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Heavy Metals Concentration in Soil-Plant-Animal Continuum under Semi-Arid Conditions of Punjab, Pakistan

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Abstract.- To determine the lead and cadmium flow in soil-plant-animal continuum under semi-arid conditions, the present study was carried out at a Livestock Experimental Station. The experimental work was performed on soil, forage plants and animal blood plasma samples were taken at four different time periods with one month interval. These samples were analyzed for Pb and Cd by atomic absorption spectrophotometer. Higher concentration of Pb was found in soil and forage than the blood plasma. On the other hand, cadmium in soil, forage and blood plasma was found as non-significant due to its accumulation in low concentration. It also indicated lower concentrations of Pb and Cd in blood plasma which were at marginal deficient levels. Based on this study, it can be suggested that the value of Pb in soil was greater than the critical levels, while Cd concentration in soil, forage or blood plasma and Pb concentration in forage and blood plasma were within the non-toxic range. So no need of supplementation of livestock with a mixture of these elements in this (semi-arid) climatic region, however, toxic level of Pb in soil is of major concern.

Key words: Lead, cadmium, forage, blood plasma, metal status.

INTRODUCTION

Minerals are associated with every life function through the production of hormone, enzymes and regulate metabolism by their actions as coworker. The complexities related to mineral nutrition and metabolism may associate with the treatment of a mineral related disease (Khan *et al*., 2006, 2007). Lead (Pb) has direct role in metabolic pathways; however, it causes toxicity when exceeded from its critical levels as well as due to the deficiencies of other trace elements (Farmer and Farmer, 2000; Bibi *et al*., 2014). The soils which have low level of lead than the toxic concentration are safe for animals and plants (Ross, 1994). Low concentration of Pb in soil may possibly be due to the use of canal water for irrigation instead of using sewage or industrial wastewater, therefore, forage plant may grow on such type of soils accumulates less concentration of Pb and thus posing no threats for grazing livestock.

The Pb level in pasture-soil depends on intrinsic nature of soil and extrinsic factors through anthropogenic activities leading to an increase in the

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level of Pb than the critical level (Osweiler, 1996; Shad *et al.,* 2014). The pastures, which are near the industrial area or close to major road ways, become a potential source of heavy metal toxicity. Consequently, high accumulation of these metals induces adverse effects on plant growth which may cause problem of toxicities in animals depending on these plants/forages. High concentration of Pb in soil may also be caused by air pollution, contamination from vehicles and gasoline and excessive use of industrial wastewater. Resultantly, higher level of Pb can affect all parts of the body of animals in all over the world (Radostits *et al.*, 1994; Shad *et al.*, 2013).

During recent time periods, the transfer of trace and heavy elements in the soil-crop and ultimately in animal system has attracted much more attention (Zhang *et al*., 2011; Ahmad *et al*., 2011). The reason, thereby, is the usage of crop plants grown under heavy metals contaminated soils by human and animals (Adamsa *et al*., 2004; Shad *et al.,* 2013). Therefore, mineral supplementations is required in nutrients deficient soils for optimal plant growth and production, better immunity against diseases and improve the lactation and growth of grazing animals including cows (Corah and Ives, 1991; Greene, 1997).

Kottferová and Koréneková (1998) described free-living animals as important indicators of the environmental pollution with heavy metals. Ahmad *et al*. (2010) reported sheep and cattle as indicators of the environmental pollution because they rear freely on pasture. The increase of heavy metal concentration in domestic animals may result in low reproduction and fitness problems along with cancerous and/or teratogenic diseases in animals (Bires *et al*., 1995). Oskarsson *et al*. (1995) reported low excretion of metals such as Cd, Pb and Hg in animals, especially in the offspring.

Keeping in view the importance of minerals for the grazing livestock, the present investigation was carried out to observe the critical values of minerals whose high or low concentration can cause toxicity or deficiency. The main objective of the present study was to provide the balanced mineral nutrition to the grazing livestock for the sake of high yield and productivity of meat and milk by means of supplementation and fertilizers.

MATERIALS AND METHODS

Investigated site

The present research was conducted at the Livestock Experiment Station Khizerabad, Sargodha, Punjab, Pakistan with average temperature, 7°C to 25°C. The average rainfall in this district is 180 to 200 millimeter during the months of July and August. Blood of cows, forage and soil samples were collected from the experimental station four times with one month interval. The study was made to check the transfer of minerals from soil to forage, and forage to blood plasma of 40 different Sahiwal cows.

Soil sampling

Soil samples were collected from the fields where forages were grown, on which the targeted cattle graze. Five places were selected randomly and soil was collected at the depth of 12-15 cm with the help of stainless steel auger following Sanchez (1976). Thus five samples, one from each site, were collected in such a way that all the soil layers were present in equal amounts. The sampling was done four times with the one month interval. All the air dried samples were placed in an oven at 70^{o}C for complete removal of moisture.

Forage sampling

Sahiwal cows were managed in the traditional semi-nomadic system and cows selected those were grazing on the natural growing pasture whole the year. The nature of these soils is predominantly loamy to clay. The cows graze on forage plants were mostly cultivated species. Forage samples were collected randomly from five different places, from which it was frequently given to these cows, using sterilized apparatus. The forage samples (four times at the interval of one month) were collected from the same place where soil samples were collected and on which the Sahiwal cows were grazing. Five samples were collected for each time of interval. The forage species included barseem (*Trifolium alexandrium*), sarson (*Brassica compestres*), oat (*Avana sativa*) and some other minor species.

Blood sampling

Blood samples were collected from the

jugular veins of non pregnant healthy cows belong to Sahiwal breed with the help of sterilized needle. The sample was collected in the test tubes and heparinized immediately. Blood samples were collected four times per one month interval. Five samples were collected each time of sampling interval. After blood collection, plasma was separated by centrifugation at 3000 rpm for 15-30 minutes following Koh and Babidges (1986). The obtained serum was put in small labeled vials and placed at -20°C in freezer.

Wet digestion and mineral analysis

All the samples, soil, forage plants and blood plasma were digested with sulphuric acid (H_2SO_4) and hydrogen peroxide H_2O_2 in 1:2 ratios. After cooling, the digested samples were diluted with double distilled water to final volume of 50 mL. Concentration of the metals, Pb and Cd in soil forage plants, and blood plasma samples were analyzed using atomic absorption spectrophotometer as described by Anonymous (1980).

Statistical analysis

The obtained data were statistically analyzed following Steel and Torrie (1980). The statistical significance was tested at 0.05, 0.01 and 0.001 levels of probability using the software SPSS statistical program.

RESULTS AND DISCUSSION

Lead in soil sample

Analysis of variance for the concentration of lead in soil sample varied significantly ($p < 0.001$) with respect to sampling periods (Table I). It was observed from the data that the concentration of Pb in soil was higher at first sampling interval, but it suddenly drops at second sampling period. A slight increase was observed at third sampling interval and it again decreases at fourth sampling time period. Inconsistent trends of depression and elevation was observed during this study in soil sample as shown in Figure 1a. The mean Pb concentration in soil sample ranged from 59.9 to 135.7 mg kg^{-1}, showing higher level at first sampling interval and lower at second one. The Pb concentration in soil was observed greater than the toxic level. Similarly, the concentration of Pb was found higher than 5-25 mg kg^{-1} as reported by Hayashi *et al.* (1985). Furthermore, it was studied that the concentrations of Pb in soil was found higher than those reported by Oluokun *et al*. (2007) in Nigeria.

Lead in forage sample

Analyses of variance of data showed significant ($p<0.01$) effect of time intervals on the lead concentration in forage samples (Table I). The data showed that highest level of concentration of Pb in forage was found at fourth sampling period and lowest at second period (Fig. 1b). The forage level increase and decrease at different time intervals. The mean of concentration of lead in forage samples ranged between 36.85 and 60.21 mg kg^{-1}. In the present investigation lead concentration in forage is less than the toxic level (30 mg kg^{-1}) as proposed by Ross in 1994 and Oluokun *et al.* (2007). There is no threat to livestock feed on these forages because Pb concentration is below the toxic level. So use of forages grown in less contaminated

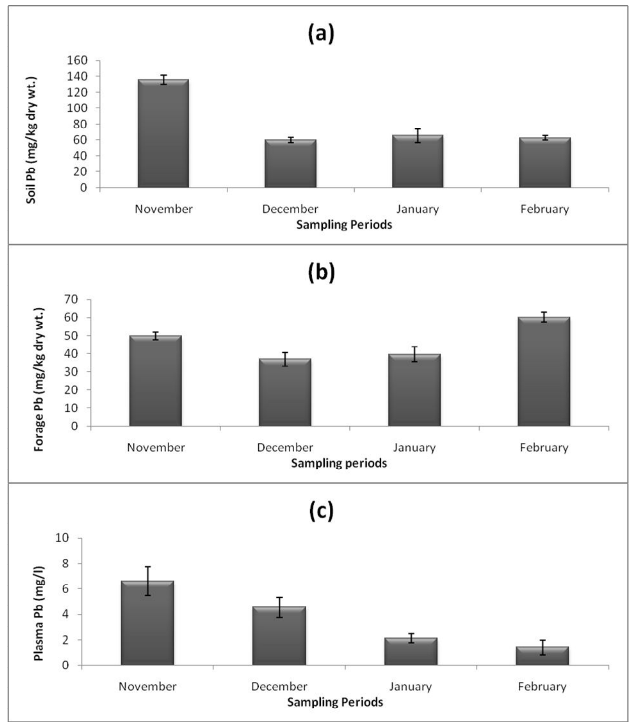

Fig. 1. Fluctuations in soil (a), forage (b) and plasma (c) concentrations of Pb at different sampling periods

Table I.- Analysis of variance for Pb and Cd concentrations in soil, forage, and blood plasma at different sampling intervals.

SOV	df	Mean Squares					
		Soil		Forage		Blood plasma	
		Pb	Cd	Pb	Cd	Pb	Cd
Sampling period	3	6685.079***	1.597*	565.969***	0.032ns	28.318**	0.000ns
Error	16	153.859	0.389	52.196	0.076	2.973	0.001

SOV, source of variation; df, degree of freedom; ***significant at 0.001 levels; **significant at 0.01 levels; * significant at 0.05 levels; ns, non-significant.

areas has been recommended to avoid high accumulation of toxic metals in the body of animals feed on these forages (Ahmad *et al*., 2010). However, in the present study the concentration of lead in forage was lower than the toxic level, so non-toxic for livestock grazing in such areas.

Lead in plasma sample

Analysis of variance of data indicated least significant effect ($p<0.05$) on all sampling intervals of lead in plasma (Table I). Present investigation showed highest level of Pb in plasma of dry Sahiwal breed at first sampling period and lowest at fourth period which showed inconsistence pattern of Pb level from first to last sampling periods (Fig. 1c). Its concentration was ranged between 1.39 and 6.6 mg L^{-1}. The concentration of lead in plasma is at lower level than the toxic level. In present investigation, concentration of Pb was higher as compared to those reported by López-Alonso *et al*. (2000). There is no threat to livestock feed on the forages grown under these soils having Pb concentration below than the toxic level (Ahmad *et al*., 2008).

Cadmium in soil sample

Analysis of variance for soil Cd concentration depicts that its concentration varied significantly ($p < 0.001$) at different sampling periods (Table I). It was observed from the data that the level of Cd in soil increased from first to second sampling interval and then a gradual decrease was observed up to fourth period. The mean concentration of Cd in soil varied from 0.62 to 1.89 mg kg^{-1} dry weights. The higher level was observed at second sampling period and lower level was found at fourth period (Fig. 2a). The values observed in present investigation are less than the critical value *i.e.*, 3.0 mg kg^{-1} observed by Kloke (1980). The literature showed that the safe limit of Cd in soil was up to 3.0 mg kg^{-1}. However, if the concentration of Cd exceed from the critical value then it become toxic as observed by Ross (1994).

Cadmium in forage sample

The concentration of Cd in forage showed non-significant effect on sampling intervals ($p>0.05$) as indicated by ANOVA (Table I). The highest value of cadmium was found in second sampling period and lowest in third period (Fig. 2b). It ranged between 0.3 and 0.5 mg kg^{-1} with consistent pattern of increase and decrease among different sampling intervals. Cd concentration in present investigation was below the toxic level as suggested by Aksoy *et al*. (1999) in Turkey. The adequate level of Cd in plants has been suggested to be around 3 mg kg^{-1} as reported by Cicek and Koparal (2004).

Cadmium in plasma sample

Analysis of variance evident that there was a non-significant effect of different sampling periods on the concentration of Cd in plasma (Table I). Concentration of Cd in plasma of dry Sahiwal breed increased and decreased gradually. The highest level was observed in third sampling period while lowest was found in second period. Its concentration was ranged from 0.05 to 0.06 mg L^{-1} (Fig. 2c). Cd and Pb are non-essential elements that are toxic to plants and animals. It is known that only a small amount of the total Pb in soils may be taken up by plants, and the translocation of Pb from roots to tops is greatly limited. The amount of Pb and Cd taken up by plants depends on the total amounts in the soil and their availability (Gowda *et al*., 2003).

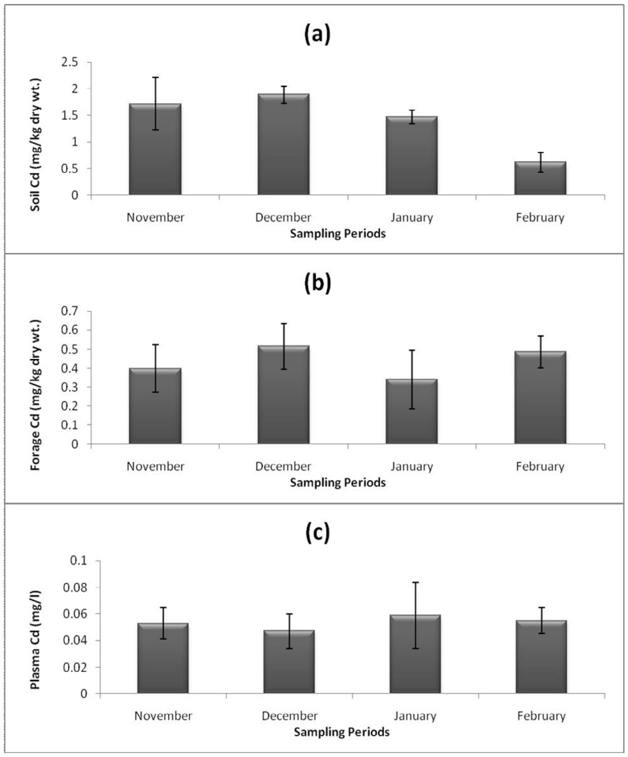

Fig. 2. Fluctuation in soil (a), forage (b) and plasma (c) concentrations of Cd at different sampling periods.

The values of Cd in plasma observed in present investigation were almost similar to those (0.06 mg L^{-1}) reported by Gowda *et al*. (2003). Plasma Cd concentration observed at different sampling intervals was found to be inconsistent as reported by Smith and Loneragon (1997). They suggested that Cd concentration up to 5 mg kg^{-1} in diet induces adverse effects on plasma. Lui (2003), in China, has described the concurrent poisoning level of lead and Cd in animals. Blood Cd concentrations were at or near the detection limit for their assay of 0.005 ppm, equivalent to 5 µg kg^{-1}. Research conducted in an industrialized area of North West Spain, Galicia reported blood concentrations of Cd in six to ten month old calves and cows (Lopez Alonso *et al.*, 2000). The mean blood Cd concentration was 0.37 and 0.45 µg L^{-1} in calves and cows, respectively. Toxic levels of Cd cause various diseases like hemorrhagic and abortion with dystrophy. Present investigation suggested that the areas under observations contained low concentration of Cd so there is rare chance of toxicity due to cadmium.

CONCLUSIONS

As for the soil, Pb concentrations were significantly higher than the Pb in plasma. However no-significant differences of cadmium concentration were recorded for forage and blood plasma. Both of the metals, Pb and Cd were indicated at non-toxic levels, so, cows under investigation are out of danger. But the information for a particular area or ecosystem is necessary to continue for future which could be useful for farmers utilizing these areas.

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