstein-Friesian, as well as in Simmental dairy-beef breed. The authors note the influence of the breed on the accumulation of copper, zinc and other chemical elements. Thus, muscle tissue of White-Backed bulls accumulate significantly ($P<0.01$) more Cu as compared to other dairy cattle (Polish Red and Polish Black-and-White) as well as dairy-beef cattle. In our study, the concentration of copper in the animals of the Black-and-White and Hereford breeds was 2-3 times higher than that in cattle from Poland. The influence of breed is shown for zinc as well. The level of zinc in the animals of the Polish Holstein-Friesian breed was significantly higher than that in Polish Red and Polish Black-and-White breeds. According to our data, the content of Zn in cattle of Western Siberia was higher than that in Polish Holstein-Friesian breed. However, in addition to pedigree, differences in the accumulation of trace elements may be associated with different types of muscles tested in our work and in the study of Domaradzki et al. (2015). Thus, Czerwonka and Szterk (2015) have shown in their experiment that zinc content in cattle can vary by two times depending on the type of muscles. In Hereford and Braford beef cattle from Uruguay, the content of zinc and copper also considerably differ in different muscles (Cabrera et al., 2010). Data on the Zn content in muscles of Hereford cattle from Siberia are comparable with data on the Zn content in the rib plate-flank and the Cu content in the tenderloin and eye of rump from the Uruguayan cattle.

Miranda et al. (2005) have shown that the copper and zinc content in muscle tissue and kidneys of calves aged 9-12 months in rural and industrial areas do not differ. However, the level of zinc and copper in the liver in these areas differed; at that, the contents of zinc and copper in liver of the animals from the rural district was higher than that in the animals of industrial district. In cattle of Western Siberia, the level of Cu and Zn in these organs and muscle tissue was similar to the data of Miranda et al. (2005). However, the accumulation of copper in the liver of Black-and-White cattle was lower by 1.5 - 2 times, whereas in the kidneys it was more than twice higher in contrast to animals from Spain.

In cattle at the age of 3 to 5 years old grown in Slovakia at three farms (Haniska, Cestice and Perin) situated within 2, 5.5 and 6 km from the smelter, a significant influence of the industrial enterprise on the level of heavy metals in muscles and liver of cattle of different ages was not revealed (Korenekova et al., 2002). However, the concentration of copper in the liver and muscular tissue in cattle from Slovakia was much higher (up to 2-5 times) than in animals from the West Siberia.

Intriguing data were obtained on the content of zinc and copper in liver and kidneys (dry weight) in cattle, grown near one of the most polluted areas of the planet – lead-zinc mine in Kabwe (Zambia) and in five other areas of Zambia (Yabe et al., 2011). Data presented by the authors of the conducted study show that the content of copper and zinc in cattle from Zambia are significantly higher than in animals from Russia. Thus, the concentration of copper in the liver and kidneys was by 2-5 times higher, while the level of zinc in the liver and kidneys was 3-6 times higher than in cattle of Siberia.

Thus, the content of copper and zinc in soil, fodders, organs and muscle tissue of Siberian cattle does not exceed maximum permissible levels.

CONCLUSION

In soils and rough fodders from different zones of Western Siberia, the contents of Cu and Zn were below MPL and RAC. The data obtained can be used for further monitoring the level of these elements in other zones of Siberia.

It is shown that the cattle breed has impact on the accumulation and distribution of zinc and copper in cattle organs and muscle tissue within the MPC. Interbreed differences in the concentration of zinc and copper may indicate a specific role of hereditary component in their accumulation. Therefore, to assess the elemental status of the animals, these factors should be taken into consideration. The obtained average population values of Cu and Zn content in the organs and muscle tissue of dairy and beef cattle could be considered as physiological and environmental standards in Western Siberian conditions. The territory of Western Siberia because of satisfactory content of the studied heavy metals in soil, fodders and muscular tissue of cattle is suitable for the production of ecologically safe meat products, including baby food.

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REFERENCES

[1] Cabrera, M., Ramos, A., Saadoun, A., and Brito, G., 2010, “Selenium, Copper, Zinc, Iron and Manganese Content of Seven Meat Cuts from Hereford and Braford Steers Fed Pasture in Uruguay,” Meat Science, 84, pp. 518–528. doi:10.1016/j.meatsci.2009.10.007.

[2] Chysyma, R.B., Bakhtina, Y.Y., Petukhov, V.L., Korotkova, G.N. and Kochneva, M.L., 2003, “Heavy Metal Concentration in Water and Soil of Different Ecological Areas of Tyva Republic,” J. De Physique. IV, Proceedings of JP XII International Conference on Heavy Metals in the Environment. Editors: C. Boutron and C. Ferrari, Grenoble pp. 301-302. DOI: 1051/jp4:20030301

[3] Chysyma, R.B., Petukhov, V.L., Kuzmina, E.E., Barinov, E.Ya., Dukhanov, Yu.A. and Korotkova, G.N. 2003, “The Content of Heavy Metals in Fodder of the Tyva Republic,”. J. De Physique IV: Proceedings of JP XII International Conference on Heavy Metals in the Environment. Editors: C. Boutron and C. Ferrari, Grenoble, pp. 297-299.

[4] Czerwonka, M. and Szterk, A., 2015, “The Effect of Meat Cuts and Thermal Processing on Selected Mineral Concentration in Beef from Holstein-Friesian Bulls,” Meat Science, 105, pp. 75-80, Retrieved from http://dx.doi.org/10.1016/j.meatsci.2015.03.011

[5] Domaradzki, P., Florek, M., Staszowska, A., and Litwińczuk, Z., 2015, “Evaluation of the Mineral Concentration in Beef from Polish Native Cattle,” Biol. Trace Elem. Res., [Epub ahead of print] DOI: 10.1007/s12011-015-0549-3

[6] García-Vaquero, M., Miranda, M., Benedito, J.L., Blanco-Penedo, I., López-Alonso, M., 2011, “Effect of Type of Muscle and Cu Supplementation on Trace Element Concentrations in Cattle Meat,” Food Chem. Toxicol., 49(6), pp. 1443-1449, DOI: 10.1016/j.fct.2011.03.041

[7] Hozo, S.P., Djulbegovic, B., and Hozo, I., 2005, “Estimating the Mean and Variance from Themedian, Range, and the Size of a Sample,” BMC Medical Research Methodology, 5, 13, Retrieved from http://dx.doi.org/10.1186/1471-2288-5-13

[8] Korenekova, B., Skalicka, M., and Nad, P., 2002, “Concentration of Some Heavy Metals in Cattle Reared in the Vicinity of a Metallurgic Industry,” Veterinarski Arhiv, 72(5), pp. 259-267.

[9] Korotkevich, O.S., Petukhov, V.L., Sebezhko, O.I., Barinov, Ye.Ye., and Konovalova, T.V., 2014, “Content of 137 Cs and 90 Sr in the Forages of Various Ecological Zones of Western Siberia,” Russian Agricultural Sciences, 40(3), pp. 195-197.

[10] Korotkevich, O.S., Zheltikova, O.A., and Petukhov, V.L., 2009, Biohimicheskie, gematologicheskie parametry i akkumulyaciya tyazhelyh metallov organah i tkanyah svinej skorospeloj myasnoj porody [Biochemical, Hematological Parameters and Accumulation of Heavy Metals in Organs and Tissues of Fast-growing Meat Breed Pigs], Proceedings of the Russian Academy of Agricultural Sciences, 4. pp 41-43.

[11] Kostial, K., 1986, “Cadmium,” In: Mertz, W. (Ed.). “Trace Elements in Human and Animal Nutrition,” (5th ed.), Academic Press, INC. San Diego.

[12] López-Alonso, M., Benedito, J.L., Miranda, M., Castillo, C., Hernández, J., and Shore, R.F., 2000, “Arsenic, Cadmium, Lead, Copper and Zinc in Cattle from Galicia,” NW Spain. Sci. Total Environ., 246 (2–3), pp. 237–248.

[13] López-Alonso, M., Benedito, J.L., Miranda, M., Castillo, C., Hernández, J., and Shore, R.F., 2002, “Contribution of Cattle Products to Dietary Intake of Trace and Toxic Elements in Galicia, Spain,” Food Addit. Contam., 19(6), pp. 533–541.

[14] Miller, I.S., Petukhov, V.L., Korotkevich, O.S., Korotkova, G.N., and Konovalov, I.S., 2013, “Accumulation of Heavy Metals in the Muscles of Zander from Novosibirsk Water Basin,” Proceeding of the 16th International Conference on Heavy Metals in the Environment, Rome, Italy, DOI: http://dx.doi.org/10.1051/e3sconf/20130111007

[15] Miranda, M., Lopez-Alonso, M., Castillo, C., Hernadez, J., and Benedito, J.L., 2005, “Effects of Moderate Pollution on Toxic and Trace Metal Levels in Calves from a Polluted Area in Northern Spain,” Environ Int., 31, pp. 543-548.

[16] Miranda, M., Benedito, J.L., Blanco-Penedo, I., López-Lamas, C., Merino, A., and LópezAlonso, M., 2009, “Metal Accumulation in Cattle Raised in a Serpentine-soil Area: Relationship between Metal Concentrations in Soil, Forage and Animal Tissues,” J. Trace Elem. Med. Biol., 23(3), pp. 231–238.

[17] Narozhnyh, K.N., Efanova, Y.V., Petukhov V.L., et al., 2013, “The Content of Lead in Some Organs and Tissues of Hereford Bull-calves,” Proceeding of the 16th International Conference on Heavy Metals in the Environment, Rome, Italy, DOI: http://dx.doi.org/10.1051/e3sconf/20130115003

[18] Petukhov, V.L., Dukhanov, Yu.A., Sevryk, I.Z., Patrashkov, S.A., Korotkevich, O.S., Gorb, T.S., and Petukhov, I.V., 2003, “Cs-137 and Sr-90 Level in Dairy Products,” J. De Physique IV: Proceedings of JP XII International Conference on Heavy Metals in the Environment. Grenoble, pp. 297-299.

[19] Petukhova, T.V., 2012, “Content of Heavy Metals in the Muscle Tissue of Cattle,” 16th International Conference on Heavy Metals in the Environment, Rome, Italy, DOI: http://dx.doi.org/10.1051/e3sconf/20130115002

[20] Yabe, J., Nakayama, S.M.M., Ikenaka, Y., Muzandu, K., Ishizuka, M., and Umemura, T., 2011, “Uptake of Lead, Cadmium, and other Metals in the Liver and Kidneys of Cattle near a Lead-zinc Mine in Kabwe, Zambia,” Environmental Toxicology and Chemistry, 30, 8, pp. 1892-1897.

[21] Talanov, G.A., and Khmelevsky, B.N., 1991, Sanitariya kormov: Spravochnik [Fodder Sanitation: a Handbook], Mocow, Agropromizdat,. 303 p.

[22] Methodical Guidelines 2.1.7.730–99, 1999, Metodicheskie ukazaniya: Gigienicheskaya ocenka kachestva pochvy naselennyh mest: utv. Glavnym gosudarstvennym sanitarnym vrachom RF [Hygienic Assessment of Soil Quality in Residential Areas,” Approved by Chief State Medical Officer of the Russian Federation 7.02.1999], enforced on 05.04.1999, Moscow, 20 p.