

Monitoring of Heavy Metal Contamination Using Tissues of Two Ardeids Chicks, Korea

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Abstract This study presents concentrations of iron, zinc, manganese, lead and cadmium in livers and kidneys of Little Egret *Egretta garzetta* (n = 10) and Black-crowned Night-Heron *Nycticorax nycticorax* (n = 10) chicks from Pyeongtaek, Gyeonggi-do, Korea. Essential element concentrations such as iron, zinc and manganese did not differ between both species chicks in livers and kidneys, but lead and cadmium concentrations were significantly greater in both tissues of Little Egret chicks (lead 2.78 ± 4.06 µg/g wet weight, cadmium 10.3 ± 12.8 kg/g wet weight) than in those of Black-crowned Night-Heron chicks (lead 0.92 ± 0.73 µg/g wet weight, cadmium 1.00 ± 1.00 kg/g wet weight). Lead and cadmium concentrations in livers of Black-crowned Night-Heron chicks were highly related to sediment and/or prey concentrations of their foraging sites in Korean studies. It shows that lead and cadmium concentrations in livers of heron chicks can reflect those of surrounding environment of breeding sites. In this study, cadmium concentrations were higher in kidneys than in livers and it's not recent high-level exposure but chronic background exposure to cadmium contamination around breeding site. Therefore, we suggest that cadmium concentrations in livers and kidneys can be used as a bioindicator of acute and/or chronic local contamination.

Keywords Little Egret chicks · Black-crowned Night-Heron chicks · Livers · Kidneys · Heavy metals

Piscivorous birds such as herons and egrets 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 tropic 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 km of the nesting colony (Kim and Koo 2007a).

The most common poisons in avian species are heavy metals and agricultural chemicals. For example, lead is one of the main causes of poisoning through direct or indirect exposure by sinkers and lead shots (Johansen et al. 2004). Lead and cadmium can be highly toxic to the organisms, affecting several physiological and endocrine systems at environmentally relevant concentrations (Martin et al. 2003).

In egret and herons, the concentration of essential heavy metals such as iron, manganese and zinc in tissues varied according to the difference of growth speed in each tissue, the amount required for tissue growth in case of essential element, but, pollution elements such as lead and cadmium were bioaccumulated with age (Honda et al. 1986).

In heron and egret chicks, differences of trace metal concentrations in feathers reported between species in a same colony (Burger et al. 1992; Kim and Koo 2007b) and among locations in Pakistan (Boncompagni et al. 2003), but they were not evaluated for growth rate of heron and

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egret chicks. Han (2001) reported diet differences of chick Little Egrets and Black-crowned Night-Herons in same site of this study. And some researchers (Burger et al. 1992; Boncompagni et al. 2003; Kim and Koo 2007b) reported differences of heavy metal concentrations among wild birds by their habitat and diet levels.

In this paper, we examine differences of heavy metal concentration in livers and in kidneys of chick Little Egrets and Black-crowned Night-Herons nesting at Pyeongtaek heronry, Gyeonggi-do, Korea, provide lead and cadmium levels of herons and egrets, and relationship of lead and cadmium concentrations among in sediment, preys and tissues from Korea.

Materials and Methods

Field examination of this study was carried out during the 2000 breeding season (April–June) at a breeding site in Pyeongtaek city (37° 02'N, 127° 02'E), Gyeonggi-do, Korea, where Little Egrets and Black-crowned Night-Herons frequently nest in the same colony. Rice paddy fields surround the colony where many herons foraged.

Chick Little Egrets ($n = 10$) and Black-crowned Night-Herons ($n = 10$) were marked by plastic ring in 1–3 days after hatching, and were captured in 20–22 days after hatching from the different nests.

The chicks were weighed (limit = 0.1 g), and the bill (limit = 0.1 mm), wing (limit = 0.1 mm) and tarsus (limit = 0.1 mm) length were measured. Chicks were euthanized by thoracic compressions and frozen at -20°C until they were dissected. Livers and kidneys were separately dissected from the body of the specimens and stored in chemically clean jars.

The concentration of iron, zinc, manganese, lead and cadmium was determined by flame atomic absorption spectrophotometry (Hitachi Z-6100), after mineralization of samples with nitric, sulfuric, and perchloric acid in kjeldahl flasks. Tissues with low lead, cadmium concentration were measured by AA spectrophotometry, after treatment with DDTC (Sodium N, N-Diethyldithio-carbamate trihydrate ($(\text{C}_2\text{H}_5)_2\text{NCS}_2\text{Na}\cdot 3\text{H}_2\text{O}$)-MIBK (Methyl Isobutyl Ketone ($\text{CH}_3\text{COCH}_2\text{CH}(\text{CH}_3)_2$)) (Honda et al. 1986). Spikes and blanks were run at a frequency of at least 5% of the total number of samples. To assess accuracy, a standard, a spike, a blank 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 ($\mu\text{g/g}$) in tissues were estimated on wet weight basis. Detection limit was 1.0 $\mu\text{g/g}$ for iron, zinc, and manganese 0.1 $\mu\text{g/g}$ for lead, and 0.01 $\mu\text{g/g}$ for cadmium.

We statistically tested for differences (growth and heavy metal concentrations) between chick Little Egrets and Black-crowned Night-Herons using t -test. We log transformed data to satisfy the homogeneity of variance assumptions of t -test (Kim and Koo 2007b). We used SPSS 10.0 version for statistical analysis.

Results and Discussion

Chick Black-crowned Night-Herons had significantly higher weight (t -test, $p < 0.01$), wing length (t -test, $p < 0.05$) and tarsus length (t -test, $p < 0.05$) than chick Little Egrets at 20–22 days after hatching, but bill length was not different between Little Egret and Black-crowned Night-Heron chicks (Table 1).

Iron, zinc and manganese concentrations did not significantly differ in livers and kidneys between Black-crowned Night-Heron chicks and Little Egret chicks (Tables 2, 3). In laboratory and natural environment, clinical signs and diagnosis of zinc poisoning in wild birds was more than 280 $\mu\text{g/dry weight g}$ in livers (Sileo et al. 2003). Manganese may become toxic at high levels (Boncompagni et al. 2003). In Barn Owls *Tyto alba*, manganese concentrations were positively correlated with liver size (Esselink et al. 1995). However, zinc and manganese concentrations in this study were far below the level associated with toxicity. Therefore, we suggest that essential elements in Korean Little Egret and Black-crowned Night-Heron chicks are within normal range for herons and egrets (Honda et al. 1986; Jeong 1998; Kim et al. 2000; Custer et al. 2007) and are maintained there by normal homeostatic mechanism.

Lead (t -test, $p < 0.05$) and cadmium (t -test, $p < 0.01$) concentrations in livers were higher Little Egret chicks than Black-crowned Night-Heron chicks (Table 2). Lead has been responsible for acute incidents of birds poisoning. However, herons and egrets generally do not ingest lead shot (Custer 2000). Lead concentrations in livers of Little

Table 1 Measurements of chick Black-crowned Night-Herons and Little Egrets after hatching 20–22 days

	Weight (g)	Bill (mm)	Wing (mm)	Tarsus (mm)
Little Egret ($n = 10$)				
Mean $\pm$ SD	325 $\pm$ 26.3	47.0 $\pm$ 0.63	120 $\pm$ 26.5	61.7 $\pm$ 1.46
Range	308–355	46.9–47.3	90.0–140	60.0–62.8
Black-crowned Night-Heron ($n = 10$)				
Mean $\pm$ SD	439 $\pm$ 56.3	48.5 $\pm$ 3.23	158 $\pm$ 22.4	69.5 $\pm$ 3.87
Range	312–540	41.8–51.4	123–185	63.0–73.4
p -value	$p < 0.01$	NS	$p < 0.05$	$p < 0.05$

NS was not significant

Table 2 Heavy metal concentrations (arithmetic mean, $\mu\text{g/g}$ wet weight) of livers in chicks of Little Egret and Black-crowned Night-Heron

	Iron	Zinc	Manganese	Lead	Cadmium ^a
Little Egret (n = 10)					
Mean $\pm$ SD	192 $\pm$ 102	46.5 $\pm$ 20.4	3.49 $\pm$ 1.44	2.78 $\pm$ 4.06	10.3 $\pm$ 12.8
Range	84.6–390	23.5–84.1	2.41–3.23	ND–15.3	ND–14.5
Black-crowned Night-Heron (n = 10)					
Mean $\pm$ SD	298 $\pm$ 49.1	65.2 $\pm$ 12.5	3.32 $\pm$ 0.85	0.92 $\pm$ 0.73	1.00 $\pm$ 1.00
Range	235–366	43.1–75.6	2.47–4.91	ND–1.97	ND–2.00
p-value	NS	NS	NS	$p < 0.05$	$p < 0.01$

^a Was $\mu\text{g/wet kg}$

NS was not significant

Egrets from Carmague, France (Cosson et al. 1988), Black-crowned Night-Heron chicks from Agassiz National Wildlife Refuge, USA (Custer et al. 2007) and Great Egrets from Korea (Honda et al. 1986) were within the range of the background level ($<2 \mu\text{g/wet weight g}$) for wild birds (Scheuhammer 1987). In this study, liver lead concentrations in all species were lower than the toxic level ($<10 \mu\text{g/wet weight g}$) for lead (Scheuhammer 1987). One Little Egret's sample exceeded the toxic level. Jager et al. (1996) reported that $10 \mu\text{g/g}$ dry weight (approximately $3.70 \mu\text{g/g}$ wet weight) lead in kidneys correspond to environmental contamination. In this study, kidney lead concentrations were lower than $3.70 \mu\text{g/g}$ wet weight in all egrets and herons.

In kidneys, lead (t -test, $p < 0.01$) and cadmium (t -test, $p < 0.01$) concentrations were much higher Little Egret chicks than Black-crowned Night-Heron chicks (Table 3). Herons generally do not accumulate cadmium (Custer 2000). In this study, the two ardeids did not exceed the background level in livers (approximately $1 \mu\text{g/g}$ wet weight) and kidneys (approximately $2.67 \mu\text{g/g}$ wet weight) of wild birds (Scheuhammer 1987). Cadmium in livers were also at background levels in Little Egrets from Carmague, France (Cosson et al. 1988), Black-crowned Night-Herons from Delaware Bay, USA (Rattner et al. 2000) and

other egrets and herons from Korea (Jeong 1998; Kim et al. 2000).

In Korean studies, lead concentrations in livers and kidneys of Black-crowned Night-Heron chicks from Siheung city (mean 4.70 and $17.6 \mu\text{g/g}$ wet weight, respectively) (Jeong 1998) were higher than those of and Gwangju city (mean 3.58 and $2.96 \mu\text{g/g}$ wet weight) (Kim et al. 2000) and this study (mean 0.92 and $1.19 \mu\text{g/g}$ wet weight). Also, cadmium concentrations in livers and kidneys from Siheung city (mean 0.09 and $0.30 \mu\text{g/g}$ wet weight) (Jeong 1998) were higher than those of Gwangju city (mean 0.04 and $0.12 \mu\text{g/g}$ wet weight) (Kim et al. 2000) and this study (mean 0.001 and $0.002 \mu\text{g/g}$ wet weight).

In sediment and/or preys, lead concentrations were relatively higher in foraging sites of the Siheung and Gwangju colony than in those of the Pyeongtaek colony. In cadmium, prey concentrations were relatively higher in the Gwangju colony than in those of the Pyeongtaek colony (Jeong 1998; Shim et al. 1998; Kim et al. 1999, 2000) (Table 4). In Siheung breeding site, Sihwa Lake is one of important foraging site of herons and egrets (Jeong 1998) and lead concentrations of water and sediments in Sihwa Lake were much higher than other wetlands in Korea (Shim et al. 1998).

Table 3 Heavy metal concentrations (arithmetic mean, $\mu\text{g/g}$ wet weight) of kidneys in chicks of Little Egret and Black-crowned Night-Heron

	Iron	Zinc	Manganese	Lead	Cadmium ^a
Little Egret (n = 10)					
Mean $\pm$ SD	77.4 $\pm$ 17.9	18.5 $\pm$ 1.65	2.37 $\pm$ 1.89	8.93 $\pm$ 5.50	18.4 $\pm$ 26.5
Range	52.2–110	15.5–20.1	1.26–3.91	2.16–18.8	ND–79.8
Black-crowned Night-Heron (n = 10)					
Mean $\pm$ SD	58.9 $\pm$ 26.9	17.4 $\pm$ 5.18	2.29 $\pm$ 0.87	1.19 $\pm$ 1.12	2.20 $\pm$ 1.10
Range	31.9–92.6	11.8–26.1	1.17–3.50	ND–2.32	ND–4.30
p-value	NS	NS	NS	$p < 0.01$	$p < 0.01$

^a Was $\mu\text{g/wet kg}$

NS was not significant

Table 4 Lead and cadmium concentrations ($\mu\text{g/g}$ wet weight) in sediment, prey and livers of Black-crowned Night-Heron chicks from Korea

	Sediment	Prey	Livers
Lead			
Siheung	41.8 ± 16.5^a	–	4.70 ± 2.92^b
Gwangju	45.6 ± 17.9^c	4.14 ± 3.04^d	3.49 ± 1.49^d
Pyeongtaek	3.67 ± 2.47	0.66 ± 0.33	0.92 ± 0.73
Cadmium			
Siheung	0.06 ± 0.58^a	–	0.09 ± 0.05^b
Gwangju	–	0.036 ± 0.026^d	0.043 ± 0.023^d
Pyeongtaek	3.00 ± 0.79	0.003 ± 0.002	0.001 ± 0.001

^a From Shim et al. (1998)^b From Jeong (1998)^c From Kim et al. (1999)^d From Kim et al. (2000)

As a result, we suggest that high lead concentrations in livers of Black-crowned Night-Heron chicks from the Siheung and Gwangju colony were related to those of their diet and the sediment concentrations of their foraging sites surrounding breeding sites. Also, cadmium concentrations in livers were related to their prey concentrations. In addition, elevated lead concentrations in ardeids were reported to lead contaminated area near downtown from Cairo, Egypt (Burger et al. 1992) and lead contaminated environment from Pakistan (Boncompagni et al. 2003).

This study did not differ in zinc, manganese and iron but did differ in lead and cadmium between species. In Pakistan (Boncompagni et al. 2003), herons and egrets partly differed significantly in eggs, while they did not differ in feathers between species in a same area. They suggested that the same feeding site caused no difference in feathers. However, cadmium and lead concentrations in feathers and tissues of herons and egrets were different to their habitat environment (Burger et al. 1992) and their diet (Kim and Koo 2007b). Therefore, we suggest that lead and cadmium difference from Pyeongtaek is attributed to different food materials of herons and egrets (Han 2001). Furthermore, detailed investigation will be necessary to analyze the prey materials of Black-crowned Night-Herons and Little Egrets.

With continued cadmium exposure, even at low sublethal dietary level, there is a continual increase in the cadmium content of the renal cortex concurrent with an increase in the renal metallothionein concentration (Scheuhammer 1987; Elliott and Scheuhammer 1997). For cadmium, liver-to-kidney concentration ratios more than 1 indicate acute exposure to relatively high dietary cadmium concentrations and less than 1 indicate chronic exposure to low levels (Scheuhammer 1987). In this study, the ratio of

cadmium liver-to-kidney ratio in herons and egrets were consistent with chronic background exposure. This trend was reported some wild birds (Elliott and Scheuhammer 1997; Jeong 1998; Kim et al. 2000, 2009). We suggest that the local and species differences of acute and/or chronic contaminations of cadmium are attributed to cadmium concentrations in their diet and environmental pollution (Kim and Koo 2007b).

Comparing lead and cadmium in chick livers and kidneys of Black-crowned Night-Herons with those of Little Egrets bred in Pyeongtaek heronry, results showed that the levels were much higher in the latter than in the former. However, because breeding success was higher of Little Egrets (62.7%) than of Black-crowned Night-Herons (50.7%) (Han 2001), therefore, we suggest that the relatively higher concentration of lead and cadmium concentrations in Little Egret chicks than in Black-crowned Night-Heron chicks did not affect the reproductive success of these.

Iron, zinc and manganese concentrations did not significantly differ in livers and in kidneys between Black-crowned Night-Heron and Little Egret chicks. However, lead and cadmium concentrations were significantly higher in Little Egret chicks than in Black-crowned Night-Heron chicks in both tissues. We suggested that lead and cadmium difference from Pyeongtaek heronry was attributed to different food items of herons and egrets. In Little Egret and Black-crowned Night-Heron chicks, lead and cadmium concentrations were within the background levels for wild birds. It seems unlikely that the extent of metal contaminant exposure threatens physiological and biological processes of Korean herons and egrets. In Korean studies, lead and cadmium concentrations in the liver of Black-crowned Night-Heron chicks were highly related to those in sediment and/or preys of their foraging sites. Therefore, we suggest that liver lead and cadmium concentrations of egrets and herons can reflect environment of breeding sites. In this study site, the breeding success was 62.7% on Little Egrets and 50.7% on Black-crowned Night-Herons. Therefore, lead and cadmium concentrations in both species did not affect the reproductive success of these.

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