Pol. J. Environ. Stud. Vol. 21, No. 5 (2012), 1257-1260

Original Research

Lead Toxicity Evaluation in Rams Grazing on Pasture during Autumn and Winter: A Case Study

Zafar Iqbal Khan[1]*, Muhammad Ashraf[2,3], Kafeel Ahmad[1], Alireza Bayat[4], Muhammad Khalid Mukhtar[1], Syed Ali Hassan Naqvi[1], Rab Nawaz[1], Mian Jahan Zaib[1], Muneeba Shaheen[1]

[1]Department of Biological Sciences, University of Sargodha, Sargodha, Pakistan
[2]Department of Botany, University of Agriculture, Faisalabad, Pakistan
[3]Department of Botany and Microbiology, King Saud University, Riyadh, Saudi Arabia
[4]Animal Production Research, MTT Agrifood Research Finland, FI-31600, Jokioinen, Finland

Received: 10 May 2011
Accepted: 28 December 2011

Abstract

A study was conducted to appraise the levels of lead (Pb) in soils, pastures, and plasma of 20 rams raised in a farm alleged to have metal contamination. Samples of soil, pasture, and ram blood were taken four times with a one-month interval during autumn and winter, from October to January, and analyzed for Pb by atomic absorption spectroscopy. Considerable statistically significant differences were found in soil, pasture, and plasma Pb contents as a function of sampling time. The mean Pb values in soil ranged from 17.9 to 28.8 mg/kg, in forage from 9.92 to 16.4 mg/kg, and in plasma from 0.058 to 0.086 mg/l with a steady dwindle with sampling times. Forage Pb concentrations found in the present study were higher than the decisive Pb level for animals, but lower than the toxic limits for forage, signifying no anticipation of toxicity present in rams grazing therein. A significant positive correlation also was found between soil and forage and between forage and blood plasma Pb during this study. Overall, soil, forage, and rams contained tolerable amounts of Pb, so there is no risk of Pb toxicity presently to the rams being reared at the animal farm under study.

Keywords: soil, lead, pasture, blood plasma, ram, Pakistan

Introduction

It is now widely reported that ruminant metabolism undergoes considerable changes due to varying climatic conditions, topography, and types of diets fed to animals [1-3]. One of the fundamental issues in nutrition of animals is the balanced supply of all components, including trace elements. Thus, appraisal of the status of different trace elements present in animal diet is necessary to attain maximum production. However, the supply of most trace elements, if slightly exceeding the optimal level, causes considerable toxicity to animals. On the other hand, scarcity of these elements may cause metabolic disorders and can make unambiguous deficiency diseases [4-6]. Copper has a manifold function in animal metabolism, including regulation of the activities of a variety of enzymes and in the oxidation-reduction process [7]. For instance, the phagocytic ability of neutrophils was enhanced when copper was fed to copper-deficient goat kids [8]. Copper management can cause noteworthy increases in hemoglobin and serum copper levels.

*e-mail: zikhan11@gmail.com

Lead (Pb) is among the most common heavy metals that cause toxicity to animals and humans [9]. This is because the metal occurs commonly in most materials related to different types of industries as well as in air, water, plants, and animal feeds. Although Pb, like many other heavy metals, has no prominent role in the body, its presence in the body can ultimately lead to deficiencies of other trace metals [10, 11]. It has been reported that excess Pb may perturb the metabolism of purines and enhance oxidative stress in patients with pollenosisin, in addition to reducing the enzymatic activity in the body [12].

Many livestock farms wherein forage is grown are prone to frequent applications of a multitude of chemical compounds, e.g. insect repellents and fungicides that contain a variety of metals, including Pb. Therefore, such chemicals contaminate the surroundings as well as the animals being reared therein. Thus, the premier objective of this study was to investigate the levels of Pb in the plasma of rams, pastures, and soils in a livestock farm in the district of Sargodha, Punjab, Pakistan, where forages and tributary water used by ruminants are treated with fertilizers, pesticides, and sewage water.

Materials and Methods

Study Area

The study was conducted during October, November, and December 2008, and January 2009 at the Livestock Station, Khizerabad, Central Punjab, Sargodha (32º8'0"N 73º7'0"E). The description of this farm and comprehensive procedure has been given elsewhere [13]. Briefly, 2,000 sheep of the Kajli breed and nearly 1,400 cattle of the Sahiwal breed are being reared therein. The location, management, forage types, sampling procedure, sample preparation for analytical work, and statistical evaluations have been described in a previous publication [14].

Animals and Sample Collection

Twenty uncastrated male Kajli sheep, known as rams and representative of a group of 100, were used in this study. Samples were taken from mature rams older than two years old and weighing 45 to 50 kg. The rams were sampled simultaneously with soil and forage sampling during autumn and winter, from October to January. Blood samples (10 ml) were taken from the jugular vein using 20 ml heparinized vacutainer test tubes. The tubes containing blood were kept in an ice-packed cold box from the field and immediately transported to the laboratory of the Department of Biological Sciences, University of Sargodha. The samples were centrifuged at 3,000 rpm for 15 min. and then the plasma was harvested into clean test tubes. The tubes containing plasma were placed in a freezer at -20ºC until Pb analysis.

Soil and forage samples were collected during autumn and winter, from October to January, from three different sites within the farm which was frequently allowed to be grazed. At the chosen sites, forages were taken very carefully at 15 cm height from the ground to simulate the grazing behavior of rams. At the same sites within the pasture where forages were sampled, the soil was dug with auger to approximately 15 cm depth, and samples were taken. The soil samples were thoroughly mixed and placed in clean plastic bags, ground well with a pestle and mortar, and stored in tight polythene bags until Pb analysis.

Analytical Work

For the determination of Pb levels in soil, forage, and plasma samples, the aqua regia digestion was carried out in 250-ml glass beakers covered with watch glasses. A well-mixed sample (1.0 g) of ground soil or forage was digested in 15 ml of aqua regia on a hot plate for 2 h and 30 min at 120ºC. After evaporation, when the material was almost dry, the sample was diluted with 20 ml of 2% nitric acid (v/v with H_2O). After filtering through Whatman filter paper #42, the solute was diluted to 100 ml with distilled water and stored until analysis.

The Pb concentrations in soil and forage were determined using an atomic absorption spectrophotometer (Model #AA-6300, Shimadzu, Japan). For the analysis of Pb in plasma samples, an atomic absorption spectrophotometer coupled with a graphite furnace was used (AA-6300 & GFAEXi7i, Shimadzu, Japan).

Statistical Analysis

The data of Pb concentrations in soil, forage, and blood plasma were subjected to analysis of variance using SPSS computer software for testing the significance at 0.05, 0.01, and 0.001. Correlations among soil, forage and plasma Pb concentrations were also drawn using the same software.

Results

Soil Pb concentration decreased ($P<0.05$) gradually from October to January (28.8 to 17.9 mg/kg, respectively; Fig. 1). The highest forage Pb concentration was observed at the first sampling and the lowest at the final sampling

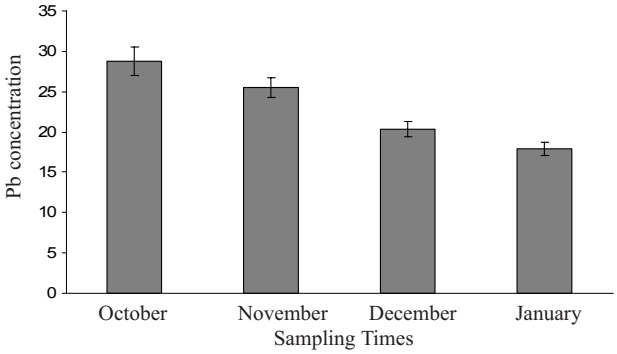

Fig. 1. Soil Pb concentrations (mg/kg dry weight; Bars represent means with standard errors) during autumn and winter, from October to January.

(P<0.05) with a gradual decrease in sampling time from October to January (16.4 to 9.9 mg/kg; Fig. 2). Mean blood plasma Pb concentrations ranged from 0.058 to 0.086 mg/l. The plasma Pb concentration decreased consistently with time progress from October to January (P<0.001, Fig. 3).

A significant and positive correlation (0.5550) was found between soil and forage Pb concentrations, but a low and negative correlation (-0.0060) was found between soil and blood plasma Pb concentrations. Correlation between forage and blood Pb concentrations was also positive and significant (0.5745).

Discussion

Decreasing Pb concentration in soil from October to January is probably due to either leaching into depth by irrigation or exhaustion of lead in soil by absorption by forage. The values of soil Pb concentration found in this study were far lower than the established level of 50 mg Pb/kg soil, a level reported to be suitable for forage cultivation [15]. Bowen [16] reported that in India, the soil Pb concentrations varied from 2-200 mg/kg, particularly in those areas which are of an uncontaminated nature. Generally, the Pb concentrations in soil range from 5 to 25 mg/kg and are optimum for plant growth [17]. Thus, the concentrations observed in the present study are within this range.

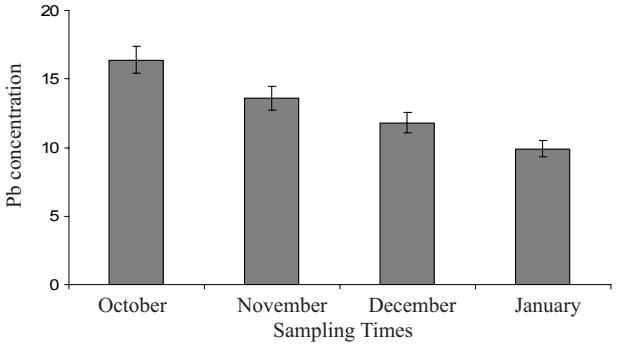

Fig. 2. Forage Pb concentrations (mg/kg dry weight; Bars represent means with standard errors) during autumn and winter, from October to January.

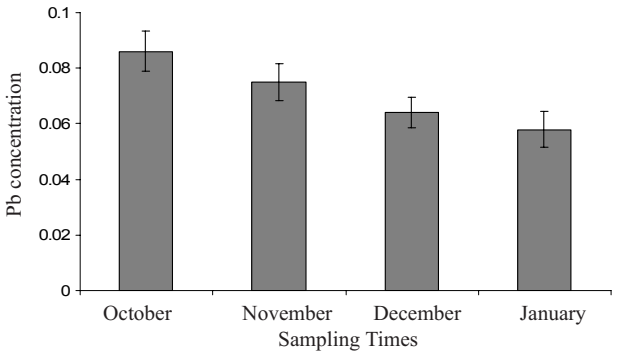

Fig. 3. Concentrations of Pb in blood plasma of rams (mg/l; Bars represent means with standard errors) during autumn and winter, from October to January.

Furthermore, the soil Pb concentrations recorded in the present investigation are consistent with those reported earlier by Mlay and Mgumia [18] in Turkey, which ranged from 10.8 to 30.5 mg/kg and considered optimum for forage crop requirement by the same authors. According to the criteria suggested by Ross [19], the concentration of Pb in soil from 100-400 mg/kg is considered toxic, and soil Pb concentrations in our investigation did not exceed this upper limit and had been found far below the toxic limit for plants. Based on these findings, there is no need for soil amendment with Pb-containing fertilizers at this animal ranch.

Mean forage Pb concentration in this study was higher than the critical value of 0.05-3 mg/kg proposed by Tokalioglu et al. [20]. Higher values of forage Pb were also reported earlier by Li et al. [21]. High levels of forage Pb in the present study can be ascribed to the soil Pb being of reasonable amount. Maximum tolerable levels of Pb by animals is 30 mg/kg, and Pb poisoning is more frequently observed in farm animals [22]. In our investigation, forage allowance of Pb is less than tolerable levels and thus presently there are no threats of Pb toxicity to the rams being reared at this farm.

Plasma Pb values in the present study were found in the range of reference level of 0.01-0.20 mg/l as recommended by Puls [23], indicating no toxicity danger for ruminants. The plasma Pb levels in the rams were reasonably higher compared to those (0.01 mg/l) reported by Lopez et al. [24] in animals in Galicia, Spain, indicating that the rams were prone to high amounts of Pb being taken efficiently from the soil and forages. The Pb levels in blood plasma of rams were in conformity with those reported earlier by Yazar et al. [25] in blood plasma of Angora goats in Turkey ranging from 0.043-0.094 mg/l. The possibility for the rams to obtain extra Pb through ingestion of feed, soil, and water cannot be ruled out due to the fact that this metal tends to bio-accumulate in the body [26]. Pb may enter the body not only through absorption in the gastrointestinal tract, but also through the respiratory tract and skin. The majority of absorbed Pb can be accumulated in the skeleton and it appears to be relatively immobile, and absorbed Pb is stored in the liver and kidneys [27]. The levels of Pb in tissues and fluids have been shown to increase with increasing exposure to Pb [28]. The low levels of plasma Pb in this investigation may have been due to its deposition in various organs, although its concentration in the forage consumed by rams were higher. The threats of fertilizers and organic manures as sources of Pb that can affect animals and humans have been reported in other parts of the world [29, 30].

Conclusions

Based on the soil, forage, and plasma analyses for Pb, it can be concluded that the levels of this metal in the blood plasma of rams and soil were lower than the reference values, while forage contained higher amounts of Pb. However, the Pb concentrations are still below the toxic levels for animals at the animal farm. Thus, presently there

is no threat of toxicity to the ruminants. However, there is a need to continuously monitor the Pb status in animals, soils, and forages in order to avoid toxicity to animals, as this metal has mostly toxic effects rather than being essential for ruminant growth.

References

1. TRIPATHI M. K., MONDAL D., KARIM S. A. Growth, haematology, blood constituents and immunological status of lambs fed graded levels of animal feed grade damaged wheat as substitute of maize. J. Anim. Physiol. Anim. Nutri. **92**, 75, **2008**.
2. FRELICH J., PECHAROVA E., KLIMES F., SLACHTA M., HAKROVA P., ZDRAZIL M. Landscape management by means of cattle pasturage in the submountain area of the Czech republik. Ecology, **25**, S115, **2006**.
3. JELINEK P., GAJDUŠEK S., ILLEK J. Relationship between selected indicators of milk and blood in sheep. Small Rumin. Res. **20**, 53, **1996**.
4. MASSANYI P., NAD P., TOMAN R., KOVACIK J. Concentrations of cadmium, lead, nickel, copper and zinc in various muscles of sheep. Die Bodenkultur **52**, 255, **2001**.
5. RAJCAKOVA L., GALLO M., MLYNAR R. Accumulation of copper, zinc, lead and cadmium in grass with high imission load and its effect on the production of grass mass. Slovak. J. Anim. Sci. **36**, 221, **2003**.
6. CERMAK B., LAD F., KLIMES F., JILEK R., KOBES M. Dynamic of nutrients quality characteristic of pasture in different altitude in South Bohemian region.Slovak J. Anim. Sci. **39**, 99, **2006**.
7. SKRIVANOVA V., SKRIVAN M., TUMOVA E., SEVCIKOVA S. Influence of dietary vitamin E and copper on fatty acid profile and cholesterol content of raw and cooked broiler meat. Czech J. Anim. Sci. **49**, 71, **2004**.
8. SOLAIMAN S. G., CRAIG JR T. J., REDDY G., SHOEMAKER C. E. Effect of high levels of Cu supplement on growth performance, rumen fermentation, and immune responses in goat kids. Small Rumin. Res. **69**, 115, **2007**.
9. ROBERTS J. R. Metal toxicity in children. In: Training manual on pediatric environmental health; Putting it into practice. Emeryville, CA: Children's Environmental Health Network http://www.cehn.org/cehn/trainingmanual/pdf/manual-full.pdf [Cited on 7th May 2011] **1999**.
10. FARR G. The Hair Tissue Mineral Analysis/The trace metals http://www.becomehealthynow.com/article/hairchiro/123 [Cited on 7th May 2011] **2001**.
11. FARR G. Why heavy metals are a hazard to your health. http://www.becomehealthynow.com/ebookprint.php?id=122 [Cited on 7th May 2011] **2004**.
12. SZYCZEWSKI P., SIEPAK J., NIEDZIELSKI P., SOBCZYŃSKI T. Research on heavy metals in Poland. Pol. J. Environ. Stud. **18**, 775, **2009**.
13. KHAN Z. I., AHMAD K., ASHRAF M., NAQVI S. A. H., KASHAF S., EJAZ A., VALEEM E. E. Evaluation of potassium status of rams: The use of pasture and blood plasma as indicators under semi-arid environmental conditions in Punjab, Pakistan. Pak. J. Bot. **43**, 3933, **2010**.
14. KHAN Z. I., ASHRAF M., AL-QURAINY F., AHMAD K., GONDAL S., FARDOUS A. Studies on the transfer of copper from soil to pastures at different sampling periods: A case study of a semiarid region (Sargodha) in Pakistan. Biol. Trace Elem. Res. **2010**.
15. KABATA-PENDIAS A., PENDIAS H. Trace Elements in Soils and Plants. 3rd ed.; CRC Press Inc.: Boca Raton, USA, **2000**.
16. BOWEN H.J.M. Trace Elements in Biochemistry. 1st ed.; Academic Press: New York, USA, **1996**.
17. FRANK C.L.U. Basic toxicology. Fundamentals, target organs, and risk assessment. 2nd ed.; Hemisphere Publishing Corporation: NY, pp. 361, **1991**.
18. MLAY P.S., MGUMIA Y.O. Levels of lead and copper in plasma of dairy cows, pastures, soil and water from selected areas of Morogoro suburbs, Tanzania. Livestock Research for Rural Development. Volume 20, Article #60. Retrieved November 26, 2010, from http://www.lrrd.org/lrrd20/4/mlay20060.htm. **2008**.
19. ROSS M.S. Source and form of potentially toxic metals in soil plant system. In: Toxic metals in soil-plant system (ed Ross MS). John Wiley: Chichester. pp. 3-25, **1994**.
20. TOKALIOGLU S., KARTAL S., GUNIS A. A. Determination of heavy metals in soil extracts and plant tissues at around of zinc smelter. Int. J. Environ. Analyt. Chem. **80**, 210, **2000**.
21. LI Y., MCCRORY D. F., POWELL J. M., SAAM H., JACKSON-SMITH D. A survey of selected heavy metal concentration in Wisconsin dairy feeds. J. Dairy Sci. **88**, 2911, **2005**.
22. ANNONYMOUS Mineral tolerance of domestic animals. National Academy of Sciences, National Research Council, Washington, DC. **1980**.
23. PULS R. Minerals levels in animal health; diagnostic data. 2nd ed.; Sherpa International: Clear Brook, BC, Canada. **1994**.
24. LOPEZ A.M., BENEDICTO J. L., MIRANDA M., CASTILLO C., HERNANDEZ J., SHORE R. F. Arsenic, cadmium, lead, copper and zinc in cattle from Galicia, Spain. The Sci. Total Environ. **246**, 237, **2000**.
25. YAZAR E., ALTUNOK V., EROGLU T. Concentrations of some elements in blood serum of Angora goats. Medycyna Wet. **62**, 1249, **2006**.
26. MULCHI C. L., ADAMU C. A., BELL P. F., CHANEY R. L. Residual heavy metal concentrations in sludge amended coastal plain soils. I. Comparison of extractants. Commun. Soil Sci. Plant Anal. **22**, 919, **1991**.
27. REICHLMAYR-LAIS A.M., KIRCHGESSNER M. Handbook of nutritionally essential mineral elements (O'Dell B.L., Sunde R.A., Eds.), Marcel Dekker: New York, pp. 479. **1997**.
28. MCDOWELL L.R. Minerals in animal and human nutrition. 2nd ed.; Amsterdam: Univ. Florida, **2003**.
29. RADOSTITS O.M., GAY C.C., BLOOD D.C., HINCHLIFF K.W. Veterinary medicine; a textbook of the disease of cattle, sheep, pigs, goats and horses. 9th ed.; WB Saunders: London pp. 1599-1603, **2000**.
30. MOTTO H. L., DAINES D. H., CHILKO D. M., MOTTO C. K. Lead in soils and plants: Its relationship to traffic volume and proximity to highways. Environ. Sci. Technol. **4**, 232, **1970**.