Relationships of Metals Between Feathers and Diets of Black-Tailed Gull (*Larus crassirostris*) Chicks

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Abstract In birds, metal contaminants in feathers are influenced by prey concentrations and environmental quality. In Black-tailed Gull chicks, Cd, Pb and Cu concentrations were strongly correlated between feathers and stomach contents. Between feathers and livers, Pb, Zn and Fe concentrations were significantly correlated. Cd concentrations were within the range of other seabirds and within the background level for bird feathers (<2 μ g/ g dw). At the lighthouse, eight chicks exceeded the background for Pb level in feathers (>4 μ g/g dw). Elevated Pb concentrations might be attributed to ingestion of paintbased chips and natural (soil and rocks) sources. There is evidence that the analyzed birds suffered from acute toxicity, including high levels of pecking from conspecifics and increased mortality from elevated Pb levels. It seems likely that these birds might experience negative health effects from this increased Pb exposure. As a result, Blacktailed Gull chick feathers are a very useful monitoring tool for assessing Cd, Pb and Cu contamination. Essential elements such as Fe, Zn, Mn and Cu were all within the acceptable range of normal concentrations for seabird species including gulls and may be maintained by normal homeostatic mechanisms.

Keywords Black-tailed Gull chicks \cdot Feathers \cdot Stomach contents \cdot Pb \cdot Threshold level

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J. Kim e-mail: herons@khu.ac.kr Birds that forage in aquatic environments are more vulnerable to contaminants than terrestrial birds because contaminants can quickly move in water and can be stored in bottom sediments and intertidal zones. Metal concentrations can be varied in birds because of their differing feeding habits. They affect birds both indirectly by reducing food supplies and directly by affecting reproduction, behavior, and the hematopoietic and nervous systems (Burger and Gochfeld 2000a, b).

Bird feathers can be used as bioindicators of metal contamination because: (1) birds sequester metals in their feathers, (2) the proportion of body burden in feathers is relatively constant for each metal, (3) a relatively high proportion of the body burden of certain metals is stored in the feathers (Burger and Gochfeld 2009), and (4) feather concentrations of metal contaminants are highly correlated with the diet of wild birds including seabirds (Kim and Koo 2007; Paiva et al. 2008). Breast feathers are well-known as the best bioindicator of whole-body burdens, and the feathers are easy to collect non-invasively and to store for decades or longer, making them especially useful for establishing temporal and spatial patterns without impacting populations (Burger and Gochfeld 2009).

In Korea, Black-tailed Gulls (*Larus crassirostris*) are permanent residents and breed on uninhabited islands. Some breeding sites, including Hongdo Island, are listed as a natural monument and are protected by the Korean government. Approximately 30,000 pairs of Black-tailed Gulls breed on Hongdo Island. One objective of this study was to determine levels of two non-essential elements, Cd and Pb, and four essential elements, Cu, Mn, Zn and Fe, in feathers of Blacktailed Gull chicks. Other objectives were to examine the effects of environmental contamination on chick metal concentrations and to examine the correlation between longterm exposure in feathers and short-term exposure in livers.

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A dilapidated lighthouse is present on Hongdo Island. Because of visible paint chips associated with the structure, we hypothesized that lead and cadmium concentrations would be higher in gull chicks from nests near the lighthouse.

Materials and Methods

Twenty individual Black-tailed Gull chicks and stomach contents were collected from Hongdo Island (34°32'N latitude, 128°43'E longitude), Gyeongsangnam-do, Korea, in 2007.

At Hongdo Island, we collected twenty breast feather samples from twenty gull chicks. Multiple breast feathers (>10 feathers per sample) were included in each sample. We also collected stomach contents (n = 20) from each chick. For Pb and Cd concentrations, we selected two study sites: a "reference" site (n = 10) with no known source of Pb contamination, and "lighthouse" site (n = 10). The ground of the lighthouse site contained visible leaded paint chips. We collected birds nesting close to (<5 m, lighthouse site), and distant from, the lighthouse (>50 m, reference site) and analyzed Pb and Cd levels.

Black-tailed Gull chicks were marked with plastic rings 1–3 days after hatching and were recaptured 20–22 days after hatching. Body mass (0.1 g), culmen (0.1 mm) and tarsus (0.1 mm) length were measured. Chicks were euthanized by thoracic compression and frozen at -20° C until necropsy. These birds were later thawed and the breast feathers and livers carefully removed from the body and weighed (±0.1 g). Both tissues were dried in an oven for 24 h at 105°C and weighed (±0.1 g). Both terpresentative of the plumage as a whole and do not reflect molt sequence as do flight feathers (Burger and Gochfeld 2009). All metal concentrations (µg/g) in feathers, livers and stomach contents were estimated on a dry weight (dw) basis.

Fe, Zn and Mn concentrations were determined by flame atomic absorption spectrophotometry (AAS; Hitachi Z-6100), after mineralization of samples with nitric, sulfuric, and perchloric acid in Kjeldahl flasks. Tissues with low Pb and Cd concentrations were measured by AAS following treatment with DDTC (sodium *N*,*N*-diethyldithio-carbamate trihydrate ((C₂H₅)₂NCS₂Na·3H₂O)-MIBK (methyl isobutyl ketone (CH₃COCH₂CH(CH₃)₂) (Kim and Koo 2007). Seven or more spikes and blanks were included in the analysis (about 20 % of the total number of samples). A spike, a blank, a standard and a sample were run in triplicate in each analytical run. Spikes recoveries ranged from 94 % to 106 %. Recovered concentrations of the samples were within 5 % of the certified values. Detection limits were 1.0 µg/g dw for Fe, Zn and Mn, 0.1 µg/g for Pb, and 0.01 µg/g for Cd. Because some of the variables were not normally distributed, nonparametric tests were used for statistical analyses. Differences in metal concentrations between lighthouse and reference sites for Black-tailed Gull chicks were assessed using Mann–Whitney's *U*-test. And, we used Kolmogorov–Smirnov test for normality. We present geometric means, 95 % confidence intervals, arithmetic mean and standard deviation in tables and text. Correlations among metal concentrations between feathers and stomach contents and between feathers and livers from Hongdo Island were assessed using Pearson correlations (r). Statistical analyses were carried out using SPSS 12.0 version with a significance level of p < 0.05.

Results

Cd, Pb, Cu, Mn, Zn and Fe concentrations were detected in all feather and stomach content samples of Black-tailed Gull chicks collected at Hongdo Island in 2007 (Table 1). Cd (r = 0.483, p < 0.05), Pb (r = 0.731, p < 0.01) and Cu (r = 0.479, p < 0.05) concentrations were significantly correlated between the feathers and stomach contents for gull chicks (Fig. 1). In contrast, Mn, Zn and Fe concentrations were not correlated.

Pb (r = 0.569, p < 0.01), Zn (r = 0.485, p < 0.05) and Fe (r = 0.501, p < 0.05) concentrations were significantly correlated between the feathers and livers of Black-tailed Gull chicks. Cd, Mn and Zn concentrations were not correlated between feathers and livers.

Feathers of Black-tailed Gull chicks at the lighthouse site had significantly higher Cd concentrations than at the reference site. In contrast, Cd concentrations in stomach contents were not significantly different between the two sites. Pb concentrations in the stomach contents of gull chicks at the lighthouse site were significantly higher than at the reference site. However, Pb concentrations in feathers were not significantly different between the two sites (Table 2).

Discussion

In seabirds including gull species, metal contaminant concentrations such as Cd, Pb and Hg in tissues were influenced by their prey concentrations (Riget et al. 2000; Paiva et al. 2008). Metal concentrations can be influenced by contamination of environmental media including water, sediment and air (Janssens et al. 2001; Dauwe et al. 2002; Boncompagni et al. 2003). Also, regarding essential elements, Cu can fluctuate by diet and environmental contamination from water, sediment and industrialization (Boncompagni et al. 2003; Tiller et al. 2005; Custer et al.

	Cadmium	Lead	Copper	Manganese	Zinc	Iron
Feathers $(n = 20)$						
Geomean	0.07	2.56	4.48	0.91	214	50.1
CI	0.04-0.11	1.14-3.98	2.44-6.53	0.39-1.43	119-309	21.3-78.9
Mean \pm SD	0.08 ± 0.03	3.24 ± 1.75	4.67 ± 1.39	1.19 ± 0.95	217 ± 33.7	65.8 ± 38.4
Stomach contents	(n = 20)					
Geomean	0.17	19.9	9.43	53.4	107	1,672
CI	0.09-0.25	8.38-31.5	4.89-14.0	19.0-87.8	41.7-172	757–2,587
Mean \pm SD	0.19 ± 0.10	26.4 ± 20.8	10.4 ± 4.18	78.6 ± 61.9	141 ± 95.4	$2,088 \pm 1,184$

Table 1 Concentrations (geometric mean, 95 % confidence intervals (CI) and mean \pm SD, $\mu g/g$ dw) of heavy metals in feathers and stomach contents of Black-tailed Gull chicks





Table 2 Cadmium and lead
concentrations (geometric
mean, 95 % CI and
mean \pm SD, μ g/g dw) in
stomach contents and livers at
the lighthouse and reference
sites

results between sites ^b NS was not significant

able 2 Cadmium and lead		Cadmium	Cadmium		Lead			
iean, 95 % CI and		Stomach contents	Feathers	Stomach contents	Feathers			
\pm SD, $\mu g/g dw$) in omach contents and livers at	Lighthouse site $(n = 10)$							
e lighthouse and reference	Geomean	0.19	0.10	78.0	3.13			
tes	CI	0.06-0.33	0.04-0.16	13.2–143	0.51-5.76			
	Mean \pm SD	0.22 ± 0.12	0.10 ± 0.01	105 ± 89.0	4.24 ± 1.86			
	Reference site $(n = 10)$							
	Geomean	0.15	0.05	13.7	2.08			
	CI	0.05-0.25	0.02-0.09	3.95-23.4	0.70-3.47			
Mann–Whitney's U-test	Mean \pm SD	0.16 ± 0.06	0.06 ± 0.02	15.7 ± 7.68	2.24 ± 0.87			
NS was not significant	p value ^a	NS ^b	< 0.01	<0.001	NS			

2007; Horai et al. 2007; Kim and Koo 2007). Also, in Cu contaminated areas, some birds had elevated Cu levels and there were attributed to high Cu contamination (Tiller et al. 2005, Horai et al. 2007). In this study, we found strongly positive relationships of Cd, Pb and Cu between feathers and stomach contents in Black-tailed Gull chicks. For Pb and Cd, effects of prey concentration have been reported in some birds (Ferns and Anderson 1994, 1997; Kim and Koo 2007; Kim et al. 2010). As a result, we suggest that feathers of Black-tailed Gull chicks can reflect local contamination surrounding the breeding colony and can be a very useful monitoring tool for assessing Cd, Pb and Cu contamination. Pb exposure in seabird chicks, but not adults, was associated with ingesting paint chips from a building, and Pb

poisoned chicks from lead-based paint chips previously have been reported (Sileo et al. 1990; Work and Smith 1996). We found high Pb accumulation in feathers and stomach contents at the lighthouse site. Elevated Pb levels in the stomach content of gull chicks from the lighthouse may have resulted from mixing of identified (diets) and unidentified (lead-based paint chips) stomach contents before ingesting and natural (soil and rocks) sources by packing. Elevated Pb concentrations in birds can have negative effects such as decreased egg size, eggshell thickness, and sperm quality, and increased nestling mortality in wild birds (Dauwe et al. 2004).

Cd levels from this study were within the range of those of other seabird studies including gull species, chicks and/ or adults (Burger and Gochfeld 2000b; Burger and Gochfeld 2001; Agusa et al. 2005; Custer et al. 2007; Malinga et al. 2010). Greater than 2 μ g/g dw Cd concentration in feathers was considered as a threshold level of abnormal poisoning that may have an adverse effect on kidneys (Burger and Gochfeld 2000b). Elevated Cd concentrations in feathers were associated with reduction of bone growth rates and fledgling success (Spahn and Sherry 1999). At lower levels, Cd toxicity can cause sublethal and behavioral effects (Burger and Gochfeld 2000b). For Cd in seabird species worldwide, including this study, no seabird species exceeded the background level of 2 μ g/g dw.

Lead and Cd concentrations in Black-tailed Gull chicks were higher or tended to be higher near the lighthouse. Lead concentrations in stomach contents and Cd concentrations in feathers were significantly higher in birds from the lighthouse. Although not significant, the trend was for higher Pb concentrations in feathers and higher Cd concentrations in stomach contents.

In Black-tailed Gull chicks from this study, Pb levels found were greater than in gull and seabird species, chicks and/or adults (Burger and Gochfeld 2000a, b; 2001; Agusa et al. 2005) and were similar to those of Kelp Gull Larus dominicanus adults from Namibia (Burger and Gochfeld 2001), but were lower than those of Laysan Albatross Phoebastria immutabilis droop-winged chicks (mean 16.9 µg/g dw) from Midway Atoll (Burger and Gochfeld 2000b). In feathers, Pb levels of 4 μ g/g dw are known to be a threshold level for toxicity (Burger and Gochfeld 2000b). Elevated Pb levels can cause several behavioral toxicities, neurological alterations, behavioral deficits and growth slowdown (Burger and Gochfeld 2000a, b). In this study, 8 of 20 individuals (40.0 %) exceeded the background level for Pb in feathers, all from the lighthouse site $(4.24-6.35 \ \mu g/g \ dw)$. This further supports the association between Pb concentration and the ingestion of lead paint chips.

In seabird chicks, including gull species, Pb and Cd concentrations were influenced by differences in trophic

level, contamination of food items and foraging sites (Riget et al. 2000; Borgå et al. 2006). Overall, the mean concentration of Cd and Pb reported for gull chicks that nested on Hongdo Island were below the threshold levels for lead and cadmium toxicity in feathers for birds, except for lead concentrations in some individuals at the lighthouse site. As a result, Pb levels of gull chicks at the lighthouse site may negatively affect physiological and ecological parameters. There is evidence that the analyzed birds suffered from acute toxicity such as increased conspecific attacks and increased mortality for elevated Pb levels (Lee 2003). It seems likely that these birds might experience negative health effects from this increased Pb exposure.

Relationships of metals between feathers and livers have been reported for Cd and Pb in shorebird species (Kim and Oh 2012) and Cd in Black-tailed Gulls (Agusa et al. 2005). We found relationships of Pb, Zn and Fe between feathers and livers. The strong correlations between feathers and livers for metal contaminants such as Cd and Pb might be attributed to the small influence of external contamination. Feathers can reflect Cd and Pb concentrations in livers and can be a suitable surrogate for non-lethal monitoring of environmental contamination by metals instead of livers.

The concentrations of Fe, Zn, Mn and Cu quantified in feathers of Black-tailed Gull chicks were below those commonly associated with toxicity. Additionally, bioaccumulation patterns of these elements were consistent with those seen in other birds (Borgå et al. 2006). In this study, Fe, Zn, Mn and Cu concentrations in feathers were within the range from earlier reports of various seabird species including gull species, suggesting that these essential element levels represent either background or normal physiand ecological levels ological for physiological requirements and element metabolic regulations (Burger and Gochfeld 2000b, 2001; Agusa et al. 2005; Custer et al. 2007; Malinga et al. 2010). In conclusion, Fe, Zn, Mn and Cu levels from this study are within the acceptable range of normal concentrations for seabird species, including gull species, and may be maintained by normal homeostatic mechanisms (Agusa et al. 2005).

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