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METAL AND ORGANOCHLORINE CONTAMINANTS IN TISSUES OF NESTLING WADING BIRDS (CICONIIFORMES) FROM SOUTHERN FLORIDA

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Abstract.—Nine species of nestling ciconiiforms collected from southern Florida were tested for concentrations of lead, cadmium, and copper in the liver, and chlorinated hydrocarbon concentrations in the brain. Geographic location effected both lead and cadmium concentrations in the liver with highest concentrations found in samples from the freshwater Everglades area. Neither nestling size nor diet had an effect on liver lead, cadmium or copper concentrations. Chlorinated hydrocarbon concentrations were found most frequently and in the highest concentrations in pooled brain samples of nestlings collected in the Lake Okeechobee area. Lead intoxication (liver lead > 2 μg/g) may have contributed to or caused the deaths of two Tricolored Herons (*Egretta tricolor*) and one Roseate Spoonbill (*Ajaia ajaja*) nestling. Several individuals with high concentrations of one contaminant also had other contaminants in high concentrations.

Because nestling ciconiiform birds are confined to their nest and fed a variety of vertebrate and invertebrate foods from wetland habitats, they are often considered good biomonitors for local contamination of such habitats (Custer and Mulhern 1983). As part of a study to look at the effects of diseases on a declining population of ciconiiforms in southern Florida, 962 birds found dead were examined (Spalding and Forrester 1991). Ninety-seven nestlings were tested for lead, cadmium, copper, and/or chlorinated hydrocarbon concentrations. Mercury concentrations from this same group of birds were reported elsewhere (Spalding et al. 1994, Sundlof et al. 1994, Beyer et al. 1997).

METHODS

Liver (n = 50) and brain (n = 80) samples were selected from 97 nestlings for analysis based on species, size, and location. Larger nestlings (61% of the sample) were selected within a species and location because they had a greater opportunity to accumulate contaminants. Dead nestlings were collected from colonies on the edges of and on islands in Lake Okeechobee, the freshwater Conservation Areas of the Everglades, Rodger's River Bay Colony (mangrove estuary island on the western coast of Everglades National Park), and mangrove islands in Florida Bay. Samples were collected during 1987 to 1991. Roseate Spoonbill (*Ajaia ajaja*) brain samples, tested for organochlorine compounds from

Florida Bay, were divided between the eastern half and the western half of the bay because a difference was suspected for mercury contamination. Nestlings were placed in one of two size categories based upon adult bill length divided by two. Nestlings also were placed in one of three forage groups based upon their predominant diet of large fish (Great Blue Herons *Ardea herodias*, Great White Herons, *A. h. occidentalis*, and Great Egrets, *Ardea albus*), small fish (Snowy Egrets, *Egretta thula*, Tricolored Herons, *E. tricolor*, Little Blue Herons, *E. caerulea*, and Reddish Egrets, *E. rufescens*), and small fish and arthropods (White Ibises, *Eudocimus albus*, and Roseate Spoonbills) (Bancroft et al. 1991, Frederick 1995).

Frozen liver was evaluated for metal contaminants using cold vapor atomic absorption spectrophotometry as described in Sundlof et al. (1994). The lower level of detection for metals was 0.05 µg/g for lead and copper, and 0.01 µg/g for cadmium. Frozen brain (15 pooled samples from 80 birds) and the entire carcass from one nestling with a deformed bill were analyzed for chlorinated hydrocarbons by the Mississippi State Chemical Laboratory, Mississippi State University, using the modified Micro Method for sample sizes below 4 g (Manual of analytical methods for the analysis of pesticides in humans and environmental samples, EPA-600/8-80-038, June 1980, Section 5, A[2]). GC/Mass spectrometry was used to confirm p, p'-DDE. Results are reported in µg/g wet weight, as are those discussed, unless otherwise noted. The lower level of detection for most organochlorines was 0.01 µg/g, and 0.05 µg/g for both toxaphene and PCBs.

Lead, copper, and cadmium concentrations were log transformed prior to analysis to better meet the assumption of homogeneity of variance. An ANOVA was conducted on the transformed data to test for differences among the main effects of location, diet, and age using an (α-level of 0.050. Tukey's means comparison procedure was used to determine significant differences between levels of the significant main effects (Ott 1993). Differences in concentrations of p,p'-DDE in pooled brain samples between locations were tested using ANOVA and Tukey's means comparison procedure on the logarithmically transformed data. Subsequent results are presented as geometric means and the ranges or 95% confidence intervals.

RESULTS

Lead concentrations in ciconiiform nestling livers averaged 0.32 µg/g (range = 0.09-6.3 µg/g) and were below 1 µg/g except for three samples (Table 1). These were two Tricolored Heron nestlings (one about 1 week old, the other less than 5 days) from two colonies in the central Everglades (6.3 and 3.1 µg/g) and a Roseate Spoonbill nestling (about 2 weeks old) from eastern Florida Bay (2.8 µg/g). Geographic location had a significant ($P = 0.010$) effect on lead concentration. Livers of nestlings collected in the Everglades had a significantly higher mean concentration of lead (0.50 µg/g) than livers of nestlings collected in mangrove areas (0.22 µg/g). Liver lead concentrations of nestlings collected in Lake Okeechobee and Florida Bay (0.27 and 0.43 µg/g, respectively) were not significantly different from concentrations in nestlings from the Everglades or mangrove areas. Size did not have a significant ($P = 0.931$) effect on lead concentrations in livers of nestlings. Although there were no significant differences ($P = 0.053$) in liver lead concentrations between diets, nestlings fed on large fish (0.25 µg/g) tended to have lower mean lead concentrations than nestlings fed on small fish or small fish and arthropods (0.41 µg/g and 0.36 µg/g, respectively).

Table 1. Concentrations of lead, copper and cadmium in livers of nestling Ciconiiforms collected in southern Florida, 1987-91.

Area Species	n	Geometric mean (range) µg/g wet weight					
		Lead		Copper		Cadmium	
Everglades							
Great Egret	1	0.16		NA[b]		0.07	
Tricolored Heron	4	1.4	(0.27-6.3)	23	(2.7-170)	0.22	(0.08-0.68)
White Ibis	4	0.22	(0.18-0.28)	27	(18-55)	0.07	(0.02-0.21)
All species[c]	9	0.50A[a]	(0.21-1.2)	25	(10-59)	0.12A	(0.06-0.24)
Florida Bay							
Great White Heron	5	0.32	(0.19-0.51)	20	(5.3-57)	0.10	(0.04-0.21)
Roseate Spoonbill	6	0.55	(0.18-2.8)	17	(6.4-46)	0.08	(0.02-0.40)
All species[c]	11	0.43AB	(0.28-0.68)	18	(11-30)	0.09AB	(0.05-0.16)
Lake Okeechobee							
Great Blue Heron	6	0.28	(0.20-0.44)	5.5	(1.5-13)	0.05	(0.02-0.10)
Great Egret	6	0.26	(0.18-0.33)	43	(22-220)	0.07	(0.05-0.11)
Little Blue Heron	4	0.28	(0.19-0.52)	23	(4.1-44)	0.06	(0.04-0.09)
White Ibis	2	0.25	(0.24-0.27)	20	(8.8-45)	0.03	(0.03-0.04)
All species[c]	18	0.27AB	(0.24-0.31)	17	(9.2-29)	0.06B	(0.04-0.07)
Mangrove Estuary							
Great Egret	5	0.18	(0.09-0.32)	6.1[d]	(2.4-15)	0.07	(0.06-0.09)
Snowy Egret	5	0.26	(0.16-0.41)	21	(11-58)	0.08	(0.03-0.14)
Tricolored Heron	2	0.20	(0.16-0.24)	12	(3.6-37)	0.15	(0.13-0.17)
All species[c]	12	0.22B	(0.17-0.28)	12	(6.8-22)	0.08AB	(0.07-0.11)

[a]Means with different letters differ significantly ($P < 0.050$).
[b]Not analyzed.
[c](95% confidence interval).
[d]$n = 4$.

Liver cadmium concentrations averaged 0.08 µg/g (range = 0.02 - 0.68 µg/g). Geographic location had a significant ($P = 0.026$) effect on cadmium concentrations in the livers of nestling ciconiiforms. Livers of nestlings collected in the Everglades had a significantly higher mean concentration of cadmium (0.12 µg/g) than nestlings collected from Lake Okeechobee (0.06 µg/g). Cadmium concentrations of nestlings collected in mangroves (0.08 µg/g) or Florida Bay (0.09 µg/g) were not significantly different from cadmium concentrations of Everglades or Lake Okeechobee nestling livers. The highest cadmium concentrations were found in the same individuals with the highest lead concentrations for two Tricolored Heron nestlings from two colonies in the central Everglades(0.36 and 0.68 µg/g), and a Roseate Spoonbill nestling from eastern Florida Bay (0.40 µg/g). Size or diet did not have a significant effect on cadmium concentrations in livers ($P = 0.406$ and $P = 0.097$, respectively).

Table 2. Organochlorine concentrations in pooled brain tissue of ciconiiform nestlings from colonies in southern Florida, 1987-91.

Colony site Species	No. birds pooled	Organochlorine (µg/g, wet weight) HCB	Oxychlordane	Heptachlor-Epoxide	t-Nonachlor	p,p' DDE
Lake Okeechobee						
Great Blue Heron	5	0.01	0.03	0.03	0.05	0.38
Great Egret	10	ND[b]	0.03	0.03	0.04	0.80
Snowy Egret	4	ND	0.04	0.04	0.03	0.95
White Ibis	2	ND	0.02	0.02	0.01	0.40
all species[c]		<0.01	0.03	0.03	0.04	0.58A[a]
Central Everglades						
Great Egret	1	0.01	ND	0.07	0.05	1.30
Tricolored Heron	2	ND	ND	ND	ND	0.07
Snowy Egret	2	ND	ND	ND	ND	0.06
all species		<0.01	<0.01	0.01	0.01	0.18A
Mangrove Estuary						
Great Egret	5	0.01	0.02	0.02	0.02	0.11
Snowy Egret	10	ND	ND	ND	ND	0.05
all species		<0.01	0.01	0.01	0.01	0.07AB
Florida Bay						
Great White Heron	10	ND	ND	ND	ND	0.02
Reddish Egret	2	ND	ND	ND	ND	0.02
Great Egret	2	ND	ND	ND	ND	ND
Tricolored Heron	5	ND	ND	ND	ND	0.01
Roseate Spoonbill						
Western Bay	10	ND	ND	ND	ND	0.02
Eastern Bay	10	ND	ND	ND	ND	ND
all species		<0.01	<0.01	<0.01	<0.01	0.01B

[a]Means with different letters were significantly different (P < 0.050).
[b]ND = not detected. Lower level of detection = 0.01 µg/g, 0.05 µg/g for toxaphene and PCBs. One-half the lower level of detection was used for calculating means of compounds not detected. a-BHC, G-BHC, ß-BHC, d-BHC, G-chlordane, toxaphene, PCB, o,p'-DDE, a-chlordane, dieldrin, o,p'-DDD, o,p'-DDT, p,p'-DDD, p,p'-DDT, endrin, cis-nonachlor, mirex, endosulfan I, endosulfan II, and aldrin were not detected in any of the samples. Percent lipid range = 5.10 - 9.10; Percent moisture range = 78.9 - 85.0.
[c]Adjusted mean.

Copper concentrations ranged from 1.5 to 225 µg/g, with a mean of 17 µg/g. There were no significant differences in copper concentrations between geographic locations, sizes or diets (P = 0.587, P = 0.854, P = 0.570, respectively).

Organochlorine concentrations were low in the pooled brain tissue samples from six colony locations, except for a few cases (Table 2). One large Great Egret nestling from the Everglades that was severely emaciated had 1.3 μg/g DDE in brain. Location did have a significant effect on p,p'-DDE concentrations in the pooled brain samples (P = <0.001). Brain samples collected from Lake Okeechobee (0.58 μg/g) and the Everglades (0.18 μg/g) had significantly higher levels of p,p'-DDE than samples collected in Florida Bay (0.01 μg/g). Brain samples collected from mangrove estuary (0.07 μg/g) did not have significantly different p,p'-DDE levels from any of the other sites. Overall, Great Egrets had higher concentrations of organochlorines, and compounds were detected more commonly, than in the other five species examined. DDE was detected in all samples except Roseate Spoonbills in eastern Florida Bay and Great Egrets in Florida Bay. In addition to DDE, four other chlorinated hydrocarbon compounds were found: hexochlorobenzene, oxychlordane, heptachlor-epoxide and t-nonachlor. These compounds were found more commonly, and in greater concentrations at Lake Okeechobee compared to other locations. These compounds were generally more common at more northern sites and were not detected in any of the Florida Bay samples. Twenty additional compounds (Table 2), were not detected in any of the samples. The entire carcass of a single Little Blue Heron with a deformed bill from Lake Okeechobee did not contain detectable concentrations of any of the organochlorine compounds listed in Table 2.

Discussion

Three contaminants, lead, cadmium, and DDE, were found in higher than expected concentrations in livers and brains of nestling wading birds found dead in southern Florida. Contaminants measured in birds that are found dead, as in our study, should be higher than those measured in birds collected alive if the contaminant has an effect on survival of the bird. Unfortunately we rarely had data for such comparisons.

Liver lead concentrations where highest in samples from the freshwater Everglades. In a study of White Ibis and Anhingas (*Anhinga anhinga*) at a landfill just east of the Everglades, elevated lead concentrations were found in carcasses of both species, especially in White Ibis (annual means of 0.34-1.5 μg/g over 4 years with a highest value of 3.1 μg/g in a pooled sample of 2 nestlings) that feed in the landfill (Rumbold 1997). The highest lead concentration (0.52 μg/g) found in wading bird eggs in Florida also was from White Ibis at the same landfill site (Rumbold 1997). One adult White Ibis collected in Collier County in 1971 had 9.0 μg/g lead in muscle tissue (Ogden et al. 1974). Lead concentrations in livers from the Everglades and Florida Bay in this study were higher than in nestling Louisiana Herons (mean = 0.27, highest = 0.53 µg/g) and Cattle Egrets (*Bubulcus ibis*) (mean = 0.35, highest = 0.47 in one study and mean = 0.59, highest = 1.05 µg/g) from Galveston Bay, Texas (Hulse et al. 1980; Cheney et al 1981).

Hoffman et al. (1985) experimentally dosed American Kestrels (*Falco sparverius*) with lead and suggested that greater than 2 µg/g lead in the liver may be associated with impaired growth, whereas greater than 5 µg/g in the liver may impair survival. If this applies for nestling wading birds, then one of the Tricolored Herons (6.3 µg/g) may have died from lead poisoning. Lead concentrations are elevated in fish and aquatic invertebrates collected near industrialized areas, urban areas, and ponds with lead shot (Eisler 1988).

Furness (1996) suggested that cadmium poisoning may occur above 40 µg/g in liver. Our findings and those of others for Florida birds are well below this concentration. As mentioned above, we found three instances of high lead mirrored by high cadmium in liver. The highest cadmium concentration found in eggs of wading birds collected in Florida was 0.05 µg/g (Ogden et al. 1974). An adult White Ibis collected in Collier Co. in 1971 had 9.0 µg/g cadmium in breast muscle (the same bird had high lead concentrations noted above). Burger et al. (1993) found 0.20 µg/g (geometric mean, dry weight) cadmium in the feathers of 15 nestling Wood Storks (*Mycteria americana*) from Florida. Cadmium concentrations in liver were higher in southern Florida, especially in the Everglades, than in Tricolored Heron (mean = 0.031, highest = 0.038 µg/g) and Cattle Egret nestlings (mean = 0.029, highest = 0.046 in one study and mean = 0.021, highest = 0.023 µg/g) in Galveston Bay Texas (Hulse et al. 1980; Cheney et al. 1981). Residues in liver that exceed 10 µg/g are considered to probably have cadmium contamination (Eisler 1985). Reported background cadmium concentrations are 0.06 µg/g in soil and 0.08 µg/g in fresh water (Jenkins 1981). Heinz and Haseltine (1983) found behavioral effects in Black Ducks (*Anas rubripes*) dosed with cadmium but did not report liver concentrations.

Custer and Mulhern (1983) found that copper in livers of prefledgling Black-crowned Night-herons (*Nycticorax nycticorax*) in three colonies on the northeastern coast of the U.S varied significantly between colonies (range = 23.1 - 381 µg/g, dry weight. Comparable dry weight values for nestlings in our study ranged between 5.8 - 893 µg/g, dry weight. The significance of these values is unknown; copper is an essential element. Reported background copper concentrations are 20 µg/g in soil and 0.01 µg/g in fresh water (Jenkins 1981). Frank and Borg (1979) found over 1000 µg/g copper, along with high concentrations of other metals, in the livers of Mute Swans (*Cygnus olor*); only one had signs of intoxication.

Most data of organochlorine residues in ciconiiforms are from eggs (Ohlendorf et al. 1979). Rumbold et al. (1996) reported as much as 1.6 μg/g DDE in eggs, and 0.11 μg/g in carcasses, of White Ibis nestlings near the landfill east of the Everglades. Nesbitt et al. (1981) found a mean of 0.05 μg/g DDE in the brains of nestling Brown Pelicans (*Pelecanus occidentalis*) from Florida in 1971-72. Thus, it was surprising to find a mean of 0.69 μg/g DDE in the brains of nestling wading birds in the Lake Okeechobee area 20 years later. This finding suggests that DDT use was greater, continued later in this area, or that wading birds accumulate more DDE than pelicans due to diet differences. Concentrations of DDE as high as 28 μg/g were found in the brain of an individual Great Blue Heron suspected to have died from dieldrin (6.6 μg/g) poisoning in Florida (Ohlendorf et al. 1981). Generally birds with combined DDT and DDD concentrations of 30 μg/g or greater in brain do not survive (Stickel et al. 1966). We found no studies of the sublethal effects of DDE at the low concentrations that we found for nestling brains in our study.

Individual birds with high concentrations of one contaminant were likely to have high concentrations of another contaminant. We found this in several cases with lead and cadmium, and with mercury and DDE. The same Great Egret from the central Everglades that had 1.3 μg/g DDE in brain, had the highest liver mercury concentration (19 μg/g) found among nestlings (Sundlof et al. 1994). This generalization applied only for individual birds and not for groups of birds from different locations. For example, the highest mean brain concentrations of DDE were found in birds from the Lake Okeechobee area, whereas, mean liver mercury concentrations were lowest in the same area (Sundlof et al. 1994).

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