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Mercury in Liver, Kidney, Feather and Muscle of Seabirds from Major Wetlands of the Caspian Sea, Iran

J. Aazami · A. Esmaili-Sari · N. Bahramifar · M. Ghasempouri · M. Savabieasfahani

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Abstract Concentration of mercury in tissues of the great cormorant (n = 18), mallard (n = 18), and coot (n = 15) of the Caspian Sea were determined. Liver mercury in the great cormorant, mallard, and coot were (5.7 ± 0.91; 0.3 ± 0.02; 0.09 ± 0.02). Kidney levels were (3.6 ± 2.24; 0.26 ± 0.03; 0.08 ± 0.02); feather (8.7 ± 0.8; 1.04 ± 0.16; 0.23 ± 0.15) and muscle were (2.26 ± 2.04; 0.11 ± 0.01; 0.03 ± 0.02) respectively. Mercury Tolerable Daily Intake limit is set at 5 μg g^{-1}. But even at levels that are currently considered "tolerable", mercury poisoning can occur in children and young who consume polluted game meat regularly.

Keywords: Mercury · Great cormorant · Mallard · Coot · Caspian Sea

Habitat destruction, as a result of anthropogenic pollution, is a serious challenge. Metal pollution is a well-known culprit and often causes irreparable damage to the environment. Mercury (Hg) pollution is especially worrisome because Hg levels have increased as much as two to tenfold during the past one and half centuries (Swain et al. 1992). Most alarming, is the fact that despite seriousness of the worldwide Hg pollution, no international legally-binding agreement exists to curb Hg release.

Wetlands that are among the most important habitats for breeding and foraging of various wildlife species, are threatened by Hg pollution. Many wildlife species that reside in wetlands are chronically exposed to Hg, which can lead to adverse behavioral, developmental, neurological, hormonal, and reproductive effects (Scheuhammer et al. 2007). The coastal wetlands of the Caspian Basin include many shallow and saline water pools, which attract a variety of birds and support great biodiversity. Anzali and Gomishan wetlands are home to more than 400 species of birds that are unique to the Caspian Sea area, they host many resident and migrating birds and provide important nesting habitat and stopovers.

Anzali Wetlands (37°28'N, 49°25'E) are an area of about 200 km^2 (Fig. 1). Anzali Wetlands, and other area wetlands, have been acknowledged for the important role they play in ecology of migratory birds and as especial habitat for waterfowl. Industrial, agricultural and domestic discharges enter the Eastern Anzali which supports a variety of plant-life (water depth between 0.8 to 1.5 m). Over six million $m^3$of polluted water enter Anzali, and there are no control of physicochemical parameters of these waters which can increase nutrient load and heavy metal content of the Anzali. Temperatures of Anzali range from 4.5°C in February to 27.5°C in August. Gomishan Wetlands (54°53'N, 37°9'E) consist of a series of narrow, brackish lagoons that lay on the eastern side of the Caspian Sea. Gomishan extends to Turkmenistan where it becomes a protected area for wildlife conservation. The average depth of Gomishan is 100 cm (max 250 cm) and the temperatures range between 4.5°C in February to 30.5°C in July. Over a hundred species of waterbirds and twenty species of fish have been spotted in these wetlands. East to

J. Aazami · A. Esmaili-Sari · N. Bahramifar · M. Ghasempouri
Tarbiat Modares University, School of Natural Resource and Marine Sciences, P.O. Box 46414-356, Noor, Mazandaran, Iran

N. Bahramifar
Department of Chemistry, Payame Noor University, Noor, Mazandaran, Iran

M. Savabieasfahani (✉)
P.O. Box 7038, Ann Arbor, MI 48107, USA
e-mail: mozhgan_savabieasfahani@hotmail.com

Fig. 1 Anzali and Gomishan Wetlands are indicated in *black*. These wetlands are located along the coast of the Caspian Sea, where birds of this study (great cormorant, mallards, and coot) were obtained in March of 2009

Gomishan is a vast area of low-lying plains with salt-tolerant vegetation. The recent rise of almost 2 m in the Caspian Sea level has flooded these plains and increased the size of Gomishan to $\sim$ 200 km^2.

We know that wetlands are ''hot spots'' for organic Hg production since they contain an abundance of dissolved organic matter and humic acid. Wetlands can also drastically affect concentration, speciation, and transport of Hg (Wang et al. 2002). Since wetlands play an important role in biogeochemical cycling of Hg, habitat conservation and preservation efforts, if implemented in wetland areas, can have significant positive outcomes.

Birds in this study, the great cormorant (*Phalacrocorax carbo*), mallard (*Anas platyrhynchos*), and coot (*Fulica atra*) are an important food source for the local population and provide a major source of protein. We opted to study Hg in these three species of aquatic birds to determine their safety for human consumption. Furthermore, we wanted to compare Hg pollution in the great cormorant, mallard, and coot of the Caspian region with values reported from other geographical regions. Our study was also partially in response to the increasing interest in bird conservation in Iran; as part of a worldwide recognition that maintaining and protecting habitat integrity and diversity is critical in preventing further loss of habitat, environmental degradation, and ecosystem destabilization and collapse.

Materials and Methods

In March 2009, under license of Environmental Protection Agency of Gilan and Gorgan, a total of fifty-one birds were shot and removed from Anzali (n = 23) and Gomishan (n = 28). They included great cormorant (n = 18); mallard (n = 18); and coot (n = 15). Birds were transported to the laboratory on ice. Sexual dimorphism in mallards allowed phenotypic identification of the sexes. In great cormorant and coot, sex was determined by identifying the sex organ during dissection. Bird's weigh and length were recorded. On the same day, liver, kidney, pectoral muscle, and breast feather were removed and kept in clean plastic bags in -20°C for later use. Sample preparation followed Spalding et al. (1994) with some modifications. Feathers were washed vigorously in deionized water alternated with acetone, then air dried overnight. All other tissues were placed in freeze-dryer for 48 h; then homogenized by Polytron Homogenizer.

Mercury was measured using a LECO AMA 254 Advanced Mercury Analyzer (USA). Accuracy of total Hg analysis was checked by running three samples of Standard Reference Materials (SRM), National Institute of Standard and Technology (NIST), SRM 1633b, SRM 2709 and SRM 2711 in seven replicates. Recovery varied between 94.8% and 104.2%. The detection limit was 0.001 mg/kg in dry weight.

Statistical Package for the Social Sciences software (SPSS version 17) was used for all statistical analysis. Data were tested for normality using Shapiro–Wilk's test. Data were not normally distributed; therefore a non-parametric test was used for analysis. Mercury concentrations in liver, kidney, feather, and muscle were tested for mean differences between species using Mann–Whitney *U* test. Probability level of $p \leq 0.05$ was set to indicate statistical significance.

Results and Discussion

Weight and length of birds are reported in Table 1. For all three species, Hg concentration and range are reported in Table 2. Table 3 tabulates *p* values for Mann–Whitney U tests; showing significance when comparing Hg concentrations among all species and tissues. Mercury concentrations differed significantly between the three species (Table 3). In general, metal exposure and tissue distribution in birds varies based on the route of exposure and duration of exposure (chronic or acute). Field and experimental studies in birds suggest that tissue distribution and accumulation of Hg also varies depending on growth stage, breeding status, molting and migration (Honda et al. 1986).

In this study, the fish-eating cormorants had higher Hg than the other two primarily herbivorous species, corroborating that species higher on the food chain, accumulate higher levels of contaminants. Over all, seabirds are particularly vulnerable to toxic effects of metals because they are long lived and are often at the top of their food chains. In the

Table 1 Biometrics, weight (g) and length (cm) of the great cormorant, mallard, and coot (mean ± SD) from the wetlands of the Caspian Sea, Iran

Species	Sex	Length (cm)	Weight(g)
Great cormorant (n = 18)	F (n = 7), M (n = 11)	95 ± 2 (89 – 97)	2111 ± 53 (1981 – 2256)
Mallard (n = 18)	F (n = 9), M (n = 9)	63 ± 2 (60 – 65)	1037 ± 30 (986 – 1098)
Coot (n = 15)	F (n = 6), M (n = 6) U (n = 3)	32 ± 0.1 (30 – 34)	598 ± 15 (589 – 602)

The range of values is indicated in the parentheses

F = Female, M = male, U = Unknown

Table 2 Mercury concentration (mean ± SD) (mg/kg dry weight) in the tissues of the great cormorant, mallard, and coot from the wetlands of the Caspian Sea, Iran

Species	Liver	Kidney	Feather	Muscle
Great cormorant	5.7 ± 0.91 (7 – 4)	3.6 ± 2.24 (8 – 1.4)	8.7 ± 0.8 (10 – 8)	2.3 ± 2 (8 – 1)
Mallard	0.3 ± 0.02 (0.51 – 0.08)	0.26 ± 0.03 (0.45 – 0.07)	1.04 ± 0.16 (1.6 – 0.5)	0.11 ± 0.01 (0.16 – 0.03)
Coot	0.09 ± 0.02 (0.14 – 0.05)	0.08 ± 0.02 (0.1 – 0.05)	0.23 ± 0.15 (0.3 – 0.1)	0.03 ± 0.02 (0.08 – 0.01)

The range of values is indicated in the parentheses

Table 3 Tabulated *p* values for Mann–Whitney U tests

		Great cormorant				Mallard				Coot			
		Liver	Kidney	Feather	Muscle	Liver	Kidney	Feather	Muscle	Liver	Kidney	Feather	Muscle
Great cormorant	Liver	_	0.02[b]	0.03[b]	0.02[b]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
	Kidney	_	_	0.05[b]	0.04[b]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
	Feather	_	_	_	0.03[b]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
	Muscle	_	_	_	_	<0.001[c]	<0.001[c]	0.003[b]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
Mallard	Liver	_	_	_	_	_	0.86[a]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
	Kidney	_	_	_	_	_	_	<0.001[c]	0.01[b]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
	Feather	_	_	_	_	_	_	_	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
	Muscle	_	_	_	_	_	_	_	_	<0.001[c]	<0.001[c]	<0.001[c]	<0.001[c]
Coot	Liver	_	_	_	_	_	_	_	_	_	0.78[a]	0.01[b]	<0.001[c]
	Kidney	_	_	_	_	_	_	_	_	_	_	0.003[b]	<0.001[c]
	Feather	_	_	_	_	_	_	_	_	_	_	_	<0.001[c]
	Muscle	_	_	_	_	_	_	_	_	_	_	_	_

[a] Not significant

[b] *p*–value < 0.05

[c] *p*–value < 0.001

great cormorant and mallard Hg concentration was highest in the liver followed by kidney, feather, and muscle; in coot Hg levels were highest in the feather > liver ∼ kidney > and muscle. There is conflicting data about differences in metal body burden of the sexes, but we saw no differences in metal body burden between the sexes. Table 4 is a review of the current literature on mercury levels in various tissues of the same species from other geographical locations.

In agreement with earlier reports the great cormorant in this study had significantly more Hg in the liver than in the kidney; indicating that in wild caught birds, liver Hg are consistently greater than kidney, which in turn exceed those in the muscle (Horai et al. 2007). Other studies, however, have shown that Hg in kidney is ≥ liver. In this study, liver (0.3 ± 0.02) and kidney (0.26 ± 0.03) had similar levels of Hg in the mallard; as they did in the coot (liver 0.09 ± 0.02; kidney 0.08 ± 0.02) (Table 2). A positive correlation was found between liver and kidney Hg in the mallard ($r = 0.76$, $p < 1001$) (Fig. 2). Coot had least Hg; this may be due to its diet (feeding on low trophic level mollusks and aquatic plants), its smaller size, and shorter life-span.

Muscle was the tissue with least Hg in all birds. At this time of the year, birds are least active and oxygen is abundant in the muscle tissue. Freadman (1979) suggests

Table 4 A review of the current literature on mercury levels in various tissues of the same species from other geographical locations; a comparison with this study

Location	Tissue	Mean	SD	References
Great Cormorant (*Phalacrocorax carbo*)				
Japan	Feather[b]	2.85	0.6	(Saeki et al. 2000)
Japan	Kidney[b]	4.05	2.18	
Japan	Liver[b]	3.39	1.39	
Japan	Muscle[b]	1.27	0.35	
Japan	Feather[a]	2.8	2.7	Nam et al. (2005)
Japan	Kidney[a]	14	9	
Japan	Liver[a]	12	9	
Japan	Muscle[a]	1.1	0.3	
Moravia	Muscle[b]	1	0.3	(Houserova et al. 2005)
Moravia	Liver[b]	10	8.3	
Moravia	Kidney[b]	2.7	2.1	
Iran	Kidney[c]	9.25	–	Mazloomi et al. (2008)
Iran	Liver[c]	8.32	–	
Iran	Feather[c]	4.42	–	
Iran	Muscle[c]	2.06	–	
Iran	Feather[b]	8.68	0.8	This study
Iran	Liver[b]	5.67	0.91	
Iran	Kidney[b]	3.59	2.24	
Iran	Muscle[b]	2.26	2.04	
Mallard (*Anas platyrhynchos*)				
United States	Liver[b]	0.47	0.94	Gerstenberger (2004)
United States	Muscle[b]	0.06	0.06	
Iran	Feather[b]	1.04	0.16	This study
Iran	Liver[b]	0.3	0.02	
Iran	Kidney[b]	0.26	0.03	
Iran	Muscle[b]	0.11	0.01	
Coot (*Fulica spp*)				
United States	Liver[a]	0.06	–	White et al. (1986)
United States	Liver[b]	0.22	0.31	Gerstenberger (2004)
United States	Muscle[b]	0.03	0.01	
Iran	Feather[b]	0.5	0.01	Zolfaghari et al. (2009)
Slovakia	Leg skeletal muscles[b]	0.01	–	Gasparik et al. (2010)
Iran	Feather[b]	0.23	0.15	This study
Iran	Liver[b]	0.09	0.02	
Iran	Kidney[b]	0.08	0.02	
Iran	Muscle[b]	0.03	0.02	

[a] :mg/kg w w, [b]: mg/kg d w, [c]: mg/kg undefined

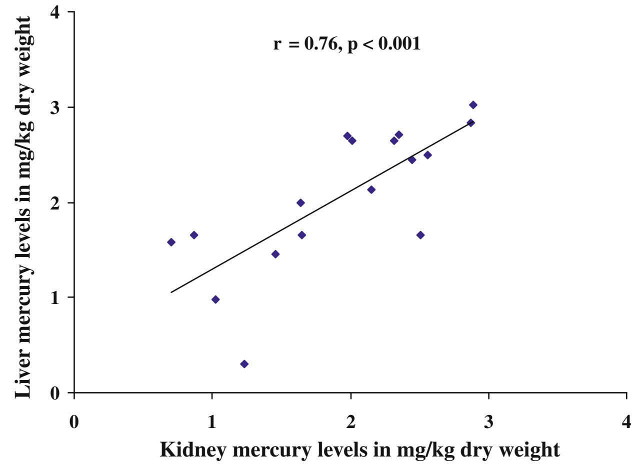

Fig. 2 Correlation between liver and kidney mercury (values are mean ± SD, mg/kg dry weight) in the mallard (n = 18) obtained from wetlands of the Caspian Sea in March 2009

that oxidation of Hg compounds and their rapid excretion can rid the muscle tissue from Hg, while in other tissues, where oxidation is low, Hg levels are higher. Over all, muscle is described as a storage site for Hg while liver has a buffering role in Hg accumulation. After liver capacity for withholding Hg runs out, Hg can be diverted to the muscle, at which time its concentration in the muscle starts increasing. Henny et al. (2002) theorizes that as exposure to methylmercury increases, so does the percentage of hepatic inorganic Hg, which indicates greater hepatic demethylation. As a result of metallothioneins' binding and immobilization of inorganic Hg, liver concentrations of Hg relative to muscle may increase.

Mercury levels of 5 mg/kg dry weight in feathers is often associated with impaired reproduction (Burger and Gochfeld 2000); the great cormorant (8.68 ± 0.78) of this study seems to be the only bird at risk of reproductive impairments.

Mercury in the great cormorant and mallard of the Caspian wetlands was higher than values reported form the Czech Republic, Japan and the United States. Mercury in coot was similar to what has been reported from Slovakia and the United States.

The World Health Organization has set 5 μg g^{-1} as the tolerable daily intake limit for Hg, while the European Union has sets the safety level for Hg at 0.5 μg g^{-1} of fresh weight. Yet other investigators argue that even at levels currently considered "tolerable", Hg poisoning can occur in children and young adults who consume polluted game meat regularly (Nriagu et al. 1992). We conclude that consumption of these species may therefore be hazardous to public health in Iran.

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References

Burger J, Gochfeld M (2000) Metal levels in feathers of 12 species of seabirds from Midway Atoll in the northern Pacific Ocean. Sci Total Environ 257:037–052

Freadman M (1979) Role partitioning of swimming musculature of striped Bass morone saxatilis walbaum and bluefish, pomatomus saltatrix L. J Fish Biol 15:417–423

Gasparik J, Vladarova D, Capcarova M, Smehyl P, Slamecka J, Garaj P, Stawarz R, Massanyi P (2010) Concentration of lead, cadmium, mercury and arsenic in leg skeletal muscles of three species of wild birds. J Environ Sci Health, Part A: Toxic/Hazard Subst Environ Eng 45:818–823

Gerstenberger S (2004) Mercury concentrations in migratory waterfowl harvested from Southern Nevada Wildlife Management Areas, USA. Environ Toxicol 19:035–044

Henny C, Hill E, Hoffman D, Spalding M, Grove R (2002) Nineteenth century mercury: hazard to wading birds and cormorants of the Carson River, Nevada. Ecotoxicology 11:213–231

Honda K, Min B, Tatsukawa R (1986) Distribution of heavy metals and their age-related changes in the eastern great white egret, Egretta alba modesta, in Korea. Arch Environ Contam Toxicol 15:185–197

Horai S, Watanabe I, Takada H, Iwamizu Y, Hayashi T, Tanabe S, Kuno K (2007) Trace element accumulations in 13 avian species collected from the Kanto area, Japan. Sci Total Environ 373:512–525

Houserova P, Hedbavny J, Matejicek D, Kracmar S, Sitko J, Kuban V (2005) Determination of total mercury in muscle, intestines, liver and kidney tissues of cormorant (*Phalacrocorax carbo*), great crested grebe (*Podiceps cristatus*) and Eurasian buzzard (*Buteo buteo*). Vet Med Czech 2:61–68

Mazloomi S, Esmaeili A, Ghasempoori S, Omidi A (2008) Mercury distribution in liver, kidney, muscle and feathers of Caspian sea common cormorant (*Phalacrocorax carbo*). Res J Environ Sci 2:433–437

Nam D, Anan Y, Ikemoto T, Okabe Y, Kim E, Subramanian A, Saeki K, Tanabe S (2005) Specific accumulation of 20 trace elements in great cormorants (*Phalacrocorax carbo*) from Japan. Environ Pollut 134:503–514

Nriagu J, Pfeiffer W, Malm O, de SOUZA C, Mierle G (1992) Mercury pollution in Brazil. Nature 356:389

Saeki K, Okabe Y, Kim EY, Tanabe S, Fukuda M, Tatsukawa R (2000) Mercury and cadmium in common cormorants (*Phalacrocorax carbo*) Enviro Pollut 108:249–255

Scheuhammer A, Meyer M, Sandheinrich M, Murray M (2007) Effects of environmental methylmercury on the health of wild birds, mammals, and fish. Ambio 36:12–19

Spalding M, Bjork R, Powell G, Sundlof S (1994) Mercury and cause of death in great white herons. JWM 58:735–739

Swain E, Engstrom D, Brigham M, Henning T, Brezonik P (1992) Increasing rates of atmospheric mercury deposition in midcontinental North America. Science 257:784

Wang Q, Liu R, Lu X, Li Z (2002) Progress of study on the mercury process in the wetland environment. Adv Earth Sci 17:881–885

White D, King K, Mitchell C, Mulhern B (1986) Trace elements in sediments, water, and American coots (Fulica americana) at a coal-fired power plant in Texas, 1979–1982. Bull Environ Contam Toxicol 36:376–383

Zolfaghari G, Esmaili-Sari A, Ghasempouri SM, Baydokhti RR, Hassanzade Kiabi B (2009) A multispecies-monitoring study about bioaccumulation of mercury in Iranian birds (Khuzestan to Persian Gulf): Effect of taxonomic affiliation and trophic level. Environ Res 109:830–836