Bulgarian Journal of Agricultural Science, 27 (No 3) 2021, 593–599

Determination of heavy metals in liver and skeletal muscles of pigs and calves: experience from Bosnia and Herzegovina

Edhem Haskovic[1], Tidza Muhic-Sarac[2], Maja Lukic[1], Maja Marjanovic[1], Sabina Zero[2] and Erna Islamagic[1*]

[1]*University of Sarajevo, Department of Biology, Faculty of Science, 71 000 Sarajevo, Bosnia and Herzegovina*
[2]*University of Sarajevo, Department of Chemistry, Faculty of Science, 71 000 Sarajevo, Bosnia and Herzegovina*
[*]*Corresponding author:* erna.islamagic@hotmail.com

Abstract

Haskovic, E., Muhic-Sarac, T., Lukic, M., Marjanovic, M., Zero, S. & Islamagic, E. (2021). Determination of heavy metals in liver and skeletal muscles of pigs and calves: experience from Bosnia and Herzegovina. *Bulg. J. Agric. Sci., 27 (3)*, 593–599

Certrain heavy metals, as trace elements, have a significant role in biochemical processes of living systems. However, when present in quantities exceeding those required for accomplishing their biological functions, they can have a toxic effect. Given that, meat and meat products have an important role in human diet the objectives of the present study was to estimate Cr, Cu, Mn, Fe, Ni, Cd, Pb and Zn concentrations in liver and muscle tissues of pigs and calves from Bosnia and Herzegovina; moreover to compare the obtained values in order to determine the potentially same pattern of heavy metals accumulation and contamination. After acid digestion,concentration of metals was determinated by atomic absorption spectrometry – flame technique (FAAS). Chromium and manganese levels in pig's muscle tissue were higher compared to liver tissue concentration of analyzed metals, while zinc had higher content in calf's muscle. Levels of copper and manganese were higher in calf's hepatic tissue compared to muscle, and the same results for manganese, iron and zinc was obtained from analysis of pig's tissues.

Keywords: heavy metals; FAAS; pig; calf; liver; muscles

Introduction

Heavy metals are a term used to describe the group of elements associated with potential toxic effects on biological systems (Duffus, 2002). Their redox properties provided them with a possibility to avoid the control mechanisms of the cell resulting in interaction with proteins, displacement of other metals from their binding sites and generating reactive oxygen species which eventually leads to cell destruction (Flora et al., 2008). However, some heavy metals, as essential nutrients have a significant role in biochemical and physiological processes in human body regulating enzymatic activity and controlling metabolic and signaling pathways (Fraga, 2005). For instance, zinc is trace element crucial for the function of more than 300 enzymes (Marreiro et al., 2017), chromium and manganese are involved in carbohydrate and lipid metabolism (Aschner & Aschner, 2005; Pechova & Pavlata, 2007), while iron and copper affect erythropoiesis regulating hemoglobin metabolism (Collins et al., 2010).

Analysis of metal concentration in livestock show that farm animals have not only proved to be a good biomonitor species for metal contamination and indicator of environmental pollution (Korenekova et al., 2008), but can also have a harmful effect on human health through the food chain (Duruibe et al., 2007), as heavy metals have a tendency to accumulate. Given that intensive animal farming is based on quantity, accumulation of toxic chemicals in industrial livestock can be the result of a compounded feed to which mineral supplements with considerable amounts of toxic metals are added (EFSA, 2004). Considering the availability of these toxicants, maximum admissible levels for meat and offal have been established by the European Union (Commission Regulation, EC, 2006).

The previous studies of heavy metal concentration in cattle and pig's tissue show that liver, kidneys and muscle take central place in heavy metal metabolism (Falandysz, 1993; Sedki et al., 2003). In terms of distribution, these toxicants tend to accumulate in liver tissue with an exception of zinc for which muscles represent a major storage site in cattle (Alonso et al., 2010; Jorhem et al., 1989; Kottferova & Korenekova, 1995). Generally, some heavy metals are essential, trace elements with nutritional benefits in biological systems, with an exception of cadmium, lead, mercury and arsenic since there is no level of these elements that appears to be necessary to the body (Flora et al., 2012; Thevonod, 2010). Results of previous studies show that heavy metal accumulation is not always associated with age (Lopez-Alonso et al., 2007). Studies of heavy metal concentration in pig and cattle's tissues in past decades found only in rare cases exceeded the maximum acceptable levels as specified **(**Lopez-Alonso et al., 2007).

Monitoring and assessment of the heavy metals concentration in the animal tissues is usually carried out in developed countries as it has been proven that their toxicity is a major threat and several health risks are associated with it (Jaishankar et al., 2014). However, information regarding metal concentrations in muscle and liver tissue of pig and cattle are not available in Bosnia and Herzegovina. The objective of the present study was to determinate Cr, Cu, Mn, Fe, Ni, Cd, Pb and Zn concentrations in liver and muscle tissues of pigs and calves and to contribute to the establishment of a database of toxic metals on the territory of Bosnia and Herzegovina. Also, we compared the obtained values within selected species to determine potential same pattern of heavy metals accumulation and contamination.

Materials and Methods

Sampling

In this study we analyzed concentration of heavy metals in a total of 40 liver and muscle samples of pig and calves. Study was performed in Laboratory of Biochemistry and Physiology and Laboratory of Analytical Chemistry at Faculty of Science, University of Sarajevo, Bosnia and Herzegovina. Ten liver and ten muscle samples of each species were collected from butcheries in Sarajevo in the period of May to June 2017. The research included both female and male individuals, approximately 5 months (calves) and eight months (pigs) old. All samples were packed individually in plastic bags and transported to the laboratory for analysis.

Chemical and statistical analysis

The concentration of Cr, Cu, Mn, Fe, Ni, Cd, Pb and Zn were determined by the flame atomic absorption spectrometry using AA240FS atomic absorption spectrometer, Varian, Australia. Sample wet-mineralization was performed using modified method of Welna et al. (2011). This method is based on an acid wet-mineralization of 5 g (fresh sample) of liver or muscle tissue in 20 mL of 65% HNO_3 (Merck, Germany) and heating on a hot plate until complete sample digestion. Subsequently, 5 mL of 30% H_2O_2 (Merck, Germany) was added and samples were heated for approximately 5 minutes. The resulting solutions were filtrated through filter paper white ribbon, quantitatively transferred to a 50 mL volumetric flask and stored at room temperature for further analysis. To check the accuracy of the method, blank samples were analyzed at the same time and with the same manner of the pig and calf's samples. The instrument was calibrated with multi-element aqueous standards, prepared from 1000 mg/l certified atomic absorption standard solutions of Cr, Cu, Mn, Fe, Ni, Cd, Pb and Zn (CetriPUR, Merck, Germany). Samples were carefully handled to avoid contamination. Glassware was properly cleaned, and the reagents were of analytical grade. Double distilled deionized water was used throughout the study.

Student's t-test was performed to determine the statistically significant differences regarding heavy metal concentrations in different tissues (liver and muscle) and between different species (calf and pig) at a significance level of 0.05. The detection limits (LOD) were calculated on the basis of three times the standard deviation of the blank signal.

Results

Presented study shows results of heavy metal (Cr, Cu, Mn, Fe, Cd, Pb, Ni and Zn) concentration in muscle and liver tissues of pig (n=20) and calf (n=20) determined by flame atomic absorption spectrometry (FAAS) in order to establish a database of toxic metals on the territory of Bosnia and Herzegovina and to estimate the risk of contamination.

Average values and t-test results of the comparison between two different tissues within same species for pig and calf are presented in Table 1. The concentrations of heavy metals in muscle and liver tissues of pig and calf are expressed in mg/kg wet weight (fresh sample). Results showed statistically significant differences between Cr, Cu, Mn, Fe, Ni and Zn concentrations in liver and muscle tissue of pig. As for the results obtained by calf tissues analysis, difference in Cu, Fe, Ni, Cd and Pb levels were statistically significant. In general, iron had the highest concentration of all analyzed metals in liver – 130.86 mg/kg and 39.47 mg/kg for pig and calf respectively, while cadmium (0.09 mg/kg for pig and 0.10 mg/kg for calf) and chromium (0.26 mg/kg for pig and 0.13 mg/kg for calf) levels in both tissues were the lowest.

Descriptive statistics and t-test results for heavy metals concentration in muscle tissue of both analyzed species presented in Table 2 showed statistically significant differences in Cr, Mn and Zn concentration between species. Lead concentration in pig's muscle varied from non-detectable by using FAAS (10 % of samples) to 1.88 mg/kg. In calf muscle chromium, manganese, cadmium and lead concentrations ranged from non-detectable by using FAAS to 0.48 mg/kg (Cr), 0.17 mg/kg (Mn), 0.19 mg/kg (Cd) and 1.80 mg/kg (Pb).

Table 3 presents descriptive statistics and t-test results for heavy metal concentrations in liver tissue of both analyzed species with statistically significant differences in Cu, Mn, Fe and Zn concentrations. Lead concentration in pig's liver tissues ranged from non-detectable by using FAAS (40% of samples) to 2.41 mg/kg. Chromium, nickel, cadmium and lead levels in calf's liver tissues varied from non-detectable by using FAAS to 0.42 mg/kg (Cr), 1.09 mg/kg (Ni), 0.21 mg/kg (Cd) and 1.67 mg/kg (Pb).

The highest concentration among all analyzed metals in muscle tissues of pig and calf was the concentration of zinc (18.80 mg/kg and 26.92 mg/kg, respectively), while the highest level of iron was detected in liver tissues of both species (130.86 mg/kg and 39.47 mg/kg, respectively). Cadmium concentration in liver of pig and calf was the lowest (0.09 mg/kg and 0.10 mg/kg, respectively), and as for the

Table 1. Average values and t-test results of heavy metal concentration in liver and muscle tissue of pig and calf (mg/kg fresh sample)

Metal	Pig tissue (n=20), mg/kg			Calf tissue (n=20), mg/kg		
	Muscle	Liver	p	Muscle	Liver	P
Cr	0.49	0.26	$p < 0.05$	0.24	0.13	$p > 0.05$
Cu	0.91	6.67	$p < 0.05$	0.78	20.45	$p < 0.05$
Mn	0.26	2.07	$p < 0.05$	0.10	1.57	$p > 0.05$
Fe	10.78	130.86	$p < 0.05$	14.11	39.47	$p < 0.05$
Ni	1.13	0.72	$p < 0.05$	0.98	0.56	$p < 0.05$
Cd	0.13	0.09	$p > 0.05$	0.14	0.10	$p < 0.05$
Pb	1.26	0.98	$p > 0.05$	1.24	0.85	$p < 0.05$
Zn	18.80	29.84	$p < 0.05$	26.92	25.48	$p > 0.05$

Table 2. Comparative values of heavy metal concentration in muscle tissue of pig and calf (mg/kg fresh sample)

Muscle tissue (n=20)	Species	Cr, mg/kg	Cu, mg/kg	Mn, mg/kg	Fe, mg/kg	Ni, mg/kg	Cd, mg/kg	Pb, mg/kg	Zn, mg/kg
Mean value	Pig	0.49	0.91	0.26	10.78	1.13	0.13	1.26	18.80
	Calf	0.24	0.78	0.10	14.11	0.98	0.14	1.24	26.92
STDEV	Pig	0.13	0.42	0.06	4.67	0.22	0.05	0.50	5.37
	Calf	0.14	0.24	0.06	4.60	0.45	0.04	0.47	5.64
Max. value	Pig	0.78	1.53	0.35	17.83	1.40	0.20	1.88	28.77
	Calf	0.48	1.22	0.17	21.74	2.17	0.19	1.80	40.20
Min. value	Pig	0.29	0.26	0.16	4.89	0.86	0.05	0.59	10.81
	Calf	0.07	0.35	0.01	6.97	0.40	0.08	0.45	18.13
CV(%)	Pig	27.12	46.03	23.27	43.36	19.03	37.90	39.39	28.58
	Calf	58.51	31.08	56.88	32.57	45.35	31.41	38.19	20.95
T - test		$p<0.05$	$p>0.05$	$p<0.05$	$p>0.05$	$p>0.05$	$p>0.05$	$p>0.05$	$p<0.05$

Table 3. Comparative values of heavy metal concentration in liver tissue of pig and calf, mg/kg fresh sample

Liver tissue (n=20)	Species	Cr, mg/kg	Cu, mg/kg	Mn, mg/kg	Fe, mg/kg	Ni, mg/kg	Cd, mg/kg	Pb, mg/kg	Zn, mg/kg
Mean value	Pig	0.26	6.67	2.07	130.86	0.72	0.09	0.98	29.84
	Calf	0.13	20.45	1.57	39.47	0.56	0.10	0.85	25.48
STDEV	Pig	0.13	1.80	0.38	7.41	0.22	0.07	0.75	3.27
	Calf	0.15	12.02	0.19	13.94	0.30	0.05	0.36	5.36
Max. value	Pig	0.51	9.73	2.66	140.12	1.11	0.24	2.41	35.02
	Calf	0.42	49.60	1.87	61.54	1.09	0.21	1.67	37.67
Min. value	Pig	0.07	3.39	1.27	117.01	0.44	0.02	0.39	22.82
	Calf	0.01	6.07	1.28	19.48	0.08	0.01	0.37	17.26
CV (%)	Pig	49.99	27.02	18.55	5.67	30.31	75.85	77.12	10.96
	Calf	111.86	58.81	12.30	35.33	54.13	53.48	42.45	21.03
T - test		$p>0.05$	$p<0.05$	$p<0.05$	$p<0.05$	$p>0.05$	$p>0.05$	$p>0.05$	$p<0.05$

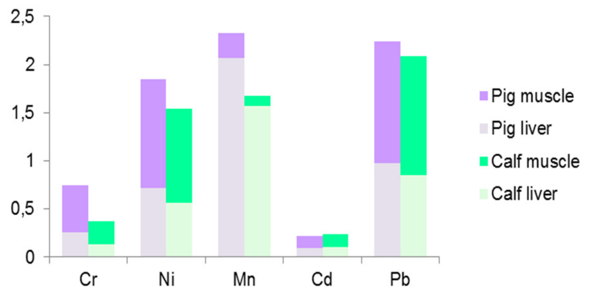

Figure 1. The average levels of heavy metals (Cr, Ni, Mn, Cd, Pb) in pig and calf's liver and muscles (mg/kg fresh sample)

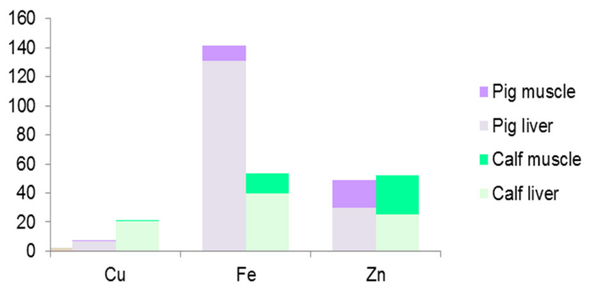

Figure 2. The average levels of heavy metals (Cu, Fe, Zn) in pig and calf's liver and muscles (mg/kg fresh sample)

results obtained by muscle tissue analysis, the lowest concentration is obtained for cadmium (0.13 mg/kg) in pig's tissue and manganese (0.10 mg/kg) in calf's tissue (Figure 1 and Figure 2).

The obtained values of LOD for the studied metals are: Cr (0.17 mg/kg); Cu (0.19 mg/kg); Mn (0.12 mg/kg); Fe (3.15 mg/kg); Ni (0.33 mg/kg); Cd (0.10 mg/kg); Pb (1.54 mg/kg) and Zn (0.21 mg/kg).

Discussion

In this study the concentration of heavy metals in the liver and muscles of the two selected species (pig and calf) was determined. The aim was to determine the concentration of the selected metals in these tissues and to make a comparison of the obtained values within the same, but also different species.

Cadmium and lead concentrations in pig muscle tissue were higher compared to liver tissue concentrations, but these differences were not statistically significant (Table 1). Obtained results for cadmium are not in accordance with those previously reported in literature. Comparative studies carried out in Finland (Falandysz, 1993) and Spain (Lopez-Alonso et al., 2007) reported higher concentration of cadmium in liver tissue but lower cadmium concentration in both liver and muscle compared to results obtained in this research. Lead concentrations were higher in muscle tissue but in 40% of liver tissue samples lead level was not detected by using FAAS, while lead concentration in 90% of muscle samples exceeded maximum permissible limit. Data presented in previous studies show higher lead concentration in liver compared to muscle (Falandysz, 1993). Lopez-Alonso et al. (2007) reported 100 times lower concentration in liver and muscle compared to results obtained in this study. Concentration of essential metals analyzed in this study was within safe range (NRC, 2005) and in general was similar to results previously reported in literature. Obtained results showed that zinc, iron, manganese and copper concentrations were higher in hepatic tissue (Table 1). Same distribution was reported in previous studies (Falandysz, 1993; Jorhem et al., 1989; Lopez-Alonso et al., 2007; Tomović et al., 2011). Chromium and nickel concentrations were higher in muscle tissue (Table 1) and Lopez-Alonso et al. (2007) also reported higher concentrations of these trace elements in muscle tissue. Higher nickel concentrations in muscle tissue were reported in Serbia (Tomović et al., 2011), while Jorhem et al. (1989) noted similar distribution in both analyzed tissues.

As already stated, the present research included analysis of heavy metal concentration in muscle and liver tissue of calf. Results show statistically significant difference in lead and cadmium concentration between these two tissues with higher levels in muscle (Table 1). Previously reported data show different distribution and significantly lower levels of lead in both tissues (Khalafalla et al., 2011; Kottferova & Korenekova, 1995; Okareh & Oladipo, 2015). Distribution of cadmium between tissues was not in accordance with literature data (Kottferova & Korenekova, 1995; Okareh & Oladipo, 2015), but levels of this metal, in general, were in agreement with those previously reported (Khalafalla et al., 2011; Korenekova et al., 2002.). Results of our research regarding essential elements (Table 1) in calf were similar to those previously reported. In terms of distribution, copper, manganese and iron concentrations were higher in liver tissue of calf (Khalafalla et al., 2011; Korenekova et al., 2002), while different distribution of chromium, zinc and nickel was earlier reported (Kottferova & Korenekova, 1995; Kramer et al., 1983; Sedki et al., 2003). Results of presented research showed that chromium, copper, manganese and zinc levels were in accordance with results of previous comparative studies. In contrast, nickel concentration in both analyzed tissues was higher compared to values listed in literature (Okareh & Oladipo, 2015, Oymak et al., 2017).

Result of cadmium concentration of liver and muscle in our study show almost the same concentrations of this toxic metal in both species (Table 2, Table 3). However, these levels are not in accordance with literature data and represent a potential health risk since maximum admissible levels for meat established by the European Union have been exceeded. European Commission (2006) proposed safe permissible level for cadmium in offal 0.05 mg/kg and in meat 0.5 mg/kg. There are several reasons to explain this highly toxic level of cadmium in analyzed tissues. We can assume that these drastically different data between studies are result of different living habitats and unrelated life conditions given that industry promotes heavy metal accumulation (Korenekova et al., 2002). However, we do not have any information regarding cadmium levels in livestock in Bosnia and Herzegovina in order to compare and evaluate whether these high concentration were the result of commercial livestock feed or environment pollution. It is possible that higher cadmium levels can be related to intensive production farming which is based on giving zinc and copper supplements. EFSA (2010) reported that high copper supplementation of feed increases the risk of cadmium accumulation in liver and kidney of pigs. Also, different analytical methods and chemicals used in mentioned studies could influence on different outcomes of research.

Results of the present study regarding lead concentration showed higher concentration of this metal in pig tissues (Table 1) but levels of lead in both species were significantly higher compared to results noted in recent literature. Average lead concentration in pig and calf's tissues exceeded maximum permissible levels of lead in muscle (0.05 mg/kg) and offal (0.1 mg/kg) established by EU (Commission Regulation, EC,, 2006). In 70% of calf liver and 90% of calf muscle samples lead levels were higher than specified safe permissible levels. As well as for cadmium, we can assume that these differences are reflecting different environment conditions and extended exposure to lead. Anthropogenic activities such as vehicular traffic are the most common lead sources (Orisakwe et al., 2017). Extended exposure and intoxication reduces lead deposition in bones and promotes binding of lead to proteins creating a form in which they are able to reach other organs (Ellah & Yahia, 2009). In general, ruminants are more resistant to toxic effects of this metal (Humphreys, 1991). It is reported that kidneys, bones and liver are predominant organs of lead accumulation and intoxication could be lethal (Spivey Fox, 1987).

As for the essential elements, different concentration among species could be consequence of different feeds and supplementation requirements. Significant differences in digestive system may be the cause of different distribution of trace elements since it has been reported that digestion and absorption are more effective in livestock (Niemi et al., 1991). Zinc concentration in muscle tissue of calf was significantly higher (Table 2) which could be related to the role of muscle in zinc accumulation in livestock. It has been reported that calf muscle possesses zinc concentrations similar to those in the liver (Orisakwe & Oladipo, 2017). Chromium levels in our pig samples were higher than those in calf, with significant difference for muscle chromium concentrations (Table 2). Chromium is an essential element for livestock animals with wide-spread use as a supplement. Authors (Pechova & Pavlata, 2007) reported that chromium supplementations of sows have positive effect on the size of litter on birth. That is one of the reasons chromium is added to livestock feed which could be the cause of greater concentrations of this metal in pig tissue in this study. Nickel concentrations in muscle and liver of pig were higher compared to calf tissues but these differences were not statistically significant (Table 2, Table 3). Nickel is essential element for ruminants given that effects the activity of ruminal urease, so Ni requirements appears to be higher in ruminants than in other animals (Miranda et al., 2009). Mangan levels in both analyzed pig tissues were significantly higher compared to results obtained from calf tissue samples (Table 2, Table 3). Manganese is essential element for pigs important for reproduction of these animals (Rasbech, 1969). Results of this study regarding copper concentration show significantly higher levels of copper in calf liver (Table 3), while concentrations of this mineral were higher in pig muscle but with no significant differences (Table 2). These interspecies differences could be result of the fact that ruminants in general show capacity to accumulate copper in the liver when this element is supplied in excess while pigs do not accumulate copper in this organ even when this element is supplied at levels above physiological requirement (Lopez-Alonso et al., 2017). Given that iron is deposited in liver (Anderson, 2005), higher levels of this metal in hepatic tissue of both species (Table 1) were expected. However, interspecies differences regarding iron concentration in this organ were statistically significant (Table 3). Lower iron levels in calf liver could be result of cadmium accumulation effecting iron absorption in livestock (Korenekova et al., 2002).

Conclusions

Based on results of the present study and available literature data we can only assume possible reasons for obtained disparity in analyzed tissues between species. There are several arguments which could explain these levels of metals, such as animal gender and age, environment pollution and presence of industry or different feeding habits. However, in order to evaluate whether the observed differences are result of different physiological structure of pig and calf or living conditions, we should perform further, more extensive, analysis. Higher number of samples would probably contribute to statistically more credible results, which would be better reflection of heavy metal content in analyzed tissues. Heavy metal biomonitoring is carried out in developed European countries as widely used technique for assessment of environment pollution. Scandinavian countries made great effort in past decades to reduce heavy metal emission which led to reduction of animal exposure to these toxicants. Introduction of these control measures of heavy metals content in foodstuff would minimize the risk of potential contamination, given that meat of pig and calf is the most consumed by Bosnia-Herzegovinian population.

Acknowledgements

We would like to thank Aida Piplica for technical support.

References

Alonso, M. L., Benedito, J. L., Miranda, M., Castillo, C., Hernandez, J. & Shore, R. F. (2000). Arsenic, cadmium, lead, copper and zinc in cattle from Galicia, NW Spain. *Science of the Total Environment, 246(2),* 237-248.

Anderson, G. J. & Frazer, D. M. (2005). Hepatic iron metabolism. *Seminars in Liver Disease, 25(4),* 420-432.

Aschner, J. L. & Aschner, M. (2005). Nutritional aspects of manganese homeostasis. *Molecular Aspects of Medicine*, *26(4),* 353-362.

Collins, J. F., Prohaska, J. R. & Knutson, M. D. (2010). Metabolic crossroads of iron and copper. *Nutrition Reviews, 68(3),* 133–147.

Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs (Text with EEA relevance) *Official Journal of the European Union*, *364*, 5-24.

Duffus, J. H. (2002). "Heavy metals" a meaningless term? *Pure and Applied Chemistry, 74(5),* 793-807.

Duruibe, J. O., Ogwuegbu, M. O. C. & Egwurugwu, J. N. (2007). Heavy metal pollution and human biotoxic effects. *Indian Journal of Pharmaceutical Sciences*, *2(5),* 112-118.

EFSA (2004). Opinion of the scientific panel on contaminants in the food chain on a request from the Commission related to cadmium as undesirable substance in animal feed. *EFSA Journal*, *102*, 1-119.

Ellah, M. R. A. & Yahia, D. (2009). Interrelationship between lead and cadmium contents in hair and internal organs of female buffaloes. *Journal of Veterinary Medicine*, *6(2),* 159-162.

Falandysz, J. (1993). Some toxic and essential trace metals in swine from Northern Poland. *Science of the Total Environment, 136(1-2),* 193-204.

Flora, G., Gupta, D. & Tiwari, A. (2012). Toxicity of lead: a review with recent updates. *Interdisciplinary Toxicology, 5(2),* 47-58.

Flora, S. J., Mittal, M. & Mehta, A. (2008). Heavy metal induced oxidative stress and its possible reversal by chelation therapy. *Indian Journal of Medical Research*, *128(4),* 501.

Fraga, G. C. (2005). Relevance, essentiality and toxicity of trace elements in himan health. *Molecular Aspects of Medicine, 26(4),* 235-244.

Humphreys, D. J. (1991). Effects of exposure to excessive quantities of lead on animals. *British Veterinary Journal, 147(1),* 18-30.

Jaishankar, M., Tseten, T., Anbalagan, N., Mathew, B. B. & Beeregowda, K. N. (2014). Toxicity, mechanism and health effects of some heavy metals. *Interdisciplinary Toxicology, 7(2),* 60-72.

Jorhem, L., Sundstrom, B., Astrand, C. & Haegglund, G. (1989). The levels of zinc, copper, manganese, selenium, chromium, nickel, cobalt, and aluminium in the meat, liver and kidney of Swedish pigs and cattle. *Zeitschrift für Lebensmittel Untersuchung und Forschung, 188(1),* 39-44.

Khalafalla, F. A., Ali, F. H., Schwagele, F. & Abd-El-Wahab, M. A. (2011). Heavy metal residues in beef carcasses in Beni-Suef abattoir, Egypt. *Veterinaria Italiana, 47(3),* 351-361.

Korenekova, B., Skalicka, M. & Nad, P. (2002). Concentration of some heavy metals in cattle reared in the vicinity of a metallurgic industry. *Veterinarski Arhiv*, *72(5),* 259-268.

Kottferova, J. & Korenekova, B. (1995). The effect of emissions on heavy metals concentrations in cattle from the area of an industrial plant in Slovakia. *Archives of Environmental Contamination and Toxicology, 29(3),* 400-405.

Kramer, H. L., Steiner, J. W. & Vallely, P. J. (1983). Trace element concentrations in the liver, kidney, and muscle of Queensland cattle. *Bulletin of Environmental Contamination and Toxicology*, *30(1),* 588-594.

Lopez-Alonso, M., Miranda, M., Castillo, C., Hernandez, J., Garcia-Vaquero, M. & Benedito, J. L. (2007). Toxic and essential metals in liver, kidney and muscle of pigs at slaughter in Galicia, north-west Spain. *Food Additives & Contaminants, 24(9),* 943-954.

Marreiro, D. D. N., Cruz, K. J. C., Morais, J. B. S., Beserra, J. B., Severo, J. S. & Oliveira, A. R. S. (2017). Zinc and oxidative stress: current mechanisms. *Antioxidants, 6(2),* 24.

Miranda, M., Benedito, J. L., Blanco-Penedo, I., Lopez-Lamas, C., Merino, A. & Lopez-Alonso, M. (2009). Metal accumulation in cattle raised in a serpentine-soil area: relationship between metal concentrations in soil, forage and animal tissues. *Journal of Trace Elements in Medicine and Biology*, *23(3),* 231-238.

National Research Council (2005). Mineral Tolerance of Animals, 2nd ed. National Academies Press, Washington (USA).

Niemi, A., Venalainen, E. R., Hirvi, T., Hirn, J. & Karppanen, E. (1991). The lead, cadmium and mercury concentrations in muscle, liver and kidney from Finnish pigs and cattle during 1987–1988. *Zeitschrift für Lebensmittel Untersuchung und Forschung*, *192(5),* 427-429.

Okareh, O. T. & Oladipo, T. A. (2015). Determination of heavy metals in selected tissues and organs of slaughtered cattle from Akinyele Central Abattoir, Ibadan, Nigeria. *Journal of Biology, Agriculture and Healthcare*, *5(11),* 124-129.

Orisakwe, O. E., Oladipo, O. O., Ajaezi, G. C. & Udowelle, N. A. (2017). Horizontal and vertical distribution of heavy metals in farm produce and livestock around lead-contaminated goldmine in Dareta and Abare, Zamfara State, Northern Nigeria. *International Journal of Environmental Research and Public Health*, 3506949.

Oymak, T., Ulusoy, H. I., Hastaoglu, E., Yilmaz, V. & Yildirim, V. S. (2017). Some heavy metal contents of various slaughtered cattle tissues in Sivas - Turkey. *Journal of the Turkish Chemical Society*, *4(3),* 721-728.

Pechova, A. & Pavlata, L. (2007). Chromium as an essential nutrient: a review. *Journal of Veterinary Medicine (Praha), 52(1),* 1.

Rasbech, N. O. (1969). A review of the causes of reproductive failure in swine. *British Veterinary Journal*, *125(12),* 599-616.

Sedki, A., Lekouch, N., Gamon, S. & Pineau, A. (2003). Toxic and essential trace metals in muscle, liver and kidney of bovines from a polluted area of Morocco. *Science of the Total Environment, 317(1),* 201-205.

Spivey Fox, M. R. (1987). Assessment of cadmium, lead and vanadium status of large animals as related to the human food chain. *Journal of Animal Science, 65(6),* 1744-1752.

Thevenod, F. (2010). Catch me if you can! Novel aspects of cadmium transport in mammalian cells. *BioMetals*, *23(5),* 857-875.

Tomović, V. M., Petrović, L. S., Tomović, M. S., Kevrešan, Ž. S. & Džinić, N. R. (2011). Determination of mineral contents of semimembranosus muscle and liver from pure and crossbred pigs in Vojvodina (northern Serbia). *Food Chemistry*, *124(1),* 342-348.

Welna, M., Szymczycha-Madeja, A. & Pohl, P. (2011). Quality of the trace element analysis: sample preparation steps. Avilable at: https://www.intechopen.com/books/wide-spectra-of-quality-control/quality-of-the-trace-element-analysis-sample-preparation-steps (accessed 15 May 2018).

Received: February, 11, 2020; *Accepted:* March, 2, 2020; *Published:* June, 2021