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STUDIES OF LEAD RESIDUES IN BONES AND FEATHERS OF GRIFFON VULTURES

Petar Stamberov, Tandzhu Mehmedov

University of Forestry, Faculty of Veterinary Medicine, Sofia, Bulgaria
E-mail: pstamberov@ltu.bg

ABSTRACT

The aim of the present research is to study the content of lead in feathers and bone samples in eight carcasses of Griffon Vultures (*Gyps fulvus, Habitz* 1870) from a region in southwestern Bulgaria. In order to exclude surface lead contamination, the feather samples were preliminary examined by X-ray fluorescent spectroscopy. The samples of lead concentration in feathers and bones were analyzed by the methods of ICP-MS Spectrometry. The results displayed residues of lead in the feathers and bones only in some individuals.

Key words: Griffon vultures, lead, samples, bones, feathers, ICP-MS Spectrometry.

Introduction

Incidents of lead poisoning in birds have been reported from all around the world (Fisher et al. 2006; Bounas et al. 2016). Birds of prey are one of the main groups affected by exposure to lead impacting several threatened species (Mateo, 2009). Like other vultures, the Griffon vulture is a scavenger, that consume died game species may ingest lead fragments or shot embedded in their prey's flesh (Mateo et al. 1997; Pain et al.1997).

The Griffon Vulture (*Gyps fulvus, Habitz 1870*) has a large breeding range, extending over Europe, the Middle East and North Africa. A reintroduction Vultures Return in Bulgaria LIFE08NATY/BG/278 Project is underway since 2010 in Stara Planina Mountain, Bulgaria (Deinet et al.2013; Botha et al. 2017).

The feathers must be used with care because the complex, fine structure is highly prone to accumulating surface contamination (Burger, 1993; Dauwe et al., 2003; Golden et al., 2011; Jenny et al., 2015). Lead accumulates in feathers trough active or passive diffusion from the blood into the feather follicle (Cardiel et al., 2011).

Lead in bone has been physiologically incorporated over the lifetime of the bird (Franson and Pain, 2011). Following absorption, lead is deposited in a range of tissues, primarily liver, kidney and bone. Whilst lead in liver and kidney has a turnover rate of weeks to months, lead is retained in bone for years, thus reflecting both acute and lifetime chronic exposure from all sources (Garcfa-Fermindez et al. 1997; Pain et al.2005).

In this article, we analyzed the concentrations of lead in the bones and feathers of the griffon vultures in order to determine residues of lead and evaluate the extent to which this species may be subject to lead poisoning.

Materials and methods

The feathers and bone samples in eight carcasses of Griffon Vultures from a region in southwestern Bulgaria in cooperation with the Wildlife Rescue Center "Green Balkans" in Stara Zagora were obtained. The cause of death was illegal poisoning with carbamate pesticides.

The collected feathers were examined by X-ray fluorescence spectrometry (*XRF)* for detection of exogenous incorporated lead. X-ray diffraction spectrometer *Fischerscope*® with high-resolution

CCD color camera for optical monitoring of the measurement location along the primary beam axis were used. Then feathers were washed in running deionized water and air dried. Then the feathers were cutting and isolated in airtight plastic bags and refrigerated at 0-4 ° C for later analysis.

Bone samples of approximately 3–5 g were extracted from the femur of each of the vultures. The bones were removed from the muscles and bone marrow with scalpel and bone scissors. Then the bones were dried to a constant weight (0.5–1 g) and stored in sterile containers and refrigerated at -20 ° C until the lead analysis.

The survey was conducted in the Laboratory of Ecology and technical tests "*Aquaterratest*" Sofia. The samples were analyzed by the methods of inductively coupled plasma spectrometry. The spectrometers Varian Vista-MPX CCD Simultaneous ICP OES, Varian Australia, and Plasma Quant MS S-NR 105000-AQ032, *Analytik Jena AG*, Germany were used. The detection limit was 0.1 mg/kg dry-weight.

Results

The preliminary X-ray fluorescence spectrometry (XRF) excludes the presence of exogenous incorporated lead in none of the feather samples were examined (Fig. 1).

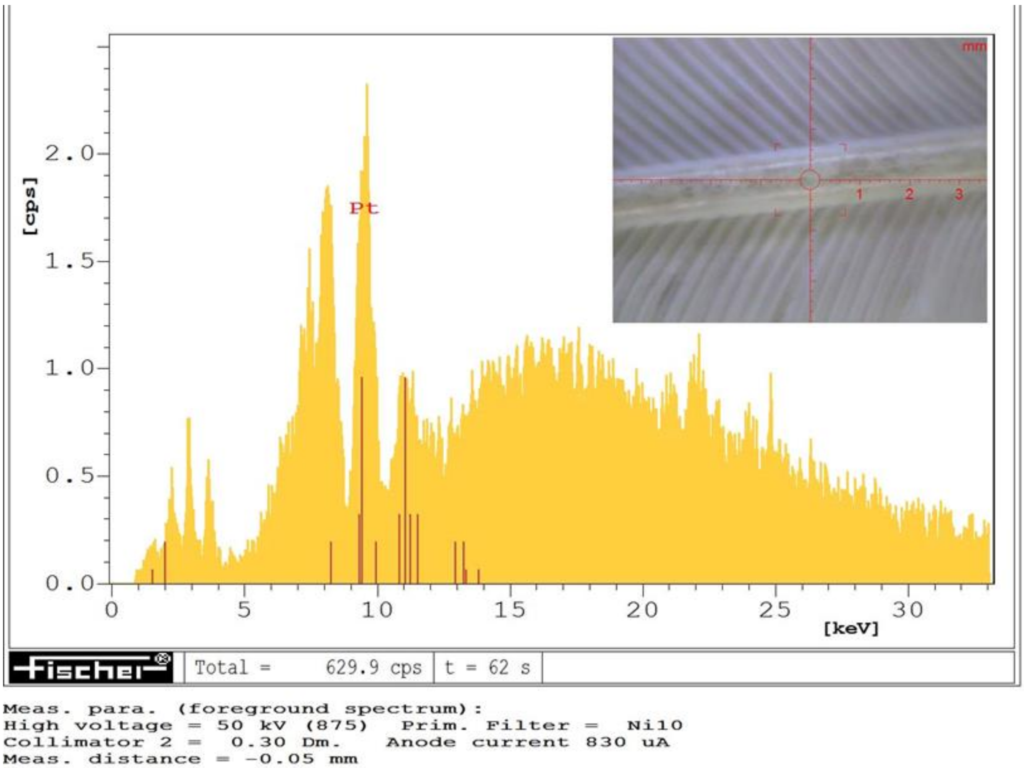

Figure 1: XRF of surface the feather of Griffon Vultures does not show presence of lead. The presence of platinum in some samples cannot be explained. The vaguely defined peaks after 15 keV are result to the large amount of light elements such as Ca, P and Mg. The presented figure is indicative for a series of measurements of different samples.

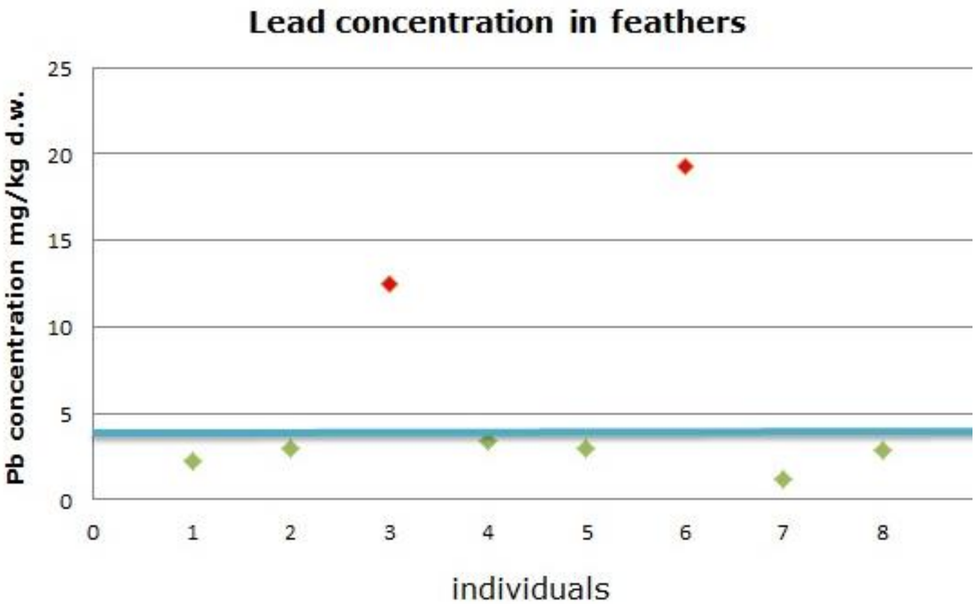

Figure 2: Concentrations of lead in feathers of griffon vultures *(Gyps fulvus, Habitz 1870).* Tolerable threshold of lead content in feathers is marked with solid blue line (4 mg/kg d.w.). The high lead concentration samples (12.49 and 19.81 mg/kg d.w) are marked with red color.

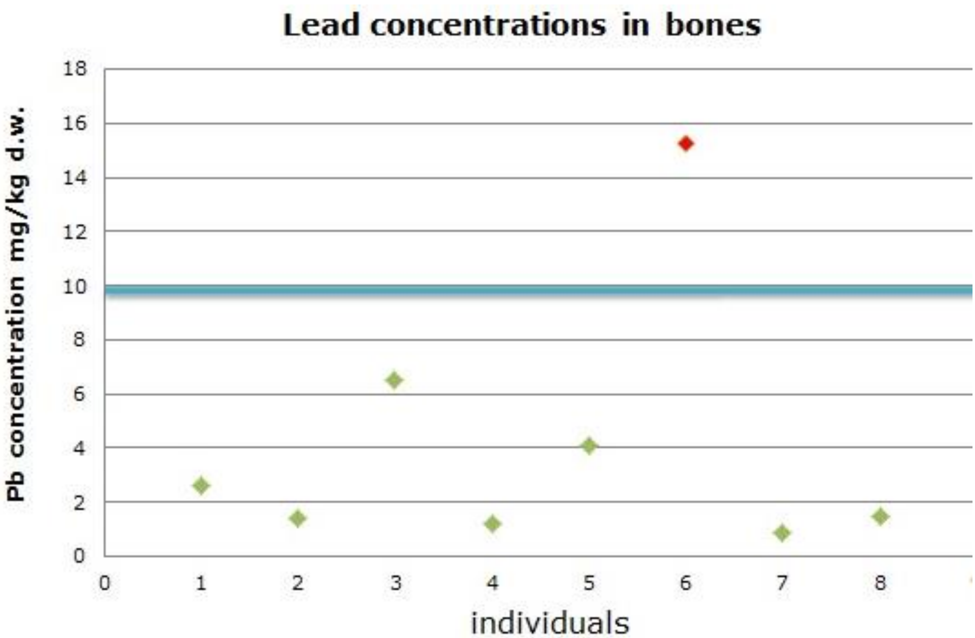

Figure 3: Concentrations of lead in femoral bones of griffon vultures *(Gyps fulvus, Habitz 1870).* Tolerable threshold of lead content in bones (<10 mg/kg d.w.) is marked with solid blue line. The sample with high lead levels is marked with red color.

Two (No 3 and No 6) of the eight feather samples showed high lead concentration of 12.49 and 19.81 mg/kg d. w. according to tolerable risk threshold of 4 mg/kg d. w. In four samples (No. 2, 4, 5 and 8) the feathers lead concentration showed about and slightly below threshold levels of 4 mg /kg d. w. The other two samples (No. 1 and 7) showed lead concentration in the feathers low of threshold levels of 4 mg /kg d. w. (Fig. 2).

The analysis of lead content in the femoral bones showed that in seven of the eighth samples the lead levels were below the background concentrations (<10.00 mg/kg d.w). A high lead concentration of 15.21 mg/kg d.w. was registered in only one (No. 6) corresponding to the high lead concentration of the same sample (No 6) in the feather. Sample No. 3 also showed some elevated lead content in bones corresponding to the high concentration of lead in the feathers of the same sample (Fig. 3).

Discussion

The distribution of lead concentrations in feathers is variable. The results should have been carefully interpreted due to the fact that, like the coat of mammals, the feathers of birds are exposed to anthropogenic pollution (Burger, 1993; Pain et al., 2005). X-ray fluorescence spectrometry has showed absence of surface lead contamination in feathers. Threshold value of > 4 mg/kg of lead concentrations in feathers is associated with adverse effects (Cardiel et al., 2011). According to Jenny et al. (2015), such presence of lead in the feathers may be a result of possible cumulative effects of lead due to repeated oral exposure.

Bone lead concentration is generally considered the best indicator of lead exposure over the total lifetime of the birds (Franson and Pain, 2011). This concentration is more difficult to interpret and threshold the value could be higher because of accumulation (Szymczak and Adrian, 1978). Lead in bones is relative immobile and excretion is very slow (Scheuhammer, 1987). The researchers recommend that lead concentrations (dry weight basis) of <10 mg/kg in bone may be considered background, and >10 mg/kg may indicate elevated lead exposure (above background levels). Birds with bone lead concentration of ≤ 20 mg/kg d.w be considered evidence of subclinical to clinical poisoning, and >20 mg/kg d.w be considered evidence of severe clinical poisoning (Mateo et al., 2003; Martin et al. 2008; Franson and Pain, 2011). Accordingly, in our case a high lead concentration in bones of one individual may be indicated for elevated lead exposure.

Conclusion

The results of this research displayed residues of lead in the feathers and bones only in some individuals. However, the feathers and bones provide a useful screening biomonitoring and potentially can be useful for future studies.

The establishment of facilities that can perform toxicological analysis of vultures is needed in our country in order to assess the threat, since any mortality that can be avoided, especially in such a small population.

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