NOTE AND RECORD

Incidence of heavy metals in feathers of birds in a humanimpacted forest, south-west Nigeria

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1 | INTRODUCTION

Hazardous wastes and heavy metal are detrimental to all life forms (Dixit et al., 2015; Sharma, Katnoria, Kaur, & Nagpal, 2015). Organic forms of heavy metals such as mercury and lead can accumulate in living tissues; the health implication ranges from low immunocompetence, failed reproduction to high mortality in a number of taxa (Falq et al., 2011; Fritsch et al., 2010; Hollamby et al., 2004; Kerby, Richards-Hrdlicka, Storfer, & Skelly, 2010; Scheifler et al., 2006; Webb & Leake, 2006). Unprecedented levels of heavy metals and their negative role in biodiversity loss and habitat degradation pose major ecological concerns (Ayangbenro & Babalola, 2017; Kibria, 2016; Sharma et al., 2015). Despite this fact, less attention is focused on monitoring the levels and impact of heavy metals in the western part of Nigeria.

Birds are promising biomonitoring species for heavy metals and xenobiotic based on their use in nondestructive avian matrices, for example feather, blood and egg; and bioavailability, and biotransference in dose-dependent responses (Becker, 2003; Furness & Greenwood, 1993; Roux & Marra, 2007; Swaileh & Sansur, 2006). Also, as feathers grow in birds, heavy metals are sequestered in the sulfhydryl group of the keratin; the metal residues remained resistant to change in older feather as blood supply stops (Burger, 1993). We investigated the incidence of heavy metals in passerine birds inhabiting a human-impacted forest in Ilara-Mokin, south-west Nigeria.

2 **METHODS**

The study area is approximately 15 km² (1,500 hectares) and located between latitude 07°20'-07°23'N and longitude 05°60'-05°90'E

Bada and Omotoriogun have equal authorship right.

within the Guinean Forests of West Africa Hotspot and Tropical Lowland Rainforest belt of south-west Nigeria. The area experiences distinct wet and dry seasons with total annual rainfall of 1,800 mm. Average monthly temperature ranged between 27 and 30°C with relative humidity <70%. The landscape is interspersed with hilly and rocky outcrops, and the forest is undergoing degradation due to farming and logging activities.

Feather samples were collected from 28 individuals of ten bird species trapped using mist-nets mounted at edges and corridors of the forest between 06:00 and 11:00 hours GMT in November 2017. The birds were identified, sexed, aged, weighed and fitted with uniquely numbered aluminium ring (SAFRING) to avoid resampling. One of the middle tail feathers was detached and kept in labelled envelope under room temperature for laboratory analysis. Feathers were collected from mainly adult birds without moult on the tail. Birds were immediately released to avoid stress.

Wet oxidation technique was used to dry and ash the feather samples at temperature range of 420-600°C. Unwashed feather samples were digested in macro-Kieldahl digestion flask using concentrated solutions of 20 ml HNO3, 10 ml H2SO4 and 10 ml $(NH_4)_2C_2O_4$ added in sequence. The product of digestion was analysed for metals using the Atomic Absorption Spectrophotometer (PerkinElmer, Model 3000). Distilled water was used as the blank sample, and there was no contamination. The concentrations of metals (µg/g) were calculated as absorbance divided by weight of feather.

Analyses were performed using the R Package version 2.15.2 (R Development Core Team, 2018). The variation in concentration of metals was tested across bird species using the Kruskal-Wallis test. The difference in detection and nondetection of metal was

TABLE 1 Average	TABLE 1 Average concentration (μg/g) with standard error (±SE) and limit of detection, LOD (mg/l), of heavy metals in feathers of bird species	ıdard error (±SE) aı	nd limit of detection	, LOD (mg/l), of heav	y metals in feathers o	of bird species		
Common name	Species	Copper (µg/g)	Lead (µg/g)	lron (μg/g)	Zinc (µg/g)	Nickel (µg/g)	Chromium (µg/g)	Cadmium (µg/g)
Olive Sunbird	Cyanomitra olivacea (n = 5) ^b	9.79 ± 9.50	55.34 ± 30.69	146 ± 144.59	27 ± 20.51	14.08 ± 8.37	39.13 ^a	ND
Collared Sunbird	Hedydipna collaris (n = 4) ^b	17.87 ± 8.59	61.71 ± 39.76	252.30 ± 308.91	57.93 ± 31.93	52.94 ^a	ND	QN
Little Greenbul	Eurillas virens ($n = 5$) ^c	1.54 ± 1.35	8.11 ± 6.20	41.23 ± 41.81	8.50 ± 6.92	10.16 ± 0.39	5.64 ^a	ND
Yellow-whiskered Greenbul	Eurillas latirostris (n = 2) ^c	2.69ª	16.39 ± 7.97	58.00 ± 71.38	5.70 ± 2.73	9.32 ^a	QN	Ŋ
Western Bluebill	Spermophaga haematina (n = 2) ^d	35.96 ± 50.16	162.29 ± 214.96	571.39 ± 787.96	181.51 ± 248.38	342.86 ^a	28.57 ^a	ND
Bronze Mannikin	Lonchura cucullata (n = 3) ^d	6.32 ± 4.52	80.49 ± 45.45	251.40 ± 315.17	38.55 ± 14.46	38.10^{a}	80.95 ^a	4.52 ^a
Black-and-White Mannikin	Lonchura bicolor (n = 2) ^d	QN	9.25 ± 8.67	13.02 ± 5.16	25.16 ± 17.45	ND	DN	DN
Orange-cheeked Waxbill	Estrilda melpoda (n = 2) ^d	QN	11.87 ± 7.36	21.75 ± 7.19	41.38 ± 20.81	ŊŊ	QN	Ŋ
Great Reed Warbler	Acrocephalus arundinaceus (n = 2) ^e	1. 08 ^a	9.03 ± 5.48	17.54 ± 7.13	80.98 ± 96.85	DN	QN	ND
Willow Warbler	Phylloscopus trochilus $(n = 1)^{e}$	ND	36.00 ^a	60.00 ^a	60.00 ^a	ND	ND	ND
	Kruskal-Wallis test	$H_{1,6} = 7.111;$ p = 0.311	$H_{1,9} = 15.35;$ p = 0.0818	H _{1,9 =} 7.005; p = 0.637	$H_{1,9=14.029};$ p = 0.121	$H_{1,5=}5.583;$ p=0.349	$H_{1,3} = 3.0;$ p = 0.392	
	Limit of detection, LOD (µg/g)	0.005	0.08	0.05	0.005	0.05	0.04	0.01
	WHO (1985)	3	0.05	0.30	5.00	4	0.15	0.005
	USEPA (1987)	0.1	0.1	0.0058	0.0766		0.05	0.008
	FEPA (2003)	<1.0	<1.9		<1.0			<1.0
Note: Included is the te	Note: Included is the test result of metal concentration among the species using the Kruskal-Wallis test and the permissible limit for metals concentration in biological tissues according to WHO USEPA	among the species I	ising the Kriiskal-Wa	illis test and the nerm	issible limit for metals o	concentration in hi	ological tissues accord	ing to WHO LISEPA

Note: Included is the test result of metal concentration among the species using the Kruskal-Wallis test, and the permissible limit for metals concentration in biological tissues according to WHO, USEPA and FEPA.

Abbreviations: n, number of individuals used for calculating averages; N, number of species.

^aRepresents values which are not average. ND represents nondetection of metal.

^bFamilies and feeding guilds are indicated as Nectariniidae and nectivore.

^cPycnonotidae and omnivore

^dEstrildidae and granivore.

^eSylviidae and insectivore.

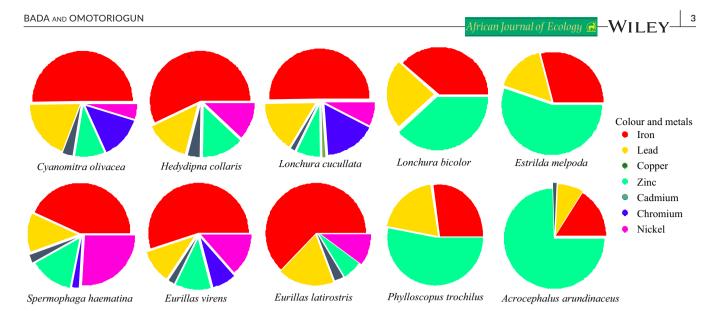


FIGURE 1 Pie chart showing percentage concentration of metals represented by colours in the feather of bird species sampled from forest in Ilara-Mokin, Nigeria

determined using chi-square test; correlation between metal concentrations was tested using Kendall's correlation.

3 | RESULTS AND DISCUSSION

The average concentrations (μ g/g) and limit of detection (μ g/g) of metals including the Kruskal–Wallis test results are presented in Table 1. With limit of detection between 0.005 and 0.08 μ g/g, higher concentration and variation of metals were found across bird species, ranging as follows: copper (1.1–35.96 μ g/g, N = 7); lead (8.1–62.3 μ g/g, N = 10); iron (13.0–571.4 μ g/g, N = 10); zinc (5.7–181.5 μ g/g, N = 10); nickel (9.4–342.9 μ g/g, N = 6); and chromium (5.6–81.0 μ g/g, N = 4). Cadmium with concentration 4.2 μ g/g was only detected in bronze mannikin *Lonchura cucullata*. Lead, iron and zinc were detected in all the birds (Table 1; Figure 2) but copper, nickel, chromium and cadmium in <70% of the birds. Iron had higher concentrations except black-and-white mannikin *Lonchura bicolor*, orange-cheeked waxbill *Estrilda melpoda* and great reed warbler *Acrocephalus arundinaceus* where values were lower compared

with zinc. western bluebill *Spermophaga haematina* showed elevated levels of metals except in chromium (Table 1; Figure 1). There was variance in the detection and nondetection of metals (χ^2 = 78.66, *df* = 6, *p* < 0.001, Figure 2a); and correlation between iron and zinc (*r* = 0.1963, *p* = 0.0718; Figure 2b); iron and lead (*r* = 0.5219, *p* < 0.05; Figure 2c); and lead and zinc (*r* = 0.3099, *p* = 0.0104; Figure 2d).

The concentrations of metals observed corroborate finding from several studies (Demibras, 1999; Gushit, Turshak, Chashda, Abba, & Nwaeze, 2016; Roux & Marra, 2007; Uluozlu, Tuzen, Mendil, & Soylak, 2009) and were higher than the permissible limits for biological tissues (EPA, 1987; FEPA, 2003; WHO, 1985). The varying metal levels in the birds may be due to the chemical properties of metals, as well as the trophic level, feeding guilds, physiology and behaviour. Most metals are insoluble in water, soluble in fats and resistant to biological and chemical degradation. Temperature and pH levels influence metal bioavailability and rate of absorbate diffusion through cellular membranes, therefore increasing their toxicity in living tissue (Markich, Brown, Batley, Apte, & Stauber, 2001; Olaniran, Balgobind, & Pillay, 2013; Worms, Simon, Hassler, & Wilkinson, 2006).

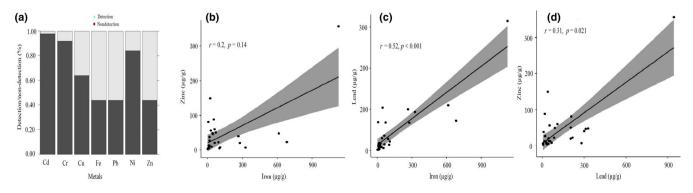


FIGURE 2 Percentage detection and nondetection of metals cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), lead (Pb), nickel (Ni) and zinc (Zn) across feathers of 28 individuals of birds (a) and the correlation between metals found in the birds with the correlation coefficient and p-values (b) Zn and Fe, (c) Pb and Fe and (d) Zn and Pb, of bird sampled from a human-impacted forest in Ilara-Mokin, Nigeria

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Furthermore, the metal levels across feeding guilds (Table 1) suggest differential exposure risks and may be pointer to widespread cases of metals in food chain in the area. The birds specialised on different food types, for example nectar, fruits, seed and insects, and may have acquired metals through contaminated food sources. However, feather age affects their metal sequestration rates and older feather tends to have higher metal loading than young feathers; this can influence metal in feather (Furness & Greenwood, 1993). The correlation among some of the metals may reflect similar chemical properties and behaviour in their bioaccumulation and detoxification processes.

The anthropogenic sources of heavy metals are more bioavailable due to their soluble and mobile reactive forms (Dixit et al., 2015). Agrochemicals are probable sources of heavy metals in an agricultural landscape; the area is intensively farmed using fertilisers, herbicides and pesticides. Pesticides are major source of lead, cadmium, copper and chromium; also, there are glyphosate-based herbicides that contain metals. Vehicular emission and industrial influent may be additional sources; the south-west axis of Nigeria is an industrialised region. External contamination of feathers by heavy metal is a problem (Borghesi et al., 2016; Dolan et al., 2017), and the high concentration of metal in this study may be influenced by external contamination. We recommend increasing species coverage and broadscale study of the area in the future.

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