HEAVY METALS IN SOME SPECIES OF WATERFOWL OF NORTHERN ITALY

Authors: Carpenè, E., Serra, R., and Isani, G.

Source: Journal of Wildlife Diseases, 31(1) : 49-56

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/0090-3558-31.1.49

Journal of Wildlife Diseases, 31(1), 1995, pp. 49–56

HEAVY METALS IN SOME SPECIES OF WATERFOWL OF NORTHERN ITALY

E. Carpenè, R. Serra, and G. Isani
Department of Biochemistry, Veterinary Biochemistry Section, University of Bologna, via Tolara di Sopra 30, Ozzano Emilia, Bologna, Italy

ABSTRACT: Concentrations of heavy metals (zinc, copper, cadmium, and iron) were measured in several tissues (brain, gizzard, leg-muscle, heart, breast-muscle, intestine, liver and kidney) of moorhens (*Gallinula chloropus*), black-headed gulls (*Larus ridibundus*), and coots (*Fulica atra*) collected between autumn 1985 and spring 1989 in northern Italy. Cadmium concentrations in the liver and kidney of water-rails (*Rallus aquaticus*) and in five species of Anatidae collected also were measured. High mean (±SD) copper levels were detected in aerobic muscles such as heart (38 ± 5 μg/g dry weight (DW)) and pectoral muscles (35 ± 7 μg/g DW). Compared to other tissues, the iron content of brain was rather low and constant, with a mean value of 160 ± 17 μg/g DW in moorhens, 157 ± 60 μg/g DW in black-headed gulls, and 157 ± 25 μg/g DW in coots. Iron concentrations in tissues of moorhens from the Reno River were significantly higher than those from the Sile River. Cadmium was detectable only in the liver and kidney; there was a linear relationship between cadmium levels in these two organs. The highest mean (±SD) cadmium concentrations were present in the kidney of black-headed gull (30 ± 20 μg/g DW).
Key words: Tissue distribution, wildfowl, zinc, copper, iron, cadmium.

INTRODUCTION

Birds accumulate high concentrations of toxic metals (Burgher and Gochfeld, 1985). Past studies often focused on lead, cadmium, and mercury. Lead generally has an anthropogenic origin, such as lead-containing gasoline, lead shot, or fishing weights (White and Stendell, 1977). Cadmium is a ubiquitous heavy metal whose abundance in the biosphere has increased due to various human activities (Cain et al., 1983); habitats near wastewater outfalls containing high levels of cadmium have shown substantial perturbations among their aquatic animal populations (Yost, 1984). Mercury may be naturally present in the environment (Leonzio et al., 1986) or introduced as a consequence of pollution (Lindsay and Dimmick, 1983). Less attention has been paid to essential heavy metals among birds; depending on their concentration in the tissue, heavy metals can exert a toxic or a beneficial effect. Each bird species has a characteristic metal content in its tissues depending on the amount of metal taken up in the diet (Cain et al., 1983) and the biology of the species (Goede et al., 1989). For strictly nonessential metals such as cadmium, mercury, or lead, when tissue levels are low, it is possible that their effects are not necessarily negative. Our objective was to compare levels of zinc (Zn), copper (Cu), iron (Fe), and cadmium (Cd) in several species of waterfowl from northern Italy; we defined Zn, Cu, and Fe as essential metals, and Cd as a nonessential metal. The sampling area was a major center for various kind of industries and for intensive agriculture in northern Italy. In the case of the moorhen (*Gallinula chloropus*), there were enough specimens to compare the influence of the geographic distribution on the metal composition. Some of the birds were collected along the Reno River in a place which was supposedly unpolluted.

MATERIALS AND METHODS

Forty-eight specimens of waterfowl were collected. Birds were shot during the hunting season (Anatidae and Rallidae); five Laridae were found dead due to trauma. Birds were collected between 1985 and 1989 in and near the Venetian Lagoon (45°33′N, 12°31′E) and Sile River (45°36′N, 12°20′E). Additional moorhens were collected from the Reno River (44°18′N, 11°07′E). We collected black coots (*Fulica atra*), mallards (*Anas platyrhynchos*), common pochards (*Aythya ferina*), garganeys (*Anas quer-*

TABLE 1. Bird species, collection sites, metals analyzed, and samples pooled, northern Italy, 1985 to 1989.

Species	Location	Total animals collected	Number of pools	Animals per pool	Metals analyzed[a]
Moorhen	Reno River	8	2	4	Cd, Zn, Cu, Fe
	Sile River	12	3	4	Cd, Zn, Cu, Fe
Black-headed gull	Venice Lagoon	5	5	1	Cd, Zn, Cu, Fe
Water rail	Reno River	1	1	1	Cd
Green-winged teal	Venice Lagoon	3	1	3	Cd
Garganey	Venice Lagoon	3	1	3	Cd
Common pochard	Venice Lagoon	6	2	3	Cd
Black coot	Venice Lagoon	6	2	3	Cd, Zn, Cu, Fe
Pintail	Venice Lagoon	1	1	1	Cd
Mallard	Venice Lagoon	3	3	1	Cd

[a] Cd, cadmium; Zn, zinc; Cu, copper; Fe, iron.

quedula), green-winged teal (*Anas crecca*), moorhens, black-headed gulls (*Larus ridibundus*), pintails (*Anas acuta*), and water rails (*Rallus aquaticus*) (Table 1). Moorhens, black-headed gulls, and coots were analyzed for Zn, Cu, Fe, and Cd content in eight tissues: brain, gizzard, leg-muscle, heart, breast-muscle, intestine, liver, and kidney. The other six species were sampled only for Cd in liver and kidney. All birds appeared to be healthy with the exception of one underweight pintail. Tissue samples from 0.5 to 2.0 g were removed, placed in polyethylene vials and lyophilized. Samples of the same tissue from several birds were pooled for analysis (Table 1); samples from black-headed gulls were analyzed individually. The lack of uniformity of the number of analyses for each species was due to the impossibility of obtaining more representative specimens. The lyophilized tissues (100 to 300 mg) were placed in platinum crucibles and heated overnight at 480 C. The ash residue was dissolved in 1 N hydrochloric acid. Metals were analyzed by flame atomic absorption spectrophotometry (AAS) using an IL-11 spectrophotometer (Thermo Jarrell Ash Corporation, Franklin, Massachusetts, USA). The accuracy of the method was evaluated by calibration with international standards from the International Atomic Energy Agency (IAEA) (Principality of Monaco: MA-A-2 (fish), MA-M-1 (oyster), MA-A-1 (copepod), and MA-M-2 (mussel)). The concentrations found with our methods fell in the confidence interval given by IAEA. All statistical analyses were performed using an SPSS program for MS WINDOWS Release 6.0 (SPSS, 1993). A one-way analysis of variance (ANOVA) procedure was performed to determine the variability of the observations within each group as well as the variability between the group means. When significant differences between mean values of metal concentrations from species or from tissues were found, we used the Scheffé test ($P = 0.05$) to determine which means were significantly different from each other. Student's t-test was used to evaluate differences between means from Reno and Sile River moorhens; differences with $P < 0.05$ were considered significant. A regression analysis between renal and hepatic Cd concentrations also was computed.

RESULTS

The metal levels in the waterfowl were characteristic of the tissues analyzed; Fe generally had the highest values followed by Zn and then Cu (Tables 2, 3, 4, and 5). Among muscles, the gizzard was rich in Zn, whereas other muscle tissues had high Cu values, except for the black-headed gull leg-muscle (Table 4). Brain tissue had low levels of heavy metals, especially Fe. Tissues connected with metal uptake, storage, and excretion (intestine, liver, and kidney) had large fluctuations.

Among moorhens, the highest Zn levels were found in the gizzard (141 ± 5 μg/g DW) and lowest in breast muscle (50.1 ± 3.5 μg/g DW) (Table 2). For Cu, the highest and lowest values were found in the breast muscle (38.1 ± 4.8 μg/g DW) and in the gizzard (9.2 ± 1.3 μg/g DW), respectively; if gizzard data are excluded, Cu was higher in muscle than in the other tissues (Table 2). Iron levels varied between a minimum (160 ± 17 μg/g DW) in brain and a maximum (2,270 ± 2,750

TABLE 2. Mean (±SD) zinc, copper, and iron concentrations (μg/g dry weight) of moorhens from the Sile and Reno Rivers in northern Italy, 1985 to 1989.

Tissues	Zinc	Copper	Iron
Brain	66.5 ± 3.3[a] (n = 4)	16.9 ± 3.6 (n = 3)	160 ± 17 (n = 4)
Gizzard	141 ± 5[b] (n = 5)	9.2 ± 1.3 (n = 4)	347 ± 21[a] (n = 5)
Leg-muscle	67 ± 12 (n = 5)	28.2 ± 8[a] (n = 4)	197 ± 91 (n = 5)
Heart	106 ± 10[a] (n = 5)	28.0 ± 3.3[ab] (n = 4)	560 ± 200 (n = 5)
Breast-muscle	50.1 ± 3.5 (n = 5)	38.1 ± 4.8[ab] (n = 3)	411 ± 67[a] (n = 4)
Intestine	109 ± 27[b] (n = 5)	13.8 ± 3.2 (n = 5)	550 ± 390 (n = 5)
Liver	120 ± 9[b] (n = 5)	18 ± 7 (n = 5)	2,270 ± 2,750 (n = 5)
Kidney	95 ± 10 (n = 5)	18.9 ± 2.7 (n = 5)	570 ± 250[a] (n = 5)

[a] Significantly different from black-headed gull.
[b] Significantly different from coot.

μg/g DW) in the liver (Table 2). Iron concentrations in tissues of moorhens from the Reno River were higher than those from the Sile River, but due to high individual fluctuations only breast-muscle and intestine values were significantly different; no significant differences were found for Cu, whereas differences were found in heart and breast-muscle for Zn (Table 3).

Among black-headed gulls the highest Zn levels were found in the gizzard (132.7 ± 4.3 μg/g DW) and the lowest in breast-

TABLE 3. Mean (±SD) zinc, copper, and iron concentrations (μg/g dry weight) of moorhens from the Sile and Reno Rivers, northern Italy, 1985 to 1989.

Tissues	Zinc		Copper		Iron	
	Sile	Reno	Sile	Reno	Sile	Reno
Brain	65 ± 2 (n = 2)	68.1 ± 4.3 (n = 2)	16.7 ± 5.1 (n = 2)	17.3 (n = 1)	152 ± 18 (n = 2)	168 ± 17 (n = 2)
Gizzard	140 ± 6 (n = 3)	142 ± 6 (n = 2)	8.6 ± 1.2 (n = 2)	9.8 ± 1.6 (n = 2)	348 ± 28 (n = 3)	345 ± 14 (n = 2)
Leg-muscle	62 ± 13 (n = 3)	75.8 ± 8.7 (n = 2)	23.6 ± 5.1 (n = 2)	32.6 ± 9.4 (n = 2)	141 ± 26 (n = 3)	283 ± 87 (n = 2)
Heart	113 ± 2[a] (n = 3)	95.0 ± 0.7 (n = 2)	26 ± 3 (n = 3)	31.5 (n = 1)	479 ± 99 (n = 3)	697 ± 297 (n = 2)
Breast-muscle	50.5 ± 0.7[a] (n = 2)	54.2 ± 0.5 (n = 2)	37.2 ± 6.5 (n = 2)	39.69 (n = 1)	353 ± 2[a] (n = 2)	468 ± 16 (n = 2)
Intestine	108 ± 13 (n = 3)	113 ± 51 (n = 2)	12.1 ± 3.2 (n = 3)	16.2 ± 0.2 (n = 2)	288 ± 102[a] (n = 3)	955 ± 253 (n = 2)
Liver	118 ± 10 (n = 3)	122 ± 10 (n = 2)	19.8 ± 3.9 (n = 3)	15.2 ± 11.8 (n = 2)	485 ± 58 (n = 3)	4,940 ± 2,520 (n = 2)
Kidney	90.0 ± 2.3 (n = 3)	103 ± 15 (n = 2)	19.2 ± 0.7 (n = 3)	18.5 ± 5.3 (n = 2)	421 ± 199 (n = 3)	797 ± 67 (n = 2)

[a] Significantly different from the corresponding value for Reno River as determined by Student's t-test, $P < 0.05$.

TABLE 4. Mean (±SD) zinc, copper, and iron concentrations (μg/g dry weight) of black-headed gulls from Venice Lagoon in northern Italy, 1985 to 1989.

Tissues	Zinc	Copper	Iron
Brain	49.0 ± 3.3[a,b] (n = 4)	9.3 ± 1.8[a] (n = 4)	157 ± 60 (n = 4)
Gizzard	132.7 ± 4.3[a] (n = 5)	5.9 ± 0.9[b] (n = 4)	117 ± 32[a,b] (n = 5)
Leg-muscle	99 ± 22 (n = 5)	7.1 ± 1.7[a,b] (n = 4)	164 ± 32 (n = 5)
Heart	77 ± 10[a,b] (n = 5)	15.6 ± 3.4[a,b] (n = 4)	431 ± 124 (n = 4)
Breast-muscle	45.0 ± 7.5 (n = 5)	14.3 ± 1.5[a,b] (n = 5)	298 ± 45[b] (n = 5)
Intestine	134 ± 12[a] (n = 4)	14.6 ± 7.5 (n = 4)	130 ± 34 (n = 5)
Liver	91 ± 22[a] (n = 5)	19.3 ± 4.2 (n = 5)	2,240 ± 260 (n = 5)
Kidney	104 ± 24 (n = 5)	24.5 ± 7.4 (n = 5)	1,140 ± 320[a,b] (n = 5)

[a] Significantly different from moorhen.
[b] Significantly different from coot.

muscle (45 ± 7.5 μg/g DW) (Table 4). For Cu, the highest and lowest values were found in the kidney (24.5 ± 7.4 μg/g DW) and in the gizzard (5.9 ± 0.9 μg/g DW), respectively (Table 4). Iron levels varied from a minimum of 117 ± 32 μg/g DW in the gizzard to a maximum of 2,240 ± 260 μg/g DW in the liver (Table 4).

Among coots, the highest Zn levels were found in the liver (203 ± 15 μg/g DW) and the lowest in the intestine (54 ± 7.1 μg/g DW) (Table 5). For Cu, the highest and lowest values were found in the heart (43.6 ± 3.3 μg/g DW) and in the intestine (3.9 ± 0.1 μg/g DW), respectively (Table 5). Iron levels varied from a minimum of 157 ± 25 μg/g DW in the brain to a maximum of 570 ± 130 μg/g DW in the kidney (Table 5).

There were several significant interspecies differences in metal concentrations when single tissues and total loads were compared (Tables 2, 4, 5). For Zn, interspecific significant differences (P = 0.05) were found in brain, gizzard, heart, intestine, and liver; for Cu, in the brain, breast-muscle, gizzard, heart, and leg-muscle; and for Fe, in breast-muscle, gizzard and kidney (Tables, 2, 4, 5). For example, higher levels of Cu and Fe were present in the heart and in the two skeletal muscles of moorhens and coots as compared to the black-headed gull tissues. Adding the Cu averages of the three tissues we obtained a total for each species, respectively, of 94, 114, and 37 μg/g DW; doing the same for Zn, we obtained: 223, 291, and 221 μg/g DW, respectively. These values were used to compute a Cu:Zn ratio of 0.42, 0.39, and 0.16 for moorhen, coot, and black-headed gull, respectively.

Cadmium was detectable (≥0.1 μg/g DW) only in the liver and kidney of the nine avian species analyzed (Table 6). In all species, Cd was higher in the kidney than in the liver. The mallard had the lowest Cd levels in both tissues, whereas the highest values were found in the black-headed gull. Due to large individual fluctuations in Cd content, no significant differences among species were found. Cadmium concentrations from the liver (n = 20) were plotted against those from the kidney (n = 20) and a positive correlation between the two organs was found, $y = 0.43 + 0.09x$, where y is defined as the

TABLE 5. Mean (±SD) zinc, copper, and iron concentrations (μg/g dry weight) of black coots from Venice Lagoon in northern Italy, 1985 to 1989.

Tissues	Zinc	Copper	Iron
Brain	67.0 ± 6.4[a] (n = 2)	10.9 ± 2.0 (n = 2)	157 ± 25 (n = 2)
Gizzard	170[a,b] (n = 1)	11.5 ± 3.2[a] (n = 2)	389 ± 127[a] (n = 2)
Leg-muscle	109 ± 13 (n = 2)	31.6 ± 1.3[a] (n = 2)	288 ± 30 (n = 2)
Heart	124[a] (n = 1)	43.6 ± 3.3[a,b] (n = 2)	452 ± 10 (n = 2)
Breast-muscle	57.2 ± 1.0 (n = 2)	38.7 ± 3.0[a] (n = 2)	364.0 ± 5.6 (n = 2)
Intestine	54.0 ± 7.1[a,b] (n = 2)	3.9 ± 0.1 (n = 2)	NA[c]
Liver	203 ± 15[a,b] (n = 2)	24.1 ± 5.6 (n = 2)	271 ± 810 (n = 2)
Kidney	82.7 ± 7.4 (n = 2)	16.1 ± 3.4 (n = 2)	570 ± 130[a] (n = 2)

[a] Significantly different from black-headed gull.
[b] Significantly different from moorhen.
[c] NA, not analyzed.

concentration of Cd in μg/g DW of liver, and x is defined as the concentration of Cd in μg/g DW of kidney (r = 0.92, P < 0.001).

DISCUSSION

It is important to analyze trace metal levels in tissues of different birds because metal patterns can vary widely with the organ or the species. Few data are available for metal concentrations in birds from northern Italy (Renzoni et al., 1986). Most concentrations of physiological heavy metals found in the tissues of aquatic birds in this study fell within the range of those reported by other authors (Szefer and Falandysz, 1987).

We believe that the levels of Fe, Zn and Cu can be linked to the physiological role played by these metals. Iron is widely utilized by molecular structures in redox systems as heme proteins and in nonheme enzymes; its biochemical role requires a fine-tuned regulation because the free forms are very toxic (Gutteridge, 1987). Excess Fe is stored in non-toxic form mainly in the liver, in the ferritin molecule; thus it is common to find high levels of Fe in the liver, which can present physiological variations of the metal in connection with the egg laying process (Saiz et al., 1990). Due to Fe segregation into ferritin and hemosiderin, hemosiderosis in birds does not appear to compromise health (Ward et al., 1988). High levels of hepatic Fe also can be connected with lead poisoning

TABLE 6. Mean (±SD) cadmium concentrations (μg/g dry weight) in kidney and liver of aquatic birds from northern Italy, 1985 to 1989.

Species	Number evaluated	Liver	Kidney
Moorhen	5	0.67 ± 0.13	1.22 ± 0.56
Black-headed gull	5	4.0 ± 3.3	32 ± 26
Water-rail	1	NA[a]	2.0
Green-winged teal	1	0.9	1.4
Garganey	1	0.6	3.2
Common pochard	2	0.39 ± 0.28	1.9 ± 0.1
Black coot	2	0.45 ± 0.07	0.95 ± 0.35
Pintail	1	0.64	2.0
Mallard	3	0.19 ± 0.04	0.63 ± 0.22

[a] NA, not analyzed.

(Ochiai et al., 1992) and Zn poisoning (Droual et al., 1991). As expected, in these birds the highest Fe levels were found in the liver, with the exception of coot. Birds of the Order Galliformes tend to have lower Fe levels than those of the Orders Passeriformes, Coraciiformes and Anseriformes (Ward et al., 1988). We observed that moorhens from the Reno River contained higher Fe concentrations than moorhens from the Sile River (Table 3), and we believe that diet may have influenced the tissue Fe concentration. A small ironworks close to the collection site on the Reno River could account for these differences.

Zinc is as important as Fe for the roles that it plays at the biochemical level. The predominance of Zn in the gizzard is rather interesting and the small standard deviation could be indicative of a specific function of the metal in this organ (Tables 2, 4, and 5). In the central nervous system we found similar Zn values (from 49 $\pm$ 3 to 67 $\pm$ 6 μg/g DW) as reported for fish (from 56.8 $\pm$ 8.7 to 64.4 $\pm$ 2.3 μg/g DW) (Carpenè et al., 1994) and mammals (Prohaska, 1987). These results add evidence to the importance of a tight homeostasis of metal ions for brain function, including neurotransmission, as proposed by Xie and Smart (1991).

In tissues, Cu probably occurs in several oxidation states. Free Cu can be relatively toxic and, like other heavy metals (Cd, Zn), it can be sequestered by metallothionein and accumulated in the liver at high concentrations. Thus, hepatic Cu can vary widely, depending on the diet (Droual et al., 1991) and on bioavailability (Zanetti et al., 1991). As for Fe, the aerobic muscles (heart and pectoral muscle) were rich in Cu which is a cofactor of cytochrome oxidase, a respiratory protein found in large amounts in aerobic muscle. Black-headed gulls had Cu levels in muscle significantly lower than the corresponding values of moorhens and coots (Tables 2, 4, and 5). This fact could be related to a lower proportion of oxidative fibers leading to lower oxidative capacity, which in turn resulted in a lower Cu:Zn ratio (0.16) in the muscular tissues. An analogous situation was found by Caldow and Furness (1993) who compared fiber types in the muscle pectoralis of the great skua (*Catharacta skua*) and the herring gull (*Larus argentatus*); the oxidative and glycolytic activities of the muscle of the great skua were significantly higher than those of the homologous muscle of the herring gull. The authors proposed that this difference enabled the former species to be a more effective aerial kleptoparasite than the latter species.

Deficiency or excess of Zn, as well as Cu and Fe can cause serious neurological disorders (Prohaska, 1987), and metal concentrations apparently are kept more constant than in other tissues. Probably an efficient brain barrier can control not only the total amounts but also the regional distribution, which for Cu in mammals can range from 9.8 μg/g DW in the corpus callosum to a maximum of 201 μg/g DW in the locus coeruleus and for Zn from 28.8 in the centrum semiovale to 107 μg/g DW in the hippocampus (Prohaska, 1987). When metal levels rise, it is likely that the synthesis of metallothionein can control the amount of the free ions even in the brain (Ebadi et al., 1989). Based on our data, we do not believe that birds were exposed to high levels of Cu or Zn. In ducks, liver values of 8.5 to 48.1 μg/g fresh weight (FW) have been reported by Burgher and Gochfeld (1985).

The presence of detectable levels of Cd in the liver and kidney of most of the investigated species (Table 6) probably was due to the biosynthesis of metallothionein, a low molecular weight protein rich in cysteine that is easily induced by heavy metals. Even if its role is still matter of discussion (Kägi and Kojima, 1987), there is no doubt that when Cd is chelated by metallothionein, the metal toxicity is decreased. Cadmium-metallothionein also was isolated from chicken liver (Weser et al., 1973) and liver and kidney of Japanese quail (*Coturnix coturnix japonica*) (Yamamura and Suzuki, 1984) injected with $CdCl_2$. Generally, renal Cd levels are higher than hepatic ones because Cd-metallothionein is first synthesized in the liver and then accumulated in the kidney (Webb, 1987). In Manx shearwaters (*Puffinus puffinus*), concentrations of Cd in the kidney reached high values (60 to 480 μg/g DW) resulting in nephrotoxic lesions (Nicholson et al., 1983). The high levels of Cd that we have found in the gulls could be because these birds also fed on refuse dump areas contamined with Cd. However, Cd concentrations in gulls were far from those reported for Manx shearwater and in the same range of those reported for dunlins (*Calidris alpina*) in which no adverse effect was observed (Goede et al., 1989). From a pollution point of view, black-headed gulls can redistribute heavy metals in short time from dump areas to the lagoons where they stay overnight.

ACKNOWLEDGMENTS

We thank Prof. C. Porceddu for helpful discussion on statistical analysis, Dr. T. L. Coombs for insight comments, and Mr. E. Perisinotto for providing part of the duck samples.

LITERATURE CITED

BURGHER, J., AND M. GOCHFELD. 1985. Comparison of nine heavy metals in salt gland and liver of greater scaup (*Aythya marila*), black duck (*Anas rubripes*) and mallard (*A. platyrhynchos*). Comparative Biochemistry and Physiology 81C: 287–292.

CAIN, B. W., L. SILEO, J. C. FRANSON, AND J. MOORE. 1983. Effects of dietary cadmium on mallard ducklings. Environmental Research 32: 286–297.

CALDOW, R. W. G., AND R. W. FURNESS. 1993. A histochemical comparison of fibre types in the M. pectoralis and M. supracoracoideus of the great skua *Catharacta skua* and the herring gull *Larus argentatus* with reference to kleptoparasitic capabilities. Journal of Zoology (London) 229: 91–103.

CARPENÈ, E., O. CATTANI, G. P. SERRAZANETTI, G. FEDRIZZI, AND P. CORTESI. 1990. Zinc and copper in fish from natural waters and rearing ponds in northern Italy. Journal of Fish Biology 37: 293–299.

———, B. GUMIERO, G. FEDRIZZI, AND R. SERRA. 1994. Trace elements (Zn, Cu, Cd) in fish from rearing ponds in Emilia-Romagna region (Italy). The Science of the Total Environment 141: 139–146.

DROUAL, R., C. U. METEYER, AND F. D. GALEY. 1991. Zinc toxicosis due to ingestion of a penny in a gray-headed chachalaca (*Ortalis cinereiceps*). Avian Diseases 35: 1007–1011.

EBADI, M., V. K. PALIWAL, T. TAKAHASHI, AND P. L. IVERSEN. 1989. Zinc metallothionein in mammalian brains. *In* Metal ion homeostasis: Molecular biology and chemistry, D. Winge and D. Homer (eds.). A. R. Liss, Inc., New York, New York, pp. 257–267.

GOEDE, A., A. NYGARD, M. DE BRUIN, AND E. STEINNES. 1989. Selenium, mercury, arsenic and cadmium in the lifecycle of the dunlin, *Calidris alpina*, a migrant wader. The Science of the Total Environment 78: 205–218.

GUTTERIDGE, J. M. C. 1987. Lipid peroxidation: Some problems and concepts. *In* Oxygen radicals and tissue injury, B. Halliwell (ed.). The Upjohn Company, Bethesda, Maryland, pp. 9–19.

KÄGI, J. H. R., AND Y. KOJIMA. 1987. Chemistry and biochemistry of metallothionein. *In* Metallothionein II, J. H. R. Kägi and Y. Kojima (eds.). Birkauser Verlag, Basel, Switzerland, pp. 25–61.

LEONZIO, C., C. FOSSI, AND S. FOCARDI. 1986. Lead, mercury, cadmium and selenium in two species of gull feeding on inland dumps, and in marine areas. The Science of the Total Environment 57: 121–127.

LINDSAY, R. C., AND R. W. DIMMICK. 1983. Mercury residues in wood ducks and wood duck foods in eastern Tennessee. Journal of Wildlife Diseases 19: 114–117.

NICHOLSON, J. K. M., D. KENDALL, AND D. OSBORN. 1983. Cadmium and mercury nephrotoxicity. Nature 304: 633–635.

OCHIAI, K., K. JIN, C. ITAKURA, M. GORYO, K. YAMASHITA, N. MIZUNO, T. FUJINAGA, AND T. TSUZUKI. 1992. Pathological study of lead poisoning in whooper swans (*Cygnus cygnus*) in Japan. Avian Diseases 36: 313–323.

PROHASKA, J. R. 1987. Functions of trace elements in brain metabolism. Physiological Reviews 67: 858–901.

RENZONI, R., S. FOCARDI, C. FOSSI, C. LEONZIO, AND J. MAYOL. 1986. Comparison between concentrations of mercury and other contaminants in eggs and tissues of Cory's shearwater *Calonectris diomedea* collected on Atlantic and Mediterranean islands. Environmental Pollution Series A 40: 17–35.

SAIZ, M. P., M. T. MARTI, M. T. MITJAVILA, AND J. PLANAS. 1990. Sexual and age variations of organ iron content in Shaver chickens. British Poultry Science 31: 339–349.

SPSS. 1993. SPSS for Windows Base System user's guide, release 6.0. SPSS Inc., Chicago, Illinois, 828 pp.

SZEFER, P., AND J. FALANDYSZ. 1987. Trace metals in the soft tissues of scaup ducks (*Aythya marila*) wintering in Gdansk Bay, Baltic Sea. The Science of the Total Environment 65: 203–213.

WARD, R. J., T. C. IANCU, G. M. HENDERSON, J. R. KIRKWOOD, AND T. J. PETERS. 1988. Hepatic iron overload in birds: Analytical and morphological studies. Avian Pathology 17: 451–464.

WEBB, M. 1987. Toxicological significance of metallothionein. *In* Metallothionein II, J. H. R. Kägi and Y. Kojima (eds.). Birkauser Verlag, Basel, Switzerland, pp. 25–61.

WESER, U., H. RUPP, F. DONAY, F. LINNEMANN, W. VOELTER, W. VOETSCH, AND G. JUNG. 1973. Characterization of Cd, Zn-thionein (metallothionein) isolated from rat and chicken liver. European Journal of Biochemistry 39: 127–140.

WHITE, D. H., AND R. C. STENDELL. 1977. Waterfowl exposure to lead and steel shot on selected hunting areas. The Journal of Wildlife Management 41: 469–475.

XIE, X., AND T. G. SMART. 1991. A physiological role for endogenous zinc in rat hippocampal synaptic neurotransmission. Nature 349: 521–524.

YAMAMURA, M., AND K. T. SUZUKI. 1984. Induction and characterization of metallothionein in the liver and kidney of Japanese quail. Comparative Biochemistry and Physiology 77B: 101–106.

YOST, K. J. 1984. Cadmium, the environment and human health: An overview. Experientia 40: 157–164.

ZANETTI, M. A., P. R. HENRY, C. B. AMMERMAN, AND R. D. MILES. 1991. Estimation of the relative bioavailability of copper sources in chicks fed on conventional dietary amounts. British Poultry Science 32: 583–588.

Received for publication 8 June 1994.