

Heavy Metal Distribution in Chicks of Two Heron Species from Korea

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Abstract This study presents concentrations of iron, manganese, zinc, copper, lead, and cadmium in tissues of black-crowned night heron (*Nycticorax nycticorax*) ($n = 8$) and grey heron (*Ardea cinerea*) ($n = 9$) chicks from Pyeongtaek heronry, Gyeonggi-do, Korea, 2001. Iron (respectively, 45.8 ± 24.0 $\mu\text{g}/\text{wet g}$, 155 ± 123 $\mu\text{g}/\text{wet g}$), zinc (38.3 ± 5.34 $\mu\text{g}/\text{wet g}$, 50.9 ± 14.0 $\mu\text{g}/\text{wet g}$), and copper (9.93 ± 2.26 $\mu\text{g}/\text{wet g}$, 30.2 ± 12.9 $\mu\text{g}/\text{wet g}$) concentrations in feathers, manganese concentrations in livers (3.26 ± 0.68 $\mu\text{g}/\text{wet g}$, 1.50 ± 0.58 $\mu\text{g}/\text{wet g}$), kidneys (1.43 ± 0.27 $\mu\text{g}/\text{wet g}$, 0.84 ± 0.34 $\mu\text{g}/\text{wet g}$), and bones (1.34 ± 0.50 $\mu\text{g}/\text{wet g}$, 3.17 ± 1.31 $\mu\text{g}/\text{wet g}$) were different between black-crowned night heron and grey heron chicks. Lead concentrations in bones (0.11 ± 0.04 $\mu\text{g}/\text{wet g}$, 0.61 ± 0.42 $\mu\text{g}/\text{wet g}$) and cadmium concentrations in liver (13.5 ± 2.30 $\mu\text{g}/\text{wet kg}$, 10.3 ± 1.59 $\mu\text{g}/\text{wet kg}$), kidney (6.61 ± 2.54 $\mu\text{g}/\text{wet kg}$, 2.31 ± 1.29 $\mu\text{g}/\text{wet kg}$), and muscle (5.25 ± 5.91 $\mu\text{g}/\text{wet kg}$, 1.37 ± 0.90 $\mu\text{g}/\text{wet kg}$) differed between chicks of the two heron species. The differences of heavy metal concentrations in tissues in herons and egrets were reported to other similar studies. Heavy metal concentrations for both heron species were at background levels. In both species, lead concentrations were higher in livers than in bones and cadmium concentrations were higher in livers than in kidneys. We suggest that it is not chronic exposure but acute exposure to lead and cadmium contamination around breeding site that leads

to these observations. Therefore, lead and cadmium concentrations in tissues can be used as a bioindicator of acute local contamination.

Introduction

Piscivorous birds are susceptible to bioaccumulation of pollutants mainly through the consumption of contaminated food. Moreover, pollutants such as lead and cadmium have been associated with breeding failures in some egrets and herons (Burger et al. 1992). Because herons are high on the trophic level of the food web, they are useful as a bioindicator species for environmental monitoring. Concentrations of environmental contaminants in tissues of herons can be used to evaluate the chronic or acute exposure. Because heron chicks are fed with prey collected within a few kilometers of the nesting colony (Erwin et al. 1991; Kim et al. 1998; Kim and Koo 2007c), their tissue concentrations reflect local contamination, particularly of the aquatic ecosystem (Furness and Greenwood 1993; Kim and Koo 2007a, b).

Pollutants such as lead and cadmium at environmentally relevant concentrations can have adverse effects on various physiological systems, including endocrine systems (Stoica et al. 2000; Martin et al. 2003). Previous studies have shown that heavy metals can also influence the reproduction and general health of birds (Janssens et al. 2003; Dauwe et al. 2004). Lead and cadmium is toxic to birds in both laboratory and field studies. Lead-exposed birds demonstrate decreased body weight and reduced reproduction (Burger et al. 1986). Toxic effects of cadmium on birds include decreased egg production, kidney damage, testicular damage, and altered behavioral response (Furness 1996).

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Black-crowned night herons (*Nycticorax nycticorax*) and grey herons (*Ardea cinerea*) migrate for breeding to Korea in late March or in early April and return to southern Japan or southeast Asia for wintering in late September or early October. They forage for their prey, such as fish, amphibians, reptiles, and invertebrates, in farming areas, streams, and rivers.

Many studies have recommended herons and egrets as bioindicators for heavy metals in aquatic systems and local pollution around breeding sites (Fasola et al. 1998; Connell et al. 2002; Boncompagni et al. 2003; Kim and Koo 2007a, b). In previous studies (Kim and Koo 2007a, b), we demonstrated the correlation between the heavy metal concentrations in the diet and livers of black-crowned night heron and grey heron chicks and the diet and feathers of those two species. However, we did not discover whether the lead and cadmium contaminations were due to acute or chronic exposure. In this paper, we estimate the distribution and difference of heavy metal in various tissues and seek to answer the question of whether lead and cadmium contamination were acute or chronic exposure of black-crowned night heron and grey heron chicks from the Pyeongtaek heronry, Gyeonggi-do, Korea.

Materials and Methods

Study Site and Sampling

This study was conducted during the 2001 breeding season (April–June) at a breeding colony in Pyeongtaek city (37° 02' N, 127° 02' E), Gyeonggi-do, Korea. Black-crowned night herons and grey herons frequently nest in the same colony. There were 63 pairs of black-crowned night herons and 300 pairs of grey herons in 2001. Pyeongtaek colony is surrounded by agricultural land, including a grape orchard. Rice paddy fields where many herons foraged surround the colony. The distance from breeding sites to additional foraging sites were 3.9 km to Jinwee stream and 6.8 km to Anseong and Jinwee stream confluence area, respectively.

Black-crowned night heron ($n = 8$) and grey heron ($n = 9$) chicks were marked with plastic rings 1–3 days after hatching in different nests and were recaptured 20–22 days after hatching. Chicks were weighed (± 0.1 g), and the bill (± 0.1 mm), wing (± 0.1 mm), and tarsus (± 0.1 mm) length were measured. Chicks were euthanized by thoracic compressions and frozen at -20°C until they were dissected. Livers, kidneys, muscles, bones, and breast feathers were separately dissected from the body, weighed (± 0.1 g) and stored in chemically clean jars.

Metal Analysis

The concentrations of iron, manganese, zinc, copper, lead, and cadmium were determined by a flame atomic absorption spectrophotometry (Hitachi Z-6100), after the mineralization of samples with nitric, sulfuric, and perchloric acid in Kjeldahl flask. Tissues with low lead and cadmium concentration were measured by atomic absorption (AA) spectrophotometry, after treatment with sodium N,N-diethyldithiocarbamate trihydrate [DDTC, $(\text{C}_2\text{H}_5)_2\text{NCS}_2\text{Na}\cdot 3\text{H}_2\text{O}$]–methyl isobutyl ketone [MIBK, $\text{CH}_3\text{COCH}_2\text{CH}(\text{CH}_3)_2$] (Honda et al. 1982). Spikes and blanks were run on a frequency of at least 5% of the total number of samples. To assess accuracy, a spike, a blank, a standard, and a sample were run in triplicate in each analytical run. Spikes recoveries ranged from 94% to 105%. Recovered concentrations of the certificated samples were within 5% of the certificated values. All element concentrations ($\mu\text{g/g}$ or $\mu\text{g/kg}$) in tissues were estimated on a wet weight basis.

Statistical Analysis

We statistically tested for differences between black-crowned night heron and grey heron chicks using the *t*-test. Data were log-transformed to obtain normal distribution that satisfied the homogeneity of variance assumptions of the *t*-test (Custer et al. 2003). We used SPSS version 12.0 for statistical analysis. We present arithmetic mean (\pm standard deviation, SD) and geometric means in the tables.

Results

Iron, Zinc, and Copper Concentrations in Tissues

Iron, zinc, and copper concentrations in livers, kidneys, muscles, and bones were not significantly different between black-crowned night heron and grey heron chicks. However, concentrations in feathers were higher in grey heron chicks than in black-crowned night heron chicks ($p = 0.001$) (Tables 1, 2, and 3).

Manganese Concentrations in Tissues

Manganese concentrations in muscles and feathers did not differ between black-crowned night heron and grey heron chicks (Table 4). However, concentrations in livers ($p < 0.001$) and kidneys ($p = 0.002$) were higher in black-crowned night heron than in grey heron chicks. Manganese

Table 1 Iron concentrations ($\mu\text{g}/\text{wet g}$) in black-crowned night heron and grey heron chicks from Pyeongtaek, Korea, 2001

	Livers	Kidneys	Muscles	Bones	Feathers**
Black-crowned night heron ($n = 8$)					
Mean \pm SD	251 \pm 182	70.9 \pm 11.7	55.8 \pm 10.2	32.3 \pm 11.8	45.8 \pm 24.0
Min–max	106–627	50.1–84.8	38.6–71.8	15.5–44.9	27.3–95.2
Geomean	210	70.0	54.9	30.0	41.6
Grey heron ($n = 9$)					
Mean \pm SD	250 \pm 187	75.7 \pm 22.9	56.9 \pm 14.1	28.1 \pm 13.7	155 \pm 123
Min–max	89.4–600	52.3–97.5	43.1–83.5	13.8–57.0	59.1–420
Geomean	198	72.8	55.5	25.5	124
p value ¹	NS	NS	NS	NS	0.001

All samples were detected

** $p < 0.01$

¹ p value for t -test comparing between species

NS = not significant

Table 2 Zinc concentrations ($\mu\text{g}/\text{wet g}$) in black-crowned night heron and grey heron chicks from Pyeongtaek, Korea, 2001

	Livers	Kidneys	Muscles	Bones	Feathers*
Black-crowned night heron ($n = 8$)					
Mean \pm SD	40.2 \pm 4.77	18.3 \pm 5.79	16.0 \pm 3.63	40.9 \pm 20.2	38.3 \pm 5.34
Min–max	32.5–45.7	13.3–28.4	11.5–20.7	20.2–78.2	29.8–46.5
Geomean	40.0	17.6	15.6	37.0	38.0
Grey heron ($n = 9$)					
Mean \pm SD	39.2 \pm 22.1	22.8 \pm 6.76	14.8 \pm 2.44	44.4 \pm 10.8	50.9 \pm 14.0
Min–max	19.4–81.5	13.0–33.9	11.4–19.4	21.5–61.4	33.2–74.7
Geomean	34.3	21.9	14.6	42.9	49.3
P value ¹	NS	NS	NS	NS	0.027

All samples were detected

* $p < 0.05$

¹ p value for t -test comparing between species

NS = not significant

concentrations in bones were higher in grey heron chicks ($p = 0.008$).

Lead Concentrations in Tissues

Lead concentrations in livers, kidneys, muscles, and feathers were not significantly different between black-crowned night heron and grey heron chicks (Table 5). However, concentrations in bones were higher in grey heron chicks than in black-crowned night heron chicks ($p = 0.001$).

Cadmium Concentrations in Tissues

Cadmium concentrations in feathers did not differ between black-crowned night heron and grey heron chicks

(Table 6). However, concentrations in liver ($p = 0.002$), kidney ($p = 0.001$), and muscle ($p = 0.046$) were higher in black-crowned night heron than in grey heron chicks.

Discussion

In this study, iron, zinc, and copper concentrations differed in feathers of two heron species in Korea, but did not in other tissues. Manganese concentrations in liver, kidney, and bones differed between black-crowned night heron and grey heron chicks. Wild birds had species-specific bioaccumulation of heavy metal during the growth stages of chicks (Honda et al. 1986). Concentrations of essential heavy metals such as iron, manganese, zinc, and copper in chick tissues varied according to the difference of growth

Table 3 Copper concentrations ($\mu\text{g}/\text{wet g}$) in black-crowned night heron and grey heron chicks from Pyeongtaek, Korea, 2001

	Livers	Kidneys	Muscles	Bones	Feathers**
Black-crowned night heron ($n = 8$)					
Mean \pm SD	55.5 \pm 31.4	10.9 \pm 1.69	4.14 \pm 1.34	1.69 \pm 0.58	9.93 \pm 2.26
Min–max	24.3–98.1	7.66–12.5	2.86–6.40	0.58–2.46	6.76–12.8
Geomean	47.9	10.8	3.97	1.59	9.70
Grey heron ($n = 9$)					
Mean \pm SD	33.9 \pm 15.5	10.8 \pm 2.38	5.86 \pm 1.94	2.02 \pm 0.98	30.2 \pm 12.9
Min–max	22.0–61.1	6.62–13.3	3.10–9.34	0.81–2.96	14.4–48.9
Geomean	31.4	10.5	5.58	1.81	27.7
P value ¹	NS	NS	NS	NS	<0.001

All samples were detected

** $p < 0.01$

¹ p value for t -test comparing between species

NS = not significant

Table 4 Manganese concentrations ($\mu\text{g}/\text{wet g}$) in black-crowned night heron and grey heron chicks from Pyeongtaek, Korea, 2001

	Livers**	Kidneys**	Muscles	Bones**	Feathers
Black-crowned night heron ($n = 8$)					
Mean \pm SD	3.26 \pm 0.68	1.43 \pm 0.27	0.57 \pm 0.32	1.34 \pm 0.50	27.0 \pm 29.8
Min–max	2.49–4.20	1.00–1.89	0.16–1.15	0.89–2.04	3.50–88.2
Geomean	3.20	1.41	0.49	1.26	16.1
Grey heron ($n = 9$)					
Mean \pm SD	1.50 \pm 0.58	0.84 \pm 0.34	0.32 \pm 0.18	3.17 \pm 1.31	21.0 \pm 16.3
Min–max	0.80–2.54	0.37–1.55	0.14–0.75	0.65–4.21	4.31–56.8
Geomean	1.41	0.78	0.28	2.77	16.2
p value ¹	<0.001	0.002	NS	0.008	NS

All samples were detected

** $p < 0.01$

¹ p value for t -test comparing between species

NS = not significant

rate for each tissue, the essential element requirements for each tissue, the physiologically required amount for the gradual increase of the movement of nestling, the necessary amount for feather formation and growth, the ossification which appeared with bone growth, and the concentration dilution by growth rate of chicks (Honda et al. 1986; Burger and Gochfeld 1993).

Concentrations of iron, manganese, copper, and cadmium were relatively high in livers and kidneys, but zinc and lead were not. The highest accumulation of manganese was in feathers. High concentrations of manganese, zinc, and lead were found in bones and feathers. This has also been reported in other wild bird species (Hulse et al. 1980; Cheney et al. 1981; Mateo et al. 2003). In general, zinc and lead were accumulated in calcareous tissues such as bone and eggshell (Underwood 1971; Sorensen 1991) and their

concentrations were higher in bones than in liver (Honda et al. 1986; Kim et al. 2000). In this study, zinc concentrations were higher in bones than in liver but lead concentrations were not.

We reported in previously studies that concentrations of essential elements such as iron, zinc, manganese, and copper in livers and feathers were not related with diet concentrations in black-crowned night heron and grey heron chicks (Kim and Koo 2007a, b). Zinc is an essential elements in birds and the threshold level for zinc toxicosis is 1,200 $\mu\text{g}/\text{g}$ (Gasaway and Buss 1972); Takekawa et al. (2002) found a negative relationship between zinc and body mass. Manganese may become toxic at high levels (Boncompagni et al. 2003). In barn owls (*Tyto alba*), manganese concentrations did not vary with body condition (e.g., protein and fat reserve), but were positively

Table 5 Lead concentrations ($\mu\text{g}/\text{wet g}$) in black-crowned night heron and grey heron chicks from Pyeongtaek, Korea, 2001

	Livers	Kidneys	Muscles	Bones**	Feathers
Black-crowned night heron ($n = 8$)					
Mean \pm SD	1.72 \pm 0.64	0.07 \pm 0.06	0.25 \pm 0.16	0.11 \pm 0.04	0.33 \pm 0.11
Min–max	1.12–3.04	0.01–0.80	0.12–0.63	0.08–0.17	0.21–0.52
Geomean	1.64	0.05	0.22	0.10	0.31
Grey heron ($n = 9$)					
Mean \pm SD	1.97 \pm 1.10	0.03 \pm 0.01	0.15 \pm 0.10	0.61 \pm 0.42	0.25 \pm 0.27
Min–max	0.84–4.02	0.02–0.04	0.04–0.37	0.06–1.52	0.17–0.41
Geomean	1.74	0.03	0.12	0.44	0.24
P value ¹	NS	NS	NS	0.001	NS

All samples were detected

** $p < 0.01$

¹ p value for t -test comparing between species

NS = not significant

Table 6 Cadmium concentrations ($\mu\text{g}/\text{wet kg}$) in black-crowned night heron and grey heron chicks from Pyeongtaek, Korea, 2001

	Livers**	Kidneys**	Muscles*	Bones	Feathers
Black-crowned night heron ($n = 8$)					
Mean \pm SD	13.5 \pm 2.30	6.61 \pm 2.54	5.25 \pm 5.91	4.82	116 \pm 55.5
Min–max	10.8–16.7	3.45–11.3	ND–17.8	ND–4.82	5.91–136
Geomean	13.4 (8) ^a	6.23 (8)	3.27 (6)	4.82 (1)	97.5 (8)
Grey heron ($n = 9$)					
Mean \pm SD	10.3 \pm 1.59	2.31 \pm 1.29	1.37 \pm 0.90	16.3 \pm 18.1	272 \pm 399
Min–max	8.41–12.7	0.46–5.05	ND–1.76	ND–59.1	97.7–1,403
Geomean	10.2 (9)	1.96 (9)	1.10 (8)	8.91 (8)	178 (9)
p value ¹	0.002	0.001	0.046	–	NS

^a Number of samples detected

* $p < 0.05$; ** $p < 0.01$

¹ p value for t -test comparing between species

ND was not detected

NS = not significant

correlated with liver size (Esselink et al. 1995). High copper contamination was reported without any sign of toxicity (Clausen and Wolstrup 1978; Kim et al. 1996) and copper concentrations in birds were elevated in areas of human activity and intensive copper use (Eisler 2000). High copper concentrations in livers of wild birds were associated with low protein and fat reserve (Esselink et al. 1995). However, zinc, manganese, and copper concentrations in this study were far below the level associated with toxicity. Therefore, we suggest that essential elements in two Korean heron species are within the normal range for herons and wild birds (Honda et al. 1986; Lee et al. 1987; Kim et al. 1996; Kim et al. 2000; Rattner et al. 2000; Custer et al. 2007) and are maintained there by the normal homeostatic mechanism.

Heavy metal concentrations in livers, feathers, eggs, and eggshells showed significant difference between herons and egrets in many studies (Burger et al. 1992; Burger and Gochfeld 1993; Fasola et al. 1998; Spahn and Sherry 1999; Connell et al. 2002; Boncompagni et al. 2003; Ayaş 2007; Kim and Koo 2007a, b) and in this study. Researchers suggested that the differences attributed to differences in diet concentrations in herons and egrets, environment pollution in breeding site, and migration conditions.

Lead has been responsible for acute incidents of birds poisoning. In this study, lead concentrations in livers and kidneys were not different to the two heron species chicks, and were within the background level of lead in wild birds (Clark and Scheuhammer 2003). Lead concentrations in livers of herons and egrets were within the range of the

background level (Hulse et al. 1980; Cheney et al. 1981; Custer and Mulhern 1983; Blus et al. 1985; Honda et al. 1986; Lee et al. 1987; Cosson et al. 1988; Husain and Kaphalia 1990; Hontelez et al. 1992; Rodgers 1997; Kim et al. 2000; Custer et al. 2007; Kim and Koo 2007a). However, in Korea, black-crowned night heron chicks and great egret adults from the Siheung colony and black-crowned night heron chicks from the Gwangju colony were within the a range consistent with lead exposure (Jeong 1998).

In this study, cadmium concentrations in liver and kidney in black-crowned night heron and grey heron chicks did not exceed the background level of wild birds (Scheuhammer 1987) and other studies also did not exceed the background concentration (Hulse et al. 1980; Cheney et al. 1981; Custer and Mulhern 1983; Blus et al. 1985; Nims 1987; Cosson et al. 1988; Husain and Kaphalia, 1990; Hontelez et al. 1992; Guitart et al. 1994; Mora and Anderson 1995; Rattner et al. 2000; Custer et al. 2007; Kim and Koo 2007a).

Lead and cadmium are often selected for ecotoxicological assessment in herons and egrets because of their toxicities (Fasola et al. 1998; Connell et al. 2002; Boncompagni et al. 2003). In heron and egret chicks, lead and cadmium concentrations were affected by the local contamination (sediment, air, and diet) of their colonies (Burger et al. 1992; Spahn and Sherry 1999; Rattner et al. 2000; Boncompagni et al. 2003; Kim and Koo 2007a, b). We suggest that lead and cadmium concentrations in herons and egrets reflect the local lead and cadmium contaminations surrounding the breeding site. Therefore, lead and cadmium concentrations in heron and egret chicks are good bioindicators of local pollution.

Lead poisoning in waterfowl is related to the ingestion of lead shot or sinkers (Scheuhammer and Norris 1996). However, herons and egrets were not found to be suffering lead poisoning because they do not ingest lead shot or sinker (Custer 2000). Scheuhammer (1987) reported that dietary lead concentrations less than 100 µg/g dry weight did not cause significant reproductive impairment in wild birds. Lead-contaminated diet did not affect osprey (*Pandion haliaetus*) reproduction in Idaho, USA (Henny et al. 1991; Clark et al. 2001). Therefore, we do not expect lead to present a hazard to the two heron species: lead concentrations in diet were far below the thresholds for adverse effects (Kim and Koo 2007a, b).

The critical organ in chronic cadmium toxicity is generally considered to be the kidney (Nordberg 1971). With continued cadmium exposure, even at a low sublethal dietary level, there is a continual increase in the cadmium content of the renal cortex concurrent with an increase in renal metallothionein concentration (Scheuhammer 1987). For cadmium, liver-to-kidney concentration ratios greater

than 1 indicate acute exposure to relatively high dietary cadmium concentrations and ratios less than 1 indicate chronic exposure to low levels (Scheuhammer 1987). The ratio was greater than 1 in this study, but other studies have detected ratios less than 1 (Honda et al. 1986; Kim et al. 2000) for Korean egrets and herons. We suggest that this can be attributed to cadmium being higher in the diet at this colony than in other areas in Korea.

In conclusion, heavy metal concentrations differed between the two heron species studied. Lead and cadmium levels were within the range of the background level and their concentrations did not suggest a threat to the reproductive success of black-crowned night herons and grey herons. Lead concentrations were higher in liver than in bones and cadmium concentrations were higher in livers than in kidney. Therefore, lead and cadmium contaminations were the result of acute not chronic exposure at this study site.

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