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MONOGRAPH

Testing heavy metals biomonitoring through birds of prey as top predators

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ABSTRACT The Experimental Zooprofilactic Institute of Sicily (Italy) has great opportunities and validated methods (accreditated by an important national control unit like ACCREDIA) for biodiversity monitoring and analysis of fauna samples. Fauna recovery centers of Bosco di Ficuzza (Palermo) and Colli San Rizzo (Messina) routinely send dead specimens to the Institute to perform every category of analysis. Valuating heavy metals content in top predators is a strong instrument for territory and biodiversity health monitoring to take actions for management. In this study some specimen members of *Buteo buteo* (Linneus, 1758) species were investigated for Pb and Cd content. The methodology used for determination was performed by Inductively Coupled Plasma Mass Spectrometry (ICP-MS) method. Results show that in sample 1 Pb levels exceed cut-off values in organs: the liver accumulate a concentration of 4.668 mg/Kg and the muscle 0.200 mg/Kg. For sample 2, Pb and Cd were absent in both organs. Sample 3 reveals a very high concentration of Pb in the liver, with a value of 2.928 mg/Kg; this is probably due to a reiterated presence in the diet of prey killed by lead pellets. Liver of sample 3 accumulated even an high concentration of Cd (3.948 mg/Kg) justified by prey consumption, as rock partridge, rats or rabbits that probably feed on plants treated with agrochemicals.

KEY WORDS Common buzzard; heavy metals; bioaccumulation; top predators.

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INTRODUCTION

The accumulation of heavy metals in the soil can be a threat to ecosystems. It is mainly due to dispersion of lead shot and fishing weights in the territory, placing anthropogenic activities like hunting as the cause of a very specific pollution problem (Pain & Amiard-Triquet, 1993).

Water birds represent the animals prone to primary lead toxicosis, mistaking lead shot for seeds or other kind of food; follow raptors eating animals killed by gunfire or affected by primary toxicosis like water birds (Scheuhammer & Norris, 1996). Cadmium has proved to be the most dangerous trace element in food and environment for its high toxicity and long persistence. It can also be transferred by atmosphere to soil and then it enters in biological structures. When cadmium is dissolved, it can be absorbed by plants and percolate in the subsoil, contaminating internal waters (Cabrera et al.,

1998). Top predators can be an important source of information on contamination of a study area comprehensive of their home range, information that can include how, when and where pollutants are transferred to organisms like plants or animals (Jager et al., 1996).

The Experimental Zooprophyilactic Institute of Sicily has great opportunities and validated methods (accredited by the national control unit ACCREDIA) for biodiversity monitoring and analysis of fauna samples. The fauna recovery centers of Bosco di Ficuzza (Palermo) and Colli San Rizzo (Messina) routinely send specimens to the Institute to perform every category of analysis for both medical and veterinary purpose. Measuring heavy metals concentrations in top predators is a strong indicator for territory and biodiversity health, a monitoring tool to take management and conservation actions for natural systems and human healthcare.

In this study 3 specimens of common buzzard (*Buteo buteo* Linnaeus, 1758) were analyzed for heavy metals content to detect potential bioaccumulation in them.

MATERIAL AND METHODS

Samples digestion

The digestion process of the samples was carried out according to Lo Dico et al. (2015): about 1 g of the samples were transferred into previously decontaminated PTFE (poly-tetrafluoroethylene-tetrafluoroethylene) vessels with 3 ml of Ultrapure nitric acid 60% (V/V) and 5 ml of water. The samples digestion was carried out using a microwave digester Multiwave 3000 (Anton-Paar, Graz, Austria).

ICP-MS analysis

The elements were determined using a 7700x series ICP-MS (Agilent Technologies, Santa Monica CA, USA). The sample extracted were pumped by a peristaltic pump from tubes arranged on an autosampler ASX-500 Series (Agilent Technologies, Santa Monica (CA), USA), combined with a quartz cyclonic spray chamber (water-cooled 2° C). A calibration curve based on 8 standard additions (BlankCal - 0.01 – 0.05 – 0.1 – 0.5 – 1 – 5 – 10 – 50 µg/L) was made to evaluate the linearity. A pool of digested samples was used for this test. The linearity of the calibration curve was considered acceptable for $r^2 > 0.999$. The instrumental/method limits of detection and quantification (LOD and LOQ) were calculated by the 3σ and 10σ approach. Reagent blanks were used for the calculation of instrumental LOD and LOQ, whereas digested samples were used for the calculation of method LOD and LOQ. The trueness of the method was assessed using the recovery of 3 concentration levels according to Lo Dico et al. (2015). An acceptance limit between 90 and 110% was chosen.

Statistical analysis

All the results under the LOQ of the method were considered for the statistical analysis as half of the LOQ values, according to Helsel (2005). All the As, Cd and Pb levels were expressed as a wet weight (w.w.) and converted to dry weight (d.w.), according to Jerez et al. (2010), for the comparison with other studies reported in the literature. The conditions of normal distribution and homogeneity of variances of the data have not been met, therefore a Wilcoxon rank-sum test was carried out to evaluate toxic metals contents differences between muscle and adipose tissues and between sexes. The relationship between the modified K index of the loggerhead turtles and toxic metals contents was examined via Spearman's correlation method with R®3.0.3 software.

RESULTS

Results (Table 1) show that in sample 1 Pb levels exceed cut-off values in organs: the liver accumulate a concentration of 4.668 mg/Kg and the muscle 0.200 mg/Kg. For sample 2, Pb and Cd were absent in both organs. Sample 3 reveals a very high concentration of Pb in the liver, with a value of 2.928 mg/Kg; this is probably due to a reiterated presence in the diet of prey killed by lead pellets. Liver of sample 3 accumulated even a high concentration of Cd (3.948 mg/Kg) justified by prey consumption, as rock partridge, rats or rabbits that probably feed on plants treated with agrochemicals.

Sample	Type of sample	Pb mg/Kg	Cd mg/Kg
Common buzzard 1	Muscle	0.200	0.000
	Liver	4.668	0.000
Common buzzard 2	Muscle	0.000	0.000
	Liver	0.000	0.000
Common buzzard 3	Muscle	0.000	0.000
	Liver	2.928	3.948

Table 1. Pb and Cd content in muscle and liver belonging to 3 specimens of common buzzard (*Buteo buteo* Linneus, 1758).

DISCUSSION

According to literature (Scheuhammer, 1987), Pb present in plumage is probably a reflection of external pollution and it not depends on diet, while its high concentration in internal organs is directly related to ingestion of lead pellets during consumption of prey killed by them. In this scenario, hunting pressure prefigures itself as a harmful attitude to wildlife and to the welfare of territory. Use of pesticides that may leave residues is known to be dangerous for the health of all life forms (EFSA). This study, indeed, shows only a small part of the effect of this behavior in agriculture but also confirm the role of top predator as bioindicator of pollutants accumulation in the territory.

Some granivorous birds can mistake lead shot for food particles or grit and ingest them. Raptors can then predate birds and ingest Pb residues that have been accumulated in some tissues or organs of their prey (Pain et al., 1995).

CONCLUSIONS

Raptors, as top predators, are confirmed to be very good bioindicator of heavy metals accumulation in wildlife and in their territory of hunting.

Lead accumulated by shooting in water bodies can be ingested by waterfowl but actually there are no restrictions on the use of Pb shot. This can constitute a risk for both humans and wildlife (Pain et al., 1995, 2005, 2007, 2009, 2010).

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