Research Article

Mercury Concentrations in Tissues of Osprey From the Carolinas, USA

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ABSTRACT Mercury (Hg) contamination is believed to be one of the most significant pollution hazards to wildlife in the southeastern United States, yet comprehensive studies of Hg contamination of piscivorous raptors are rare in this region. We analyzed total Hg (THg) concentration in tissues of 39 osprey (*Pandion haliaetus*) primarily from coastal counties of South Carolina, USA, to describe tissue distribution of Hg and to determine whether age or sex influenced Hg accumulation. To determine whether Hg poses health risks to osprey breeding in this region, we also measured selenium in all tissues and the percentage of THg that was methylated in a subset of individuals. Osprey with adult plumage tended to have higher and more variable Hg concentrations in their tissues than younger birds. Whereas highest concentrations of Hg were found in liver and kidney of older birds, chicks had highest concentrations in keratinized tissues. Mercury concentrations were correlated between feathers and soft tissues, but talon concentrations of Hg were better correlated with organs than to feathers in most cases. Contrary to previous studies on birds, we found no relationship between Hg concentration in primary feathers and the sequence in which the feather was molted. We attribute this observation to the irregular and protracted molting pattern of osprey. Also contrary to other studies, feather concentrations of Hg were considerably lower than concentrations in liver and kidney. Osprey with high concentrations of Hg in their livers and kidneys accumulated as much as 99% of it as Hg(II), suggesting that demethylation and sequestration of Hg(II) may be even more critical to mitigating adverse effects than it is for other birds that eliminate most of their Hg burden in feathers. In addition, selenium was co-sequestered with Hg in the liver and kidneys and may further mitigate any adverse effects. Based on these findings, we suggest that most osprey in this region are not currently at risk of Hg toxicosis, but recommend that additional ecotoxicological studies be performed to monitor risk to osprey in this coastal region facing heavy development. We also suggest that concentrations of Hg in talon and claw may serve as important indicators of previous exposure and provide useful information for natural resource managers seeking to assess health risks to birds. (JOURNAL OF WILDLIFE MANAGEMENT 71(6):1819–1829; 2007)

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Mercury (Hg) is one of the most important environmental pollutants affecting wildlife populations around the globe. This highly toxic metal is readily methylated and transferred through food webs where it bioaccumulates to very high concentrations in long-lived organisms and top-level predators (Fitzgerald and Clarkson 1991, Facemire et al. 1995). Due to long-range atmospheric transport, Hg is globally ubiquitous (Weiner et al. 2003) and thus poses threats to wildlife in remote and otherwise undisturbed locations (Fitzgerald et al. 1998). When high concentrations of Hg are bioaccumulated, wildlife often experience neurological symptoms due to the propensity for methylmercury (MeHg) to cross the blood–brain barrier (Heinz 1996). At considerably lower concentrations, bioaccumulation of Hg can impair reproduction by altering behaviors, such as nest attendance (Barr 1986), and through decreased reproductive output, possibly due to endocrine disruption (Hammerschmidt et al. 2002, Drevnick and Sandheinrich 2003). Moreover, maternal transfer of Hg can disrupt early development, resulting in reduced embryonic survival, increased mortality in young, and neurological impairment and behavioral abnormalities in surviving offspring (Heinz 1976, 1979, 1996; Finley and Stendell 1978).

Mercury tends to be highly bioavailable in aquatic habitats that are critical breeding and foraging grounds for a variety of wildlife. Because MeHg readily bioaccumulates in fish (Bloom 1992), piscivorous wildlife are particularly at risk of exposure (Wiener and Spry 1996). For example, common loons (*Gavia immer*) are relatively long-lived obligate piscivores that are capable of accumulating high concentrations of Hg in their tissues (Evers et al. 2003). Some larger birds such as the osprey (*Pandion haliaetus*) share these characteristics and also tend to eat larger fish than other avian obligate piscivores. Larger fish tend to bioaccumulate higher concentrations of Hg than smaller fish (Wiener and Spry 1996), placing these raptors of conservation concern at substantial risk in Hg-contaminated systems (Häkkinen and Häsänen 1980).

Mercury contamination is believed to be the single most pervasive pollution hazard to wildlife in the southeastern United States (Facemire et al. 1995), yet comprehensive studies of Hg contamination of piscivorous raptors are rare in this region compared to portions of Europe, the northern United States, and Canada. In response to this important knowledge gap, the current study describes Hg accumu-

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lation in osprey collected primarily from coastal counties of South Carolina, USA. Specifically, we addressed the following 7 questions: 1) Do total Hg (THg) concentrations vary among 10 types of tissue collected from osprey? 2) Because natural resource managers must often rely on nonlethal methods to assess risk to osprey and other raptors, are THg concentrations correlated among nondestructive tissues (feathers and talons) and target organs? 3) Does the position of a primary feather in the molting sequence influence its THg concentration? 4) Does age affect THg accumulation patterns in osprey tissue? 5) Among birds with adult plumage, does sex affect THg accumulation patterns in osprey tissue? 6) Are THg concentrations in osprey tissues from this region comparable to those of osprey from other regions of the globe? 7) Are THg concentrations in osprey breeding in this region high enough to be of toxicological concern? To address this last question we also describe the relationship between selenium (Se) and Hg among tissues and the percentage of Hg that was methylated, both of which influence Hg toxicity.

STUDY AREA

The medical clinic of the International Center for Birds of Prey (ICBP) admits approximately 400 ill or injured raptors per year, including 20–25 osprey annually. For this study, we utilized 39 osprey from 11 counties in South Carolina and 1 county in North Carolina, USA: Beaufort, Berkeley, Charleston, Chesterfield, Colleton, Dorchester, Georgetown, Horry, Jasper, Newberry, and Sumter counties, South Carolina, as well as Brunswick County, North Carolina. All birds were dead upon arrival at ICBP, died during medical treatment, or were euthanized at the center between 20 May 2003 and 24 May 2005. Osprey carcasses were immediately frozen at the ICBP and then transported on ice to the Savannah River Ecology Laboratory where they were stored frozen until dissection. Collection of osprey was in conformance with appropriate permits administered to the ICBP.

METHODS

Study Species

In the United States, osprey breed most extensively along the eastern, western, and Gulf coasts as well across the Great Lakes region. Inland populations also occur along rivers, lakes, and reservoirs where appropriate nesting habitat and fish abound. The majority of osprey breeding in the United States migrate south during the winter, where they spend up to 7 months in Central and South America (Poole 1989, Poole et al. 2002). On the eastern seaboard, the entire breeding population is believed to winter in the West Indies (22%) or South America (78%; Poole and Agler 1987).

Osprey can live up to 25 years, but <10% of birds generally live past the age of 12 (reviewed in Poole et al. 2002). Their young are semi-precocial and are fed fish by parents that are generally collected within several kilometers of the nest, but in some cases osprey may focus on productive foraging areas at greater distances (e.g., 14 km; Hagan 1986). Fledging occurs within 50–55 days of hatching, but independence from parental feeding usually takes several weeks to months (Poole et al. 2002). Hatch-year birds are characterized by dark brown dorsal contour feathers with buffy white borders. By 18 months of age, immature plumage becomes indistinguishable from that of adult birds; the buffy dorsal plumage is replaced by solid brown contour feathers (Johnsgard 1990).

Tissue Collection

We weighed thawed carcasses and recorded the wingspan, total length, foot length, and beak depth. We characterized birds as 1) chicks, 2) hatch-year birds, or 3) subadult or adult (hereafter referred to as ad) based on plumage characteristics. We subjectively assessed body condition (K) of birds by palpating tissue depth at the keel, and we assigned a relative rank of I–IV (I = severely emaciated, IV = obese).

We collected 6 tissue samples from carcasses that may function as useful, nondestructive metrics of Hg exposure. We collected primary feathers numbers 1, 5, and 10 (numbered from innermost first molted to outermost last molted) from the left wing of each bird to assess the effect of molt sequence on feather Hg concentration. In 5 cases, we collected primary feathers from the right wing due to confounding injuries to the left wing. We also removed 10–20 contour feathers from the left breast and left scapular regions. Finally, we clipped a 5-mm portion of the talon from digits 2 and 3 on the left foot of each bird (i.e., front-facing, innermost talons; Proctor and Lynch 1993). The quantity of talon collected was limited to that which could be collected noninvasively (i.e., without disturbing the talon's blood supply).

We rinsed feathers and talons prior to analysis following a modified protocol of Burger and Gochfield (1993). We agitated feathers and talons in trace metal–grade acetone for 30 seconds. We then rinsed tissues 3 consecutive times in 18-MΩ deionized water before storing them frozen.

After feather removal, we dissected birds to obtain soft-tissue samples. We removed portions of the liver (approx. 20 g), kidney (approx. 3 g), and brain (approx. 6 g) from each bird. We removed an approximately 20-g sample of muscle from the left breast of each osprey. We froze all soft tissues at −70° C and stored them until analysis. Gonads were visible in 35 birds, but in many cases sexes still remained indistinguishable because birds were either immature or gonads were regressed. In cases where we could not confirm the sex visually, we removed a portion of the gonad and preserved it in 10% neutral buffered formalin. We embedded gonads in paraffin and sectioned them at 5 μm. We stained thin sections with hematoxylin and eosin and we confirmed sexes using light microscopy at 100–400× magnification.

Digestion and Hg and Se Analysis

We digested 25–500 mg of dried feathers and lyophilized soft tissues, and approximately 40 mg of dried talon, by adding concentrated trace metal–grade nitric acid (2.5–5.0 mL) to samples in fluoropolymer digestion vessels prior to

digestion in a microwave (MARS-5; CEM Corp., Matthews, NC) according to United States Environmental Protection Agency (EPA) method 3052 (EPA 1996). After digestion, we brought samples to a final volume of 10 mL with 18-MΩ deionized water.

We performed Hg and Se analysis by inductively coupled plasma mass spectrometry (ICP-MS; Perkin Elmer, Norwalk, CT) on diluted samples according to the EPA method 6020a (EPA 1998). We calibrated the instrument daily using calibration standards covering a range of 0.02–10 μg/L prepared by serial dilution of National Institute of Standards and Technology (NIST) traceable primary standards. For quality-control purposes, we included certified reference material (TORT-2; National Research Council of Canada [NRCC], Ottawa, ON, Canada) and reagent blanks in the digestion and analysis procedure. Mean percent recoveries for Hg and Se in tissue reference material were 92% and 108%, respectively. Method detection limits for Hg and Se varied according to sample mass, but ranged from 2.6 ng Hg/g to 46.1 ng Hg/g and 5.4–90.9 ng Se/g dry mass. We reported concentrations of Hg and Se in tissues on a dry-mass basis. In cases where we compared our Hg concentrations to soft-tissue concentrations that were reported on a wet-weight basis in the literature, we converted these values to dry mass, assuming 80% moisture content (i.e., we multiplied the concentration by 5).

Mercury Speciation

We analyzed subsamples ($n = 4$ individuals per age class) of brain, muscle, kidney, liver, and scapular feather for MeHg. We performed mercury speciation analysis on tissue subsamples according to the method of Liang et al. (1994) as modified by Hammerschmidt and Sandheinrich (2005). Briefly, we weighed approximately 50 mg of dried homogenized tissue or feathers into 15-mL polypropylene centrifuge tubes to which we added 5–10 mL of 4.7-M trace metal–grade HNO_3. We sealed and heated the tubes to 60° C in a convection oven for approximately 12 hours. We centrifuged the resulting digests at 1,000× g for 20 minutes to remove any insoluble material. This procedure quantitatively releases both Hg(II) and MeHg, which are the only Hg species found in biological tissues (Liang et al. 1994, Hammerschmidt and Sandheinrich 2005). We then analyzed aliquots of the supernatants by aqueous phase ethylation followed by room temperature precollection and isothermal gas chromatography with cold vapor atomic fluorescence detection (Liang et al. 1994). We then determined total mercury in the extracts using ICP-MS as described above in order to determine percent MeHg. Both THg and MeHg standards were NIST traceable. We processed and analyzed the certified reference materials TORT-2, DORM 2, and DOLT 2 (lobster hepatopancreas, dogfish muscle, and dogfish liver, respectively; NRCC, Ottawa, ON, Canada), as well as blank samples and samples spiked with standards along with the unknowns. Mean percent recoveries for certified reference materials ranged from 94% to 96%. The method detection limit for MeHg was 0.16 ng Hg/g dry mass. Spike recovery averaged 101%.

Statistical Analysis

Because we were unable to collect ≥1 tissue type from approximately 25% of osprey in our study, we did not use multivariate statistics (e.g., multivariate analysis of variance) in order to avoid elimination of these birds from our analysis. Instead, we compared Hg concentrations in each type of tissue among age classes using analysis of variance (ANOVA) followed by Tukey's pair-wise comparisons. Among birds with adult plumage, we also examined the effect of sex on Hg concentration in each tissue using ANOVA. Because studies have shown that Hg concentrations often vary among primary feathers sampled from the same bird, we also used a series of paired t-tests to compare Hg concentrations among primary feathers numbers 1, 5, and 10. Concentrations of Hg in all of these models were log transformed prior to analysis to meet assumptions of parametric statistics. In our models examining age class and sex, we used a sequential Bonferroni adjustment to accommodate multiple, non-independent comparisons.

We used Pearson correlation coefficients to assess relationships of THg concentrations between tissue types, as well as with body size. We performed correlations on $\log(x + 1)$–transformed data because examination of scatterplots suggested that log transformation was required to more closely approximate assumptions of normality and linearity. Again, we used a sequential Bonferroni adjustment to accommodate multiple comparisons. Because no measures of body size (mass, wingspan, length, or foot length) were related to Hg concentrations in tissues ($\bar{x}\ r = 0.05$, $r \leq 0.47$ in all cases), we did not include body size in any of our analyses.

We handled Se and MeHg data as follows. For each tissue, we examined the relationship between THg and Se using Pearson correlation coefficients. The small number of samples analyzed for MeHg precluded statistical comparisons among age classes. However, it was clear qualitatively that percent MeHg was much lower in the kidney and liver of adult birds than in younger age classes (see Results). Therefore, we examined the relationship between percent MeHg and (log) THg in liver and kidney across age classes using linear regression.

RESULTS

Mean THg concentrations (μg/g dry wt) varied among tissues, but the hierarchical sequence of concentrations varied according to age class. In all age classes, breast and scapular feathers had the lowest THg concentrations (Figs. 1–3; Table 1). In chicks, mean THg concentrations were highest in talon, followed by liver and primary feathers. Muscle, kidney, and brain concentrations of THg were generally low in chicks. In hatch-year birds, liver supplanted talon as the tissue with greatest THg concentrations, followed by muscle and primary feathers (Figs. 1–3; Table 1). In contrast, among birds with adult plumage, mean kidney concentrations of THg exceeded concentrations

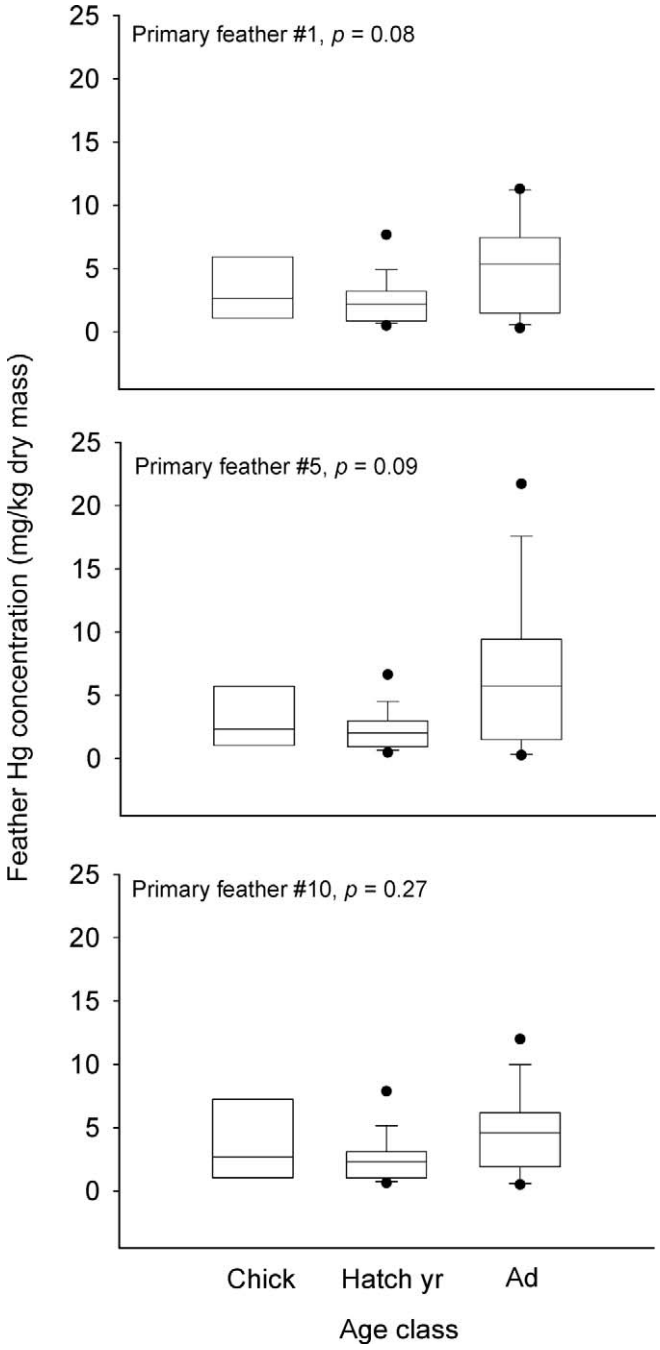

Figure 1. Box plots of total mercury (Hg) concentrations in primary feather numbers 1, 5, and 10 from 3 age classes of osprey collected in the Carolinas, USA, from May 2003 to May 2005. Results of statistical comparisons are presented on each graph.

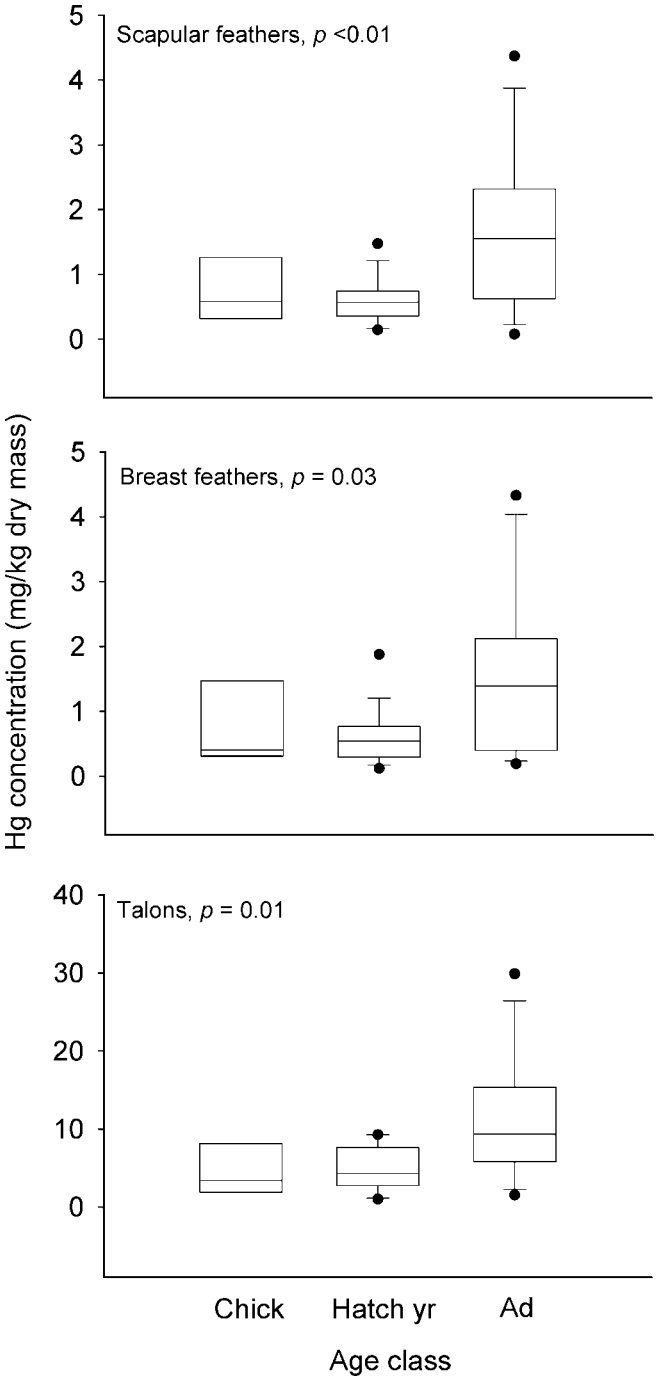

Figure 2. Box plots of total mercury (Hg) concentrations in scapular feathers, breast feathers, and talons from 3 age classes of osprey collected in the Carolinas, USA, from May 2003 to May 2005. Results of statistical comparisons are presented on each graph. After Bonferroni adjustment, none of these statistical comparisons were significant, but in each case there was a noteworthy tendency for adult osprey to differ from hatch-year birds.

found in the liver and talon, followed by primary feathers, muscle, and brain.

There was a general trend for adults to have higher and more variable THg concentrations than chicks and hatch-year birds (Figs. 1–3), but this tendency was only statistically significant for liver ($F_{2,36} = 11.02$, $P < 0.001$) and kidney ($F_{2,36} = 13.81$, $P < 0.001$) after Bonferroni adjustment. Age class did not significantly affect THg concentration in any of the primary feathers (in all cases $F \leq 2.67$, $P \geq 0.083$), muscle ($F_{2,36} = 3.00$, $P = 0.062$), or brain ($F_{2,34} = 3.21$, $P = 0.053$). Although not statistically significant after correction for multiple comparisons, there was a noteworthy tendency of older birds to accumulate more THg in scapular feathers ($F_{2,36} = 5.66$, $P = 0.007$), breast feathers ($F_{2,35} = 4.02$, $P = 0.027$), and talons ($F_{2,36} = 4.88$, $P = 0.013$) compared to hatch-year birds. Chicks and hatch-year birds had similar concentrations of THg in all tissues.

Among the 17 osprey with adult plumage, we were able to

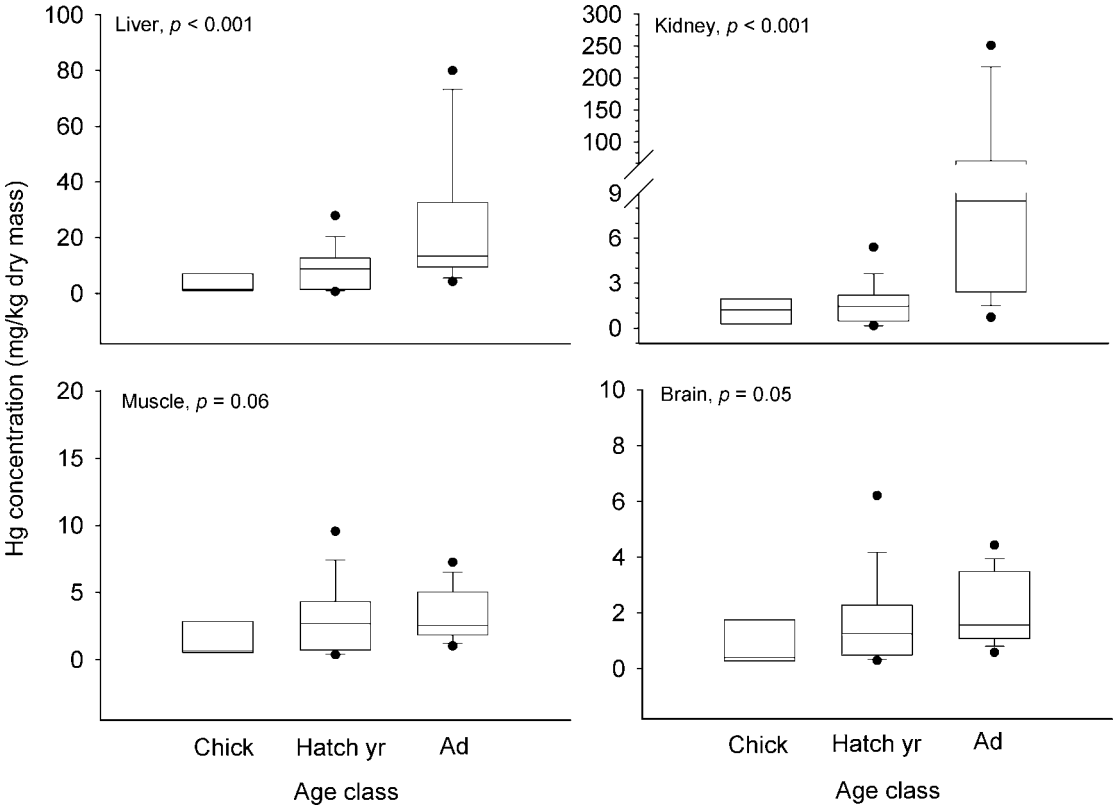

Figure 3. Box plots of total mercury (Hg) concentrations in soft tissues from 3 age classes of osprey collected in the Carolinas, USA, from May 2003 to May 2005. Results of statistical comparisons are presented on each graph. For liver and kidney, common superscripts identify age classes that did not differ from one another statistically.

confirm sexes on 7 females and 8 males. Within each sex, tissue concentrations of THg were highly variable, ranging by as much as 9-fold for specific tissues. After we corrected for multiple comparisons, there was no statistically significant difference between sexes in THg concentration in any tissue ($0.022 \leq P \leq 0.906$).

Mercury concentrations were significantly correlated between every tissue type analyzed, even after our conservative adjustment of α, but the strength of the 45 correlations varied widely (Table 2). Mercury concentrations in the 5 feather types were strongly correlated with one another (in all cases $r > 0.67$, $P < 0.001$). Talons exhibited comparable correlation with each feather type (in all cases $r > 0.67$, $P < 0.001$). However, Hg concentrations in talons were more closely related to Hg concentrations in liver, muscle, and brain ($r = 0.67$–0.73, $P < 0.001$) than any of the feather types were to these 3 soft tissues ($r = 0.38$–0.65; Table 2). Although talon concentrations were also related to kidney concentrations ($r = 0.55$, $P < 0.001$), the strongest relationship between kidney and a nondestructive tissue was with scapular feathers ($r = 0.69$, $P < 0.001$). Mercury concentrations were similar among the primary feather numbers 1, 5, and 10 (in all cases, $P > 0.68$; Fig. 1). Among these 3 feathers, primary number 1 was typically most strongly correlated with Hg concentrations in soft tissues (Table 2). Mercury concentrations were also correlated between soft tissues, and in some cases these relationships were extremely strong (e.g., muscle vs. brain; $r = 0.96$, $P < 0.001$).

Median percent MeHg was fairly high and similar (72–98%) in feather and muscle of osprey, and intermediate (62–72%) in brain tissue. However, percent MeHg varied substantially among age classes for both kidney and liver (Table 3). Whereas median percent MeHg ranged from 52% to 69% in liver and kidney of young birds, the percent MeHg in the same tissue from adult birds was consistently lower (median 9–29%). When we combined age classes for regression analysis, there was no relationship between THg and percent MeHg in liver ($r^2 = 0.17$, $P = 0.18$). However, there was a strong negative relationship between THg and percent MeHg in the kidney ($r^2 = 0.74$, $P < 0.001$). There were also strong relationships between Se and THg in both liver and kidney ($P < 0.001$; Table 4; Fig. 4), but no relationship was evident between these elements in any other tissue analyzed (Table 4).

DISCUSSION

Comparison of Tissues

Our study represents the only detailed study of Hg accumulation in osprey from the southeastern United States and one of the most comprehensive reports of Hg accumulation in osprey feathers and soft tissues from any portion of the species' range. The bulk of previous studies on Hg accumulation in osprey have focused on concentrations in eggs and feathers, with relatively few reporting concen-

Table 1. Summary of mercury concentrations reported in feathers and soft tissues of osprey, 1968–2007.[a]

Age	$\bar{x}$[b]	Range	Comments	Study
Feathers				
Chick	0.8	0.3–2.1	Scapular feathers	Current study
	0.8	0.3–2.1	Breast feathers	Current study
	2.1	1.8–2.7	Mantle feathers; mean is from multiple yrs, range is based on 1 yr of data	Hughes et al. 1997
	3.3	1.1–5.2	Mantle feathers	Hughes et al. 1997
	3.4	NR[c]	Mantle feathers	Hughes et al. 1997
	3.5	0.9–9.0	Primary feathers nos. 1, 5, 10	Current study
	4.6	1.6–6.8	Mantle feathers	Hughes et al. 1997
	7.0	2.1–26.5	Conglomerate of feather types	DesGranges et al. 1998
	7.4	5.8–10.1	Mantle feathers	Hughes et al. 1997
	11.0	7.6–17.0	Mantle feathers	Hughes et al. 1997
	12.6	4.2–20.9	Mantle feathers	Häkkinen and Häsänen 1980
	21.8	11.8–51.2	Mantle feathers	Häkkinen and Häsänen 1980
	37.4	5.5–101.0	Conglomerate of feather types	DesGranges et al. 1998
Hatch yr	0.6	0.1–1.5	Scapular feathers	Current study
	0.6	0.1–1.9	Breast feathers	Current study
	2.4	0.5–7.9	Primary feathers nos. 1, 5, 10	Current study
Ad	1.5	0.2–4.3	Breast feathers	Current study
	1.6	0.1–4.4	Scapular feathers	Current study
	3.0–16.5	NR	Feather type and age of birds NR; 5 means from museum specimens estimated from figure	Johnels et al. 1968
	5.2	0.3–21.7	Primary feathers nos. 1, 5, 10	Current study
	6.7	5.3–7.6	Wing and tail feathers	Hughes et al. 1997
	16.5	1.2–68.0	Conglomerate of feather types	DesGranges et al. 1998
	21.1	7.5–47.9	Wing and tail feathers	Hughes et al. 1997
	28.8	17.3–40.2	Wing and tail feathers	Hughes et al. 1997
	58.1	5.3–193.0	Conglomerate of feather types	DesGranges et al. 1998
Liver				
Chick	0.7	0.2–1.3		DesGranges et al. 1998
	1.5	NA[d]	Single bird	Wiemeyer et al. 1980
	2.6	2.0–3.1		Wiemeyer et al. 1987
	3.4	NA	Single bird	Noble and Elliot 1990
	3.5	0.8–10.3		Current study
	3.6	1.0–8.5		DesGranges et al. 1998
	4.0	0.8–7.1		Häkkinen and Häsänen 1980
Hatch yr	8.6	0.7–27.9		Current study
	13.5	1.8–31.0		Wiemeyer et al. 1980
	71.6	17.0–175.0		Wiemeyer et al. 1987
Ad	15.5	NA	Single bird	Noble and Elliot 1990
	22.9	1.4–65.0		Wiemeyer et al. 1987
	24.4	4.3–80.0		Current study
	74.5	3.9–337.5		Wiemeyer et al. 1980
Kidney				
Chick	0.9	0.4–2.1		DesGranges et al. 1998
	1.2	0.3–2.0		Current study
	3.3	0.8–6.8		Häkkinen and Häsänen 1980
	5.3	1.4–11.8		DesGranges et al. 1998
Hatch yr	1.6	0.2–5.4		Current study
	9.2	1.0–32.5		Wiemeyer et al. 1980
Ad	49.1	0.7–251.0		Current study
	225	6.0–650.0	$n = 3$	Wiemeyer et al. 1980
Muscle				
Chick	0.4	0.1–0.7		DesGranges et al. 1998
	1.5	0.3–4.2		Current study
	1.6	0.5–3.4		Häkkinen and Häsänen 1980
	1.8	0.5–3.9		DesGranges et al. 1998
Hatch yr	2.9	0.4–9.6		Current study
Ad	3.4	1.0–7.3		Current study

trations in soft tissues, especially in older birds. A compilation of extant data (Table 1) reveals that mean feather concentrations in osprey from the coastal region of the southeastern United States are among the lowest ever documented in osprey. However, feather concentrations in our study spanned up to a 10-fold range within each age class, suggesting substantial variability in environmental conditions and exposure across our study area. Such a wide range of Hg concentrations is expected in adult birds that migrate and may thus encounter different levels of contamination on the wintering grounds. However, the fact that feathers from young birds spanned an equally large

Table 1. Continued.

Age	$\bar{x}$[b]	Range	Comments	Study
Brain				
Chick	0.2	0.1–0.4		DesGranges et al. 1998
	0.8	0.3–2.0		Current study
	1.0	0.2–1.9		DesGranges et al. 1998
	1.5	0.4–2.9		Häkkinen and Häsänen 1980
Hatch yr	1.6	0.3–6.2		Current study
Ad	2.1	0.6–4.4		Current study

[a] All concentrations reported as μg/g dry mass.
[b] In cases where data reported as wet mass, we converted to dry mass, assuming 80% moisture.
[c] NR = not reported.
[d] NA = not applicable.

fold-range of concentrations suggests that there are hot spots of Hg contamination or enhanced bioavailability along the South Carolina coast. Some of our results for soft tissues further support this contention. For example, one of the chicks we sampled had the highest concentration of THg in its liver ever documented in an osprey chick, even compared to osprey from areas of known industrial inputs (Table 1).

Our study generally supports previous studies that suggest body feathers are better indicators of Hg exposure than flight feathers (Furness et al. 1986, Furness and Camphuysen 1997), but to our knowledge, our study is the first to report talon or claw concentrations of any contaminant from any bird species. Of the 10 tissues we analyzed, we found that talons contained the third highest concentrations of Hg (all age classes combined) and were up to 8 times more contaminated with Hg than various types of feathers. Like feathers, talons are keratinized tissues and would therefore be expected to accumulate Hg during formation. However, the keratinization of talons is different from that of feathers (Sawyer et al. 2005), which may influence the relative affinity of these 2 tissues for Hg. Importantly, Hg concentrations in talons were usually more strongly correlated with Hg concentrations in soft tissues than were concentrations in feathers. The strength of the correlation between concentrations of Hg in talons and other tissues may stem from the fact that unlike feathers, talons grow continuously (Bearhop et al. 2003). Thus, talons are likely representative of the recent body pool of Hg (i.e., within a few months of sampling), whereas Hg in a feather is reflective of the amount of Hg that was in circulation in the blood when a particular feather formed (Burger 1993). In birds with complex molting or migration patterns, feather concentrations can sometimes be difficult to interpret, and talons may therefore provide useful complementary information regarding Hg exposure. Based on our findings and the fact that small portions of talons can be sampled with temporary disturbance to the bird, future studies of the use of talons and claws as a nondestructive index of Hg exposure will be important for establishing this technique as a monitoring tool.

Another key finding from our study was the lack of relationship between molt sequence and Hg concentration of primary feathers of osprey. This result contrasts with extensive previous work on birds with discrete molting periods, which suggests that feathers formed earlier in the molting process have higher concentrations of Hg than those molted later (e.g., Furness et al. 1986). This pattern is believed to occur as MeHg mobilized from the labile body pool during molting decreases as the molt proceeds, reducing the total amount of Hg available for deposition in feathers later in the molting sequence. We suggest that this pattern of Hg deposition in feathers was not present in osprey due to their *Staffelmauser* molting pattern. Osprey exhibit an irregular molt of the primary feathers, in which molting occurs in successive waves throughout the year, starting at primary number 1 and preceding to primary number 10, and only ceases during breeding and migration (Prevost 1983, Poole et al. 2002). After molting ceases for

Table 2. Pearson correlation matrix describing the relationships between 10 types of tissue sampled from osprey collected in the Carolinas, USA, from May 2003 to May 2005.

Tissue type	Primary feather no. 1	Primary feather no. 5	Primary feather no. 10	Scapular feathers	Breast feathers	Talon	Kidney	Liver	Muscle	Brain
Primary feather no. 1										
Primary feather no. 5	0.74									
Primary feather no. 10	0.86	0.67								
Scapular feathers	0.85	0.85	0.83							
Breast feathers	0.82	0.84	0.84	0.98						
Talon	0.67	0.78	0.70	0.85	0.83					
Kidney	0.59	0.59	0.34	0.69	0.67	0.55				
Liver	0.58	0.56	0.42	0.65	0.63	0.73	0.80			
Muscle	0.48	0.38	0.41	0.53	0.53	0.67	0.49	0.86		
Brain	0.50	0.42	0.48	0.60	0.58	0.70	0.57	0.87	0.96	

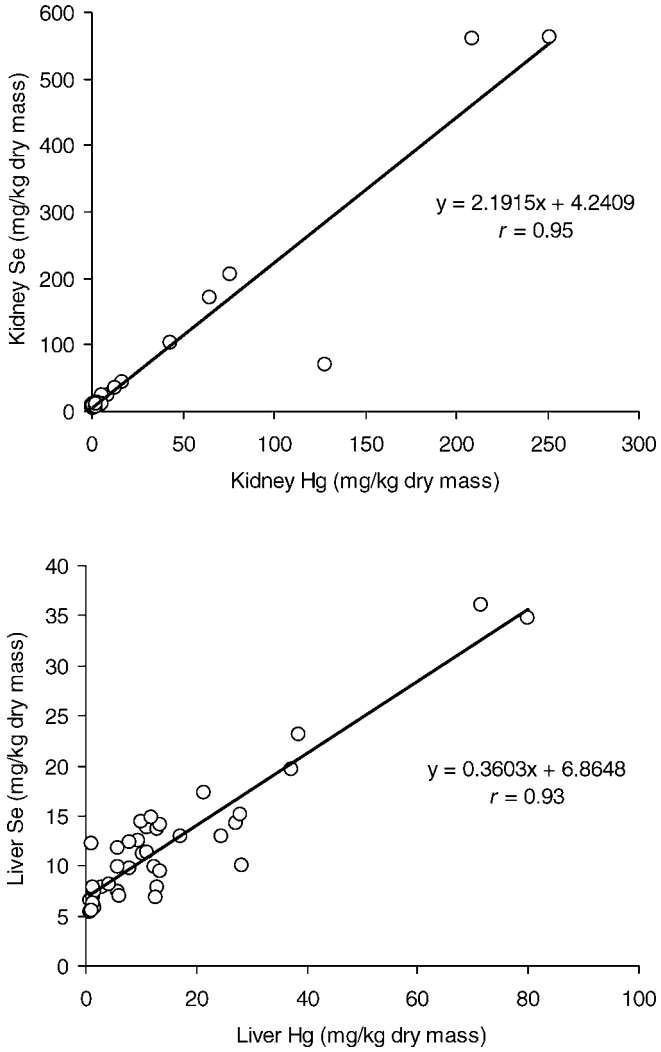

Figure 4. Relationship between selenium (Se) and total mercury (Hg) in the kidney and liver of 39 osprey collected in the Carolinas, USA, from May 2003 to May 2005.

these activities, it resumes at the feather where it stopped. In addition, new waves of molting are initiated before the preceding wave is completed. This pattern of molt is common among large soaring birds, including many raptors, and enables birds to replace each primary feather over the course of a year (Prevost 1983). Because feather replacement in osprey occurs over such a protracted period of time, on both the wintering and breeding grounds, and in overlapping waves, it follows that a sequential pattern of Hg deposition in feathers should not be detected. Our interpretation of the results is supported by Johnels et al. (1968) who found that Hg concentrations among primary feathers collected from a single adult osprey varied according to its migration location but not by molt sequence.

Perhaps most intriguing, we found that THg concentrations in osprey feathers were low relative to concentrations in their liver and kidney, particularly in adults. This observation contrasted with numerous studies that demonstrated that birds fed MeHg typically have higher concentrations of Hg in their feathers than in target organs (e.g., Heinz 1976, Finley and Stendall 1978, Spalding et al. 2000). Because osprey are obligate piscivores, they are likely exposed to more MeHg than Hg(II) in the environment (Wiener and Spry 1996). Thus, the patterns of THg accumulation in feather and soft tissues in osprey was the opposite of what would be expected based on the aforementioned studies. We hypothesize that our findings can be explained by tissue- and age-specific differences in Hg speciation, which was supported by the subsample of osprey tissues analyzed for MeHg. Adult birds with the highest concentrations of THg in liver and kidney sequestered as much as 90–99% of it in the less toxic, inorganic form (Hg(II)). This finding can be interpreted several ways, the most parsimonious of which is that demethylation of Hg resulted in sequestration of Hg(II) in soft tissues and therefore proportionally low concentrations of MeHg are transferred to feathers (Kim et al. 1996*a*). Alternatively, osprey may have progressively accumulated the small quantities of Hg(II) they encounter in their diet over their lifetime (Thompson and Furness 1989). Regardless of the mechanism by which this pattern arose, there is increasing evidence that bird species vary widely in the proportion of Hg(II) sequestered in their detoxification organs (Fimreite 1974, Thompson and Furness 1989, Kim et al. 1996*a*, Houserová et al. 2007). Future studies examining characteristics (e.g., feeding strategy, molting pattern, lifespan) of birds with differential Hg sequestration capacity will shed insight into the evolution of mechanisms for eliminating MeHg and how these strategies relate to relative ecological risk.

Table 3. Median and range of percent methylmercury (MeHg) in liver and kidney from 2 age classes (n = 4/age class) of osprey collected in the Carolinas, USA, from May 2003 to May 2005.

Tissue	Age class	Median % MeHg	Range % MeHg
Liver	Chick	52	25–69
	Hatch yr	55	53–59
	Ad	29	10–51
Kidney	Chick	69	50–87
	Hatch yr	68	52–89
	Ad	9	1–71

Table 4. Pearson correlation coefficients describing the relationship between selenium and total mercury in 10 types of tissues from osprey collected in the Carolinas, USA, from May 2003 to May 2005.

Tissue type	r
Primary feather no. 1	0.24
Primary feather no. 5	0.29
Primary feather no. 10	0.27
Scapular feathers	0.29
Breast feathers	0.17
Talon	0.18
Kidney[a]	0.95
Liver[a]	0.93
Muscle	0.35
Brain	0.23

[a] Statistically significant correlations.

Effects of Age and Sex

In general, adult birds tended to have more variable and higher mean concentrations of THg in tissues compared to chicks and hatch-year birds. The effect of age was only significant for liver and kidney, but there were strong trends of THg concentration increasing with age in talon, scapular feathers, and breast feathers. These findings are consistent with predictions based on the bioaccumulative nature of Hg and the behavior of osprey. Birds accumulate Hg over their lifetime, and thus adults have longer time windows for accumulation of Hg in soft tissues compared to chicks (Furness et al. 1990, Thompson et al. 1991, DesGranges et al. 1998; but see Gochfeld and Burger 1987). Young osprey that have not yet migrated should have tissue concentrations indicative of local exposure conditions. In contrast, healthy subadult and adult osprey along the Atlantic coast migrate south to overwinter. Thus, highly variable Hg concentrations in older birds may be attributable to exposure to Hg on the wintering grounds, presumably in Central or South America (Poole et al. 2002; T. Murphy, South Carolina Department of Natural Resources [SCDNR], personal communication).

We were unable to detect any significant effect of sex on Hg concentrations in tissues of birds with adult plumage. This finding contrasted with previous work that suggested females sometimes have lower concentrations of Hg in their tissues due to elimination through their eggs (Braun and Gaskin 1987, Lewis et al. 1993, Becker et al. 2002). Given the high variability in Hg concentrations within each sex, it is likely that our sample sizes (n = 7–8/sex) were simply insufficient to detect an effect of sex if it existed.

Toxicological Significance

In the majority of osprey, feather and brain concentrations of THg were below levels associated with health hazards in birds (Scheuhammer 1991, Burger and Gochfield 1997). Scheuhammer (1991) concluded that concentrations of Hg exceeding 20μg/g in the primary feathers of piscivorous birds were indicative of birds that experienced reduced reproductive success. Only one bird from our sample population (ad osprey no. 23 = 21.7 μg/g) exceeded this feather concentration. Scheuhammer (1991) also concluded that Hg concentrations ≥75 μg/g dry mass in brain tissue were associated with signs of neurointoxication and eventual death in birds. In our sample of osprey, Hg concentrations only averaged 0.8–2.1 μg/g dry mass among age classes. However, one hatch-year osprey was discovered perched on a mailbox in a residential area, exhibited neurological symptoms (e.g., trembling), and was unresponsive to human approach. The bird died within 12 hours of capture and had the highest concentrations of brain Hg from our sample population (6.2 μg Hg/g). Interestingly, this same bird had unremarkable feather concentrations of Hg (i.e., less than or equal to the mean concentration in our sample population), but its talon concentration of Hg was more than twice the average for its age class. This observation supports our contention that talons may be more representative than feathers of the recent body pool of Hg.

In contrast to feathers and brain, liver and kidney concentrations of THg were high enough to suggest risk of adverse effects in several cases. Threshold liver concentrations of THg are debated in the literature, ranging from 25 μg/g dry mass (Zillioux et al. 1993) to 100–150 μg/g dry mass (Thompson 1996) for adverse effects in birds, and concentrations exceeding 200 μg/g being directly lethal (Scheuhammer 1987). Seven birds with adult plumage (>40% of this age class) and one hatch-year bird exceeded the more conservative criterion, but no osprey had liver concentrations of Hg exceeding the latter 2 criteria. Although threshold criteria for kidney tissue are not reported as clearly in the literature, 3 adult osprey had kidney concentrations exceeding 100–200 μg/g dry mass (128 μg Hg/g, 209 μg Hg/g, and 251 μg Hg/g). However, the toxicological significance of THg concentrations is best interpreted in conjunction with information on Hg speciation and the concentration of Se present in tissues (Henny et al. 2002). In the subsample of osprey examined for Hg speciation, adult birds with high THg in their liver and kidneys had very low percent MeHg, which decreases their likelihood of experiencing adverse effects (Henny et al. 2002). Moreover, there were strong linear relationships between THg and Se in both the kidney and liver, but not other tissues, indicating co-sequestration of these elements. Selenium has well documented protective effects against Hg toxicity, possibly by assisting in sequestration of Hg(II) as HgSe (Pelletier 1985, Cuvin-Aralar and Furness 1991, Kim et al. 1996*b*, Henny et al. 2002). Previous work has shown that adult fish-eating birds, such as snowy egrets (*Egretta thula*) and double-crested cormorants (*Phalacrocorax auritus*), can accumulate very high concentrations of THg (geometric means as high as 675 μg Hg/g dry mass) without overt effects due to efficient demethylation and sequestration of Hg(II) with Se in the kidneys and liver (Henny et al. 2002). Although quantification of biological effects was beyond the scope of the current study, the observed patterns of demethylation and Se accumulation suggested that adult osprey may be able to tolerate accumulation of fairly high Hg concentrations.

MANAGEMENT IMPLICATIONS

Our results suggested that most osprey sampled along the South Carolina coast are not currently in danger of adverse effects related to Hg exposure. This appears particularly true for younger birds. However, 2 of the 6 chicks that we collected along the South Carolina coast had tissue concentrations of Hg suggestive of local contamination. Based on this observation, we advocate that future monitoring programs and placement of nesting platforms along this coastal region consider the spatial distribution of nests in relation to potential sources of Hg (e.g., industry inputs) and habitats of differing Hg bioavailability (e.g., estuaries, reservoirs). In contrast to the low levels of Hg found in most young birds, several adult osprey had high concentrations of THg in liver and kidney, suggesting the potential for serious health effects including reproductive

anomalies. However, most of the Hg in the liver and kidneys of these birds appeared to be inorganic and co-sequestered with large amounts of Se, potentially mitigating any adverse effects. Finally, our findings indicated that traditional feather sampling may not be as effective for monitoring osprey as it has been in seabirds. Talons may provide an additional nonlethal biomonitoring tool that will provide complementary information for natural resource managers assessing health risks to osprey.

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