TRACE ELEMENTS AND METAL CONTENT IN THE FEATHERS OF THE NORTHERN BALD IBIS (*GERONTICUS EREMITA*)

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Abstract. Long-lived bird species, such as the Northern Bald Ibis (*Geronticus eremita*), are exposed to heavy metals through air, water and food because they are at the highest point in the food supply chain. Birds accumulate heavy metals in their bodies, organs, and in their feathers. The article aimed to investigate the trace element and heavy metal content of Northern bald ibis feathers using ICP-MS, as an endangered sentinel species, in order to both determine the contamination level of the area and evaluate the risk the species is facing. The study included 34 samples of feathers obtained from bald ibises released into their natural habitat. The results of the study showed that the average values of the trace elements ($^{11}Na^{+}$, ^{24}Mg, $^{20}Ca^{+}$, $^{39}K^{+}$, P, $^{27}Al^{+}$, $^{56}Fe^{+}$) which are necessary for the survival of living organisms were above 100 µg g-1, the second group of metals ($^{205}Ti^{+}$, ^{55}Mn, $^{63}Cu^{+}$, $^{66}Zn^{+}$, ^{82} Br) which are toxic to the body in high amounts were between 8.24-22.4 µg g^{-1} and those which are highly toxic at very low doses ($^{51}Sb^{+}$, ^{60}Ni, As, ^{60}Co, $^{138}Ba^{+}$, $^{208}Pb^{+}$, $^{111}Cd^{+}$, Mo, ^{75} Se) were lower than 1 µg g^{-1}. This study of the Northern Bald Ibis species, animals threatened by extinction due to environmental pollution, investigated their levels of heavy metal pollution in the feathers and indicates that Bald Ibis are possible bioindicators of environmental trace element contamination.

Keywords: *bioindicator, environmental pollution, heavy metal toxication*

Introduction

As a result of human activities, pesticides, industrial products and waste materials containing heavy metals impair the biological integrity of the ecosystem in various ways. Heavy metal pollution of the soil occurs in industrial and mining areas, through the use of fertilizers, drain waters, pesticides, waste products of the coal or oil industry and air pollution. These metals include $^{24}Mg^{+}$, $^{31}P^{+}$, $^{39}K^{+}$, $^{56}Fe^{+}$, $^{11}Na^{+}$, $^{20}Ca^{+}$, $^{63}Cu^{+}$, $^{66}Zn^{+}$, ^{75} Se and Ag, which are essential for the human body, but can be toxic in high amounts. Other metals, such as $^{60}Ni^{+}$, $^{111}Cd^{+}$, $^{138}Ba^{+}$, $^{27}Al^{+}$, $^{52}Cr^{+}$, ^{55}Mn, $^{208}Pb^{+}$, ^{82}Br, ^{60}Co, ^{205}Ti and $^{51}Sb^{+}$ are toxic even at very low amounts.

Animals ingest heavy metals by consuming feedstuffs, grass, and water, or from the application of veterinary drugs, or by licking mineral blocks, or paints containing heavy metals (Kara et al., 2016; Govind and Madhuri, 2014; Das et al., 2009). Due to higher sensitivity levels, birds are usually considered to be bio-indicators for monitoring the effects of environmental pollution (Yohannes et al., 2017).

Since long-living bird species such as the Bald Ibis are at the top of the food chain, they are more exposed to heavy metals through air, water and food. When the metals are digested they are either stored in or excreted from the body. Metals that accumulate in the organs or feathers of the birds may also be passed on to eggs (Dauwe et al., 2000).

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Feathers contain blood vessels during the growth period and heavy metals in the blood are passed to the feathers and then become isolated there after keratinization. Thus feathers provide valuable information on the level of heavy metals in birds' blood. Since the 1960s, feathers have been used as bio-indicators for monitoring heavy metal exposure of birds, and it has been reported that feathers which are directly exposed to environmental pollution may have higher heavy metal contents due to exogenous contamination (Rutkowska et al., 2018).

Thus, there is a need for studies to determine the toxic effects of environmental pollution in order to maintain biological diversity and protect sentinel species such as the Bald Ibis (Bauerová, 2017; Da Silva et al., 2017; Dolan et al., 2017; Borgesi et al., 2016).

Numerous studies have been conducted on birds of prey to determine the residuals of environmental pollutants such as Pb, Cd, and Hg and pesticides (İnangi et al., 2019; Carneiro et al., 2018; Espín et al., 2016). The ingestion of heavy metals by animals has been reported to cause intoxication, resulting in behavioral disorders, impairment of feathering or decreased breeding and hatchability (Kara et al., 2016).

The Bald Ibis population in Turkey declined to a size near extinction due to the intensive use of pesticides (Dichlorodiphenyltrichloroethane - DDT) against grasshoppers between 1955 and 1960. Survivors did not lay eggs for several years and nestlings hatched were weak. In the offspring of subsequent generations, deformations of the beak and feet were observed (Akyıldız et al., 2005). In the red list published by the International Union for the Conservation of Nature (IUCN), the Bald Ibis was classified as an endangered species. There are three different breeding centers in different locations of the world, including the Birecik district of Turkey, Fas in Morocco and Palmyra in Syria (*Fig. 1*). The life span of the Bald Ibises is about 25-30 years. Bald Ibises in Birecik district are housed in wooden nests or in nests carved into calcite rocks (Tel and Keskin, 2012).

The General Administration of Nature Conservation and National Parks, with the support of the World Wildlife Foundation, established a breeding station in the Birecik district of Sanliurfa province. The Bald Ibises are kept semi-captive, eating pests such as snakes, grasshoppers, insects, scorpions, snails, lizards or snapdragons in the fields near the Euphrates River, when they are out of the breeding station in the summer. They are fed on a diet of raw lean meat, whey, cooked egg, and carrot when they enter the breeding station in winter (Mundan and Cetin, 2012).

There have been few studies on the factors threatening the survival of the Bald Ibis. Tel and Keskin (2012) investigated the prevalence of Yersinia spp. and Aeromonas hydrophyla, while Tel et al. (2013) reported the presence of Salmonella spp. Campylobacter spp. and the absence of Chlamydia psitaci in feces samples of Bald Ibisies. Recently, Spergser et al. (2018) reported the cultivable microbiota isolated from different organs of Northern Bald Ibises. As exposure to heavy metals, even at low concentrations, may negatively affect reproduction of birds by decreasing egg production and hatchability, and increasing the mortality of hatchlings (Malik and Zeb, 2009; Scheuhammer, 1987), it is important to monitor the heavy metal exposure status of endangered species, such as the Bald Ibis. However, to the best of our knowledge, there has been no previous report on the heavy metal exposure status of Bald Ibises.

Therefore, the objective of this study was to determine metal and heavy metal contents of feather samples of Bald Ibises raised in the Birecik District of Sanliurfa, Turkey, in order to estimate predict the risk of heavy metal exposure of this species. In the future, Bald Ibises might be an ecological indicator for trace element exposure in pollinated ecosystems and studies determining the source of exposure might help develop conservation strategies for protecting this important and charismatic wildlife species

Materials and methods

This study was carried out with the permission and support of the Turkish General Directorate of Nature Conservation and National Parks (Approval No: 72784983.04-42845).

Sample collection

Feather samples of 34 individual birds (17 male and 17 female) were obtained from the wings or tails of birds raised semi-captive at the Bald Ibis breeding station in Birecik district of Şanliurfa province, Turkey (37.01.29 N:37.58.38 E). The feather samples were placed in plastic bags and stored at -20 °C until analysis. Whole feather samples were used for chemical analysis after the removal of dust or feces residues with distilled water. (Costa et al., 2013) Collection of the feather samples was approved by the General Directorate of Nature Conservation and National Parks in the framework of the protocol signed by Harran University and the General Directorate of Nature Conservation and National Parks (Protocol No: 26130895-030.03)

Figure 1. Settlement and habitat of the bald ibis and map image of Birecik (Turkey) and Palmira (Syria)

Chemicals and standard solutions

Chemicals including 60% hydrogen peroxide (H_2O_2), 37% hydrochloric acid (HCl) and 65% nitric acid (HNO_3) were purchased from Merck (Darmstad, Germany). Stock standard solutions for each element were purchased from Agilent Technologies, Japan (Lot number: 10-160YPYZ). The 99.9980% Argon gas was supplied by Linde Gases (Linde Group, Turkey)

Microwave acid digestion

After the feather samples were weighed they were washed with distilled water. The samples were put into a Teflon microwave vessel and 2 ml 60% H_2O_2, 3 ml 37% HCl and 1 ml 65% nitric acid (HNO_3) were added to each sample. The samples were left overnight at room temperature for slow digestion. The samples were treated in a microwave oven (Cem Mars 5) at 180 °C and 800 W for 1 h, then transferred to a 50 mL tube and distilled water was added up to 50 mL.

Analysis of essential element and heavy metal content on ICP-MS

Essential element and heavy metal content of the digest was assessed using an ICP-MS device inductively coupled plasma-mass spectrometer with an Auto Sampler and nebulizer (Agilent, 7500ce Octopole Reaction System, Japan) (Mikoni et al., 2017). Calibration of the method was applied using an internationally validated standard. The accuracy of the device and the method was achieved by measuring a certified reference.

Statistical analyses

Statistical calculations were performed using SPSS 22.0 software (SPSS Inc., Chicago, USA). Conformity of the data to normal distribution was assessed with the Shapiro Wilk test. The Mann-Whitney-U test was applied to variables not showing normal distribution. Correlations between numerical variables were determined using the Spearman rank correlation coefficient. The result of the hierarchical clustering was presented in a dendrogram (*Fig. 2*). A value of $p < 0.05$ was considered statistically significant. The correlations between the elements found in the samples were calculated while the significance value was evaluated.

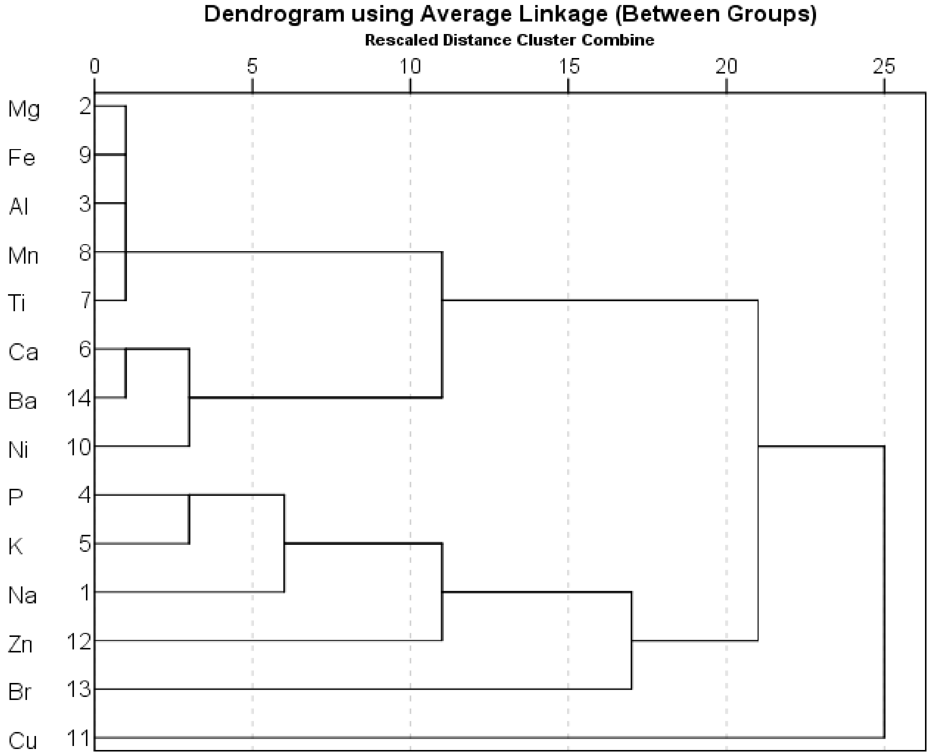

***Figure 2.** Hierarchical clustering results of feather samples (dendrogram)*

Results

The mean concentrations of the essential elements and heavy metals measured in the feather samples are presented in *Table 1.*

Table 1. *Concentrations of essential elements and heavy metals in the feather samples (µg g-^{1}, dw, a dry weight basis)*

Elements	Minimum	Maximum	Mean	Std. error	Std. deviation	CV (%)
Na	1151.90	4601.24	2048.83	124.74	727.25	35.49
Mg	194.87	1181.50	365.96	28.64	167.00	45.63
Al	11.37	1160.74	246.52	34.73	202.50	82.14
P	87.37	1238.64	248.78	32.41	188.97	75.958
K	0.13	2178.72	168.31	70.57	411.41	244.43
Ca	850.62	7486.56	2200.42	217.01	1265.12	57.49
Ti	6.67	72.69	22.74	1.96	11.41	50.17
Mn	1.13	46.12	8.54	1.33	7.76	90.87
Fe	53.93	1664.62	337.58	48.10	280.44	83.07
Ni	0.06	23.37	1.86	0.68	3.94	211.83
Cu	0.09	6.10	2.89	0.21	1.22	42.21
Zn	16.07	210.57	67.88	7.57	44.15	65.04
Br	0.02	35.03	8.24	1.31	7.66	92.96
Ba	0.26	21.01	3.48	0.62	3.61	103.74

Concentrations of the essential elements (Na, Mg, Ca, K, P, Al, Fe) were detected to be higher than 100 µg g^{-1} while those of Ti, Mn, Cu, Zn and Br were between 8.24 and 22.4 µg g^{-1}. Of the heavy metals, As was detected in 5 samples with concentrations varying from 44.77 to 2083.05 µg g^{-1} while Co was found in only 2 samples with concentrations of 344.24 and 848.31 µg g^{-1}. Se and Pb were detected in 1 sample each, at concentrations of 447.01 and 119.66 µg g^{-1}, respectively. Detectable concentrations of Sb, Ni, Mo and Pb were not observed. A significant difference in the metal contents between the sexes was observed only for Cu ($p = 0.024$) while no difference was observed in the contents of other metals ($p > 0.05$)

Other toxic elements were found in the values of Co (227-842 µg g^{-1}), As (45-30 836 µg g^{-1}), Se (0-447 µg g^{-1}), and Pb (0-119 µg g^{-1}). The Sb, Cd and Mo values were determined under the measurable value.

The correlation coefficients between individual metal contents of the samples are shown in *Table 2*. High and significant correlations were observed especially among Mg, Al, Ti, Mn, Fe and Ni contents. The hierarchical clustering results among the metal contents are presented in *Figure 2.*

In this study, significant correlations from moderate to high correlation coefficients between essential elements were observed. For example, moderate positive correlations were determined between Na and Br and between Na and P ($r = 0.473$ $p = 0.01$) ($r = 0.566$, $p = 0.001$) and a weak positive correlation was determined between Na and K content (r: 0.388; p: 0.023). In addition, Ca content was moderately correlated with Ni ($r = 0.518$, $p = 0.02$) and Ba ($r = 0.861$, $p = 0.001$) and with Fe ($r = 0.401$, $p = 0.019$) and Mn ($r = 0.412$, $p = 0.016$) and a weak significant negative correlation with Cu (r = -0.378 $p = 0.027$).

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Table 2. *Correlations between the metal contents of the feather samples*

	Na	Mg	Al	P	K	Ca	Ti	Mn	Fe	Ni	Cu	Zn	Br
Mg	0.026	1											
Al	-0.103	0.947^{**}	1										
P	0.566^{**}	-0.079	-0.206	1									
K	0.388^{*}	0.205	0.195	0.072	1								
Ca	0.313	0.480^{**}	0.395^{*}	0.176	-0.035	1							
Ti	0.094	0.920^{**}	0.948^{**}	-0.099	0.204	0.565^{**}	1						
Mn	-0.233	0.889^{**}	0.896^{**}	-0.199	0.111	0.412^{*}	0.819^{**}	1					
Fe	-0.081	0.960^{**}	0.971^{**}	-0.219	0.22	0.401^{*}	0.909^{**}	0.939^{**}	1				
Ni	-0.022	0.906^{**}	0.872^{**}	-0.061	0.246	0.518^{**}	0.844^{**}	0.942^{**}	0.903^{**}	1			
Cu	-0.221	-0.341^{*}	-0.233	-0.113	0.073	-0.378^{*}	-0.297	-0.374^{*}	-0.314	-0.444^{**}	1		
Zn	0.106	-0.246	-0.224	-0.107	-0.202	-0.203	-0.142	-0.209	-0.21	-0.23	-0.255	1	
Br	0.473^{**}	0.118	0.079	0.215	0.425^{*}	0.293	0.197	0.051	0.144	0.138	0.048	-0.243	1
Ba	0.162	0.571^{**}	0.582^{**}	0.029	0.033	0.861^{**}	0.717^{**}	0.557^{**}	0.547^{**}	0.608^{**}	-0.292	-0.108	0.272

Fe is a trace element found in complex with other metals. In this study, Fe showed a positively strong correlation with Ni ($r = 0.903$, $p = 0.01$), a medium correlation with Ba ($r = 0.547$; $p = 0.01$), and weak negative correlation with Cu ($r = -374$ P: 0.029).

When the correlations of Mg were examined, there was a weak negative correlation with Cu values ($r = -0.342$; $p = 0.049$), while there was a moderate positive correlation with Ca values ($r = 0.480$ $p = 0.04$), and a strong positive correlation with Al, Ti, Mn, Fe, Ni, and B ($r = 0.947$ $p = 0.01$, $r = 0.920$; 0.881; 0.960; 0.904; 0.571, $p = 0.001$, respectively).

A strong positive correlation was found between Mn and Fe and Ni ($r = 0.939$; 0.942 and $p = 0.01$). There was a moderate positive correlation with Ba ($r = 0.55$; $p = 0.01$).

A strong positive correlation was found between Al, Mn, Fe, Ni and Ba ($r = 0.971$; 0.948; 0.872; 0.582; $p = 0.001$ respectively). There was a moderate positive correlation with Ca ($r = 0.395$; $p = 0.021$).

As all birds were between 1 and 4 years of age, no statistically significant difference was found between age and heavy metal levels ($p > 0.05$). In addition, gender analyzes in bald ibises can only be performed genetically by Realtime PCR and RFLP-PCR. No statistically significant difference was found between the genders and heavy metal levels in the analyses ($p > 0.05$).

Discussion

Due to the accumulation of metals in feathers they reflect both the physiological status of the animal and the environmental conditions. Therefore, feathers are valuable materials for monitoring the effect on birds of environmental pollution with heavy metals. This study reports for the first time the metals and heavy metals contents of feather samples from the Bald Ibis.

The natrium (1151-4601, 2048 ± 727 $\mu g\ g^{-1}$) and K (0.13-2178 $\mu g\ g^{-1}$, 168 ± 70.57) content detected in the feather samples of Bald Ibises were similar to those reported for Na (1087-3950 $\mu g\ g^{-1}$) and K (84-285 $\mu g\ g^{-1}$) in the Greater Flamingo (Borgessi et al., 2016).

Calcium plays a role in muscle contractions, enzyme activation and cardiac rhythm (Zamberlin et al., 2012). The mean Ca content in the feather samples of Bald Ibises were higher than that reported by Hanson and Jones (1968) for geese (600-2800 µg g^{-1}) from different localities of USA and lower than that (3151.87–4087.67) in red-breasted flycatchers (*Ficedula parva*) as reported by Hanc et al. (2017). The higher level of Ca in the present study could have been due to the higher Ca content of the rocks where the Bald Ibises are housed. Bald Ibises are housed either in wooden nests or in holes carved into limestone rocks within the breeding station. Limestone found around Sanliurfa mostly consists of formations of middle upper Eocene age, comprising limestone (Fonsi Formation) or clayey and chalky limestone (Gaziantep Formation) (Richardson, 1991, cited by Canakci et al., 2007). Turgut et al. (2008) reported that Sanliurfa stone samples contain a high amount of Ca and lower amounts of Al, Fe, Mg, S and Si, while they contain no Na, K and Cl.

Mg plays an important role in protein metabolism, blood pressure regulation and neuromuscular transmission. The Mg content in the feather samples (365.96 ± 28.65 µg g^{-1}) in the present study was lower than that reported by Borghesi et al. (2016) for the Greater Flamingo (463-1843) and higher than that reported by Hanc et al. (2017) in red-breasted flycatchers (*Ficedula parva*) (174.58 µg g^{-1}).

This difference may be related to the feeding patterns of the bird species studied and the environment in which they live. Flamingos feed on small plankton-like animals in wetlands. The Bald Ibises (*Geranticus eremita*) outside of the breeding station feed on insects, snails, etc, and the nesting area includes calcium-rich limestone rocks. This difference might also be related to the age of the bird as the basic elements were expected to be higher because the birds in this study were younger and still in the growth and development period.

Potassium plays an active role in cellular activities, in nerve conduction and bone metabolism. The higher K content of the Bald Ibis feathers associated with environmental pollution, demonstrated that the water, soil and plants in the habitats. Sodium plays a role in the acid-base balance in extracellular fluid. Mean content of Fe in the feather samples in the present study (337.58 ± 48.10) was found to be higher than the reported values of 48.9 ± 5.8, 185.84 ± 18.47 and 52.79 ± 50.69 µg g^{-1} for Common moorhens (*Gallinula chloropus*) (López-Perea et al., 2019), Italian Sparrows (*Passer italiae*) (Innangi et al., 2019) and Anna's humming birds (*Calypte anna*) (Mikoni et al., 2017) respectively. No significant difference in the Fe content between the sexes was observed ($p > 0.05$). The higher Fe content of the Bald Ibis feathers could be attributed to species differences associated with nutritional behavior. For example, moorhens eat plant seeds and invertebrates (Zamani-Ahmadmahmoodi, 2010) while the Bald Ibises in this study consumed both small vertebrates and invertebrates in addition to the diet supplied in the breeding station which contained lean meat. However, the differences may also arise from the geographical distribution of the same species. Innangi et al. (2019) reported that the feathers of Italian Sparrows contained a 9-fold lower level of Fe than those of House Sparrows in Southern Africa.

Based on the observation that Mn together with Zn and Ca is abundant in black-coloured feathers, and that healthier birds have more coloured feathers, Innangi et al. (2019) suggested that higher levels of Mn might indicate a healthier bird. The higher Mn content in the feather samples in this study could therefore be attributed to the black-coloured plumage of Bald Ibises.

The mean Ba content detected in this study (3.48 ± 0.62 µg g^{-1}) was lower than the value reported by Adout et al. (2007) for Feral Pigeons (4.84 ± 1.31-16.7 ± 8.1) and Hooded Crows (*Corvus corone cornix*) in rural (15.6 ± 7.36) or industrial areas (18.3 ± 6.8). This can be thought to be due to industry in the regions close to the study area. Barium can pharmacologically replace Ca (John et al., 2005) ions in the keratin structure of bird feathers and high Ba levels can result in health problems in young birds (Hanc et al., 2011).

The results of this study showed a high correlation between Ba content and Ca content in the feather samples ($r = 0.861$, $p = 0.001$). It has been suggested that environmental pollution with Ba is caused by dust from road aggregate material (Brumbaugh et al., 2006). The lower Ba content in the Bald Ibis feather samples could therefore be explained by the distance of the breeding station from main roads.

The mean Zn content (67.88 ± 7.57 µg g^{-1}) in this study was found to be similar to that reported for birds raised in regions near to industrial areas. López-Perea et al. (2019) reported a mean Zn level of 71.11 µg g^{-1} in the feather samples of common moorhens living in wetlands irrigated with industrial waste water. Dolan et al. (2017) observed the highest levels of Zn (82.1 mg/kg) in the feathers of Northern Goshawks living in regions near to industrial areas in Norway and Spain. Mikoni et al. (2017) found 123.79 mg g^{-1} on the feathers of free-flying Anna's humming birds in California, which was approximately double the values in the current study. The authors attributed the high level of Zn in this species to the pollution of the nectar and flies they eat by Zn contained in dish washing detergents. Innangi et al. (2019) reported Zn content of 106.26 µg g^{-1} in the feather samples of free-flying birds in a region close to an industrial zone in southern Italy. Borghesi et al. (2016) reported levels of 77.44 mg kg^{-1} in bird colonies living near industrial zones in France, Spain and Italy Zinc is an important and necessary element for the keratinization process in birds, and is also used in veterinary medicine as an emetic and antiseptic drug or as an antagonist drug against copper sulphate toxicity. As excessive ingestion of Zn is toxic, environmental pollution with Zn represents a risk for wildlife (Sundaresan et al., 2008; Honda et al., 1986)

The mean value Ni found in the current study (1.86 ± 0.67 µg g^{-1}) was higher than the values found by Borghesi et al. (2016) (1.058 µg g^{-1}) and López-Perea (2019) (0.087 µg g^{-1}) and lower than that reported by Mikoni et al. (2017) (3.28 µg g^{-1}). Thus, the Ni content reported in this study was within the range previously reported in literature. However, Ni is a metal extensively used in industry and industrial activities, and isa major source of environmental pollution. The town of Birecik, where the study was conducted, is close to the industrial city of Gaziantep. The mean concentration of Ni in the sediments at different locations of the Euphrates River has been reported to be 0.16-0.35 µg g^{-1} (Oymak et al., 2009). Therefore, disposal of industrial waste in the Euphrates River may be the cause of Ni exposure of the birds.

Al can be transmitted from the environment to both natural and anthropogenic sources and its toxicity varies according to the target organ. The mean Al content found in the present study (246.52 ± 34.73) was higher than the values found by Borghesi et al. (2016) (3.71 µg g^{-1}) and Dolan et al. (2017) (23 µg g^{-1}). Mn was determined in the range of 20- 491 µg g^{-1} and these values were higher than all other studies [Lopez et al., 2019, (3.93-89 µg g^{-1}), İnnangi et al., 2019, (0.33-23.83 µg g^{-1}) and Mikoni et al., 2019, (0.33-23.83 µg g^{-1}]. Excessive amounts of Mn have been reported to be life-threatening and high amounts of Mn taken by ingesting sediments can cause acute and chronic toxicity (Sánchez-Virosta et al., 2015).

In some studies, the Cu content of feather samples gas been found to be 1.97-149.9 µg g^{-1} (Mikoni et al., 2017), 5.04-17.04 µg g^{-1} (López et al., 2019), 6.59 ± 4.72 µg g^{-1} (Bauerová et al., 2017), and 2.68 µg g^{-1} (İnnangi et al., 2019). In this study, the Cu value was determined between 5 -500 µg g^{-1}. This value is higher than those reported by other study groups. Cu is widely used in veterinary medicine as an anthelmintic antiperspirant. Pollution from water, sediment and industrialization affects the amount of Cu in birds. The high Cu concentration determined in this study may reflect the pollution of the habitat in which the animal lives.(Custer et al., 2008) This shows that there is a continuing risk of environmental contamination for the Bald Ibis, which is at risk of extinction.

Arsenic is a toxic element. Acute and chronic intoxication may be seen with excessive intake of this metal (Borghesi et al., 2016; Sanches Virost et al., 2015). In the current study, the mean arsenic levels in the feather samples were 486 ng/g in males and 1451 ng g^{-1} in females although the differences between the sexes were not significant. Bauerová et al. (2017) reported cyanide levels of 1510 ng g^{-1}, Borghesi et al. (2016), 819 ng g^{-1}, and López-Perea et al. (2019) 378 ng g^{-1}. Arsenic is bound to sulphur-rich proteins in feathers (Murphy et al., 1990).

Other toxic elements determined in the current study were Se 447.01 ng g^{-1}, Pb 119.66 ng g^{-1}, and Co 227.96 -848.31 ng g^{-1}. The presence of toxic heavy metals in bird feathers may be due to the contamination of their feathers with external pollution. It has also been reported in other studies that pollution can occur with these metals (Al, Co, Ni, Cu, Fe, Zn, Mn, Ag, Tl, Pb and Cd) during the growth period of feathers (Dauwe et al., 2003). Exposure of birds to Pb, Se, Cd, Zn and As causes reproductive failure, growth disorder, and behavioral changes (Binkowski et al., 2013; Govind and Madhuri, 2014; Alvárez et al., 2013). With the exception of Cu, no significant difference was observed in terms of other elements. Most studies on different bird species have reported no significant difference between the sexes in the metal content of feather samples (Markowski et al., 2013; Squadroneet al., 2016).

The fact that high and significant correlations were observed among Mg, Al, Ti, Mn, Fe and Ni contents might indicate a common physiological or environmental basis. Given that trace elements can move though food chains by a variety of routes.

Conclusion

This is the first study to determine heavy metal pollution in the feathers of the Bald Ibis, which is an endangered bird. In this study, the presence of heavy metals such as Pb, As, Br, Ni, Zn, which are important for the determination of environmental pollution, demonstrated that the water, soil and plants in the habitats of the people are exposed to metal pollution because of industrial, domestic and agricultural activities. The heavy metal limits determined in this study, are at a level that threatens the life of the Bald Ibis and reflects the pollution of the habitat they live in, and that the continued risk of environmental contamination threatens the continuation of the species. This study indicates that Bald Ibis are possible bioindicators of environmental trace element contamination. The only way to ensure the continuity of the species of Bald Ibis and transfer them to future generations is to protect the ecosystem they live in and eliminate or reduce the threats such as the heavy metal elements identified. In order to do this, there is a need for national and international co-operation with local people so that habitat and birds will be protected together in the local community.

As a result, we recommend further studies to understand the effects of the heavy metals on the population of the species Bald Ibis.

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