

Tissue distribution and correlations of heavy metals in wild birds from Southern Turkey: an ecologically important region on the west Palearctic migration route

Mustafa Yipel (✉ musyip@hotmail.com)

Hatay MKÜ: Hatay Mustafa Kemal Universitesi <https://orcid.org/0000-0002-6390-9313>

İbrahim Ozan TEKELİ

Hatay MKÜ: Hatay Mustafa Kemal Universitesi

Cafer Tayer İŞLER

Hatay MKÜ: Hatay Mustafa Kemal Universitesi

Muhammed Enes ALTUĞ

Hatay MKÜ: Hatay Mustafa Kemal Universitesi

Research Article

Keywords:

Posted Date: April 25th, 2023

DOI: <https://doi.org/10.21203/rs.3.rs-2021061/v2>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License. [Read Full License](#)

Version of Record: A version of this preprint was published at Environmental Science and Pollution Research on May 2nd, 2023. See the published version at <https://doi.org/10.1007/s11356-023-27292-8>.

Abstract

Even if some are essential for biological functions, the accumulation of heavy metals above tolerable physiological limits is potentially toxic to also wild animals. The present study aimed to investigate concentrations of environmentally relevant heavy metals (As, Cd, Cu, Fe, Hg, Mn, Pb, and Zn) in feathers, muscle, heart, kidney, and liver tissues of wild birds (golden eagle [*Aquila chrysaetos*], sparrowhawk [*Accipiter nisus*], and white stork [*Ciconia ciconia*]) from Hatay province, southern Turkey. The metal concentrations of tissues were determined by a validated ICP-OES analysis method after microwave digestion. The concentration differences of metals in species/tissues and the correlations between essential/non-essential metals were determined by statistical analysis. According to the results, Fe ($326.87 \pm 3.60 \text{ mg kg}^{-1}$) had the highest, and Hg ($0.09 \pm 0.00 \text{ mg kg}^{-1}$) had the lowest mean concentration in all tissues. Compared to the literature; Cu, Hg, Pb, and Zn concentrations were lower; Cd, Fe, and Mn concentrations were higher. The correlations between As and all essentials; Cd and Cu, Fe; Hg and Cu, Fe, Zn; Pb and all essentials were significantly positive. In the conclusion, while essential Cu, Fe, and Zn are below the threshold value and do not pose a risk, Mn is close to the threshold value. Therefore, periodically monitoring the pollutant concentrations in bioindicators is a key necessity for the early determination of biomagnification trends and prevention of potential toxic stress on wildlife ecology.

1. Introduction

Heavy metals are persistent and ubiquitous inorganic pollutants that create environmental contamination as a result of geological (such as volcanic eruptions and soil erosion) or anthropological activities (such as mining, burning fossil fuels, and pesticides) (Naccari et al., 2009, Sánchez-Virosta et al. 2015; Yamac et al. 2019). They accumulate in the tissues of terrestrial and aquatic organisms and magnify in the food chain (Naccari et al., 2009).

Depending on their biological necessity, metals are categorized as essential (chromium [Cr], copper [Cu], cobalt [Co], iron [Fe], manganese [Mn], nickel [Ni], selenium [Se], tin [Sn], vanadium [V], and zinc [Zn]) or non-essential (arsenic [As], cadmium [Cd], mercury [Hg], and lead [Pb]). Despite essentials being necessary for biological processes, their accumulation above the tolerable physiological thresholds can have toxic effects. Even at low concentrations, non-essentials can be toxic (Squadrone et al., 2018). Metals and other contaminants can cause early (within seconds to days) and late (weeks to decades) symptoms. The early symptoms include biochemical such as enzyme inhibition; physiological such as disruption of hormonal balance, and behavioral such as migration, aggression, predation, and changes in mating or feeding habits, etc., which can occur at molecular, cellular, tissue, organ, system, and organism concentrations. The late symptoms include birth, death, sex, and changes in biomass ratios and migration consequences, resulting in a decline in population growth (at the population concentration), changes in the predator-prey relationship (at the community concentration), changes in physicochemical conditions, species distribution, food cycle ratios, food chain step, and model (at the ecological concentration) (Boudou and Ribeyre, 1997; Bhat, 2013, Weis, 2014; Nikinmaa, 2014).

Due to high affinity (Cd, Cr, Cu, and Pb in feathers, nails, horns, and hooves), biotransformation (Cd in the liver), and excretion (Cd in the kidney), the accumulation of metals is higher in some tissues and organs referred to as (Mukhtar et al., 2020; Tekeli et al., 2021). Hence feathers (Zolfaghari et al., 2007; Nighat et al., 2013; Grúz et al., 2018; Squadrone et al., 2018; Mukhtar et al., 2020), liver (Kenntner et al., 2007; Pérez-López et al.,

2008; Komosa et al., 2012; Salamat et al., 2014; Mukhtar et al., 2020), kidney (Kenntner et al., 2007; Naccari et al., 2009; Mukhtar et al., 2020), muscle (van Wyk et al., 2001; Kalisińska et al., 2006; Naccari et al., 2009; Salamat et al., 2014) and heart (van Wyk et al., 2001; Mukhtar et al., 2020) are the most commonly examined tissues and organs in studies of environmental pollution-related metal accumulation in wild birds.

Wild birds are one of the most exposed species to environmental pollutants, particularly metals, through diet (because of their high trophic concentration) and environmental exposure (air, etc.) (Pérez-López et al., 2008). Thereby, birds are one of the most often subjected wild animals as bioindicators in the ecotoxicology studies (Kenntner et al., 2008; Pérez-López et al., 2008; Naccari et al., 2009; Salamat et al., 2014; Grúz et al., 2018).

One of the largest avian predators with a significant impact on biodiversity and a wide geographic range, the *Aquila chrysaetos* (Golden eagles) (*Ac*), is regarded as an indicator species linked to environmental health (Wiens et al., 2022). Pb poisoning in birds has been linked to mortality and growth retardation, yet the literature on other metal contaminations or metal interactions is limited (Herring et al., 2017). *Accipiter nisus* (Sparrowhawk) (*An*) is a small bird predator with a widespread geographic distribution in Europe and Asia and has been used as a bioindicator for metal contamination in feathers. (Dauwe et al., 2003; Meyer et al., 2009; Hervías et al., 2017; Thornton et al., 2017; Grúz et al., 2018). Due to their predation on other vertebrate and invertebrate indicators, migratory white storks (*Ciconia ciconia*) (*Cc*) are regarded as suitable ecotoxicological bioindicators (Grúz et al., 2018; Bjedov et al., 2022).

In terms of aquatic and terrestrial habitats, Hatay, a city on Turkey's northeastern Mediterranean coast, is a significant ecological zone. Ecotoxicological studies subjected to the region are limited despite being on a route for wild bird migration and serving as a refuge for other wild species. The region has intense urban, industrial, and agricultural activities due to high population, intense national and international transport (sea and land), high agricultural production, and large-capacity industrial sectors (iron-steel, fertilizer and cement, etc.). In consequence of these activities, the region is at high risk of environmental contamination. (Yipel and Tekeli, 2016; Yipel et al., 2021).

In the study, it was aimed to 1) provide the first tissue (feathers, liver, kidney, muscle, and heart) concentration data of environmental pollution-relevant metals (As, Cd, Cu, Fe, Hg, Mn, Pb, Zn) of wild bioindicators (*Ac*, *An*, and *Cc*) from an ecologically important area (Hatay, Turkey) on the Western-Palearctic migration way; 2) to reveal the potential toxic risk at the organism and population concentration for the included species; 3) determine statistically significant differences between metal concentrations in wild species tissues; 4) determine statistically significant positive and negative correlations between essential and non-essential metals; and 5) to provide reference data for future bioconcentration and biomonitoring studies.

2. Materials And Methods

2.1. Sampling

Ac (n:9; 6 male, 3 female), *An* (n:12; 5 male, 7 female), and *Cc* (n:7; 5 male, 2 female) that were found injured or dead in Hatay City, in the southern region of Turkey (Fig. 1, 36.20 N-36.16E) and submitted to the Hatay Mustafa Kemal Wild Animal Rescue and Rehabilitation Center between 2015-2021. The weight (grams) ranges from 3740 to 5655 (mean 4213), 136 to 328 (mean 164), 5860 to 9340 (mean 7170), the length (cm) ranges from 72

to 83 (for 78), 59 to 78 (mean 66), 102 to 110 (mean 107) and the wingspan (cm) ranges from 191 to 216 (for 201), 29 to 38 (mean 33), 157 to 166 (mean 160) for *Ac*, *An*, and *Cc* respectively. During the post-mortem examination of the recently dead (<12 h) adult (>1 years old) birds, feathers, liver, kidney, muscle, and heart samples were taken. The samples were stored at -20 °C until instrumental analysis.

2.2. Metal analysis

One gram of homogenized tissue digested in a microwave (CEM; Mars X press, Matthews, NC) oven with 10 mL of nitric acid (70%) in four steps (1200 W, 100% power, 15 min ramp, 200 °C, 15 min hold). An inductively coupled plasma optical emission spectrophotometer (ICP OES-Optima 3000DV Perkin-Elmer-Norwalk, USA) was used for the analysis (1400 W plasma power, 30 rpm pump speed, 12 l/min coolant flow, 1 l/min auxiliary flow, 1 l/min nebulizer flow) of digested samples. For the calibration procedure, five standard solutions (0 to 500 ppb) of the ICP multi-element standard (mg L⁻¹ in nitric acid) were prepared. All the reagents were of analytical grade (Merck, Germany). The method quality was checked and validated by the certified reference material (DORM-4; National Research Council, Canada). The recovery rates were satisfactory, ranging from 95 to 108%. The limits of detections (LOD) were As: 3.63; Cd and Cu: 0.60; Hg: 2.70; Mn and Pb: 2.10; Fe, Mn, and Zn: 0.30 and the limits of quantification (LOQ) As: 12.00; Cd and Cu: 2.00; Hg: 9; Mn and Pb: 7.00; Fe, Mn, and Zn 1.00 expressed as ng g⁻¹.

2.3. Statistical analysis

The Shapiro-Wilk and Levene's tests were performed to determine the normality and homogeneity of the data respectively. Then non-parametric Kruskal-Wallis test was performed to determine differences between the groups and significance was defined as a P value of less than 0.05. Pearson's correlation coefficient (r) test was performed to determine the correlations between the groups and significances were defined as P values of less than 0.05 (*) and 0.01 (**). A statistical program (SPSS 23.0, IBM, USA) was used for the tests.

3. Results

The mean, minimum, and maximum metal concentrations in the feathers, liver, kidney, muscle, and heart tissues of *Ac*, *An*, and *Cc* are presented in Table 1.

The difference in tissue concentrations of As in all tissues; Cd in the liver and kidney; Cu in feathers, kidney, and muscle; Fe the in liver, kidney, and heart; Hg in all tissues; Mn in liver, muscle, and heart; Pb in kidney and muscle, and Zn in feathers, kidney, muscle, and heart between the bird species were statistically significant (P<0.05). In *Ac*, As, Pb, and Zn concentrations were higher in the muscle, Cd concentration was higher in the feathers, Cu and Hg concentrations were higher in the kidney, and Fe concentration was higher in the heart. In *An*, Cu and Pb concentrations were higher in the feathers, and Cd concentration was higher in the kidney. In *An*, Hg concentrations were higher in both feathers, muscle, and heart. In *Cc*, As concentration was higher in the feathers, Hg concentration was higher in the liver, Fe concentration was higher in the kidney, Cu, Mn, and Pb concentrations were higher in the heart.

Statistically significant ($P < 0.05$, $P < 0.01$) positive correlations were determined between total tissue or organ essential and non-essential metal concentrations in that: As and Cu, Fe, Mn, Zn; Cd and Cu, Fe; Hg and Cu, Fe, Zn; Pb and Cu, Fe, Mn, Zn (Table 2). Statistically significant ($P < 0.05$, $P < 0.01$) positive correlations were determined between non-essential metals and tissues in that: feathers and all other tissues; liver and feathers, muscle, heart; kidney and feathers, muscle; muscle and all other tissues; heart and feathers, liver, muscle for As; feathers and muscle; liver and kidney, muscle, heart; kidney and liver, muscle, heart; muscle and feathers, liver, kidney; heart and liver, kidney for Cd; feathers and muscle, heart; kidney and muscle, heart; muscle and feathers, kidney; heart and feathers, liver, kidney, muscle for Hg; feathers and kidney, muscle; liver and kidney, heart; kidney and other tissues; muscle and feathers, kidney; heart and liver, kidney for Pb concentration (Table 3).

4. Discussion

In contrast to essentials, non-essential metals like As, Cd, Hg, and Pb have no biological roles as well as cause toxic effects at lower levels in birds (Burger and Gochfeld, 2000; Sánchez-Virosta et al. 2015; Maia et al. 2017; Yamac et al. 2019; Çelik et al., 2021). Despite being a metalloid, As is frequently referred to as a metal or heavy metal. While As was once widely used as a medicine, a wood preservative, and a rodenticide, but is now banned or limited (Gruz et al. 2018). As causes reproductive, developmental, and behavioral problems in birds, as well as liver and brain damage, wing discoloration, laying delays, egg weight loss, shell thinning, and structural deterioration (Sánchez-Virosta et al. 2015; Celik et al. 2021). In the present study; the highest mean concentration of As in feathers was of *Cc* (2.85 mg kg^{-1}) and significantly higher than *An* (1.08 mg kg^{-1}) ($p < 0.05$). A similar result was reported by Mukhtar et al. (2020), who found that As concentration was higher in feathers than in bone, the heart, the liver, or the kidney of predators. Marinov et al. (2019) reported As concentrations between $1.63\text{--}3.12 \text{ mg kg}^{-1}$ in feather samples of *Cc*, similar to the current study. According to Gruz et al. (2018), the feather As concentration in *An* was the lowest similar in the current study. In another study, feather As (12.95 mg kg^{-1}) concentration in *An* was ~13 times higher than the current study (Nighat et al. 2013). On the other hand, it has been stated that the As concentration in birds is less than 1 mg kg^{-1} in uncontaminated environments (Naccari et al. 2009; Yamac et al. 2019). Maximum tolerable limit (MTL) for feathers As concentrations has been reported as 30 mg kg^{-1} (Gruz et al. 2018). Consequently, the present mean concentrations are significantly lower. In the current study, kidney concentration of *Ac* was significantly higher than the other species ($P < 0.05$). Perez-Lopez et al. (2008) reported a 3 times higher liver As concentration (1.86 mg kg^{-1}) in *Ac* than in the current study (0.52 mg kg^{-1}). In a study conducted in the surface waters of the region, As concentration (0.07 mg kg^{-1}) was reported as below the legal limit (Yipel et al., 2021).

Because of its long-term persistence and high toxicity, Cd is considered one of the most toxic non-essential element in the environment (Battaglia et al. 2005). Despite Cd accumulates at a lower concentration than Hg or Pb in the food chain, can cause developmental and behavioral problems, as well as renal and testicular damage, eggshell thinning in birds (Furness 1996, Burger and Gochfeld, 2000, Çelik et al., 2021). Thus the highest concentrations accumulate in the kidneys and causes damage in the renal tubules, Cd is frequently referred to as nephrotoxic (Kalisinska et al. 2006). As parallelly, in the current study, the highest mean Cd accumulation (1.13 mg kg^{-1}) occurred in the kidney. Similar to the present study, Naccari et al. (2009), Dzugan et al. (2012), and Lee et al. (2012) reported that the highest accumulation was found in the wild predator bird kidneys and Perez-Lopez et al. (2008) reported the lowest liver Cd concentrations in *An*. Compared to the

literature, although some were higher (Lee et al. 2012; Nighat et al. 2013), Cd concentrations in wild birds were generally lower than in the current study (Kenntner et al. 2001; Kalisinska et al. 2006; Naccari et al. 2009; Gruz et al. 2018). The liver (0.077 mg kg^{-1}) and kidney (0.145 mg kg^{-1}) concentration (w.w.) reported by Kennner et al. (2007) were lower than present results in *Ac*. Liver Cd concentrations (d.w.) in *An* were reported as 0.47 and 0.604 mg kg^{-1} (Perez-Lopez et al. 2008; Komosa et al. 2012). Feathers Cd concentrations in the *An* reported as low ($0.09\text{--}0.11 \text{ mg kg}^{-1}$) (Gruz et al. 2018; Gruz et al. 2019) and as high (11.96 mg kg^{-1}) (Nighat et al. 2013) than current study. The concentrations of present study is low than the maximum tolerable limit (MTL; 10 mg kg^{-1}) of poultry feathers (Gruz et al. 2018). The liver and kidney threshold values for Cd have been stated as 3 mg kg^{-1} d.w. (0.75 mg kg^{-1} w.w.) and 8 mg kg^{-1} d.w. (2 mg kg^{-1} w.w.), respectively, which point to an environmental increase (Scheuhammer, 1987, Kalisinska et al., 2006). On the other hand, the toxic concentration has been reported to be 100 mg kg^{-1} in birds (Kalisinska et al. 2006; Lee et al. 2012).

Clearly, hunting bullets are a major source of Pb exposure in wild birds (wetland birds and raptors) (Komosa et al. 2012). Pb, one of the most toxic metals, can cause growth, immune, reproductive, behavioral problems, hemolytic anemia, and low cell volume in birds (Maia et al. 2017; Celik et al. 2021). Lead tends to accumulate in bones, feathers, hair, and other calcium-containing tissues (Gruz et al. 2018). In the present study, the highest mean Pb concentration was in feathers (2.50 mg kg^{-1}). Battaglia et al. (2005) reported that Pb accumulates in high concentration in feathers and bone tissues of raptor species, similar to the current results. When Pb accumulation rates were evaluated in terms of tissues and bird species in the current study, the feathers of *An* had the highest mean Pb content, as expected. Although bone and feathers are the target organs for Pb in birds, the highest concentrations were detected in the muscle and heart tissues in *Ac* and *Cc* respectively. In studies; high Pb accumulation in organs other than the targets (such as muscle) in wild animals is attributed to hunting activities (Chiari et al. 2015). In muscle of *Ac* the mean Pb (2.66 mg kg^{-1}) concentration was significantly higher than in *An* (1.25 mg kg^{-1}) and *Cc* (0.57 mg kg^{-1}) ($P < 0.05$). In kidney of *Ac*, mean Pb concentration was significantly higher than in *An* ($P < 0.05$). The threshold value (mg kg^{-1} w.w.) for feathers, liver, and kidney are 4, 1.5, and 3 respectively for wild birds (Guitart et al. 1994, Burger and Gochfeld 2000; Solgi et al. 2020). The liver Pb mean concentration of *Ac* (1.70 mg kg^{-1}) exceeded the threshold value (1.5 mg kg^{-1}), suggesting a potential exposure. At exceeding the threshold values, non-essential metals replace with essentials and bind to proteins, resulting in cell activity loss (Mukhtar et al. 2020). Concentrations of 30 mg kg^{-1} d.w. (7.5 mg kg^{-1} w.w.) for liver and 20 mg kg^{-1} d.w. (5 mg kg^{-1} w.w.) for kidney have been related to acute Pb poisoning in birds (Kalinska et al., 2006).

Due to Hg accumulating faster than excretion and being able to biomagnify, can reach higher concentrations in the bodies of animals at higher trophic concentrations (Gruz et al. 2019; Yamac et al. 2019). In birds, Hg causes reproductive problems, eggshell thinning, embryotoxicity, as well as impaired coordination, walking, and flying performance, weight loss, paralysis, and death (Yamac et al. 2019; Celik et al. 2021). In the present study, the highest mean Hg was found in a raptor, *An* (excluding liver and kidney). Hg concentrations of *An* were found to be significantly higher than *Ac* in feathers, muscle, and heart tissues ($P < 0.05$). Durmus (2018) has reported that although the Hg level was higher in carnivorous bird species depending on diet, it was lower in *Ac* (0.002 mg kg^{-1}), a raptor species than in herbivorous birds. According to Zolfaghari et al. (2007), the highest Hg concentration was in *An* (1.03 mg kg^{-1}), which is approximately 8 times higher than the present study (0.13 mg kg^{-1}). Similarly, according to Gruz et al. (2018), the Hg concentration of *An* in feathers was reported to be 21 times higher (2.72

mg kg⁻¹) than in the present study. While the MTL for Hg in feathers is 2 mg kg⁻¹, Hg concentrations of 5 mg kg⁻¹ have been observed to cause sublethal and reproductive abnormalities (such as reduced egg size, hatching rate, and chick survival rate) (Rothschild and Duffy 2005; Zolfaghari et al. 2007). According to Jackson et al. (2011), 2.4 mg kg⁻¹ Hg concentrations in body feathers and 3 mg kg⁻¹ Hg concentrations in feathers cause breeding processes to deteriorate (a 10% reduction in hatching success). According to Connell et al. (2002), the threshold concentration of Hg-related adverse effects in feathers is 3-5 mg kg⁻¹, with no adverse effects detected in accumulations below this concentration (3 mg kg⁻¹). Feathers Hg concentrations in the present study are considerably below these limits and pose no risk to birds. Kenntner et al. (2007) found that the liver and kidney Hg concentrations of *Ac* were 0.024 and 0.016 mg kg⁻¹ w.w., respectively, which is lower than the present study. Plessl et al. (2017) reported that the liver and muscle Hg concentrations in a waterfowl species were 0.17 and 0.05 mg kg⁻¹ w.w., respectively. Similarly, the mean muscle Hg in waterfowl birds was 0.055 mg kg⁻¹ w.w. was reported by Rothschild and Duffy (2005). A previous study reported that the liver Hg concentrations in range from 1 to 10 mg kg⁻¹ (Lucia et al. 2010). The liver Hg concentrations higher than 2 mg kg⁻¹ w.w. have been associated with adverse effects on the reproductive system, and higher than 20 mg kg⁻¹ w.w. have been associated with mortality (Hurnikova et al. 2021). Since the Hg concentrations are below these toxic limits in the current study, there is no risk of Hg toxicity in the study area.

Whether essential metals like Cu, Fe, Mn, and Zn are necessary for biological processes (enzymatic processes, energy metabolism, hemoglobin formation, antioxidant defense, growth, feather formation, reproduction, etc.), exposure above tolerable physiological limits can be toxic to the kidneys and reproduction system in birds (Gruz et al. 2018; Celik et al. 2021). Feathers, kidney and muscle Cu concentrations of *An* had significantly higher than *Ac* ($P < 0.05$). Similar, studies have reported that *Ac* has the highest Cu accumulation in feathers of all bird species (Gruz et al. 2018, Nighat et al. 2013). Feather Cu concentrations were reported to be 28.25 mg kg⁻¹ in *An* by Nighat et al. (2013). Similar results were obtained in the current study in *An* (21.58 mg kg⁻¹) and *Ac* (13.18 mg kg⁻¹). However, Gruz et al. (2018) revealed that feathers Cu concentrations (65.45 mg kg⁻¹) were three times higher in *An* than in the present. According to Komosa et al. (2012), the Cu concentration in liver of *An* was reported 14.95 mg kg⁻¹ d.w. In the present study, Cu concentration was 10.24 mg kg⁻¹ w.w. in liver of *An*. Fe concentrations in liver, heart and Mn concentrations in liver, muscle, and heart of *An* were significantly lower than *Ac* ($P < 0.05$). Fe was the most accumulated metal in tissues. Kalinska et al. (2006) reported the liver approximately 2 times higher Fe concentration (559 mg kg⁻¹ w.w.) in White-tailed Eagles than the current study. Lee et al. (2012) reported similar liver Fe concentrations (313-339 mg kg⁻¹ w.w.) in waterfowl. Lower (Kaliniska et al. 2006; Komosa et al. 2012; Lee et al. 2012) and higher (Van Wyk et al. 2001; Naccari et al. 2009) Mn concentrations have been reported for various bird species. Zn is a essential trace element for the activity of many enzymes in the body, but too much of it might cause an additional source of stress in birds who are already under stress (Perez-Lopez et al. 2008). The feathers, kidney, and muscle Zn concentrations of *Ac* than that of *Cc*, and the muscle and heart Zn concentrations of *Ac* that of *An* were found to be significantly higher in the current study ($P < 0.05$). In terms of Zn, there was no difference among species in the liver tissue. Compared to the literature, the mean Zn concentration in the feathers of the current study is lower (Van Wyk et al. 2001; Naccari et al. 2009; Perez-Lopez et al. 2008; Komosa et al. 2012; Nighat et al. 2013; Gruz et al. 2018). The reason for this is that the tissues in the current study were weighed wet, and the measured Zn concentrations in the tissues are essential for healthy tissue growth or metabolism, so it could be attributed to endogenous accumulation rather than contamination from the environment. Threshold values of soft tissue were reported as

>200 mg kg⁻¹ for Zn, >2000 mg kg⁻¹ for Fe, >9 mg kg⁻¹ for Mn, and >50 mg kg⁻¹ for Cu (Lee et al. 2012), while feather MTL values were reported as 250 mg kg⁻¹ for Cu and 500 mg kg⁻¹ for Zn (Gruz et al. 2018). As a result, Cu, Fe, and Zn concentrations in all bird species are generally below the limits, indicating no risk yet. However, Mn concentrations are close to the threshold value (>9 mg kg⁻¹) in the current study, which may pose a minor risk of damage and intoxication.

The ecosystem is seriously threatened by metal and metalloid pollution, which also causes a number of pathologies in wildlife. In addition to being present naturally, these elements are also widely dispersed in the environment as a result of anthropogenic sources (Lucia et al. 2010). Birds are useful environmental contamination indicators due to their wide geographic distribution, ability to feed at various trophic concentrations, and long lifespans. Being at the top of the food chain and exhibiting territorial and non-migratory behaviors may lead to increased toxic metal accumulation in raptor bird bodies and eggs. As a result, these organisms are excellent bioindicators for detecting the heavy metal stress on the environment (Sánchez-Virosta et al. 2015, Gruz et al. 2019). Metal concentrations in tissues of various wild bird species determined in other studies are presented in Table 4.

5. Conclusions

In conclusion, this study is the first report of the target organ and tissue (feathers, liver, kidney, heart, and muscle) concentrations and correlations of essential and non-essential metals of three wild bird species (*Aquila chrysaetos*, *Accipiter nisus*, and *Ciconia ciconia*) and of the study region (Hayat city, south Turkey). Compare to the other studies; Cu, Hg, Pb, and Zn concentrations were generally low, Cd, Fe, and Mn concentrations were high. According to these results, the concentration of Mn may be an early warning for impairment of environmental quality and may occur biological stress on wildlife. To summarize, further studies are needed to understand the harmful effects of heavy metals on physiological and behavioral alterations, as well as immunological and reproductive concerns in wild birds at the individual, population (based on effects on target systems, tissues, and organs), and ecosystem concentrations. Determining the effects of environmental pollution at the organism concentration (molecular, cellular, tissue, organ, and system) in indicator wild animals, like birds, is also important for predicting potential problems for humans and other living organisms at the population, community, or ecosystem (terrestrial and marine) concentration. As a result, regular pollutant biomonitoring on indicator wild animals, like birds, is an ecotoxicological crucial strategy to keep the regional pollution concentrations under control.

Declarations

Acknowledgments

Acknowledgments to the Hatay Provincial Directorate of Nature Conservation and National Parks of Turkey for their valuable collaboration.

Author contribution Mustafa Yipel: Concept, Design, Data Collection or Processing, Analysis or Interpretation, Literature Search, Writing. İbrahim Ozan TEKELİ: Literature Search, Writing. Cafer Tayer İŞLER: Sample Collection or Processing Muhammed Enes ALTUĞ: Sample Collection or Processing

Funding No funding was received for the study

Data availability Not Applicable

Declarations

Ethical approval Not Applicable

Consent to participate Not Applicable

Consent for publication Not Applicable

Competing interests The authors declare no competing interests

References

- Battaglia, A., Ghidini, S., Campanini, G., Spaggiari, R., 2005. Heavy metal contamination in little owl (*Athene noctua*) and common buzzard (*Buteo buteo*) from northern Italy. *Ecotoxicol. Environ. Saf.* 60(1), 61–66. <https://doi.org/10.1016/j.ecoenv.2003.12.019>
- Bhat, S., 2013. Ecotoxicology & Impact on biodiversity. *J. pharmacogn. phytochem.* 2(2): 1-19.
- Bjedov, D., Velki, M., Lackmann, C., Begović, L., Mikuška, T., Jurinović, L., Mikuška, A., 2022. Blood biomarkers in white stork (*Ciconia ciconia*) nestlings show different responses in several areas of Croatia. *J. Exp. Zool. A: Ecol. Integr. Physiol.* <https://doi.org/10.1002/jez.2588>.
- Boudou, A., Ribeyre, F., 1997. Aquatic ecotoxicology: from the ecosystem to the cellular and molecular concentrations. *Environ. Health Perspect.* 105, 21-35.
- Burger, J., Gochfeld, M., 2000. Metal concentrations in feathers of 12 species of seabirds from Midway Atoll in the northern Pacific Ocean. *Sci. Total. Environ.* 257(1), 37–52. [https://doi.org/10.1016/S0048-9697\(00\)00496-4](https://doi.org/10.1016/S0048-9697(00)00496-4).
- Celik, E., Durmus, A., Adizel, O., Uyar, H.N., 2021. A bibliometric analysis: what do we know about metals (loids) accumulation in wild birds?. *Environ. Sci. Pollut. Res.* 28(8), 10302-10334. <https://doi.org/10.1007/s11356-021-12344-8>.
- Chiari, M., Cortinovis, C., Bertolotti, M., Alborali, L., Zanoni, M., Ferretti, E., Caloni, F., 2015. Lead, cadmium and organochlorine pesticide residues in hunted red deer and wild boar from northern Italy. *Food Addit. Contam. Part A.* 32(11), 1867-1874. <https://doi.org/10.1080/19440049.2015.1087058>.
- Connell, D.W., Wong, B.S.F., Lam, P.K.S., Poon, K.F., Lam, M.H.W., Wu, R.S.S., Yen, Y.F., 2002. Risk to breeding success of Ardeids by contaminants in Hong Kong: evidence from trace metals in feathers. *Ecotoxicology.* 11(1), 49-59. <https://doi.org/10.1023/A:1013745113901>.
- Dauwe, T., Bervoets, L., Pinxten, R., Blust, R., Eens, M., 2003. Variation of heavy metals within and among feathers of birds of prey: Effects of molt and external contamination. *Environ Pollut.* 124(3), 429-436. [https://doi.org/10.1016/S0269-7491\(03\)00044-7](https://doi.org/10.1016/S0269-7491(03)00044-7).

Durmuş, A., 2018. The Mercury (Hg) concentrations in feathers of wild birds. Appl. Ecol. Environ. Res. 16(3), 2973-2981. http://dx.doi.org/10.15666/aeer/1603_29732981.

Džugan, M., Zielińska, S., Hęclik, J., Szostek, M.P., 2012. Evaluation of heavy metals environmental contamination based on their concentrations in tissues of wild pheasant (*Phasianus colchicus* L.). J. microbiol., biotechnol. food sci. 2(1), 238-245.

Furness, R.W., 1996. Cadmium in birds: Nelson Beyer, W, Heinz, GH, Redmon-Norwood, AW (Eds.), Environ Contaminants in Wild Life Interpreting Tissue Concentrations. CRC Press, Boca Raton, pp 389–404.

Grúz, A., Déri, J., Szemerédy, G., Szabó, K., Kormos, É., Bartha, A., Budai, P., 2018. Monitoring of heavy metal burden in wild birds at eastern/north-eastern part of Hungary. Environ. Sci. Pollut. Res. 25(7), 6378-6386. <https://doi.org/10.1007/s11356-017-1004-0>.

Grúz, A., Mackle, O., Bartha, A., Szabó, R., Déri, J., Budai, P., Lehel, J., 2019. Biomonitoring of toxic metals in feathers of predatory birds from eastern regions of Hungary. Environ. Sci. Pollut. Res. 26(25), 26324-26331. <https://doi.org/10.1007/s11356-019-05723-9>.

Guitart, R., To-Figueras, J., Mateo, R., Bertolero, A., Cerradelo, S., Martínez-Vilalta, A., 1994. Lead poisoning in waterfowl from the Ebro Delta, Spain: calculation of lead exposure thresholds for mallards. Arch. Environ. Contam. Toxicol. 27(3), 289-293. <https://doi.org/10.1007/BF00213161>.

Hervías, S.P., González, Y.G., Pereira, E.M., Vulcano, A., Cabral, R.C., Coelho, N.G., Delgado, G.C., 2017. The Eurasian Sparrowhawk of macaronesia (*Accipiter nisus granti*): nesting territories, phenology, and breeding success on Madeira Island, Portugal. J. Raptor. Res. 51(1), 15-24. <https://doi.org/10.3356/JRR-16-00003.1>.

Hurníková, Z., Komorová, P., Šalamún, P., Miklisová, D., Chovancová, G., Miterpáková, M., 2021. Concentration of trace elements in raptors from three regions of Slovakia, Central Europe. Pol. J. Environ. Stud. 30(6), 5577-5591. <https://doi.org/10.15244/pjoes/137329>.

Jackson, A.K., Evers, D.C., Etterson, M.A., Condon, A.M., Folsom, S.B., Detweiler, J., Cristol, D.A., 2011. Mercury exposure affects the reproductive success of a free-living terrestrial songbird, the Carolina Wren (*Thryothorus ludovicianus*). The Auk. 128(4), 759-769. <https://doi.org/10.1525/auk.2011.11106>.

Kalińska, E., Salicki, W., Jackowski, A., 2006. Six Trace Metals in White-Tailed Eagle from Northwestern Poland. Pol. J. Environ. Stud. 15(5).

Kenntner, N., Crettenand, Y., Fünfstück, H.J., Janovsky, M., Tataruch, F., 2007. Lead poisoning and heavy metal exposure of golden eagles (*Aquila chrysaetos*) from the European Alps. J. Ornithol. 148(2), 173-177. <https://doi.org/10.1007/s10336-006-0115-z>.

Kenntner, N., Tataruch, F., Krone, O., 2001. Heavy metals in soft tissue of white-tailed eagles found dead or moribund in Germany and Austria from 1993 to 2000. Environ. Toxicol. Chem: An International Journal. 20(8), 1831-1837. <https://doi.org/10.1002/etc.5620200829>.

- Komosa, A., Kitowski, I., Komosa, Z., 2012. Essential trace (Zn, Cu, Mn) and toxic (Cd, Pb, Cr) elements in the liver of birds from Eastern Poland. *Acta Vet.* 62, 579-589. <https://doi.org/10.2298/AVB1206579K>.
- Lee, D.P., Lee, K.G., Nam, D.H. 2012. Population declines and heavy metal exposure of Swinhoe's Storm Petrels (*Oceanodroma monorhis*) breeding on the southwest coast of Korea. *Mar. Pollut. Bull.* 64(12), 2645-2649. <https://doi.org/10.1016/j.marpolbul.2012.10.018>.
- List of Migratory Birds. <https://www.gpo.gov/fdsys/granule/CFR-2000-title50-vol1/CFR-2000-title50-vol1-sec10-13>
- Lucia, M., André, J.M., Gontier, K., Diot, N., Veiga, J., Davail, S., 2010. Trace element concentrations (mercury, cadmium, copper, zinc, lead, aluminium, nickel, arsenic, and selenium) in some aquatic birds of the Southwest Atlantic Coast of France. *Arch. Environ. Contam.* 58(3), 844-853. <https://doi.org/10.1007/s00244-009-9393-9>.
- Maia, A.R., Soler-Rodriguez, F., Pérez-López, M., 2017. Concentration of 12 metals and metalloids in the blood of white stork (*Ciconia ciconia*): Basal values and influence of age and gender. *Arch. Environ. Contam.* 73(4), 522-532. <https://doi.org/10.1007/s00244-017-0431-8>.
- Marinov, M., Burada, A., DoroşEncu, A., Alexe, V., Teodorof, L., Tudor, M., 2019. Report on the accumulation of heavy metals in the feathers of some wetland birds in the Danube delta (Romania). *Rom J Biol-Zool.* 64, 73-84.
- Meyer, J., Jaspers, V.L., Eens, M., de Coen, W., 2009. The relationship between perfluorinated chemical concentrations in the feathers and livers of birds from different trophic concentrations. *Sci. Total Environ.* 407(22), 5894-5900. <https://doi.org/10.1016/j.scitotenv.2009.07.032>.
- Mukhtar, H., Chan, C.Y., Lin, Y.P., Lin, C.M., 2020. Associations and predictabilities of heavy metals in avian organs, feathers, and bone using crowdsourced samples. *Chemosphere*, 252, 126583. <https://doi.org/10.1016/j.chemosphere.2020.126583>.
- Naccari, C., Cristani, M., Cimino, F., Arcoraci, T., Trombetta, D., 2009. Common buzzards (*Buteo buteo*) bio-indicators of heavy metals pollution in Sicily (Italy). *Environ. Int.* 35(3), 594-598. <https://doi.org/10.1016/j.envint.2008.11.002>.
- Nighat, S., Iqbal, S., Nadeem, M.S., Mahmood, T., Shah, S.I., 2013. Estimation of heavy metal residues from the feathers of *Falconidae*, *Accipitridae*, and *Strigidae* in Punjab, Pakistan. *Turk. J. Zool.* 37(4), 488-500.
- Nikinmaa, M., 2014. *An Introduction to Aquatic Toxicology*. Academic Press, Elsevier, USA.
- Pérez-López, M., de Mendoza, M.H., Beceiro, A.L., Rodríguez, F.S., 2008. Heavy metal (Cd, Pb, Zn) and metalloid (As) content in raptor species from Galicia (NW Spain). *Ecotoxicol. Environ. Saf.* 70(1), 154-162. <https://doi.org/10.1016/j.ecoenv.2007.04.016>
- Plessl, C., Jandrisits, P., Krachler, R., Keppler, B.K., Jirsa, F., 2017. Heavy metals in the mallard *Anas platyrhynchos* from eastern Austria. *Sci. Total Environ.* 580, 670-676. <https://doi.org/10.1016/j.scitotenv.2016.12.013>.

- Rothschild, R.F., Duffy, L.K., 2005. Mercury concentrations in muscle, brain and bone of Western Alaskan water fowl. *Sci. Total Environ.* 349(1-3), 277-283. <https://doi.org/10.1016/j.scitotenv.2005.05.021>.
- Salamat, N., Etemadi-Deylami, E., Movahedinia, A., Mohammadi, Y., 2014. Heavy metals in selected tissues and histopathological changes in liver and kidney of common moorhen (*Gallinula chloropus*) from Anzali Wetland, the south Caspian Sea, Iran. *Ecotoxicol. Environ. Saf.* 110, 298-307. <https://doi.org/10.1016/j.ecoenv.2014.09.011>.
- Sánchez-Virosta, P., Espín, S., García-Fernández, A.J., Eeva, T., 2015. A review on exposure and effects of arsenic in passerine birds. *Sci. Total Environ.* 512, 506-525. <https://doi.org/10.1016/j.scitotenv.2015.01.069>.
- Scheuhammer, A.M., 1987. The chronic toxicity of aluminium, cadmium, mercury, and lead in birds: a review. *Environ. Pollut.* 46(4), 263-295. [https://doi.org/10.1016/0269-7491\(87\)90173-4](https://doi.org/10.1016/0269-7491(87)90173-4).
- Solgi, E., Mirzaei-Rajeouni, E., Zamani, A., 2020. Feathers of Three Waterfowl Bird Species from Northern Iran for Heavy Metals Biomonitoring. *Bull. Environ. Contam. Toxicol.* 104(6), 727-732. <https://doi.org/10.1007/s00128-020-02852-7>.
- Squadrone, S., Abete, M. C., Brizio, P., Pessani, D., Favaro, L., 2018. Metals in feathers of African Penguins (*Spheniscus demersus*): Considerations for the welfare and management of seabirds under human care. *Bull. Environ. Contam. Toxicol.* 100(4), 465-471. <https://doi.org/10.1007/s00128-018-2293-9>.
- Tekeli, I.O., Yipel, M., Sengul, S.A., Sakin, F., 2021. Concentrations of metals and organochlorine pesticides in kidney, liver, and muscle tissues of wild boars (*Sus scrofa*) from Hatay Province, Eastern Mediterranean Region, Turkey. *Bull. Environ. Contam. Toxicol.* 106(2), 257-263. <https://doi.org/10.1007/s00128-020-03072-9>.
- Thornton, M., Todd, I., Roos, S., 2017. Breeding success and productivity of urban and rural Eurasian Sparrow hawks *Accipiter nisus* in Scotland. *Écoscience.* 24(3-4), 115-126. <https://doi.org/10.1080/11956860.2017.1374322>.
- Van Wyk, E., Van der Bank, F.H., Verdoorn, G.H., Hofmann, D., 2001. Selected mineral and heavy metal concentrations in blood and tissues of vultures in different regions of South Africa. *S. Afr. J. Anim. Sci.* 31(2), 57-64. <https://doi.org/10.4314/sajas.v31i2.3831>.
- Weis, J.S., 2015. Marine pollution: what everyone needs to know. What Everyone Needs to Know (R). Oxford University Press. USA.
- Wiens, J.D., Bloom, P.H., Madden, M.C., Kolar, P.S., Tracey, J.A., Fisher, R.N., 2022. Golden Eagle Occupancy Surveys and Monitoring Strategy in Coastal Southern California, United States. *Front. Ecol. Evol.* 9:665792. <https://doi.org/10.3389/fevo.2021.665792>.
- Yamac, E., Ozden, M., Kirazli, C., Malkoc, S., 2019. Heavy-metal concentrations in feathers of cinereous vulture (*Aegypius monachus* L.) as an endangered species in Turkey. *Environ. Sci. Pollut. Res.* 26(1), 833-843. <https://doi.org/10.1007/s11356-018-3649-8>

Yipel, M., Tekeli, I.O., Dikmen, B., Yarsan, E., 2021. Distribution and ecotoxicological risk assessment of heavy metals in streams of Amanos Mountains from Southern Turkey. Bull. Environ. Contam. Toxicol. 107(5), 895-903. <https://doi.org/10.1007/s00128-021-03316-2>.

Yipel, M., Tekeli, I.O., 2016. Potential toxicological risks of industrial heavy metals on wildlife ecology: a review to draw attention to an important ecological region. In International Conference on Advanced Materials and Systems (ICAMS). The National Research & Development Institute for Textiles and Leather-INCDTP. 309-313.

Zolfaghari, G., Esmaili-Sari, A., Ghasempouri, S.M., Kiabi, B H., 2007. Examination of mercury concentration in the feathers of 18 species of birds in southwest Iran. Environ. Res. 104(2), 258-265. <https://doi.org/10.1016/j.envres.2006.12.002>.

Tables

Table 1. Metal concentrations (mean±SE, mg kg⁻¹) in feathers, muscle, heart, kidney, and liver tissues of *Aquila chrysaetos* (Ac) (n:9), *Accipiter nisus* (An) (n:12), and *Ciconia ciconia* (Cc) (n:7).

Metal	Species	Feather	Liver	Kidney	Muscle	Heart
As	<i>Ac</i>	1.51±0.28 ^{ab}	0.62±0.19 ^{ab}	1.41±0.31 ^a	1.92±0.22 ^a	1.86±0.67 ^{ab}
	<i>An</i>	1.08±0.13 ^a	0.52±0.13 ^a	0.40±0.10 ^b	1.21±0.19 ^{ab}	0.73±0.12 ^a
	<i>Cc</i>	2.85±0.33 ^b	1.02±0.09 ^b	0.34±0.02 ^b	0.35±0.10 ^b	2.10±0.24 ^b
	<i>p</i> value	0.01	0.01	0.04	0.00	0.01
Total		1.66±0.19	0.68±0.09	0.71±0.14	1.22±0.16	1.43±0.25
(min.-max.)		(<LOD-4.32)	(<LOD-1.71)	(<LOD-3.16)	(<LOD-0.78)	(<LOD-6.40)
Cd	<i>Ac</i>	1.16±0.07	1.11±0.05 ^a	1.06±0.03 ^a	1.05±0.12	1.08±0.16
	<i>An</i>	0.95±0.09	0.79±0.10 ^b	1.19±0.06 ^b	0.96±0.204	0.93±0.03
	<i>Cc</i>	1.02±0.09	1.04±0.11 ^{ab}	1.11±0.07 ^{ab}	1.08±0.05	1.04±0.11
	<i>p</i> value	<i>NS</i>	0.02	0.04	<i>NS</i>	<i>NS</i>
Total		1.04±0.05	0.96±0.06	1.13±0.03	1.02±0.04	1.00±0.06
(min.-max.)		(0.50- 1.66)	(0.30-1.67)	(0.71-1.46)	(0.38-1.75)	(<LOD-1.78)
Cu	<i>Ac</i>	13.18±1.39 ^a	14.83±3.95	24.20±4.88 ^a	17.47±2.74 ^a	11.07±2.17
	<i>An</i>	21.58±1.81 ^b	10.24±0.60	10.28±1.19 ^b	11.78±0.81 ^b	14.11±1.65
	<i>Cc</i>	11.89± 1.09 ^a	11.35±1.11	10.44±1.07 ^b	12.22 ± 1.40 ^{ab}	14.63±1.57
	<i>p</i> value	0.01	<i>NS</i>	0.00	0.03	<i>NS</i>
Total		16.45±1.25	11.99±1.33	14.80±2.03	13.72±1.09	13.26±1.07
(min.-max.)		(7.48-34.70)	(2,07-43.50)	(4.50-60.60)	(5.99-37.10)	(1.33-30.40)
Fe	<i>Ac</i>	352.92±13.82	344.08±10.25 ^a	337.17±8.15 ^a	310.83±21.45	358.58±11.07 ^a
	<i>An</i>	310.58±11.53	275.42±13.85 ^b	350.50±6.65 ^{ab}	324.42±7.57	314.58±4.98 ^b
	<i>Cc</i>	323.00±17.11	312.05±14.75 ^{ab}	367.66±9.60 ^b	304.83±14.88	338.67±14.06 ^{ab}
	<i>p</i> value	<i>NS</i>	0.00	0.04	<i>NS</i>	0.00
Total		327.29±8.37	306.65±9.38	350.50±4.90	315.16±8.29	334.75±6.35
(min.-max.)		(234.50-435.50)	(164.50-404.00)	(297.50-417.50)	(164.50-390.00)	(279.00-416.00)
Hg	<i>Ac</i>	0.04±0.01 ^a	0.05±0.02 ^a	0.12±0.03 ^a	0.06±0.02 ^a	0.04±0.01 ^a
	<i>An</i>	0.13±0.01 ^b	0.09±0.01 ^{ab}	0.11±0.01 ^a	0.13±0.02 ^b	0.13±0.01 ^b
	<i>Cc</i>	0.07±0.01 ^a	0.11±0.03 ^b	<LOD	0.10±0.01 ^{ab}	0.09 ± 0.02 ^{ab}

	<i>p</i> value	0.00	0.02	0.01	0.01	0.00
Total		0.09±0.01	0.08±0.01	0.08±0.01	0.10±0.01	0.09±0.01
(min.-max.)		(<LOD-0.22)	(<LOD-0.21)	(<LOD-0.23)	(<LOD-0.11)	(<LOD-0.21)
Mn	<i>Ac</i>	10.44±0.85	10.51±0.69 ^a	11.55±0.79	11.03±1.00 ^a	11.86±1.48 ^a
	<i>An</i>	9.40±0.82	7.71±0.72 ^b	11.36±0.55	8.94±0.48 ^b	8.38±0.37 ^b
	<i>Cc</i>	11.55±1.22	10.30±1.22 ^{ab}	11.40±1.09	10.11±0.45 ^{ab}	12.59±1.21 ^a
	<i>p</i> value	<i>NS</i>	0.01	<i>NS</i>	0.02	0.01
Total		10.27±0.54	9.25±0.54	11.43±0.42	9.91±0.42	10.55±0.67
(min.-max.)		(3.73-16.56)	(3.77-15.30)	(6.00-16.10)	(4.36-14.55)	(5.90-22.60)
Pb	<i>Ac</i>	2.23±0.44	1.70±0.50	1.67±0.29 ^a	2.66±0.35 ^a	1.28±0.60
	<i>An</i>	3.31±1.47	1.40±0.21	0.96±0.11 ^b	1.25±0.12 ^b	1.39±0.19
	<i>Cc</i>	1.44±0.23	1.45±0.24	1.04±0.07 ^{ab}	0.57±0.04 ^b	1.66±0.24
	<i>p</i> value	<i>NS</i>	<i>NS</i>	0.02	0.00	<i>NS</i>
Total		2.50±0.65	1.51±0.19	1.21±0.12	1.53±0.20	1.42±0.21
(min.-max.)		(0.44-19.20)	(<LOD-5.07)	(<LOD-2.96)	(0.33-4.76)	(<LOD-5.80)
Zn	<i>Ac</i>	20.85±1.08 ^a	18.57±2.05	19.00±1.32 ^a	24.55±1.91 ^a	21.48±2.17 ^a
	<i>An</i>	17.12±1.49 ^{ab}	13.08±1.00	18.13±1.02 ^{ab}	15.29±0.89 ^b	15.08±1.24 ^b
	<i>Cc</i>	14.20±0.49 ^b	16.71±4.75	15.66±1.05 ^b	13.68±0.89 ^b	16.75±1.24 ^{ab}
	<i>p</i> value	0.01	<i>NS</i>	0.04	0.00	0.02
Total		17.59±0.87	15.75±5.60	17.79±0.69	17.87±1.15	17.55±1.04
(min.-max.)		(8.89-30.40)	(6.70-32.30)	(8.54-26.00)	(7.56-32.20)	(8.08-31.20)

^{a,b,c} Means within the same column with different letters are statistically significant ($p < 0.05$)

LOD: Limit of detection

NS: not significant

Table 2. Correlation between essential and non-essential metal concentrations in total tissues of *Aquila chrysaetos* (*Ac*) (n:9), *Accipiter nisus* (*An*) (n:12), and *Ciconia ciconia* (*Cc*) (n:7).

Metal	Cu	Fe	Mn	Zn
As	.568**	.762**	.537**	.467**
Cd	.374**	.381**	.041	.080
Hg	.294**	.194*	-.059	.355**
Pb	.498**	.582**	.418**	.314**

* (p<0.05), ** (p<0.01)

Table 3. Correlation between non-essential metals and tissue concentrations in tissues of *Aquila chrysaetos* (Ac) (n:9), *Accipiter nisus* (An) (n:12) , and *Ciconia ciconia* (Cc) (n:7).

Metal	Tissue	Feather	Liver	Kidney	Muscle	Hearth
As	Feather	-	.604**	.610**	.947**	.614**
	Liver	.604**	-	.288	.640**	.428*
	Kidney	.610**	.288	-	.653**	.020
	Muscle	.947**	.640**	.653**	-	.573**
	Hearth	.614**	.428*	.020	.573**	-
Cd	Feather	-	.322	.314	.777**	-.017
	Liver	.322	-	.659**	.441*	.660**
	Kidney	.314	.659**	-	.378*	.490**
	Muscle	.777**	.441*	.378*	-	.196
	Hearth	-.017	.660**	.490**	.196	-
Hg	Feather	-	-.040	.036	.405*	.482**
	Liver	-.040	-	-.279	-.303	-.296
	Kidney	.036	-.279	-	.384*	.461*
	Muscle	.405*	-.303	.384*	-	.619**
	Hearth	.482**	-.296	.461*	.619**	-
Ni	Feather	-	.804**	.456*	.993**	.352
	Liver	.804**	-	.514**	.784**	.203
	Kidney	.456*	.514**	-	.423*	-.034
	Muscle	.993**	.784**	.423*	-	.343
	Hearth	.352	.203	-.034	.343	-
Pb	Feather	-	.175	.604**	.610**	.143
	Liver	.175	-	.417*	.295	.888**
	Kidney	.604**	.417*	-	.893**	.485**
	Muscle	.610**	.295	.893**	-	.223
	Hearth	.143	.888**	.485**	.223	-

* (p<0.05), ** (p<0.01)

Table 4. Metal concentrations in tissues of *Aquila chrysaetos* (Ac) (n:9), *Accipiter nisus* (An) (n:12) , and *Ciconia ciconia* (Cc), and other bird species (O) in different studies

		Species, Area	Feather	Liver	Kidney	Muscle	Heart	Literature
As	mg kg ⁻¹	An, TR	1.08	0.52	0.40	1.21	0.73	Present Study
	mg kg ⁻¹	An, HU	<LOD	-	-	-	-	Gruz et al., 2018
	µg g ⁻¹	An, PK	12.95	-	-	-	-	Nighat et al., 2013
	ppm	An, ES	-	1.863	-	-	-	Perez-Lopez et al., 2008
	µg g ⁻¹	O, IT	0.48	1.31	0.23	1.02	-	Naccari et al. 2009
Cd	mg kg ⁻¹	Ac, TR	1.16	1.11	1.06	1.05	1.08	Present Study
		An, TR	0.95	0.79	1.19	0.96	0.93	
	mg kg ⁻¹	Ac, AT, DE, CH	-	0.077	0.145	-	-	Kenntner et al. 2007
	mg kg ⁻¹	An, PL	-	0.47	-	-	-	Komosa et al., 2012
	mg kg ⁻¹	An, HU	0.09	-	-	-	-	Gruz et al., 2018
	µg g ⁻¹	An, PK	11.96	-	-	-	-	Nighat et al., 2013
	ppm	An, ES	-	0.604	-	-	-	Perez-Lopez et al., 2008
	mg kg ⁻¹	O, IR	-	1.90	-	1.74	-	N. Salamat et. al., 2014
	ppm	O, AT, DE	-	0.039	0.161	-	-	Kenntner et al. 2001
	µg kg ⁻¹	O, KR	-	3.50	19.1	-	-	D.-P. Lee et al., 2012
	µg g ⁻¹	O, IT	0.11	0.21	0.55	0.09	-	Naccari et al., 2009
	µg g ⁻¹	O, PL	-	0.042	0.346	0.017	-	Kalisińska et al., 2006
Cu	mg kg ⁻¹	An, TR	21.58	10.24	10.28	11.78	14.11	Present Study
	mg kg ⁻¹	An, PL	-	14.95	-	-	-	Komosa et al., 2012
	µg g ⁻¹	An, PK	28.25	-	-	-	-	Nighat et al., 2013
	mg kg ⁻¹	An, HU	65.45	-	-	-	-	Gruz et al., 2018

1								
	mg kg ⁻¹ ₁	O, IR	-	24.07	-	27.69	-	N. Salamat et. al., 2014
	µg kg ⁻¹	O, KR	-	2.16	1.62	-	-	D.-P. Lee et al., 2012
	µg g ⁻¹	O, SAF	-	31.85	14.90	18.23	11.70	van Wyk et al., 2001
	µg g ⁻¹	O, IT	9.24	39.51	9.61	33.49	-	Naccari et al., 2009
	µg g ⁻¹	O, PL	-	4.22	4.37	2.86	-	Kalisińska et al., 2006
Fe	mg kg ⁻¹ ₁	Ac, TR	352.92	344.08	337.17	310.83	358.58	Present Study
		An, TR	310.58	275.42	350.50	324.42	314.58	
		Cc, TR	323.00	312.05	367.66	304.83	338.67	
	µg kg ⁻¹	O, KR	-	339	157	-	-	D.-P. Lee et al., 2012
	µg g ⁻¹	O, SAF	-	227.82	384.38	293.80	538.46	van Wyk et al., 2001
	µg g ⁻¹	O, PL	-	559	174.5	66.30	-	Kalisińska et al., 2006
Hg	mg kg ⁻¹ ₁	Ac, TR	0.04	0.05	0.12	0.06	0.04	Present Study
		An, TR	0.13	0.09	0.11	0.13	0.13	
		Cc, TR	0.07	0.11	<LOD	0.10	0.09	
	mg kg ⁻¹ ₁	Ac, TR	0.002	-	-	-	-	Durmuş A., 2018
	mg kg ⁻¹ ₁	Ac, AT, DE, CH	-	0.024	0.016	-	-	Kenntner et al. 2007
	mg kg ⁻¹ ₁	An, HU	2.72	-	-	-	-	Gruz et al., 2018
	mg kg ⁻¹ ₁	An, IR	1.03	-	-	-	-	Zolfaghari et al., 2007
	mg kg ⁻¹ ₁	Cc, TR	0.012	-	-	-	-	Durmuş A., 2018
	mg kg ⁻¹ ₁	O,AT	-	0.17	-	0.05	-	Plessl et al. 2017
	mg kg ⁻¹ ₁	O, IR	-	1.7-7.5	-	0.86-6.27	-	N. Salamat et. al., 2014
ppm	O, AT, DE	-	0.577	2.942	-	-	Kenntner et al. 2001	

Mn	mg kg ⁻¹	An, TR	10.44	10.51	11.55	11.03	11.86	Present Study
	mg kg ⁻¹	An, PL	-	11.58	-	-	-	Komosa et al., 2012
	µg kg ⁻¹	O, KR	-	4.41	5.89	-	-	D.-P. Lee et al., 2012
	µg g ⁻¹	O, SAF	-	11.59	4.86	1.79	3.38	van Wyk et al., 2001
	µg g ⁻¹	O, IT	2.13	9.06	1.38	0.89	-	Naccari et al., 2009
	µg g ⁻¹	O, PL	-	3.07	1.75	0.55	-	Kalisińska et al., 2006
Pb	mg kg ⁻¹	Ac, TR	2.23	1.70	1.67	2.66	1.28	Present Study
		An, TR	3.31	1.40	0.96	1.25	1.39	
	mg kg ⁻¹	Ac, AT, DE, CH	-	9.695	3.087	-	-	Kenntner et al. 2007
	mg kg ⁻¹	An, PL	-	2.07	-	-	-	Komosa et al., 2012
	mg kg ⁻¹	An, HU	1.71	-	-	-	-	Gruz et al., 2018
	µg g ⁻¹	An, PK	38.83	-	-	-	-	Nighat et al., 2013
	ppm	An, ES	-	3.029	-	-	-	Perez-Lopez et al., 2008
	mg kg ⁻¹	O, IR	-	4.59	-	3.34	-	N. Salamat et. al., 2014
	ppm	O, AT, DE	-	7.033	2.926	-	-	Kenntner et al. 2001
	µg kg ⁻¹	O, KR	-	4.45	9.68	-	-	D.-P. Lee et al., 2012
	µg g ⁻¹	O, SAF	-	14.81	12.30	7.79	9.75	van Wyk et al., 2001
	µg g ⁻¹	O, IT	1.23	14.53	0.40	0.87	-	Naccari et al., 2009
	µg g ⁻¹	O, PL	-	2.508	2.128	0.158	-	Kalisińska et al., 2006
Zn	mg kg ⁻¹	An, TR	20.85	18.57	19.00	24.55	21.48	Present Study
	mg kg ⁻¹	An, PL	-	53.87	-	-	-	Komosa et al., 2012
	mg kg ⁻¹	An, HU	133.73	-	-	-	-	Gruz et al., 2018

$\mu\text{g g}^{-1}$	An, PK	143.9	-	-	-		Nighat et al., 2013
ppm	An, ES	-	298.3	-	-	-	Perez-Lopez et al., 2008
mg kg^{-1}	O, IR	-	29.87	-	33.77	-	N. Salamat et. al., 2014
$\mu\text{g kg}^{-1}$	O, KR	-	55.1	59.0	-	-	D.-P. Lee et al., 2012
$\mu\text{g g}^{-1}$	O, SAF	-	32.21	150.03	19.65	18.68	van Wyk et al., 2001
$\mu\text{g g}^{-1}$	O, IT	60.01	137.51	66.54	52.76	-	Naccari et al., 2009
$\mu\text{g g}^{-1}$	O, PL	-	26.57	20.04	25.55	-	Kalisińska et al., 2006

AT: Austria, **DE:** Germany, **ES:** Spain, **CH:** Switzerland, **HU:** Hungary, **IT:** Italy, **KR:** South Korea, **PL:** Poland, **PK:** Pakistan, **SAF:** South Africa, **TR:** Turkey

LOD: Limit of detection

Figures

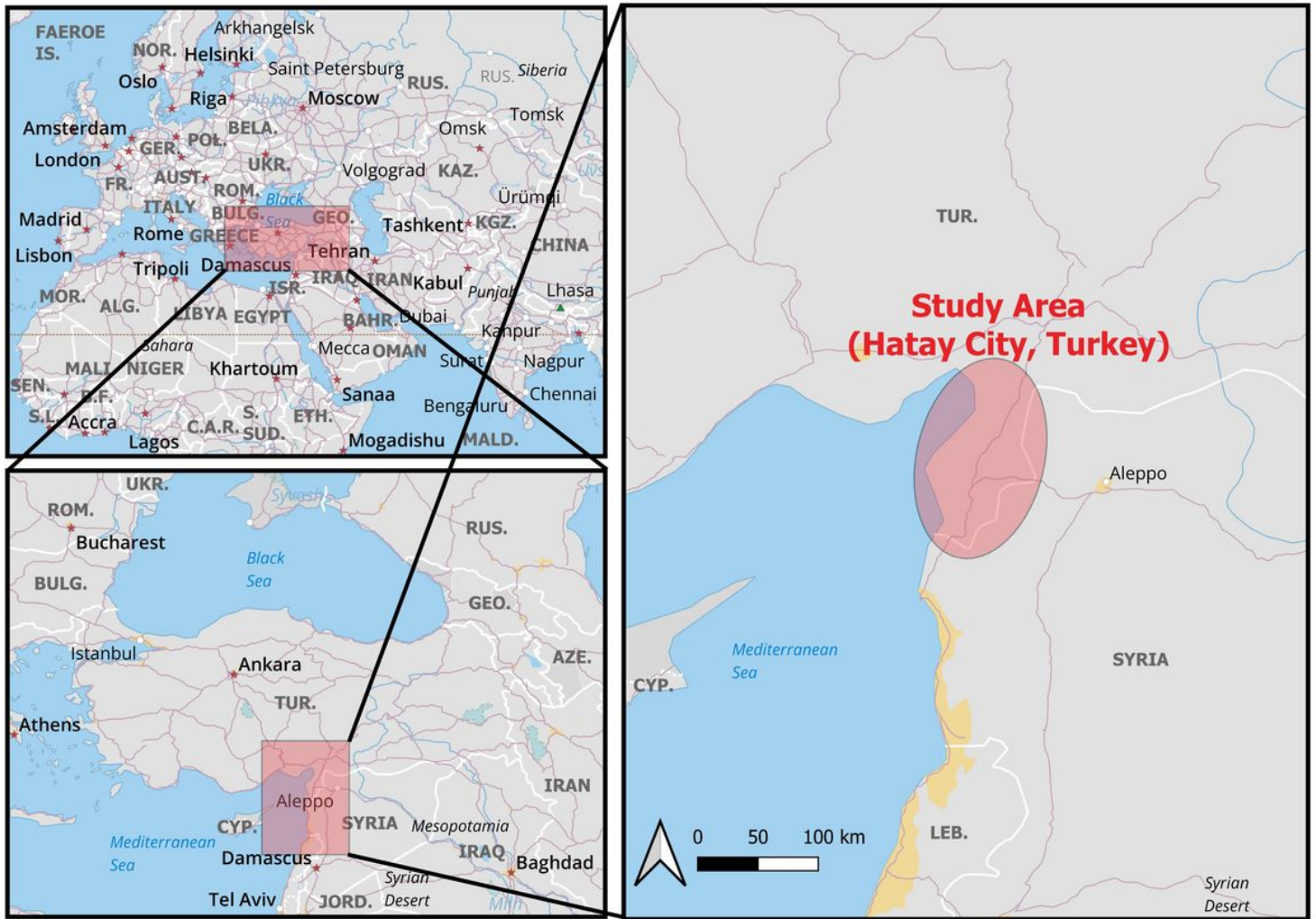


Figure 1

Ac (n:9; 6 male, 3 female), *An* (n:12; 5 male, 7 female), and *Cc* (n:7; 5 male, 2 female) that were found injured or dead in Hatay City, in the southern region of Turkey (Fig. 1, 36.20 N-36.16E) and submitted to the Hatay Mustafa Kemal Wild Animal Rescue and Rehabilitation Center between 2015-2021.