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Avian feathers as a non-destructive bio-monitoring tool of trace metals signatures: A case study from severely contaminated areas

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HIGHLIGHTS

- Trace metals were assessed in avian feathers collected from industrial areas.
- Concentrations among the highest ever found in similar samples were recorded.
- Such high concentrations of Cr, Pb, Cd may affect avian reproduction.
- Avian feathers are a convenient sampling tool for assessing metal contamination.

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ABSTRACT

The concentrations of trace metals were assessed using feathers of cattle egrets (*Bubulcus ibis*), collected within two industrial areas of Pakistan, Lahore and Sialkot. We found, in order of descending concentration: Zinc (Zn), Iron (Fe), Nickel (Ni), Copper (Cu), Cadmium (Cd), and Manganese (Mn), Chromium (Cr), Arsenic (As), and Lithium (Li), without any significant difference (except Fe, Zn, and Ni) between the two areas. The concentrations of trace metals, we recorded were among the highest ever reported in the feathers of avian species worldwide. The concentrations of Cr, Pb, Cd were above the threshold that affects bird reproductive success. The high contamination by heavy metals in the two areas is due to anthropogenic activities as well to natural ones (for As and Fe). The bioaccumulation ratios in eggs and feathers of the cattle egret, their prey, and the sediments from their foraging habitats, confirmed that avian feathers are a convenient and non-destructive sampling tool for the metal contamination. The results of this study will contribute to the environmental management of the Lahore and Sialkot industrial areas.

1. Introduction

In some developing countries, environmental degradation is increasing day by day as a consequence of the exponential growth of the human population, and of technological developments (Qadir and Malik, 2011; Xu et al., 2013; Ayyamperumal et al., 2006). Environmental contamination, by heavy metals among other chemicals, involves great health risks for all living organisms, humans and wildlife (Salzano and Angelone, 2013; Ullah et al., 2014; Klaassen, 2013). Heavy metals are non-biodegradable environmental contaminants that may accumulate in the upper levels of the food chains, in relation to the habitat and dietary preferences of each species (Boncompagni et al., 2003). Trace metals such as Chromium (Cr), Copper (Cu), Mercury (Hg), Arsenic (As), Cobalt (Co), Manganese (Mn), Selenium (Se), Zinc (Zn) and many others enter the food chain, and have the capacity to biomagnify

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(Burger and Gochfeld, 2004; Bostan et al., 2007). Exposure to these heavy metals is widely described to threaten the reproductive success of many avian species in the wild (Malik and Zeb, 2009).

The need to check for the effects of heavy metals on the environment has led to various bio-monitoring strategies, involving the use of indicators, i.e. particular organisms that may reflect the contamination of their ecosystem (Kalisinska et al., 2003; Burger and Gochfeld, 2007). Birds have been recognized since the 1960s as the potential bioindicators of environmental pollution (Erwin and Custer, 2000). Their potential is primarily due to the position of some bird species at the top of the food chain, so that any change in the lower trophic level is signaled by their response (Boncompagni et al., 2003). Many bird species live in close association with human activities, are exposed to environmental contaminants, and may suffer from the resulting toxic effects (Malik and Zeb, 2009). The concentrations of heavy metals in feathers, droppings and eggs of birds have been documented worldwide over the years (Dauwe et al., 2005; Malik and Zeb, 2009; Hashmi et al., 2013), and metal contamination was shown to affect bird populations (Gragnaniello et al., 2001; Muralidharan et al., 2004). Bird feathers offer several advantages as bioindicators of metal exposure and feather collection is non-invasive. The concentration of heavy metal is higher in feathers than in the other tissues, and hence easier to detect and quantify, because birds excrete considerable amounts of metals through feather moult (Malik and Zeb, 2009; Zamani-Ahmadmahmoodi et al., 2010). Moreover, feathers from newly born chicks indicate local contamination, derived mostly from food collected locally by their parents during the short period of egg formation and chick development (Boncompagni et al., 2003; Muralidharan et al., 2004). Waterbirds that breed in colonies provide additional advantages as bioindicators of pollution: easy sampling (Burger and Gochfeld, 2000a,b); a limited foraging range around their colony site, thus allowing inference about the source of contaminants (Burger et al., 2004); and dependence on specific habitat and prey resources (Fasola et al., 1998).

In recent years, it has become evident that environmental contamination is severe in various areas of Pakistan (Malik and Zeb, 2009), where industrial, urban and agricultural activities are major sources of pollution (Movalli, 2000; Qadir et al., 2008). For some agricultural areas, moderate concentrations of metal contamination have recently been reported using the feather of the cattle egret (*Bubulcus ibis*) as bioindicator (Malik and Zeb, 2009; Ullah et al., 2014). For the industrial areas of Pakistan, however, information on metal exposure is scarce, while, these areas might be severely contaminated by the vast quantities of urban and industrial waste from tanneries, pharmaceuticals, surgical and steel manufacturing factories. Moreover, these areas are also reported to serve as dumping sites for persistent organic pollutants (POPs) and expired chemicals after the ban imposed by Stockholm (2001).

In this paper, we assess the contamination by trace elements of prime environmental concern (As, Cd, Cr, Pb, Mn, Li, Fe and Zn) in two of the main industrial areas of Pakistan, near the cities of Sialkot and Lahore, that are presumably exposed to severe pollution, and for which we had already assessed the persistent organic pollution (Khan et al., 2013). We used as bio-indicator the cattle egret, a widespread, colonial, and predatory waterbird (Ciconiiformes: Ardeidae). In order to monitor the contaminant concentrations and their levels throughout the food chain, we collected diverse samples, cattle egret's eggs, chick feathers, its prey, and the sediments of its foraging areas in the vicinity of the breeding sites. Our results offer baseline data for South Asia, where contamination studies are still scanty; emphasize potential threats to avian populations; and provide information to the authorities concerned with the mitigation of environmental contamination, which might ultimately threaten human health in the severely polluted industrial areas of Pakistan.

2. Materials and methods

2.1. Study area

Samples were collected at two study areas, Lahore and Sialkot (Fig. 1), two major cities of Punjab Province, and, two of the major industrial hubs of Pakistan (Aftab et al., 2000). These study areas are described in detail by Khan et al. (2013). In the last decades, the exponential population growth of this region, coupled with the development of extensive industrial activities, tanneries, steel factories, leather garments and sport equipment manufacture, has resulted in widespread environmental degradation (Qadir et al., 2008). Sialkot city was reported to generate 19 million m^3 $year^{-1}$ of waste water in 2007 (World Wildlife Fund), and Lahore city generated 1350 million L d^{-1} of wastewater (Mahmood and Malik, 2014). No water treatment facility exist in these two areas, and the wastewater, drained into natural streams, ponds, open field and croplands, can be expected to produce severe environmental contamination.

2.2. Sampling plan and analytical procedures

In the Lahore area, cattle egret samples were collected near Dad Pura village, 1 km from Kalashah Kaku interchange (31°57′02″N, 74°24′14″E). This area hosts significant industrial activities, including still mills, chipboard and rubber manufactures, leather tanning and pigment factories. In the Sialkot area, another cattle egret colony (32°29′50″N and 74°32′10″E) was sampled near Khajori Wala village, 3.5 km from Sialkot city. In both study area(s), rice is the main crop, and it receives water from canals that might get polluted by the waste coming from nearby industrial areas. The egrets forage in the rice fields and in other water bodies, and are therefore exposed to the toxic metals through food intake and dust ingestion.

The samples, sediments, prey and eggs, were collected at the two study areas, using the methods detailed by Khan et al. (2013). Moreover, samples of 8–12 breast feathers were collected at two cattle egret's colonies from one chick per nest, when the chick was 10–15 d old and the external attachment of contaminants should still have been low. The feather samples were packed in polythene bags and stored in refrigerator (at −20 °C) till further analysis.

Before analysis, feathers were washed with deionized water, followed by acetone, in order to remove loosely adherent exogenous contamination. The washed feathers were put in metal-free polyethylene vials and dried in oven for 24 h at 60 °C. Dry weight was determined using a digital balance. Each sample was cut into small pieces with a stainless steel knife, and digested as described by Dauwe et al. (2002) with few modifications. All containers were soaked in 10% (v/v) HNO_3 overnight and rinsed with ultra-pure water by three times. Each sample was carefully weighed, then digested with 1 mL GR grade 65% (v/v) HNO_3 (Merck, Germany) overnight, and at the next day with 1 mL GR grade 30% (v/v) H_2O_2 (Merck, Germany). The samples, mixed and sealed in Teflon microwave digestion tubes, were digested in an accelerated microwave digestion system (Mars CEM, CEM Corporation, Matthews NC, USA) at 800 W, 120 °C for 10 min and then 800 W, 170 °C for 30 min. Trace metals were measured using a flame atomic absorption spectrophotometer (Perkin Elmer-AA240FS Fast Sequential Atomic Absorption Spectrometer). Table S-1 represents the instrumental conditions for the measurements of the analytes by FAAS. The other, samples, sediments, prey and eggs, were digested and analysed by the same method.

2.3. Quality assurance and quality control

A quality control (QC) sample was prepared by mixing aliquots of each sample, that were therefore broadly representative of the