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Risk Assessment of Chromium, Lead and Cadmium in Feed, Water and Shed Feather of Some Captive Birds at Lahore Zoo, Pakistan

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Abstract

Concentration of heavy metals lead (Pb), cadmium (Cd) and chromium (Cr) was determined in the feathers of eight birds, Emu *(Dromaius novaehollandiae)*, Ring Necked (R.N) pheasant *(Phasianus colchicus)*, Green Pheasant *(Phasianus versicolor)*, White Peafowl *(Pavo muticus)*, Pied Peafowl *(Pavo cristatus)*, Green Java Peafowl *(Pavo muticus)*, Black Shoulder Peafowl *(Pavo cristatus)* and Blue Peafowl *(Pavo cristatus)* from Lahore Zoo, Pakistan. For this purpose, water, feed and shed feathers of birds were collected from zoo. The metal accumulation pattern in studied feathers of birds was in the order: Pb>Cr>Cd. Difference in R.N pheasant was observed with an accumulation trend of Pb>Cd>Cr. Highest concentration of Cr (Mean ± S.D. 0.22 ± 0.022µg/g), Cd (0.155 ± 0.01µg/g) and Pb (0.543 ± 0.572µg/g) was detected in Emu, Black shoulder peafowl and white peafowl respectively. Lowest concentration of Pb, Cd and Cr was recorded in Ring Necked pheasant. Significant variation in metal accumulation trend was observed between water and feed of birds ($p<0.05$). Highest was in the feed compared to water with an accumulation trend of Cr>Pb>Cd. A strong positive correlation (0.70), between the accumulation of heavy metals in the feathers of birds with their feed, was observed. Concentrations of Pb, Cd, and Cr in the feathers of all the selected birds were below permissible limits of heavy metals in the feathers of birds. The

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present study concluded feathers of the bird could be used non-destructive indicators of heavy metal contamination.

Keywords: Captive birds; Feed; Heavy metals; Zoo

1. Introduction

Toxic pollutants such as heavy metals, released from agricultural, industrial and mining activities have severe impact on the ecosystem including the living organisms which are sustaining on it. Birds are more vulnerable to pollutants than mammals and are thus being used as bio-indicators for estimating heavy metal pollution of a concerned area since the 1960's (Denneman & Douben, 1993). Heavy metals are considered as one of the most dangerous group of pollutants in the environment which are a serious threat to all living organisms and among them lead is considered to be highly toxic (Roux & Marra, 2007). Heavy metals like zinc, iron, copper, manganese, have an essential role in the body of living organisms but when their concentration in the body becomes higher than the required amount, they can pose harm to the living organisms while others are toxic even when their exposure takes place in low concentration. The toxic effects of heavy metal uptake in birds include weakening of immune system, decreased body weight, reproductive dysfunction, thinning of eggshell, low growth and reproduction rates ultimately leading to the decline of population in jeopardy (Spahn & Sherry,1999; Dauwe *et al*., 2006). Lead poisoning is the most common form of poisoning in birds and can happen either acutely or chronically depending upon the properties of metal and the ability of the exposed individual to take up that toxin. Its clinical symptoms are head tilt, blindness, unable to stand, fly or walk, weight loss, dehydration, blood in droppings, weakness, depression, damage to central nervous system and death (Scheuhammer, 1987; Burger, 1995; Gochfeld, 2000).

Birds are subjected to environmental pollution studies as they are easy to monitor, study, occupy a higher trophic level being more vulnerable to bio-accumulative effects and gives information about the quality of the surroundings where the proposed study undergoes. More promising and accurate results are obtained by using living organisms as indicators of environmental pollution rather than using other physical or chemical means of analyses that can be either too costly or time consuming or a combination of both. Bird feathers are selected as indicator for monitoring heavy metal pollution as they can be sampled again and again, if required, without affecting the health of the selected individuals of birds and are useful in long period of study.

Birds are exposed to metal pollution either externally by direct physical contact or internally by consuming the heavy metal contaminated water and food (Roux & Marra, 2007). The extent to which metals are taken up by the bird depends upon the physiology of the individual (age, health status, and body size) (Grue *et al*., 1986), properties of the metals including their quantity and duration of exposure and their bio-availability (Burger, 1995). Once the metal enters the body of individual, it circulates in the whole body, gets accumulated in different body tissues or is either tucked away in the feathers of the birds (Furness *et al*., 1986).

Heavy metal concentrations in the feathers of birds have been studied worldwide. Frantz *et*

al. (2012) reported the presence of Cd, Cu, Pb and Zn in the feathers of pigeons from an urban region of Paris. Similar study reported an alarming
concentrations of Pd, Cd and Cr in the feathers of cattle egret chick from two districts, Jhang and Vehari of Punjab province in Pakistan (Ullah *et al*., 2014). Accumulation of Cr, Pb and Cd in avian feathers from Lahore and Sialkot which are the two major industrial cities of Punjab province was studied by Abdullah *et al.* (2015). Most of the work has been done on wild birds using their various body parts such as eggs, liver, kidney, blood, feathers etc. in order to estimate the level of toxic metals and the pollution which they are contributing to the environment. Recently much work has been done on birds in order to estimate the amount of organic pollutants and heavy metals by using non-destructive sampling techniques for feathers (Pilastro *et al*., 1993; Naam *et al*., 2004; Dauwe *et al*., 2006; Jaspers *et al*., 2007; Van den Steen *et al*., 2007) and faeces (Dauwe *et al*., 2000). Although various authors have used birds in their studies as bio- indicators of metal pollution, but most have focused on adult birds and birds of prey (Denneman & Douben, 1993; Esselink *et al*., 1995; Jager *et al*., 1996).

However, no study has been conducted to investigate heavy metal pollution using the feathers of captive birds in Pakistan. Due to scanty information about the accumulation of heavy metals in bird's feathers that are in imprisonment, there is a dire need to investigate their concentration within the captive birds. This shows a gap lapse which is needed to be filled. Therefore, the focus of the current study is to provide information about heavy metal concentration (Pb, Cd and Cr) in feathers of eight selected bird species from Lahore Zoo, situated near Mall road which is one of the most occupied and busiest roads of Lahore. Also, heavy metal concentrations in the feathers of Lahore Zoo birds were compared with the threshold limit of Cr, Cd and Pb as suggested by Burger & Gochfeld (2001) and Gochfeld (2000).

2. Methodology

2.1 Selection of sampling site

The feathers (shed), water and food of five peafowl species, two pheasant species and emu species were sampled in Lahore Zoological Gardens, Mall Road, Lahore (i.e. within captivity). Land birds (both with and without flight) were chosen in order to establish a link between the environmental factors responsible for the existence of selected heavy metals in the birds. Feathers from different body parts can harbor different metal concentrations. To standardize the method, shed body feathers of all the selected birds and their species were sampled. This avoided any chance of biasness in sampling, thus making the sample more precise. Heavy metals like Pb, Cr and Cd were determined using atomic absorption spectrometer. The study quantified the metals and determined the path through which metals entered the body of the birds and manifested in their feathers. The feed and water samples of these species were taken to determine the culprit responsible for the metals concentrations found in the feathers of the selected birds' species. Feathers were not plucked from the body of birds keeping in view their safety. Feathers, feed and water were sampled twice, after an interval of 15 days, and the samples were the residues for feed and water. The collected samples of feed and feathers of the birds were stored in plastic zip lock bags until chemical analysis while the water samples were taken in brown plastic bottles as brown bottles afford some protection from light. Samples were stored in refrigerator maintained at the temperature 4^{o}C after each

phase of analysis. After getting the samples the feathers were weighed and dried in an oven at 60^{o}C for 30 min to dry mass, cut into small pieces and weighed again to 100 mg and then stored in refrigerator. The feed samples were weighed, dried in oven at 70^{o}C for 30 min to dry mass, homogenized using grinder, weighed to approximately 1.5 g and then stored in refrigerator (Font *et al*., 2007) and the water samples were refrigerated.

2.2 Digestion of Samples

The Pyrex glass beakers of 100 ml were washed with diluted nitric acid and distilled water before use. Feathers were digested in 8 ml mixture of Aqua Regia (HCl: HNO_3) at 110^{o}C for 5-10 minutes on a hot plate (Font *et al*., 2007) inside a fume cabinet until brown fumes changed to white fumes of HCl and the samples became transparent. Any possible particulates were digested in the water samples by taking 100 ml of the water and adding 1ml of HCl in it. The water samples were then heated for 30 minutes on a hot plate in the fume cabinet. Feed was digested by using mixture of HNO_3-$HClO_4$; HNO_3 is taken as 10 ml and $HClO_4$ as 3 ml (Cang, 2004; Abdolgader *et al*., 2013). Samples were heated on hot plate in fume cabinet for 15-30 minutes, until white fumes of $HClO_4$ were observed and the samples became transparent. After digestion all the samples were filtered using Whatman filter paper No 42, diluted to 100 ml and stored in plastic bottles. The bottles were then placed in refrigerator at 4^{o}C until analysis for the metal concentrations. Nitric Acid (HNO_3) and Hydrochloric Acid (HCl) used in the digestion were of ultra-pure quality. Perchloric Acid ($HClO_4$) was also used. Distilled water was used for dilution and for washing of all glassware. Sodium Hydroxide (NaOH) and Acetic Acid (CH_3COOH) were used for neutralization before analysis of heavy metal concentrations and Stock Solutions of 1000 mg/L were prepared to make standard solutions for each metal analysis.

2.3 Analysis of Metal Concentrations

The samples were neutralized using NaOH and CH_3COOH with a calibrated pH meter (HI98107). The three heavy metals: lead, chromium and cadmium were analyzed by Perkin Elmer A-Analyst 800 Atomic Absorption Spectrometer, with 32 Win lab software.

2.4 Statistical Analysis

The statistics applied included calculation by basic statistics such as mean standard deviation (SD) using Microsoft Excel whereas graphs were also prepared using the same software. The map was generated using Google Earth and ARCGIS software.

3. Results

The three heavy metals selected (Chromium, Lead and Copper) were analyzed to assess the concentration level in the eight samples each of feathers, feed and water of the selected birds from the defined sampling site. These concentrations are given in Table 1. The general pattern of heavy metals observed in Emu was *Pb* > *Cr* > *Cd* in the feathers of birds. The pattern observed in R.N. Pheasant was *Pb* > *Cd* > *Cr* in the feathers of birds. It was *Pb* > *Cr* > *Cd* in Green Pheasant, White Peafowl, Pied Peafowl, Green Java Peafowl, Black Shoulder Peafowl and Blue Peafowl. Cr concentration in feathers of the selected species was recorded highest for Emu (0.220 µg/g) while lowest was for R.N. Pheasant (0.049 µg/g) as seen in Table 1. Pb concentration was recorded highest for White Peafowl (0.543 µg/g) while lowest was for R. N. Pheasant (0.117 µg/g). Cd concentration was recorded highest for Black Shoulder

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Peafowl (0.155 µg/g) while lowest was for R.N. Pheasant (0.060 µg/g).

Table 1: Heavy metal concentrations (digests) in feathers, water and feed collected from 8 species of birds collected from the Lahore Zoo.

Species	Feathers			Water			Feed		
	Cr µg/g	Pb µg/g	Cd µg/g	Cr µg/g	Pb µg/g	Cd µg/g	Cr µg/g	Pb µg/g	Cd µg/g
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
Emu *(Dromaius novaehollandiae)*	0.22 ± 0.022	0.251 ± 0.021	0.148 ± 0.011	0.038 ± 0.010	0.084 ± 0.048	0.098 ± 0.003	0.710 ± 0.039	0.566 ± 0.038	0.268 ± 0.008
R.N. Pheasant *(Phasianus colchicus)*	0.049 ± 0.001	0.117 ± 0.003	0.06 ± 0.014	0.414 ± 0.005	0.127 ± 0.005	0.098 ± 0.010	0.535 ± 0.021	0.489 ± 0.052	0.242 ± 0.008
Green Pheasant *(Phasianus versicolor)*	0.186 ± 0.015	0.19 ± 0.029	0.147 ± 0.007	0.091 ± 0.038	0.097 ± 0.037	0.108 ± 0.016	0.593 ± 0.018	0.490 ± 0.063	0.218 ± 0.018
White Peafowl *(Pavo muticus)*	0.182 ± 0.055	0.543 ± 0.572	0.12 ± 0.002	0.326 ± 0.014	0.121 ± 0.024	0.095 ± 0.011	0.353 ± 0.056	0.307 ± 0.042	0.173 ± 0.006
Pied Peafowl *(Pavo cristatus)*	0.179 ± 0.003	0.219 ± 0.004	0.122 ± 0.008	0.182 ± 0.027	0.100 ± 0.034	0.098 ± 0.007	0.626 ± 0.011	0.498 ± 0.012	0.257 ± 0.010
Green Java Peafowl *(Pavo muticus)*	0.176 ± 0.049	0.204 ± 0.03	0.125 ± 0.002	0.064 ± 0.019	0.159 ± 0.047	0.100 ± 0.008	0.568 ± 0.017	0.529 ± 0.018	0.227 ± 0.007
Black Shoulder Peafowl *(Pavo cristatus)*	0.198 ± 0.04	0.235 ± 0.043	0.155 ± 0.01	0.090 ± 0.010	0.172 ± 0.021	0.118 ± 0.012	0.418 ± 0.005	0.344 ± 0.041	0.182 ± 0.011
Blue Peafowl *(Pavo cristatus)*	0.208 ± 0.022	0.251 ± 0.024	0.143 ± 0.02	0.090 ± 0.025	0.181 ± 0.045	0.115 ± 0.010	0.623 ± 0.036	0.511 ± 0.009	0.230 ± 0.012

Chromium, Lead and Copper levels were analyzed in the eight samples of water provided to the selected species of class Aves from the selected sampling site. Cr concentration was recorded highest for R.N. Pheasant (0.414 µg/g) while lowest was for Emu (0.038 µg/g). Pb concentration was recorded highest for Blue Peafowl (0.181 µg/g) while lowest was for Emu (0.038 µg/g). Cd concentration was recorded highest for Black Shoulder Peafowl (0.118 µg/g) while lowest was for White Peafowl (0.095 µg/g).

Chromium, Lead and Copper levels were analyzed in the eight samples of feed provided to the selected species of birds from the sampling site. Cr concentration was recorded highest for Emu (0.710 µg/g) while lowest was for White Peafowl (0.038 µg/g) in feed (table 3). Pb concentration was recorded highest for Emu (0.566 µg/g) while lowest was for White Peafowl (0.307 µg/g). Cd concentration was recorded highest for Emu (0.566 µg/g) while lowest was for White Peafowl (0.307 µg/g).

The concentrations of Cr, Pb and Cd in feathers, water and feed are given in Fig 1,2 and 3 respectively. Feed and water were the same for all the birds' species in the Zoo therefore their average value in the form of mean was shown here.

Correlation analysis was carried out to find out the relations between selected heavy metals and birds species (Table 2) with significance at $p < 0.05$ and also between Cr, Pb and Cd to determine the relation between the metals (Table 3). The observation of significance ($p < 0.05$) relation was done.

ANOVA (one-way) was applied on distribution of Chromium, Lead and Cadmium (Table 4). The Cr concentration in the species' feathers and in the feed and water provided to them came out as significant at $p = 0.05$ however the observed F critical value was higher than the F value so this was neither reliable nor significant in this scenario as shown in table 7. In the species' feathers and the feed and water provided to them, the Pb concentration came out as significant at $p = 0.05$ but the F crit. value was higher than the F value so this was not reliable or significant in this case. The Cadmium concentration in the species' feathers and in the feed and water provided to them came out as significant at $p = 0.05$ and the observed F crit. value was higher than the F value so this was not significant in this situation. The result was not reliable either.

ANOVA (one-way) was applied on feathers of selected birds' species and the samples of water and feed fed to them. The heavy metals concentration in the species' feathers came out as significant at $p = 0.05$ because the P-value was less than 0.05 and the observed F value is higher than the F critical as shown in Table 4. Thus, the values are significant. In the water provided to the eight selected species of birds, the Pb concentration came out as significant at $p = 0.05$ however, the observed F crit. value was higher than the F value (also, the P-value is larger than 0.05) so this was not significant in this scenario. The Cadmium concentration in the feed provided to the selected birds came out as significant at $p = 0.05$ as the P-value is less than 0.05 and the observed F value is higher than the F crit.

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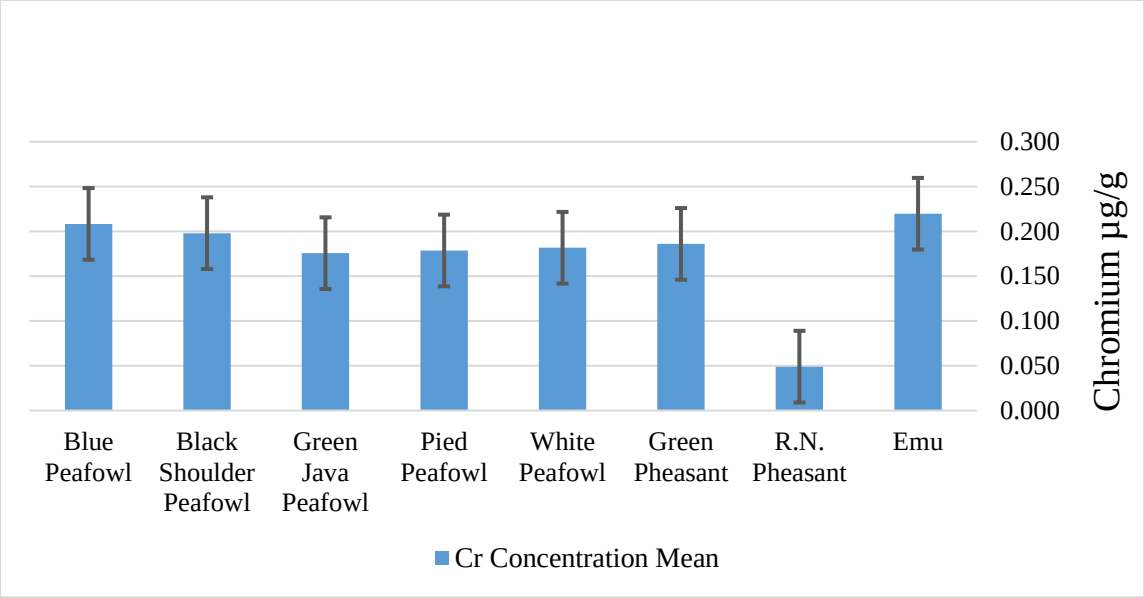

Figure 1: Concentration of Chromium in Feathers, Water & Feed of Birds

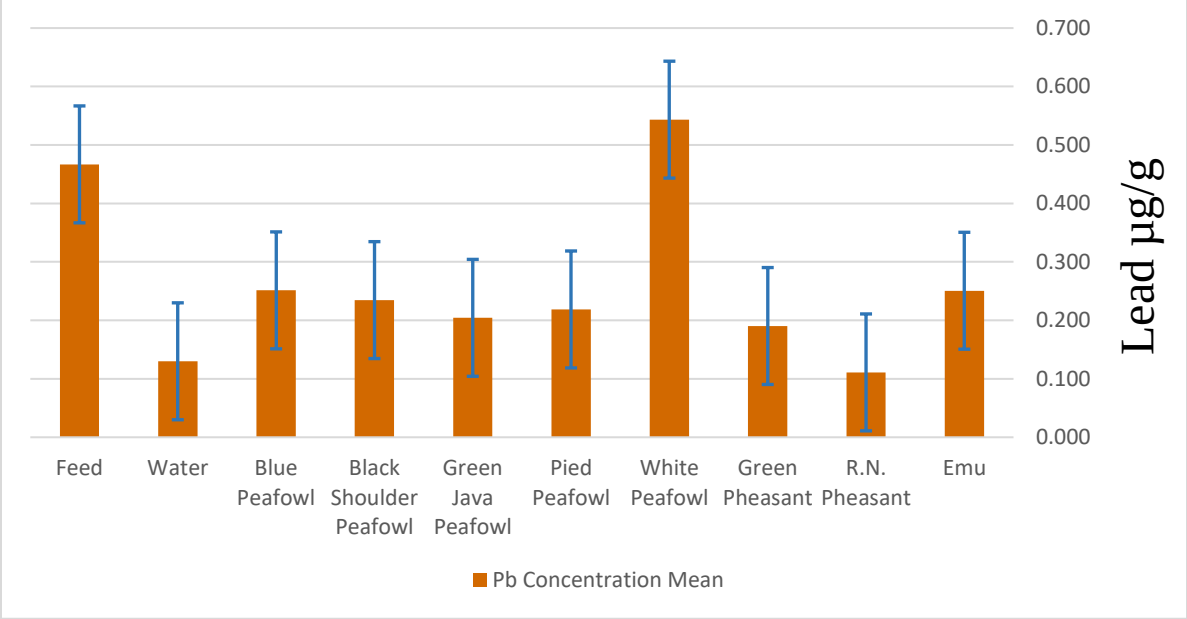

Figure 2: Concentration of Lead in Feathers, Water & Feed of Birds

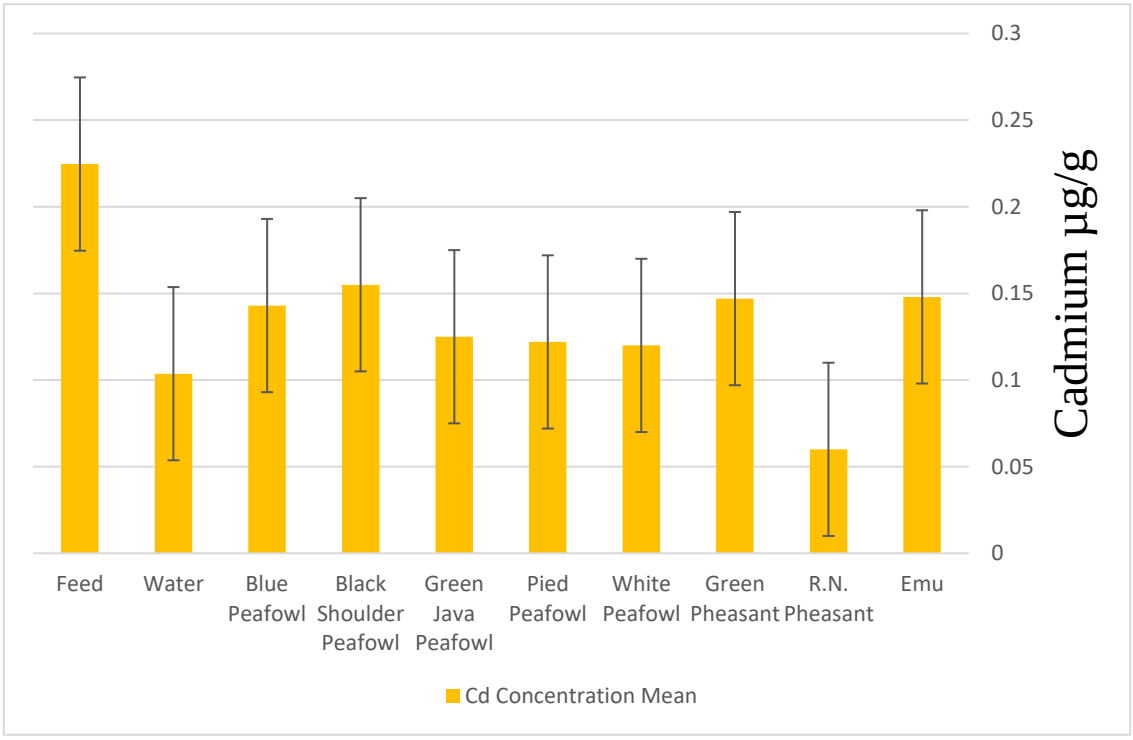

Figure 3: Concentration of Cadmium in Feathers, Water & Feed of Birds

Table 2: Correlation between concentration of metals and the bird species in Lahore Zoo

	Emu	R.N. Pheasant	Green Pheasant	White Peafowl	Pied Peafowl	Green Java Peafowl	Black Shoulder Peafowl	Blue Peafowl
Emu	1.00							
R.N. Pheasant	0.69	1.00						
Green Pheasant	0.98*	0.73	1.00					
White Peafowl	0.61	0.86*	0.65	1.00				
Pied Peafowl	0.96*	0.84*	0.97*	0.76*	1.00			
Green Java Peafowl	0.98*	0.72*	0.96*	0.61	0.95*	1.00		

Black Shoulder Peafowl	0.95*	0.60	0.93*	0.57	0.91*	0.95*	1.00	
Blue Peafowl	0.97*	0.71*	0.96*	0.62	0.96*	0.97*	0.96*	1.00

Note: Marked correlations are significant at $p < 0.05$

Table 3: Correlation between concentrations of heavy metals

		Cr			**Pb**			**Cd**		
		Feathers	**Water**	**Feed**	**Feathers**	**Water**	**Feed**	**Feathers**	**Water**	**Feed**
Cr	**Feathers**	1.00								
	Water	-0.81*	1.00							
	Feed	0.18	-0.49	1.00						
Pb	**Feathers**	0.38	0.20	-0.58*	1.00					
	Water	0.03	-0.12	-0.35	-0.04	1.00				
	Feed	-0.01	-0.39	0.95*	-0.66*	-0.26	1.00			
Cd	**Feathers**	0.95*	-0.86*	0.15	0.21	0.09	-0.04	1.00		
	Water	0.36	-0.49	-0.06	-0.24	0.64*	-0.15	0.56*	1.00	
	Feed	-0.11	-0.19	0.92*	-0.59*	-0.44	0.91*	-0.18	-0.32	1.00

Note: Marked correlations are significant at $p < 0.05$
Cr-Chromium, Pb-Lead, Cd-Cadmium

Table 4: Statistical significance of water, feed and birds species from Lahore Zoo on heavy metal concentration ascertained by ANOVA Single Factor

Metals	**F value**	**P-value**
Cr	0.208*	0.983
Pb	0.389*	0.906
Cd	0.505*	0.827
Feathers	4.7714*	0.00013
Water	0.737648*	0.657967

Feed	21.060*	3.93E-15

*significant at $p = 0.05$

4. Discussion

The world is facing a new and a gravest threat to environment, ecology, human and other species in the form of climate change. In Pakistan, rapid industrialization and urbanization over the past few decades as a result of unplanned increase in population growth has increased deforestation practices in urban cities, thus, degrading the environmental quality and making it unfit for its inhabitants. Excessive use of pesticides, fertilizers, vehicles and the resulting emissions from them, improper discharge of industrial effluents leather tanning waste, sewage water and city refuse, all are contributing to environmental contamination in the country at a larger scale (Hanif *et al*., 1987; Tahir *et al*., 1998; Aftab *et al*., 2000). As a result of this, water, air and soil becomes heavily polluted with various types of heavy metals such as As, Pb, Li, Cr, Mn, Zn,Hg and Cd (Abbas *et al*., 2004).

Birds are found almost everywhere as a result of which they are in continuous contact with human induced pollution in the environment and are therefore exposed to harmful toxins that exist in the atmosphere, and this sooner or later, shows the symptoms of those dangerous contaminants (Malik & Zeb, 2009) .Various factors are responsible for the uptake and accumulation of heavy metals in avian species which includes the age, diet and metabolism of the exposed birds (Birge *et al*., 2000). Accumulation of lead, cadmium and chromium takes place in the growing feathers of birds which restricts their harmful impacts, as these metals strongly binds to keratin in feathers and becomes functionally secluded (Malik & Zeb, 2009). Furthermore, accumulation of these heavy metals in the calcareous tissues of birds provide means for the discharge of lead from the body of female birds (Lam *et al*., 2005; Sharma & Agrawal, 2005)

Highest concentrations of lead were found in the feathers of eight selected birds compared to chromium and cadmium although chromium concentration was greater in the feed and water. Overall cadmium concentrations in all the selected birds were least compared to lead and chromium. This showed similarity with the feed and water of birds that contained least amount of cadmium in them. Metal accumulation trend in the present study of birds was reported as Pb > Cr > Cd. Difference in R.N pheasant was observed with an accumulation trend of Pb > Cd > Cr. Among all the eight selected birds, least amount of Pb, Cd and Cr pollution was observed in R.N pheasant. Cr, Cd and Pb pollution was reported highest in Emu, Black shoulder peafowl and white peafowl respectively.

In both, the feed and water, of birds, chromium concentration was higher compared to the other two heavy metals, showing more chromium pollution in them. Cadmium concentration in both the feed and water was least as compared to lead and chromium. Accumulation trend of three metals in feed and water was the same i.e. Cr > Pb > Cd. But the concentration of three metals showed significant variation between the feed and water, with feed showing notably higher concentration of chromium, lead and cadmium than water. This study suspected diet (water and feed) to be the main

source of metal exposure for birds in captivity as proposed by Lester (2014). Exposure to high amount of lead and cadmium through dietary intake can be harmful to birds (Scheuhammer, 1987). Although both feed and water were assumed to be responsible for transferring metals in the feathers of birds but feed was contributing to more metal accumulation in bird's feathers as compared to water.

Although same food and water was given to the birds but concentration of heavy metals differed from bird to bird. The reason of such difference in the accumulation rates of heavy metals in the feathers of birds might have been the age and metabolism rate of the bird. One other factor contributing to the different rates can be the bioavailability of such heavy metals. Another factor that could have played its role is the location of cages of such birds. The cage of one bird might have been more prone to the external pollution than the other.

Lead can be found anywhere in the ecosystem, in the air, water and soil. May be lead in the water given to birds comes from the plumbing material used for conveying water to the Zoo facility. Lead may also add up in the feed and water of birds which are contained in improperly glazed bowls within the cages. Food packaging may also contribute to the presence of lead in the feed of birds. Cadmium It has a long persistence rate and has very high toxicity. Cadmium can be present in air, food or water. The presence of cadmium in air can be due to natural or anthropogenic sources and if the air quality is low it would ultimately pollute the main water bodies subsequently polluting the supply lines of the water to various areas. Either way, it is harmful. Food being the significant source of cadmium can affect the birds largely. Sewage sludge in the farm fields and waste disposal on the soil might be the cause of increased cadmium concentration in the birds. Chromium is a naturally occurring element found in plants, animals, soil, rocks and volcanic dust and gases. Its presence in the environment is by both natural processes and human activities. Chromium in air exists as Cr (lll) in the form of aerosols or small particles and the area where air quality is poor it ultimately degrades the quality of water and soil and in turns deteriorates the quality of food.

Chromium and cadmium concentration in the feathers above the threshold limits poses risk to the avian population. Chromium can have drastic effects on the reproductive system and normal body functioning of various bird species (Malik & Zeb, 2009). Cadmium is not regarded a crucial element and is responsible for oviduct malfunctioning, testicular and kidney damage leading to other detrimental effects on bird's reproductive system (Malik & Zeb, 2009; Boncompagni *et al*., 2003; Burger, 2008; Eisler, 1987). Lead poisoning, having harmful impacts and being common in birds is of great concern. Lead concentration of 4 ppm in feathers of birds can harm them greatly resulting in negative impacts on bird's movement, thermoregulation and behavior, also contributing to decreased survival of young birds (Eisler, 1987; Scheuhammer, 1987; Burger, 1995; Gochfeld, 2000; Takekawa *et al*., 2002).

Like in present study, concentration of Cr and Cd in the feathers of waterfowl species in a study conducted by Kim & Oh (2014) were within the permissible limits of Cr (2.8 μg/g) and Cd (2 μg/g) reported for feathers in birds except for lead, the concentration of which in the feathers of selected birds exceeded the threshold limit value of lead (4ug/g). In the present study, mean lead and cadmium concentration in the feathers averaged 0.25 ug/g and 0.12 ug/g respectively which were well below the adverse effect levels. Similar findings were observed in the study of Burger *et al*. (2015) with an average level of lead and

cadmium in the feathers of shorebirds to be 0.5 ug/g and 0.01 ug/g respectively from 2011 to 2012. Studies conducted by Frantz *et al*.(2012), Ullah et *al*.(2014), Nighat *et al*. (2013) and Abdullah *et al*. (2015) reported significantly higher concentration of heavy metals in the feathers of birds as compared to

Table 5: Mean concentration of heavy metals (ug/g) in the feathers of birds (from 2011-2015)

Author	**Study area**	**Bird type**	**Lead**	**Cadmium**	**Chromium**
Worldwide: Dauwe et al.(2000)	Antwerp,Belgium	Great tit	4.83	0.007	-
		Blue tit	3.68	0.071	-
Tsipoura et al. (2011)	Canada	Geese	1.91	0.08	1.36
Frantz et al. (2012)	Paris Trivaux	Feral pigeon	10	0.5	-
	La Plaine	Feral pigeon	15	2	-
	Maison Blanche	Feral pigeon	20	1.0	-
	Porte de Vanves	Feral pigeon	19	0.4	-
	Parc de Choisy	Feral pigeon	8	0.2	-
	Jissieu	Feral pigeon	16	0.6	-
	Pantin	Feral pigeon	5	0.3	-
Pakistan: Nighat et al. (2013)	Central punjab	Raptors	19.52	1.83	-
	Northern punjab	Raptors	21.73	3.35	-
	Southern punjab	Raptors	18.67	1.14	-
Ullah et al. (2014)	Trimun headwork	Cattle egret	30	3.26	26.18
	Shorkot Headwork	Cattle egret	32.5	3.02	30.72
	Mailsi Headwork	Cattle egret	43.10	1.72	35.77
Abdullah et al. (2015)	Lahore	Cattle egret	297	41	21.1
	Sialkot	Cattle egret	286	37.6	19
Present study	Lahore Zoo	Emu	0.251	0.148	0.22
		Ring necked pheasant	0.117	0.06	0.049
		Green pheasant	0.19	0.147	0.186
		White peafowl	0.543	0.12	0.182
		Pied peafowl	0.219	0.122	0.179
		Green java peafowl	0.204	0.125	0.176
		Black shoulder peafowl	0.235	0.155	0.198
		Blue peafowl	0.251	0.143	0.208

values observed in present study. Results of present study were compared with results of previous research studies conducted worldwide and in Pakistan (Table 5).

There exists significant variation between the recorded concentrations of heavy metals in the feathers of present study and those values that were observed in previous research findings. This is because our present study, which includes bird species that are kept in cages and are restricted to specific environmental conditions is, however, different from other research studies that have worked on those birds which are free-living and are not restricted to only one type of habitat thus are tremendously exposed to environmental pollution compared to captive birds. Current study reported the concentration of all the selected heavy metals such as Cr, Cd and Pb in the feathers of birds well below the threshold limit of Cr, Cd and Pb as suggested by Burger & Gochfeld (2001) and Gochfeld (2000). Thus, the threshold limit for Pb, Cd and Cr in the feathers of birds was 4 µg/g, 2 µg/g and 2.8 µg/g respectively.

Almost no earlier research work conducted has documented metal concentration in feathers of avian birds that are close to or less than our observed mean concentration of metals in birds within captivity. Being the first analyses on heavy metal pollution using feathers of birds at Lahore Zoo, it is quite difficult to compare the study with previous research works. Perhaps, the efforts of comparing present study with the previously documented ones may not be very suitable due to difference in the age, diet, environmental area and metabolism of birds in the current and previous studies but still it provides some information regarding the concentration and permissible limits of heavy metals in the feathers of birds. In Pakistan, birds are least concerned organisms though they are the basic components of an ecosystem as they keep the whole system in balance. They are getting polluted by heavy metal contamination. This study is useful to monitor the environmental pollution and planning control strategies at the source.

5. Conclusion

Present work evaluated the concentration of targeted heavy metals within captive birds used as bio-indicators at Lahore Zoo, Pakistan, for which a little information already existed. No strong evidence was found that suggest the reduced immune system or body conditions of the birds at Lahore Zoo. Moreover, concentrations of selected heavy metals detected in the feather of eight birds were lower than the defined threshold limits. The present piece of work thus brings into light the concentration of heavy metals and their build up in the living organism, like birds due to growing activities of humans which are the anthropogenic causes leading to the generation of various kinds of pollutions. Present study acting as a baseline can be of considerable help in the relevant future studies for the detection of contamination caused by heavy metals in feathers of birds within captivity.

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