# Patterns and Interpretation of Mercury Exposure in Freshwater Avian Communities in Northeastern North America

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Accepted 4 December 2004

Abstract. A large data set of over 4,700 records of avian mercury (Hg) levels in northeastern North America was compiled and evaluated. As Hg emissions remain poorly regulated in the United States and Canada, atmospheric deposition patterns and associated ecological responses continue to elicit interest by landscape managers, conservation biologists, policy makers, and the general public. How avian Hg exposure is interpreted greatly influences decision-making practices. The geographic extent and size of this data set is valuable in understanding the factors that affect the exposure of Hg to birds. Featured are differences found among tissues, major aquatic habitats and geographic areas, between age class and gender, and among species. While Hg concentrations in egg and blood reflect short-term Hg exposure, Hg concentrations in liver and feather provide insight into long-term Hg exposure. Blood is a particularly important matrix for relating site-specific exposure to methylmercury (MeHg). The level of MeHg is generally 5–10x greater in adults compared to nestlings. Age also influences MeHg bioaccumulation, particularly for individuals where MeHg intake exceeds elimination. Gender is of interpretive concern when evaluating Hg exposure for species exhibiting sexual dimorphism and niche partitioning. Based on two indicator species, the belted kingfisher (Ceryle alcyon) and bald eagle (Haliaeetus leucocephalus), we found MeHg availability increased from marine, to estuarine and riverine systems, and was greatest in lake habitats. A large sample of >1,800 blood and egg Hg levels from the common loon (Gavia immer) facilitated a suitable comparison of geographic differences. Although some clusters of highly elevated Hg exposure (i.e., blood levels  $> 3.0 \ \mu g/g$ , ww and egg levels  $> 1.3 \ \mu g/g$ , ww) were associated with hydrological and biogeochemical factors known to increase MeHg production and availability, others were not. Geographic areas without a relationship between Hg exposure and biogeochemical processes were associated with emission or waterborne point sources. Differences in Hg exposure among species are primarily correlated with trophic position and availability of MeHg. Although piscivorous species were repeatedly

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shown to have some of the highest MeHg levels of the 38 species analyzed, insectivorous birds in both aquatic and terrestrial habitats (such as montane areas) were also found with elevated MeHg levels. A better understanding of the factors confounding interpretation of Hg exposure provides an effective basis for choice of indicator species and tissues according to 12 selected scenarios. This and the national need for spatiotemporal monitoring of MeHg availability require careful consideration of indicator species choice. Only then will local, regional, continental, and even global monitoring efforts be effective.

Keywords: bird; loon; methylmercury; monitoring; indicator species

#### Introduction

The ecological impact from atmospheric deposition of mercury (Hg) has emerged as a major global environmental issue. Global concerns stem from the broad geographic extent of contamination, the increasing global signal of Hg deposition, and, until recently, a general lack of regulations to control many uses and the disposal of Hg (United Nations Environment Programme, 2003). In North America, decades of increasing Hg deposition appear to have reversed in some areas (Engstrom and Swain, 1997; Schuster et al., 2002; Fevold et al., 2003), including the Northeast (Kamman and Engstrom, 2002), but the need to identify and monitor ecological changes remains a high priority (Mason et al., 2005). Federal, state and/or provincial regulation of atmospheric mercury emissions in the United States and Canada is in place for some industrial sectors (i.e., municipal and medical waste incineration), but is currently lacking for others (i.e., coalfired electrical generators and mining). Not all environmental Hg is related to atmospheric deposition. Many past and even current inputs of waterborne Hg sources occur throughout North America and the Northeast. These are related to past improper waste disposal of Hg at weapons facilities (Halbrook et al., 1999), chlor-alkali plants (Fimreite, 1974; Gardner et al., 1978; Barr, 1986; Adair et al., 2003), mercury, gold, and silver mines (Elbert and Anderson, 1998; Henny et al., 2002; Seiler et al., 2004; Weech et al., 2004) and governmental storage facilities (Moore et al., 1999) as well as current inputs from wastewater treatment plants (Glass et al., 1990).

The U.S. Environmental Protection Agency (USEPA) investigated the ecological impacts of Hg based on key wildlife species as a basis for potential regulatory actions (USEPA 1997). An outgrowth of this effort was the development of a generic wildlife

criterion value for bird and mammal species (Nichols and Bradbury, 1999). Since the USEPA Report to Congress (USEPA1997), scientific investigations on the biogeochemical process of methylmercury production and availability have dramatically improved our basic knowledge (Morel et al., 1998; Lucotte et al., 1999; Wiener et al., 2003). A better understanding of the mechanisms of Hg transfer and fate has improved the ability to predict methylmercury (MeHg) production and availability (USEPA 2002), particularly in freshwater habitats of northeastern North America (Evers and Clair, 2005). This has resulted in a greater insight into now identifying specific geographic areas and biota at greatest risk to Hg exposure and effects.

Birds are at particularly high risk to Hg toxicity because many species are at high trophic levels (e.g., susceptible to biomagnification), are longlived (e.g., susceptible to bioaccumulation), are vulnerable to neurological and reproductive impacts from elevated Hg levels, and are frequently subjected to multiple anthropogenic stressors.

### Using birds as bioindicators of MeHg availability

The use of piscivorous birds as bioindicators of MeHg availability and risk in freshwater systems is common (e.g., Fimreite, 1974; Barr, 1986; Scheuhammer, 1987; Wolfe et al., 1998; Rumbold et al., 2001; Henny et al., 2002; Evers et al., 2003), although insectivorous birds are increasingly being used as well (Wolfe and Norman, 1998; Gerrard and St. Louis, 2001; Adair et al., 2003). Historically, Hg exposure was primarily determined by killing birds and was therefore based on organs analysis (Thompson, 1996). Although collection of viable eggs continues to be a relevant lethal method widely used (Braune et al., 2001), non-lethal sampling efforts based on blood (Bowerman et al., 2002;

Evers et al., 1998; Fevold et al., 2003), feathers (Burger, 1993), and abandoned eggs (Scheuhammer et al., 2001; Evers et al., 2003) are increasingly a more frequently used approach. Since Hg concentrations in different avian tissues reflect different temporal scales of past Hg exposure, care must be taken in considering Hg pharmacokinetics when selecting the best avian tissue to match specific biomonitoring objectives.

This paper represents a three-year effort through the U.S. Department of Agriculture's Northeastern States Research Cooperative (NSRC) to comprehensively compile and synthesize bird Hg data across northeastern North America. The paper's purpose is to describe this large data set and use the information to identify and assess the importance of factors that affect exposure and bioaccumulation of Hg.

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#### Methods

#### Source data sets

We targeted the collection of Hg data in birds from aquatic freshwater systems in New England, New York, and eastern Canada (eastern Ontario to the Canadian Maritimes) (Fig. 1). The Great Lakes and Lake Champlain were not included within our data set. Only blood Hg data for belted kingfishers and bald eagles were gathered from saltwater systems; these data were used to demonstrate differences among major aquatic habitats (Fig. 2). The majority of data (>90%) were provided by BioDiversity Research Institute, Canadian Wildlife Service, U.S. Environmental Protection Agency, and the U.S. Fish and Wildlife Service.



Figure 1. Distribution of Hg sampling effort for all bird species, 1969-2003.



Figure 2. Distribution of sampling effort by habitat type for the belted kingfisher and bald eagle in Maine.

All tissue data represent analysis of total Hg on a wet weight (ww), in the case of feathers, fresh weight (fw), basis in  $\mu g/g$  (or ppm). Estimated values or ranges of the proportion of MeHg in a particular tissue are cited for each within the Discussion section. The term juvenile means young-of-year birds and adults signify individuals at least one year of age. Latin names for those species within our Hg data sets are provided in Appendix 1.

Common loon blood and egg Hg sampling locations were converted into an ESRI ArcView point shapefile (i.e., formatting georeferenced parameters in a way that can be used by spatial software). Egg Hg values were converted to adult female blood equivalency with y = 1.5544x + 0.2238 (Evers et al., 2003). A six latitudinal minute by six longitudinal minute polygon grid created in Coordinate Grid Maker 2.29 was layered on the loon data. The 6-min interval was chosen as the best resolution to balance local and regional trends. The loon Hg shapefile was spatially joined to the grid polygon where the arithmetic mean of all the points falling within a grid cell was calculated. These global means were then displayed in 1.0  $\mu$ g/g (ww) intervals.

#### Laboratory methods

The data utilized in this compilation were generated at a number of laboratories over a period of several years. Although there were some

## differences in sample preparation and analytical methods, all analyses included quality control (QC) samples to allow evaluation of accuracy and precision, and all laboratories utilized atomic absorption spectroscopy to measure Hg concentrations.

Sample types collected and submitted to the laboratories for analysis primarily included avian blood, feathers, and eggs. Blood samples were either in sealed capillary tubes or in glass or plastic vacutainer-type collection tubes. Samples that were severely clotted were not analyzed unless the entire sample could be removed from the collection tube.

Feather samples were either analyzed whole or as subsamples following homogenization. Aliquots of feathers were obtained by reducing individual feathers to small pieces with either stainless steel scissors or a Spex 6800 cryomill.

Egg samples generally required homogenization; a task that was sometimes complicated by the egg samples that were fully formed. Egg samples that were largely soft tissue were homogenized by either a Tissuemiser or a small food processor/ blender prior to subsampling. Eggs containing hard parts and feathers were homogenized with a blender or with a Spex 6800 cryomill. Only loon eggs were corrected for moisture loss

Most blood, feather, and egg samples required digestion prior to analysis. This was accomplished by following a procedure similar to EPA 245.6, in which nitric and sulfuric acids were used in conjunction with potassium permanganate and potassium persulfate to solubilize the tissue and convert any bound Hg to the free Hg<sup>2+</sup> ion (Lobring and Potter, 1991). Prior to analysis, excess KMnO<sub>4</sub> was reduced with hydroxylamine hydrochloride and the samples were made to volume with deionized water.

Analysis of digest solutions was based on the "cold vapor" atomic absorption spectroscopy method first introduced by Hatch and Ott (1968). Using either a manual or automated approach,  $Hg^{2+}$  in solution was reduced to  $Hg^{0}$  with SnCl<sub>2</sub>, the  $Hg^{0}$  was transferred to the gas phase, and the  $Hg^{0}$  -containing gas was swept into an atomic absorption cell. Mercury levels were determined by comparing sample absorbance peak heights with those of calibration standards.

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A subset of samples was analyzed by a direct determination method that did not require sample digestion (EPA 7473) (U.S. EPA, 1998). A homogenized, dry sample was placed in a tared nickel boat, weighed, and then placed into a tube furnace. A stream of O2 assisted in sample combustion and carried free or organic-bound Hg species through a heated catalyst and onto a gold trap where the free Hg<sup>0</sup> was collected. When the sample had been combusted for a sufficient length of time, the gold trap was heated and the released Hg<sup>0</sup> was carried through a pair of atomic absorption cells where it was measured. This method required samples that were particularly well-homogenized because only a small sample mass could be accommodated in the nickel boats.

Each batch of samples processed and analyzed was accompanied by a number of QC samples, including a method blank, spiked blank, certified reference material, duplicate sample, and spiked sample. Typical detection limits for data presented here were 0.0025  $\mu$ g/g (ww). Precision as measured as relative percent difference of duplicate pairs was approximately 85% and accuracy as measured by recovery of certified reference materials and spiked samples was 80%.

## Statistical analysis

Mercury concentrations are expressed as arithmetic means with standard deviations (SD) in the tables and geometric means with variation expressed as standard error (SE) in figures. Arithmetic means and SD are provided for comparative purposes with published literature. Because sample sizes were regularly small and were therefore not normally distributed, statistical analysis was conducted on the exponentiated value of the mean of the log-transformed values. Logtransformed data were normally distributed based on normal probability plot residuals. Homoscedasticity was checked with Bartlett's test, which is sensitive to the normality assumption. JMP software (SAS Institute Inc., 2001) was used to perform statistical analysis. Hypotheses were tested using one-way analysis of variance (ANOVA). Testing was followed by post-hoc tests using Tukey-Kramar honestly significant different (Tukey's test) if the ANOVA demonstrated significant differences (Zar, 1999). JMP's Tukey's test output

did not include actual probability values and instead indicated significance when numbers were positive. Therefore, only probability values "less than" and "greater than" 0.05 are shown in the Results section. Student's *t*-tests were used when comparing paired data sets. A non-parametric test, the Kruskal–Wallis One-Way ANOVA, was used in some cases to compare multiple independent groups. JMP software corrected for inequity of unbalanced data sets. We used an alpha of 0.05 for our level of significance.

#### Results

A total of 4,769 Hg concentrations representing 38 species and six tissue types are recorded within the NSRC avian database (Appendix 1–3; Fig. 1). Samples were collected between 1969 and 2003 with the majority (>84%) from 1995 to 2003. Six factors were identified as having significant influence on the interpretation of avian Hg levels. The NSRC data set was used to demonstrate how these factors influence Hg exposure.

### Influences of tissue type

Mercury data collections totaled 2,158 blood, 943 egg, 281 muscle, 1,100 feather, 239 liver, and 48 kidney samples (Appendix 1). Approximate respective inter-tissue comparative ratios based on

blood for common loons breeding in northeastern North America were 0.4:1:2:6:15 (egg:blood:muscle:feather:liver). For a site-specific subset of Hg exposure data (south-central Quebec, New England and Canadian Maritimes), there were no significant geographical differences among tissue ratios (p > 0.05; with the exception of blood) (Fig. 3). Muscle Hg levels among eight waterfowl species were categorized by four major foraging guilds during the breeding season and indicated significant differences between piscivores versus each of the other three foraging guilds and insectivores versus herbivores (Fig. 4).

Intra-and inter-tissue relationships were strongest in the following three pairings: (1) adult and juvenile blood, (2) adult female blood and egg, and (3) juvenile feather and blood. Data analyzed were based on sampling efforts that represented pairings from the same breeding territory (i.e., each pair of adult and juvenile blood Hg levels in tree swallows was from the same nesting box). Paired adultjuvenile blood Hg levels in common loons had a significant relationship ( $r^2 = 0.63$ , p < 0.01) as they did for tree swallows ( $r^2 = 0.74$ , p < 0.01). Paired adult female blood and egg Hg levels were significantly related in loons  $(r^2 = 0.79, p < 0.01)$ (based on Evers et al., 2003) and tree swallows  $(r^2 = 0.49, p < 0.01)$ . Paired eaglet feather and blood Hg levels were significantly related  $(r^2 = 0.67, p < 0.01)$  (based on Welch, 1994).



*Figure 3.* Comparison of geometric mean  $\pm$  SE of Hg levels in four tissue types for adult common loons breeding in south-central Quebec, New England, and the Canadian Maritimes (n = number of respective samples by region). The arithmetic mean  $\pm$  SD of Hg in liver is used for comparative purposes with the literature. Liver Hg values in New England are from Pokras et al. (1992).

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### Age as a factor

Five species of birds were used to demonstrate relationships in blood Hg levels between juveniles (<2 months of age) and adults (>1 year of age) (Fig. 5). Two species were insectivores (tree swallow and song sparrow) and three species were piscivores (common loon, common merganser, and

belted kingfisher). While adult tree swallows had significantly higher blood Hg levels than nestlings (p < 0.05), adult song sparrows did not have significantly different blood Hg levels than their fledged young (p > 0.05). Significantly higher blood Hg levels in adults versus juveniles were found in all three species of piscivorous birds (p < 0.05). Ratios of adult-juvenile Hg levels were: song



*Figure 4*. Comparison of geometric mean  $\pm$  SE of Hg levels in muscle among four foraging guilds of waterfowl (n = number of samples). Means not sharing a common letter are significantly different (p < 0.05). Waterfowl species represented by foraging guild are: herbivores – Canada goose; omnivores – mallard, American black duck, green-winged teal, and ring-necked duck; insectivore – common goldeneye; and piscivore – hooded merganser and common merganser.



*Figure 5.* Comparison of geometric mean  $\pm$  SE of blood Hg levels of five species for two age classes (n = number of juvenile samples, number of adult samples). Means not sharing a common letter within a given species are significantly different (p < 0.05). Species codes are: SOSP, song sparrow; TRES, tree swallow; BEKI, belted kingfisher; COME, common merganser and; COLO, common loon.

sparrow 1.7:1, common merganser 3.6:1, tree swallow 5.9:1, belted kingfisher 5.6:1, and common loon 10.6:1.

### Gender as a factor

Three species were used to demonstrate a relationship in blood Hg levels between male and female adults (Fig. 6). Blood Hg levels in male common loons were significantly higher than females (p < 0.01). In belted kingfishers and tree swallows, there was no significant difference between male and female blood Hg levels (p > 0.05).

#### Aquatic habitat comparisons

Data for the belted kingfisher and bald eagle showed relationships in MeHg availability for four major aquatic habitat categories: marine, estuarine, riverine, and lake. Only individuals sampled in Maine were used for an analysis of inter-habitat differences (Fig. 2). Both species exhibited an increasing trend in which mean blood Hg levels in marine < estuarine < riverine < lake (kingfishers, Fig. 7 and eagles, Fig. 8). Adult blood Hg levels were significantly higher in kingfishers foraging on lakes and rivers versus marine habitats (p < 0.05), but not different in rivers versus estuaries (p > 0.05). Eaglet blood Hg levels were significantly higher from nests along lakes versus those adjacent to marine (p < 0.05) and estuarine (p < 0.05) habitats. Blood Hg levels in eaglets from nests along rivers were significantly higher than those along marine habitats (p < 0.05).

### Geographic differences

Based on a standard species (common loon) and tissue types (blood and egg) and a large sample size (n = 1,882), spatial heterogeneity in MeHg availability was demonstrated across northeastern North America (Fig. 9). The proportion of mean blood and converted egg Hg concentrations (see "Source data sets" in Methods section) within a six latitudinal minute by six longitudinal minute cell grid (n = 300 cell grids) was 19.7% for 0–1 µg/g, 45.6% for 1–2 µg/g, 20.7% for 2–3 µg/g, 8.7% for 3–4 µg/g, and 5.3% for >4 µg/g.

#### Variation in species

Three sites provided an opportunity to compare Hg levels in multiple species within the same area (which avoids confounding factors related to geographical differences): two examples were on lakes and one was on a river. On Aziscohos Lake, eggs of five species (three piscivores, one insectivore, and one herbivore) were compared (Fig. 10).



*Figure 6.* Comparison of geometric mean  $\pm$  SE of blood Hg levels of three species by sex (n = number of female samples, number of male samples). Means not sharing a common letter within a given species are significantly different (p < 0.05). Species codes are: TRES, tree swallow; BEKI, belted kingfisher and; COLO, common loon.



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Figure 7. Comparison of geometric mean  $\pm$  SE of blood Hg levels for juvenile and adult belted kingfishers in Maine among four major aquatic habitat types (n = number of juvenile samples, number of adult samples). Means not sharing a common letter are significantly different (p < 0.05); comparisons are case sensitive.



*Figure 8.* Comparison of geometric mean  $\pm$  SE of blood Hg levels for juvenile bald eagles in Maine among four major aquatic habitat types (n = number of samples). Means not sharing a common letter are significantly different (p < 0.05).

Piscivore egg Hg levels were significantly higher (p < 0.05) than insectivores (tree swallows) and herbivores (wood duck). Larger piscivores tended to have higher egg Hg levels than smaller piscivores (i.e., common loon > common merganser > hooded merganser). Similar patterns were documented on Flagstaff Lake in egg Hg levels (common loon > common merganser > belted kingfisher) (Fig. 11).

Blood Hg levels were compared for 11 insectivorous birds from the Sudbury and Charles Rivers (Fig. 12). There were no significant differences in blood Hg levels between the rivers for four of the most common passerines sampled at each site (Kruskal–Wallis one-way analysis of Variance; p = 0.65). Age classes were combined because song sparrow blood Hg levels were not significantly different between breeding adults and nestlings (Fig. 5). Generally, blood Hg levels increased with body weight. For seven selected songbirds, there was a significant correlation between mean weight and blood Hg levels ( $r^2 = 0.72$ ,



Figure 9. Geographic distribution of mercury levels in adult common loon blood and blood equivalents based on eggs, 1993–2003.

p = 0.02, y = 0.0162x + 0.0054). Granivorous songbirds such as the American goldfinch had blood Hg levels significantly lower than insectivorous songbirds (p < 0.05).

### Discussion

### Selecting the correct tissue

The pharmacokinetics of MeHg and total Hg (how both forms are distributed throughout the body) are fairly well known in birds; their understanding provide insights toward selecting the correct tissue for meeting specific research or monitoring objectives. Ingestion of dietary MeHg appears to be readily absorbed into the blood (83% in common loons, Fournier et al., 2002) and is thereafter distributed to various

body tissues, namely the liver, kidney, brain, spleen, and muscle. Some tissues, such as the liver, are terminal endpoints where MeHg is largely unavailable for remobilization, whereas MeHg deposited in muscle tissue is available and remobilizes during feather molt. Blood and feather Hg samples are generally taken nonlethally. Although blood can be taken immediately after death through cardiac puncture (Henny et al., 2002), it cannot be taken through venipuncture after death for reliable comparison with living bird blood Hg levels (because of rapid and nonlinear moisture loss).

Mercury concentrations in avian tissues can indicate different (1) modes of toxic action, (2) MeHg and total Hg composition, (3) exposure timeframes, and (4) elimination abilities. Six tissues routinely used for determining Hg exposure in birds are described in our data set.



*Figure 10.* Comparison of geometric mean  $\pm$  SE of egg Hg levels among species on Aziscohos Lake, Maine (n = number of samples). Means not sharing a common letter are significantly different (p < 0.05). Species codes are: WODU, wood duck; TRES, tree swallow; HEGU, herring gull; HOME, hooded merganser; COME, common merganser and; COLO, common loon.



*Figure 11.* Comparison of geometric mean  $\pm$  SE of egg Hg levels among species on Flagstaff Lake, Maine (n = number of samples). Means with different letters are significantly different (p < 0.05). Species codes are: WODU, wood duck; BEKI, belted kingfisher; COGO, common goldeneye; COME, common merganser and; COLO, common loon.

#### Tissue interpretation

Blood is the best tissue for evaluating short-term dietary uptake. Mercury in blood is primarily MeHg (>95%) in both piscivores (Fournier et al., 2002) and insectivores (Rimmer et al., 2005). The half-life of MeHg in the blood of chicks undergoing feather molt was three days in common loons (Fournier et al., 2002) and 5–6 days in Cory's shearwaters (Monteiro and Furness, 2001). In non-molting adults, the half-life of MeHg in the blood was greater: for Cory's shearwater (*Calonectris diomedea*) the half-life was 40–60 days (Monteiro and Furness, 2001) and for the mallard was 74 days (Heinz and Hoffman, 2004). In our study, adult loon blood Hg levels, which were



*Figure 12.* Comparison of geometric mean  $\pm$  SE for blood Hg levels (adult and juvenile combined) among species on or near the Sudbury and Charles Rivers, Massachusetts (n = number of samples). Means not sharing a common letter are significantly different (p < 0.05). Species codes are: AMGO, American goldfinch; YWAR, yellow warbler; WODU, wood duck; GRCA, gray catbird; COYE, common yellowthroat; SOSP, song sparrow; SWSP, swamp sparrow; WIFL, willow flycatcher; TRES, tree swallow; RWBL, red-winged blackbird and; NOWA, northern waterthrush.

collected 60–120 days post-arrival to the breeding lake, were strongly correlated with prey fish Hg levels and therefore primarily reflected uptake of dietary Hg from the breeding lake (Fevold et al., 2003; Evers et al., 2004, Burgess and Hobson, in press; Champoux et al., in press).

As in blood, egg Hg levels are primarily in the MeHg form: in loons it was >95% (Scheuhammer et al., 2001; Evers et al., 2004), in seabirds it is >90% (Fimreite et al., 1974). Because female blood Hg levels are highly correlated with egg Hg levels for the common loon (Evers et al., 2003), eggs and their outer membranes are also pertinent tissues for predicting Hg exposure within a bird's breeding territory (Heinz and Hoffman, 2003). How predictive egg Hg levels are for the breeding territory depends on amount of time the female spent within the territory prior to egg-laying and Hg body burden levels accumulated during the winter and migration. Generally, piscivores arriving from marine overwintering areas have been exposed to lower MeHg availability than levels found on freshwater aquatic systems (Fig. 7, 8). Rapid equilibrium of dietary MeHg uptake and blood MeHg levels thereafter plays an important role in forming a strong MeHg relationship between eggs and breeding-season blood levels. The strength of this relationship is also impacted by intraclutch differences in Hg levels. In common loons, intraclutch variation between two eggs was

25% (Evers et al., 2004), in larids it averaged 39% between the first and second eggs (Becker et al., 1994), and in common merganser clutches of > 10 eggs we sometimes found variations of one order of magnitude (although nest parasitism in cavity-nesting ducks is common and it unknown if merganser clutches represent more than one female). A standardized comparison of egg Hg levels among locations therefore requires knowledge of egg laying order.

Virtually all Hg in a feather is MeHg (Thompson and Furness, 1989) and is sequestered for long time periods allowing retrospective analysis (Frederick et al., 2004). Feather Hg reflects blood Hg levels at the time of molt (Bearhop et al., 2000), however, if MeHg is depurated in the muscle tissue (as is the case for individual birds with a high dietary MeHg uptake), it is available for remobilization. Therefore, feather Hg levels reflect both site-specific dietary uptake of MeHg and body burden. Feather Hg generally reflects 70-93% of the muscle MeHg burden (Burger, 1993); therefore, there can also be chronic bioaccumulation of MeHg, particularly for highly exposed individuals (i.e., where MeHg ingestion exceeds elimination). This attribute makes feather Hg levels a relevant tissue for evaluating chronic body burdens, particularly when considering the stability of MeHg in the feathers (Appelquist et al., 1984). However, individual variation in physiological response to

Hg (Bearhop et al., 2000), as well as the broad differences in inter-species pharmacokinetics, requires careful evaluation of risk.

Four internal tissues are commonly used for Hg exposure investigations: brain, liver, kidney and muscle. Although MeHg crosses the blood-brain barrier and can have significant impacts on brain functions, brain tissue is best harvested from relatively fresh carcasses and therefore is a more difficult tissue to use in field studies. It is more commonly used for analysis in experimental studies (Heinz, 1975; Finley et al., 1979; Scheuhammer, 1988).

Liver is one of the more commonly analyzed internal tissues for Hg in birds (Sundlof et al., 1994; Augspurger et al., 1998; Pokras et al., 1998; Cohen et al., 2000). Liver and kidney filter toxins such as MeHg and effectively demethylate MeHg using selenium (Se) bonds that form a nontoxic Hg-Se protein complex (Stoewsand et al., 1974). Scheuhammer et al., (1998a) found a nearly 1:1 molar ratio of Hg:Se in the liver and kidney of common loons and common mergansers. He also demonstrated that the proportion of MeHg in the liver and kidney declined as total Hg concentrations increased (i.e., liver and kidney total Hg levels were independent of MeHg concentrations). Therefore, determining levels of currently toxic Hg in the liver and kidney requires analysis of MeHg concentrations, while concentrations of Se-bound inorganic Hg provide an indication of past MeHg exposure.

The "7:3:1 rule" is an often-used conversion factor for liver (ww), feather (fw), and muscle (ww) tissue Hg concentrations (Appelquist et al., 1985). Although Thompson et al., (1990) demonstrated the weakness of this conversion approach, these tissues in common loons of three different regions in northeastern North America follow the "7:3:1 rule" (Fig. 3).

Unlike the liver and kidney, muscle Hg levels generally have proportionally higher levels of MeHg (80–100%) in the common loon and common merganser (Scheuhammer et al., 1998a). Muscle tissue Hg levels are generally examined in waterfowl (Pearce et al., 1976; Braune et al., 1999; Cohen et al., 2000), in part, to determine potential human health risks. Our data set supports other findings that muscle Hg levels are generally less than liver and kidney (Gardiner, 1972; Gochfeld, 1980) and that piscivorous waterfowl have muscle Hg levels significantly greater than other foraging guilds of waterfowl species (Fig. 4).

### Age affects Hg exposure

A potential limitation of using birds as an indicator of Hg is the inability to identify individual age once breeding begins. The knowledge of a bird's age is critical for evaluating Hg bioaccumulation. Marking techniques, such as uniquely numbered or colored leg bands, provide a reliable method for tracking individual age and removing this limitation. Such techniques have been used successfully to evaluate time relationships with Hg. In cases where MeHg ingestion exceeds elimination, feather Hg levels increase with age (Evers et al., 1998; Rimmer et al., 2005). Scenarios where individuals can depurate and demethylate ingested MeHg at a similar annual rate of ingestion lack a positive correlation with increasing age and feather Hg levels (Furness et al., 1990; Thompson et al., 1991; Gochfeld et al., 1996; Donaldson et al., 1997; Fevold et al., 2003).

While bioaccumulation of MeHg can be a critical interpretive factor related to age in situations with high Hg exposure, there is also a common pattern for adult blood Hg levels to exceed those of unfledged juveniles in areas with even low Hg exposure (Fig. 5). Burger and Gochfeld (1997) documented adult Franklin's gull (*Larus pipixcan*) and herring gull blood Hg levels to be significantly greater than juveniles from the same colony of the same year. Differences in Hg levels between age classes (juvenile vs. adult) is dictated by (1) stage of juvenile feather molt and (2) partitioning of forage base by size of potential prey.

A major depuration route for MeHg is via the feather (Crewther et al., 1965), and therefore feathers are a useful indicator for monitoring Hg body burdens (Burger, 1993). However, the interpretation of feather Hg levels requires an understanding of feather-molt chronology (Furness et al., 1986). Blood and feather MeHg levels are highly correlated when blood is sampled during feather molt. Birds have the ability to rapidly transfer dietary uptake of MeHg from red blood cells to growing feathers (Fournier et al., 2002; Kenow et al., 2003). The physiological capacity of birds to process MeHg this way appears to be great. Fournier et al., (2002)

found loon chicks, experimentally dosed with MeHg concentrations substantially greater than those found in nature, were still able to effectively depurate much of the MeHg into emerging feathers. When juvenile feather molt ends, blood MeHg levels thereafter increase (Spalding et al., 2000a; Fournier et al., 2002). This ability to rapidly transfer blood MeHg into growing feathers partly accounts for the significant difference in blood Hg levels between adults and juveniles prior to fledging. Our data indicate that this is relevant for piscivorous and insectivorous birds. For the song sparrow, blood Hg levels of recently fledged young when compared with locally breeding adults, demonstrated no significant difference between the age classes. However, difference of age ratios among species (Fig. 5) indicates Hg differences between age classes are also dictated by other factors.

Generally, prey choice differs between adults foraging for themselves versus for their young. For example, the size of fish prey selected by juvenile common loons increases as they grow larger (Barr, 1996). We found adult blood Hg levels of piscivore species were 3.6 to 10.6 times higher than those in unfledged young (Fig. 5) and that variation suggests that adults are foraging on prey that are larger and have higher levels of Hg. In agreement, Burgess and Hobson (in press) found that adult loons fed at a higher trophic level (as indicated by stable-nitrogen isotope ratios) and had higher blood Hg levels than did juvenile loons. These differences in blood Hg and trophic levels were related to differences in body weight in the loons. Blood sampling for belted kingfishers and tree swallows typically occurs when the adults are 6 times the weight of the juveniles, while in common loons, adults are sampled when they are usually 10 times the weight of juveniles. Conversely, common mergansers are usually sampled when the adult females are relatively similar in size with the young. Such rough correlative relationships across age classes between body weight and prey-size selection are likely contributing to age class differences in Hg exposure.

### Gender affects Hg exposure

Although most studies indicate differences in Hg levels between male and female birds are not significant (Burger, 1993; Burger, 1995; Gochfeld et al., 1996), there are exceptions (Hoffman and Curnow, 1979; Braune and Gaskin, 1987; Donaldson and Braune, 1999). When there are differences in Hg levels between sexes they can be dictated by (1) depuration of Hg in eggs, (2) sexual dimorphism, and (3) niche partitioning of the forage base. Although depuration of MeHg in eggs is an important mechanism for elimination, derivation of MeHg from serum proteins and a steadystate equilibrium with dietary uptake of MeHg likely compensates for the loss of MeHg from the body burden within weeks (Furness and Greenwood, 1993) or possibly days (Kambamandi-Dimou et al., 1991). Differences in blood Hg levels between sexes of adult loons sampled > 60 days after eggs are laid regularly demonstrates other factors are involved. Choice of prey items is likely the primary factor dictating differences in Hg levels between sexes. Levels of MeHg in prey items vary according to species, trophic status, age, size, and habitat associations (Wiener and Spry, 1996).

Based on comparisons of three species, the loon exhibited significant differences of blood Hg between sexes. Loon Hg levels were also greater in liver for males vs. females (Pokras et al., 1998). Common loons are sexually dimorphic. On average, males are 21% larger than females (Evers, 2005). The larger males apparently forage on larger prey fish based on the correlative strengths between favored prey fish Hg levels (yellow perch, Perca flavescens) and their blood Hg levels (Evers et al., 2004). Larger fish generally have higher Hg levels than smaller fish of the same species from the same location (Weiner and Spry, 1996; Drysdale et al., in press; Kamman et al., 2005). Therefore, blood Hg levels are higher in male common loons because they are foraging on larger fish.

In general, blood Hg differences between male and female belted kingfishers are not significant, however, within individual pairs Hg levels are typically significantly different. Because males and females are relatively similar in size (females tend to be slightly heavier), prey size is likely not the driving factor; rather males and females partition foraging niches (Albano, 2000).

### Hg patterns in aquatic habitats

Aquatic systems are one of the more at-risk ecosystems for MeHg bioavailability because one

of the better-known methylating organisms, sulfate-reducing bacteria, inhabit this environment (Gilmour et al., 1992). To adequately compare MeHg bioavailability across four major aquatic habitat types requires a standard species, age class and sampling tissue. Both the bald eagle and belted kingfisher fit these criteria. A subset of the blood Hg data from both species in Maine indicates interspecies agreement that MeHg availability increases from marine to estuarine to riverine to lake ecosystems. Because atmospheric deposition of Hg is relatively uniform across the Maine study area (VanArsdale et al., 2005), with some significant local exceptions, interpretation of the hydrological and biogeochemical factors influencing Hg methylation and availability and their relationships with bird blood Hg levels is presented.

Although marine systems are well known for their elevated biotic MeHg levels, those levels primarily represent long-lived species with top trophic status (e.g., swordfish and shark species). A standard comparison between freshwater and in-shore marine systems documents the latter has significantly lower MeHg availability. In-shore marine systems appear to be more effective in diluting MeHg production versus freshwater systems, although in-shore habitats geochemically greatly vary in MeHg production (Hammerschmidt and Fitzgerald, 2004). Estuaries are dynamic communities that are influenced by tidal actions and varying volumes of fresh and salt water. They are generally hydrologically heterogeneous landscapes that have less of an ability to dilute Hg inputs than marine systems. Although tidal exchanges do regularly provide an important flushing mechanism that lowers MeHg availability (Lamborg et al., In Press), tidal responses vary in magnitude daily, weekly, and monthly. Methlymercury availability therefore varies tremendously within and between estuaries (Shriver et al., 2002).

Based on our analysis, the ability of inorganic Hg to be converted to MeHg and become available to biota is greater in freshwater versus saltwater habitats. Gariboldi et al., (1998) also documented prey items were higher in Hg from freshwater versus saltwater habitats based on sampling efforts with the wood stork (*Mycteria americana*). Riverine habitats tend to have higher

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levels of MeHg availability than estuaries, but tend to have lower levels when compared to lakes. Comparisons of MeHg availability between adjoining riverine and lake habitats based on crayfish, fish, and birds consistently show greater Hg exposure on lakes (Fimreite, 1974). Flushing abilities within riverine systems are a driving force for these differences.

Therefore, in coastal regions, MeHg exposure arising from atmospheric Hg deposition is generally greatest in piscivorous birds foraging on freshwater lakes. Lake hydrology and biogeochemistry largely determine the degree of aquatic MeHg exposure. Lakes with low pH (< 6.3, Meyer et al., 1995; Burgess and Hobson, in press), large areas of scrub-shrub and emergent wetlands (Kramar et al., 2005), and large areas of exposed shoreline substrate of organic or sandy soils that are frequently inundated and dried through the summer and fall (i.e., reservoirs; Evers and Reaman, 1998) are predictive of elevated blood Hg levels in the common loon. Although newly created reservoirs are well known for their ability to enhance MeHg production and availability through the decomposition of vegetation (Jackson, 1988; Lucotte et al., 1999; Gerrard and St. Louis, 2001), this phenomenon is generally viewed as short-lived (i.e., <10 years in secondary consumers) (Lucotte et al., 1999). Some reservoirs have longer lasting abilities to enhance Hg methylation and have the potential to be some of the highest risk aquatic habitats (Evers et al., 2004).

## Geographic differences exist

There are continental patterns in the availability of MeHg. Long-term sampling efforts across North America indicate a significant west to east trend exists with northeastern North America exhibiting the highest levels (Evers et al., 1998, 2003; Scheuhammer et al., 2001). Significant within-region differences are primarily driven by hydrological and biogeochemical factors and point source influences. The collection of > 1,800 blood and egg Hg levels for the common loon across New England, New York and eastern Canada enabled us to effectively evaluate areas of greatest concern (Fig. 9). Clusters of elevated MeHg availability were found in the western Adirondack Mountains of New York, north-central Massachusetts,

southeastern New Hampshire, western mountains of Maine, and a small area east of Orrington, Maine in the United States (Fig. 9). In Canada, areas with high MeHg exposure were in eastern Ontario, south-central Quebec and southern Nova Scotia. There did not appear to be smooth spatial trends in loon Hg levels across northeastern North America, as highly elevated Hg levels were scattered among low Hg concentrations in almost every region sampled.

In the Adirondack Mountains and eastern Canada, clusters of elevated MeHg availability were likely related primarily to lake acidification. It is well established that lakes with low pH contain fish with higher levels of Hg than same-size and species of fish in lakes with more circumneutral pH levels (Wiener et al., 1990; Winfrey and Rudd, 1990; Drysdale et al., in press). Oligotrohphic lakes in eastern Canada and parts of New England and New York are susceptible to increased rates of anthropogenically derived sulphur deposition (i.e., acid rain) (Driscoll et al., 2001). Although there is evidence of declining levels of atmospheric input of sulfur dioxide, base cation levels are lowered in many systems where responses in lake pH levels are lagging behind predictive models. Therefore, these acidified lakes continue to be a cause of concern for their ability to enhance MeHg productivity. Associations between lake acidity, fish Hg levels and lower common loon productivity have been observed in the U.S. and Canada (Meyer et al., 1998; Burgess et al., 1998a). Scheuhammer and Blancher (1994) predicted up to 30% of lakes in central Ontario have the potential to adversely impact common loon productivity.

Other areas of concern are related to topography and lake hydrology (e.g., western mountains of Maine) and point sources (both airborne and waterborne). Airborne sources in southern New England appear to contribute to greaterthan-expected loon Hg levels in southeastern New Hampshire (Evers, 2001) and Orrington, Maine. Waterborne point sources are well known in eastern Massachusetts, such as on the Sudbury River. There, investigations associated with the Nyanza Superfund Site have documented associated Hg contamination > 25 km downstream from the point source (Wiener and Shields, 2000).

## Species Hg exposure patterns

Differences in Hg levels among species are dictated by trophic level and availability of MeHg (i.e., aquatic vs. terrestrial and marine vs. freshwater; low exposure vs. high exposure). Trophic structure is a primary driver of variability in MeHg biomagnification (Cabana et al., 1994). Recent evidence indicates that the trophic status of an aquatic vertebrate is based primarily on the complexity, both longitudinal and vertical, of the planktivorous community (Chen et al., 2005). Methylmercury can biomagnify several orders of magnitude in aquatic ecosystems. For example, the average bioconcentration factor for the common loon in Maine lakes is  $1.37 \times 10^6$  (based on unfiltered water for total Hg) (Evers et al., 2004).

The degree of MeHg biomagnification through aquatic-based food webs is the primary reason for the multitude of Hg studies on obligate piscivores. Particular emphasis has been placed on larger species for which trophic status is potentially greatest; such species include the common loon (Meyer et al., 1995, 1998; Burgess et al., 1998a, b; Evers et al., 1998, 2003; Scheuhammer et al., 1998b, 2001; Fevold et al., 2003; Burgess and Hobson, in press; Champoux et al., in press), bald eagle (Grier, 1974; Wiemeyer et al., 1984; Frenzel and Anthony, 1989; Bowerman et al., 1994; Anthony et al., 1999; Bowerman et al., 2002), osprey (Cahill et al., 1998; DesGranges et al., 1998), wading birds (Gariboldi et al., 1998; Bouton et al., 1999; Spalding et al., 2000b; Henny et al., 2002), and seabirds (Braune, 1987; Burger and Gochfeld, 1995; Monteiro and Furness, 1995, 1997; Braune et al., 2001). Other foodweb pathways important for MeHg transfer are generally of lesser concern because trophic status of the endpoint species is generally lower than piscivores. Benthic-based MeHg transfer through bivalves has been investigated using various diving species of waterfowl (Ohlendorf et al., 1986; Henny et al., 1991; Braune et al., 1999; Cohen et al., 2000; Wayland et al., 2002), while such transfer through macroinvertebrates (larval and adults) (Bishop et al., 1995; Wolfe and Norman, 1998; Reynolds et al., 2001; Gerrard and St. Louis 2002, Adair et al., 2003) and vegetation has also been described (Fimreite, 1974; Langis et al., 1999).

Elevated MeHg bioavailability in specific terrestrial ecosystems within northeastern North America has recently been documented. Montane environments without standing water appear to have the ability to generate MeHg. Rimmer et al., (2005) documented Bicknell's Thrush blood Hg concentrations for 21 mountain locations (arithmetic mean of 0.14  $\pm$  0.08 µg/g, ww with a range of < 0.01 to 0.70 µg/g, ww) at levels similar to those found in many of the insectivorous songbirds sampled along rivers in Massachusetts (Fig. 12).

The comparison of multiple species within the same area and habitats, while using appropriate tissues and minimizing confounding factors (such as age class and sex), is the optimal approach for determining interspecies relationships of Hg exposure. Based on such past studies (Dustman et al., 1972; Fimreite, 1974; Langis et al., 1999) and our data sets, Hg exposure can be approximately predicted by foraging guilds. An all-purpose ranking from low to high Hg exposure for birds is: terrestrial herbivores, aquatic herbivores, terrestrial insectivores, benthivore-bivalves, benthivore-macroinvertebrates, small piscivores, and large piscivores.

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Exposure of Hg in scavengers and omnivores is broad and dependent on opportunistic food sources (Fimreite, 1974). Our ranking assumes MeHg availability is driven by atmospheric deposition and is not universal in application, because some habitats such as montane ones contain insectivorous birds that have Hg exposure greater or equivalent to piscivores.

### Recommended bioindicators

We recommend species and tissue types that best indicate 12 targeted scenarios (Table 1) based on the analysis of our data set, the recommendations made by the working group (USEPA Hg Mason et al., 2005), species' ubiquitous within northeastern North America, and logistical feasibility. Identified indicator species are not universal and may be only relevant to the scenario posed. Many of our chosen bioindicators are also useful for determining MeHg effects through such endpoints as long-term reproductive success. For example, bald eagle breeding populations are used in Michigan (Bowerman et al., 2002) and common

Table 1. Summary of recommended avian bioindicators, age/sex class, and tissue type for 12 scenarios in freshwater, estuarine, and terrestrial systems in northeastern North America

Scenario	Species	Age/sex <sup>1</sup>	Tissue type
Comparison of major aquatic habitat types	Belted kingfisher	Adult & fledged young	Blood & $egg^2$
	Bald eagle	Juvenile	Blood & feather
Lake $> 25$ ha	Common loon	Adult	Blood & egg
	Common merganser	Adult female	Blood & $egg^2$
Lake $< 25$ ha	Common loon	Juvenile	Blood
	Hooded merganser	Adult female	Blood
River	Common merganser	Adult female	Blood & $egg^2$
	Belted kingfisher	Adult & juvenile	Blood& $egg^2$
	Tree swallow	Adult & juvenile	Blood & egg
Estuaries	Ammodramus sparrow spp.	Adult & fledged young	Blood
Emergent wetlands	American bittern	Adult	Blood & egg
-	Virginia rail	Adult	Blood & egg
	Song sparrow	Adult & fledged young	Blood
	Red-winged blackbird	Adult & fledged young	Blood & egg
Shrub-scrub wetlands	Waterthrush spp.	Adult	Blood
	Swamp and Song sparrows	Adult	Blood
Montane areas	Bicknell's thrush	Adult & fledged young	Blood
Deciduous forest	Wood thrush	Adult & fledged young	Blood
Coniferous forest	Catharus thrush spp.	Adult and fledged young	Blood
Long-term risk in lakes	Common loon	Adult	Feather
Greatest risk in aquatic systems	Bald eagle	Adult	feather

<sup>1</sup> Juvenile = unfledged young which have yet to reach completion of feather molt and fledged young = young-of-the-year that have completed feather molt.

<sup>2</sup> When using egg tissue from these species, only use composite values for entire clutch to avoid wide intra-clutch variation.

loon breeding populations with color-marked individuals are monitored throughout New England (Evers et al., 2004) and Wisconsin (Meyer et al., 1998; Fevold et al., 2003).

Our selections are species- and genera-specific for illustrative purposes, but species with similar foraging requirements, behavior, and natural history patterns may be suitable surrogates; preferably, trophic status is similar. Evolving techniques in stable isotope analysis offer numerous applications to matching trophic status through analysis of tissues (e.g., blood, feather, egg, muscle, bone). Such techniques provide quantitative measures of trophic position (Hobson, 1993; Bearhop et al., 2000; Nisbet et al., 2002; Dominguez et al., 2003), dietary emphasis (i.e., freshwater vs. marine [Hobson, 1990; Mizutani et al., 1990; Bearhop et al., 1999], marine versus terrestrial [Hobson, 1987; Hobson and Sealy, 1991; Schmutz and Hobson, 1998]), contaminant bioaccumulation (Cabana and Rasmussen, 1994; Kidd, 1998; Atwell et al., 1998), and nutrient allocation to reproduction (Hobson et al., 1997; Hobson et al., 2000). Here in, scenarios and associated avian selections relate primarily to freshwater breeding habitats.

Selecting a standard species across multiple aquatic habitats, particularly between freshwater and saltwater ones, is difficult. The belted kingfisher is a ubiquitous species that is an obligate piscivore in all major aquatic habitats. As a burrow-nesting species, repeated access to young and adults is feasible for the kingfisher. Sampling efforts to determine site-specific exposure should focus on blood. Intraclutch variability in egg Hg levels appears to be high (Lane et al., 2004). Bald eagle pairs also commonly forage within all major aquatic habitats. Adults are difficult to capture, therefore, chicks are generally sampled to determine Hg exposure (Bowerman et al., 2002). Blood and breast feathers are the most common sampling tissues. Adult eagle feathers from the nest site can be useful for determining Hg exposure (Bowerman et al., 1994) and may reflect some of the highest Hg levels within an aquatic ecosystem. However, foraging habits of breeding pairs vary dramatically within and between breeding seasons, habitat type, and geographic area (Knight et al., 1990; Kozie and Anderson, 1991; Anthony et al.,

1999). Although either fish or birds can comprise the majority of prey remains at eagle nests, Dominguez et al., (2003) found that stablenitrogen isotope ratios showed little difference in trophic status among nests in Newfoundland.

The common loon is one of the better bioindicators of lake-specific MeHg availability as it has a top trophic position in the aquatic food web, is long-lived, and in most cases remains within its breeding territory for 4-6 months. Adult blood and egg Hg levels reflect dietary Hg exposure of breeding loons on lakes > 25 ha. Territorial pairs occupying lakes < 25 ha generally maintain and feed on more than one lake (i.e., multi-lake territories) (Piper et al., 1997). Because adult common loons with multi-lake territories rarely bring food items back to their natal lake to feed their young, blood Hg levels of juvenile loons best represent MeHg availability on their natal lake. Common mergansers are also obligate piscivores that can reflect lakespecific MeHg availability. Sampling efforts for adult females can be facilitated through the use of artificial boxes. Similar to kingfishers, intraclutch variation in egg Hg levels is high for mergansers and dictates individual egg or composite analysis (versus selecting one egg). Other high-trophic level piscivores that generally forage on lakes are not optimal lake-specific indicator candidates because (1) of their tendency to commonly use multiple waterbodies within their breeding territory, (2) they are difficult to efficiently capture and sample (e.g., great blue heron), and (3) they regularly prey on lower-trophic-level organisms such as benthicfeeding fish (e.g., osprey) and terrestrial birds and mammals (e.g., bald eagle). Instead, such species best represent MeHg availability at a watershed level. Double-crested cormorants phalacrocoraxauritus may be good indicators of multiple large lakes and other aquatic systems.

Determining mercury exposure in riverine habitats is most promising with the common merganser, hooded merganser, belted kingfisher, and tree swallow. The belted kingfisher is increasingly being used as an indicator for assessing Hg in riverine systems (Baron et al., 1997; Moore et al., 1999). Use of artificial nesting boxes on riverine habitat and experimental design interests for both piscivorous and insectivorous birds can be achieved with the hooded merganser and tree swallow.

Although most investigations of avian Hg exposure have focused on waterbodies, wetlands and strictly terrestrial habitats are increasingly being included during risk assessments. In emergent wetlands, insectivores best reflect MeHg availability. Larger-bodied insectivorous birds have greater Hg exposure than their smaller counterparts (Fig. 12). The Virginia rail (Rallus limicola) is a good indicator candidate because it is more insectivorous than the sora (Porzana carolina) and is more common and less limited by marsh size than the American bittern (Botaurus lentiginosus). Clapper rails (Rallus longirostris) in San Francisco Bay had greater body burdens of Hg than associated piscivorous birds, such as terns (U.S. Fish and Wildlife Service, 2003). The red-winged blackbird had some of the highest blood Hg levels of songbirds within a Massachusetts riverine wetland (Fig. 12). Although the red-winged blackbird and sparrow species (i.e., song and swamp) are granivores most of the year, during the breeding season they are obligate insectivores. Based on limited Hg data, both the Louisiana (Seiurus motacilla) and northern waterthrush may be insectivorous passerines at greatest risk in riverine habitats; waterthrushes forage specifically on aquatic organisms. Other relatively large-bodied, insectivorous passerines associated with northern aquatic systems, such as the rusty blackbird (Euphagus carolinus), may also be at risk; specific Hg-sensitive habitats are acidic headwater areas (Bank et al., 2005) draining recently logged coniferous catchments (Porvari et al., 2003).

Methylmercury availability in terrestrial insectivorous passerines is relatively unknown but a recent compelling study by Rimmer et al. (2005) indicates further investigations are needed. That study documented Hg levels in Bicknell's thrush and further comparison of the blood Hg level ranges show an overlap with those of eaglets; thereby indicating equivalent trophic status of a terrestrial-based insectivore with an aquatic-based piscivore. Recent investigations have demonstrated that MeHg is present in foliage (approximately 1% of the total Hg content) (St. Louis et al., 2001; Ericksen et al., 2003). Miller et al., (2005) estimated MeHg availability to terrestrial food webs using forest foliage and modeled deposition and concentrations of leaf, litterfall,

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precipitation (wet and dry), and particulate Hg in northeastern North America. Litterfall total Hg concentrations from these models were significantly correlated with the blood Hg levels of Bicknell's thrush (Rimmer et al., 2005). Conceivably MeHg in litterfall and contributions of foliar total Hg to saturated soils where potential methylation environments exist are providing an important basis for biomagnification of MeHg in invertebrates. Acidified environments further enhance methylation (Furutani and Rudd, 1980; Xun et al., 1987), and with the influence of heavy wet deposition of acid ions (i.e., acid rain), northeastern North America's landscape is generally more acidic than pre-industrial times (Driscoll et al., 2001). Soil acidification may impact bird populations in several ways (Graveland, 1998) including the depletion of soil calcium levels. Breeding birds have high demands of calcium for eggshell formation and proper juvenile growth. The widespread depletion of environmental calcium availability in northeastern North America is now linked to adverse effects on the distribution of wood thrush (Hylocichla mustelina) (Hames et al., 2002). Unfortunately, the strong link between environmental acidification with MeHg production and calcium depletion may be creating a scenario where their synergy has the potential for long-term, landscape-level impacts on insectivorous passerine populations across much of northeastern North America.

### Acknowledgements

We thank the many scientists representing agencies, universities, industry, and organizations for their data contributions. Several biologists at Bio-Diversity Research Institute were instrumental in collecting many of the tissue samples; they include, Cory Counard, Chris DeSorbo, Joseph Kaplan, Oksana Lane, James Paruk, Lucas Savoy, Kate Taylor, Keren Tischler, and Dave Yates. Pierre-Yves Daoust of the Atlantic Veterinary College provided all muscle, kidney, and liver tissues analyzed for Hg by the Canadian Wildlife Service. Hydro-Quebec contributed otherwise unavailable data. Gary Heinz and Stan Wiemeyer graciously provided rapid reviews that improved the manuscript. The U.S. Department of Agriculture's Northeastern States Research Cooperative

Appendix 1. Basic geog	graphic information,	sampling time peric	od, sample s	ize by tissue	e and data so	urce						
Species	Latin Name	Project Geographic	Geographi – Latitude	c Extent	Geographic Extent – Loi	ngitude	Sampli Durati	ng Nı	mber	of samples	by tissue	
		Area	Min	Max	Min	Max	Begin	End Blo	I poo	gg Feath	er Liver Kidney	Muscle
Common Loon	Gavia immer	ME, MA, NH, NY, VT, NB, NS, ON, PE, PO	42.350277	51.999444	-79.447777	-60.817	1986	2003 A: J:-	770 6 452	60 A: 63 J: 52	1 A: 30 A: 10 J: 8 J: 2	A: 18 J: 6
Black-crowned Nioht-Heron	Nycticorax nveticorax	ME, PQ	43.487777	48.460277	-74.57777	-68.53	1661	2003 J:	17		J: 6	
Great Blue Heron	Ardea herodias	ME, MA, PQ	45.030277	49.460277	-74.597222	-64.722222	1661	2003 A: I.	1 1	J: 8	J: 8	
Canada Goose	Branta bernicla	ME, NB, NL, BE PO	46.133	57.283	-66.867	-54.167	1989	.r 1999	2	A: 24	A: 7	A: 15
Wood Duck	Aix sponsa	ME, MA, NH, ME, BO	42.338333	49.440555	-77.480277	-66.450	1990	2003 A:	13 1	6 A: 2	A: 2	A: 4
Mallard	Anas platyrhynchos	NB, FQ ME, NH,NY, NB, NL, MS DO,	43.693888	52.917	-79.411944	-63.400	1990	2003 J:	~	A: 11 J: 1	A: 8	A: 16*
American Black Duck	Anas rubripes	NB, NL, NS, DE PO	43.783	54.167	-78.830	-53.250	1988	1993		A: 39	A: 39	A: 62*
Green-winged Teal	Anas crecca	NB, NL, BE PO	45.867	54.167	-78.838333	-53.500	1990	1992 A:	1	A: 8	A: 8	A: 24*
Ring-necked Duck	Aythya collaris	NH, NB, NI, NB, NI PO	45.500	51.994722	-79.280833	-55.083	1990	1997		A: 34 1-1	A: 34 1-1	A: 39*
Common Goldeneye	Bucephala	ME, NB, NL, NS PO	43.633	53.167	-79.083888	-56.083	1989	2003 A:	6	2. A: 32	A: 32	A: 39
Hooded Merganser	Lophodyte cucultatus	ME, MA, NH, NB, NL, PO	42.26	53.333	-79.024166	-57.833	1989	2003 A: J:	13 4	5 A: 11 J: 1	A: 17	A: 20
Common Merganser	Mergus merganser	ME, NH, NY, VT, NB, NL, NS, PO	43.5825	54.167	-79.182222	-53.500	1989	2003 A: J: -	11 2 59 2	6 A: 24 J; 32		A: 31
Osprey	Pandion haliaetus	ME, NH, PQ, NS NR NI, PF	42.968055	53.333	-78.752222	-60.433	1989	2003 A: I·	2 8	3 A: 10 1·62	A: 12 A: 4 1·11 1·9	A: 6 I· 1
Bald Eagle	Haliaeetus leucocephalus	ME, NH, NB, NS. PEI	43.836388	47.000	-71.611388	-59.920	1969	2003 J:	108 3	8 A: 9	A: 28 A: 7 J: 2 J: 2	A: 8 J: 2
American Woodcock	Scolopax minor	ME, MA	44.496666	44.496666	-69.237222	-69.237222	2000	2002 J:	+	J: 4		
Herring Gull	Larus argentatus	ME	44.76128	45.05178	-71.013383	-70.8182	1990	2003 A: J:	1 8	A: 1 J: 1		

Appendix 1. Basic geographic information, sampling time period, sample size by tissue and data source

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Species	Latin Name	Project Geographic	Geographic – Latitude	Extent	Geographic Extent – Lon	ıgitude	Samplin Duratio	g Numh	er of se	amples by	' tissue		
		Area	Min	Max	Min	Max	Begin E	ind Blood	Egg	Feather	Liver <b>k</b>	Cidney N	Muscle
Belted Kingfisher	Ceryle alcyon	ME, MA, NH VT	42.172777	46.024166	-73.224166	-68.27444	1997 2	003 A: 11 1. 183	7 16	A: 71 I· 13			
Downy Woodmocher	Picoides pubescens	MA	42.347350	42.347350	-71.382380	-71.382980	2003 2	003 A: 1					
Willow Flycatcher	Empidonax traillii	MA	42.176240	42.390130	-71.382380	-71.319020	2003 2	003 A: 1 1· 5					
Great Crested Flycatcher	Myiarchus crinitus	MA	42.28558	42.28558	-71.44901	-71.44901	2003 2	003 J: 4					
Eastern Kingbird	Tyrannus tyrannus	MA	42.17386	42.364408	-71.31916	-71.37542	2003 2	003	26				
Common Raven Tree Swallow	Corvus corax Tachycineta bicolor	ME ME, MA	44.97933 42.17325	44.97933 44.94138	-71.0215 -71.5831	-71.0215 -70.9279	2000 2 2000	000 J: 1 003 A: 53	55				
Cliff Swallow	Petrochelidon	ME	44.93642	45.15257	-71.0336	-70.4466	1999 2	J: 73 001 A: 19					
Barn Swallow	pyrrhonota Hirundo rustica	ME	44 97047	44 97047	-70 7166	-70.7166	1999 2	U:6 001 A:3					
Eastern Titmouse	Baeolophus bicolor	MA	42.347350	42.347350	-71.382380	-71.382380	2003 2	003 J: 1					
Eastern Bluebird	Sialia sialis	MA	42.30914	42.38378	-71.49121	-71.38748	2003 2	003 J:7					
Bicknell's Thrush	Catharus bicknelli	ME, VT	45.183611	45.183611	-70.264722	-70.264722	1999 2	003 A: 21		A: 18			
Gray Catbird	Dumetella	MA	42.176240	42.390130	-71.382380	-71.319020	2003 2	003 A: 2 1· 4					
Yellow Warbler	Dendroica petechia	MA	42.176240	42.390130	-71.382380	-71.319020	2003 2	003 A:4 1.5					
Northern Waterthrush	Seiurus novehoracensis	MA	42.176240	42.347350	-71.382380	-71.319020	2003 2	003 A: 2					
Common Vellowthroat	Geothlypis thrichas	MA	42.176240	42.390130	-71.382380	-71.319020	2003 2	003 A:4 1·5					
Song Sparrow	Melospiza Melodia	ME, MA	42.176240	42.390130	-71.382380	-71.319020	2001 2	003 A: 16 J: 18					
Swamp Sparrow	Melospiza georgiana	MA	42.176240	42.390130	-71.382380	-71.319020	2003 2	003 A: 5 J: 13					
Red-winged Blackbird	Agelaius phoeniceus	MA	42.175555	42.351388	-71.381111	-71.321666	2003 2	003 A: 3 J: 3					
Common Grackle Brown-headed	Quiscalus quiscula Molothrus ater	MA MA	42.34631 42.176240	42.35972 42.176240	-71.37398 -71.319020	-71.36956 -71.319020	2003 2 2003 2	003 003 J: 1	9				
Cowbira American Goldfinch	Cardueli tristis s	ME, MA	42.176240	43.731683	-71.382380	-70.566450	2000 2	003 A: 12					

Appendix 1. Continued

	r.				
Species	Blood (ww) Mean $\pm$ SD (R	ange)	Egg (ww) Mean $\pm$ SD (Range)	Feather (fw) Mean ± SD	(Range)
	Adult	Juvenile		Adult	Juvenile
common loon black-crowned night-heron	$2.04\ \pm\ 1.39\ (0.05-8.63)$	$\begin{array}{rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$	$0.78\ \pm\ 0.60\ (0.01-9.00)$	$12.7 \pm 6.6 \ (2.2 - 63.4)$	$5.4 \pm 4.8 \; (0.3 - 25.7)$
great blue heron		$0.49 \pm 0.56 (0.03 - 1.76)$	$0.09 \ \pm \ 0.04 \ (0.05 - 0.12)$		$5.2 \pm 2.0 \ (1.3 - 6.9)$
Canada goose				$0.3~\pm~0.1~(0.3-0.8)$	
wood duck	$0.05~\pm~0.04~(0.01-0.14)$		$0.12 \ \pm \ 0.23 \ (0.01 - 0.94)$	$1.6 \pm 0.3 \; (1.4 - 1.8)$	
mallard		$0.05~\pm~0.03~(0.03-0.08)$		$0.9~\pm~0.5~(0.3-1.8)$	
American black duck				$1.8 \pm 1.2 \ (0.6 - 6.6)$	
American green-winged teal				$1.3 \pm 1.4 \ (0.3 - 4.5)$	
ring-necked duck				$1.5 \pm 0.8 \; (0.3 - 3.0)$	
common goldeneye	$0.21 \pm 0.06 \ (0.15 - 0.31)$		$0.33 \pm 0.18 \ (0.09 - 0.72)$	$2.8 ~\pm~ 1.3~(0.8 - 7.0)$	
common merganser	$1.57 \pm 0.59 \ (0.74 - 2.35)$	$0.60 \ \pm \ 0.47 \ (0.03 - 2.29)$	$1.43 \pm 0.86 \ (0.28 - 3.93)$	$8.0\ \pm\ 4.1\ (3.3-17.7)$	$8.8~\pm~5.4~(3.3-31.4)$
hooded merganser	$0.88 \ \pm \ 0.55 \ (0.07 - 1.91)$	$0.68 \ \pm \ 0.30 \ (0.34 - 1.13)$	$0.64 \pm 0.44 \ (0.15 - 1.90)$	$10.4 \pm 3.8 \ (2.7 - 18.0)$	
osprey	$1.42 \pm 0.18 \ (1.29 - 1.54)$	$0.31 \ \pm \ 0.20 \ (0.03 - 0.81)$	$0.19 \pm 0.09 \ (0.06 - 0.38)$	$15.6 \pm 13.6 (0.1 - 38.3)$	$8.2~\pm~5.6~(0.1-26.5)$
bald eagle		$0.30 \ \pm \ 0.27 \ (0.01 - 1.20)$	$0.45 ~\pm~ 0.29 ~(0.03 - 1.29)$	$14.0 \pm 6.8 \ (2.8 - 24.8)$	
American woodcock		$0.03 \pm 0.01 \ (0.02 - 0.04)$			$0.2~\pm~0.1~(0.1-0.3)$
herring gull		$0.46 \ \pm \ 0.13 \ (0.28 - 0.72)$	$0.63 \pm 0.55 \ (0.01 - 1.63)$	$18.1 \pm 15.1 \ (4.4 - 57.0)$	
belted kingfisher	$0.99 \ \pm \ 0.82 \ (0.07 - 4.57)$	$0.17 \ \pm \ 0.18 \ (0.01 - 1.35)$	$0.56 \pm 0.77 \ (0.03 - 3.03)$	$7.2 \pm 7.6 \ (0.6 - 46.1)$	$8.0~\pm~5.4~(3.8-19.8)$
willow flycatcher		$0.43 \ \pm \ 0.25 \ (0.20 - 0.80)$			
great crested flycatcher		$0.09 \ \pm \ 0.02 \ (0.07 - 0.11)$			
eastern kingbird			$0.12 \pm 0.04 \ (0.04 - 0.21)$		
tree swallow	$0.41 \pm 0.21 \ (0.11 - 1.00)$	$0.07 \ \pm \ 0.03 \ (0.02 - 0.16)$	$0.19\ \pm 0.11\ (0.04-0.64)$		
cliff swallow	$0.22 \pm 0.10 \ (0.08 - 0.47)$				
barn swallow	$0.13 \pm 0.03 \ (0.11 - 0.15)$				
eastern bluebird		$0.01 \ \pm \ 0.01 \ (0.01 - 0.02)$			
Bicknell's thrush	$0.29~\pm~0.26~(0.05-0.80)$				
gray catbird	$0.13 \ \pm \ 0.07 \ (0.08 - 0.19)$	$0.05 \ \pm \ 0.02 \ (0.03 - 0.07)$			
yellow warbler	$0.04 \pm 0.03 \ (0.01 - 0.07)$	$0.04\ \pm\ 0.03\ (0.01-0.08)$			
northern waterthrush	$0.92 \ \pm \ 0.95 \ (0.25 - 1.59)$				
common yellowthroat	$0.28~\pm~0.13~(0.15-0.44)$	$0.10\ \pm\ 0.05\ (0.04-0.17)$			
song sparrow	$0.35 \pm 0.30 \ (0.08 - 1.34)$	$0.21 \ \pm \ 0.14 \ (0.01 - 0.56)$			
swamp sparrow	$0.74 \pm 0.47 \ (0.22 - 1.45)$	$0.30 \ \pm \ 0.14 \ (0.07 - 0.48)$			
red-winged blackbird	$0.67 \pm 0.71 \ (0.20 - 1.49)$	$0.90 \ \pm \ 0.38 \ (0.46 - 1.13)$			
common grackle			$0.04 \pm 0.03 \ (0.01 - 0.07)$		
American goldfinch	$0.01 \ \pm \ 0.01 \ (< 0.01 \ - \ 0.03)$				

Appendix 2. Tissue Hg levels (arithmetic mean  $\pm$  SD and range) from nonlethal sampling efforts

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Species	Liver (ww) Mean ± SD (Range)	Kidney (ww) Mean ± SD (Range)	Muscle (ww) Mean ± SD (Range)
common loon - adult	$29.7 \pm 38.5 (1.9 - 154.0)$	$39.9 \pm 25.1 (1.7 - 79.1)$	$4.10 \pm 3.40 \ (0.40 - 15.90)$
black-crowned night-heron great blue heron	$14.9 \pm 23.6 (0.3 - 92.6)$	$55.9 \pm 24.9 (11.0 - 60.7)$ $1.5 \pm 0.3 (1.2 - 1.9)$ $1.9 \pm 0.9 (0.4 - 3.3)$	$0.90 \pm 0.81 (0.20 - 2.40)$
Canada goose	$0.1 \pm 0.3 (0.1 - 0.2)$		$0.04~\pm~0.03~(0.02-0.14)$
wood duck mallard	$\begin{array}{l} 0.3 \ \pm \ < \ 0.1 \ (0.3 - 0.3) \\ 0.5 \ \pm \ 0.2 \ (0.1 - 0.8) \\ 0.6 \ \pm \ 0.4 \ (0.1 - 2.6) \end{array}$		$0.13 \pm 0.08 (0.02 - 0.26)$
American black duck American green-winged teal	$\begin{array}{r} 0.6 \pm 0.4 \; (0.1 - 2.0) \\ 0.8 \pm 0.3 \; (0.2 - 1.3) \end{array}$		$\begin{array}{r} 0.16 \ \pm \ 0.12 \ (0.03 \ - \ 0.45) \\ 0.21 \ \pm \ 0.15 \ (0.02 \ - \ 0.59) \end{array}$
ring-necked duck common goldeneve	$\begin{array}{r} 0.6 \ \pm \ 0.3 \ (0.2 - 1.5) \\ 1.5 \ \pm \ 1.8 \ (0.1 - 8.2) \end{array}$		$\begin{array}{r} 0.17 \ \pm \ 0.11 \ (0.03 - 0.47) \\ 0.33 \ \pm \ 0.20 \ (0.05 - 0.88) \end{array}$
common merganser			$1.71 \pm 1.71 \ (0.08 - 6.77)$
hooded merganser osprey – adult	$\begin{array}{r} 4.7 \ \pm \ 3.0 \ (1.1 - 12.2) \\ 10.6 \ \pm \ 11.2 \ (0.6 - 23.0) \end{array}$	$15.7 \pm 14.7 (2.3 - 36.7)$	$\begin{array}{r} 0.96 \ \pm \ 0.35 \ (0.04 - 1.92) \\ 1.5 \ \pm \ 1.5 \ (0.6 - 4.3) \end{array}$
osprey – juvenile bald eagle – adult	$\begin{array}{r} 0.9 \ \pm \ 0.4 \ (0.2 - 1.5) \\ 2.2 \ \pm \ 2.2 \ (0.6 - 11.8) \end{array}$	$1.1 \pm 0.6 (0.3 - 2.1) \\9.2 \pm 12.4 (0.7 - 33.4)$	0.1 $0.4 \pm 0.4 (0.2 - 1.5)$
bald eagle – juvenile herring gull	$\begin{array}{c} 1.2 \ \pm \ 0.9 \ (0.6 - 1.9) \\ 3.6 \ \pm \ 2.5 \ (1.0 - 8.2) \end{array}$	$0.6 \pm 0.5 (0.3 - 1.0)$	$\begin{array}{l} 0.3 \pm 0.1 \ (0.2 - 0.3) \\ 1.59 \pm 1.32 \ (0.35 - 3.98) \end{array}$

Appendix 3. Tissue Hg levels (arithmetic mean  $\pm$  SD and range) from lethal sampling efforts

provided funding to compile and synthesize the Hg databases.

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