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### Ecotoxicology and Environmental Safety

journal homepage: www.elsevier.com/locate/ecoenv

# Mercury concentrations in blood, brain and muscle tissues of coastal and pelagic birds from northeastern Canada



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ARTICLE INFO

Keywords: Sublethal effects Contaminant Mercury Neurotoxicity Aquatic birds

#### ABSTRACT

Mercury (Hg) is a toxic element which has increased in marine environments for more than a century, due largely to anthropogenic activities, and biomagnifies in food chains to harmful levels in some top predators like waterfowl and seabirds. We analysed total mercury (THg) concentrations in blood, brain and muscle tissue from healthy specimens of 13 coastal and pelagic bird species from eastern and northern Canada to provide a baseline on current concentrations, especially for brain concentrations which are highly underrepresented in the literature. We also examined within and among tissues relationships of THg concentrations within individuals. THg concentrations were generally higher in pelagic species and scavenging gulls, when compared to coastal waterfowl. Brain and muscle tissue had similar concentrations of THg in the birds examined, but both of these tissues had lower concentrations that those found in blood. Our results, and that of a previous study, suggest that body condition has a large influence on blood THg concentrations and should be considered when using blood as a sampling medium. Many of the species we examined had tissue THg above levels known to cause deleterious, sublethal effects in some species.

#### 1. Introduction

Mercury (Hg) is a toxic, non-essential element that is naturally occurring, but due to anthropogenic activities has increased in the environment (Lindberg et al., 2007; Krabbenhoft and Sunderland, 2013), notably in marine food webs (Dietz et al., 2009; Bond et al., 2015; Stenhouse et al., 2018). It has long been known to have negative effects on wildlife (and humans), and thought to deleteriously influence nervous, excretory and reproductive systems (Wolfe et al., 1998; Hoffman et al., 2011). Because global exposure of Hg is increasing and human and wildlife health are at risk, international efforts to resolve the environmental threat culminated in the Minimata Convention (http:// mercuryconvention.org), an agreement to reduce Hg emissions which came into effect in August 2017 and now requires implementation and monitoring (Evers et al., 2016).

Environmental Hg is a concern for coastal ecosystem health along northern and eastern North America, including marine birds (Goodale et al., 2008; Burgess et al., 2013). In a recent, comprehensive review on Hg and birds, Whitney and Cristol (2017) found strong evidence that Hg negatively affects several aspects of avian health. In the Gulf of Maine, many bird species have elevated levels of Hg (Goodale et al., 2008; Pollet et al., 2017; Stenhouse et al., 2018), and dovekies (*Alle alle*) wintering in coastal Newfoundland and Labrador had higher Hg exposure than during the breeding season in east Greenland; birds with higher Hg concentrations produced smaller eggs (Fort et al., 2014). Among some seabirds in Arctic Canada, there is a pattern that Hg increases with increasing latitude (e.g., Pratte et al., 2015), and overall that Hg in marine birds increased from the 1970s through the 1990s but has since plateaued and may be declining (Braune et al., 2016). Despite apparent declining trends in some metrics, Dietz et al. (2013) and Scheuhammer et al. (2015) reviewed available information and noted that Hg remains a key issue of concern for wildlife and human health across much of the circumpolar Arctic.

Recently, Fort et al. (2015) sampled Hg in tissues of seabirds that died in a massive mortality event off the coast of western France. The values they measured in apparently healthy birds were similar to what Koeman et al. (1975) had measured decades earlier, but Fort et al. (2015) found higher Hg in birds that died in winter wrecks than during

https://doi.org/10.1016/j.ecoenv.2018.04.004 Received 31 January 2018; Received in revised form 2 April 2018; Accepted 4 April 2018 0147-6513/ © 2018 Elsevier Inc. All rights reserved.

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the breeding season, suggesting that Hg could have been an aggravating factor in mortality. In trying to compare their values to wild, healthy birds, they noted that there was limited information on concentrations of Hg in the brain tissue of seabirds (see also Espín et al., 2012) – surprising, given the known deleterious, neurotoxic effects of this element, and thus we might expect particular attention on levels of Hg in brain tissue. Among Arctic wildlife, Dietz et al. (2013) also noted that there are few studies that report Hg in brains of wild birds. Hepatic and blood concentrations are much better represented in research (e.g., Braune and Scheuhammer, 2008; Hoffman et al., 2011).

In Arctic seabirds specifically, a recent study on Arctic terns (*Sterna paradisaea*) highlighted that while hepatic levels of Hg may be below levels known to cause impairment in other avian species, brain concentrations of Hg were within the range considered high for wildlife (Provencher et al., 2014). This is particularly of concern for seabirds, which are long-lived species and long distance migrants, because Hg is a known neurotoxin that can impair bird cognitive abilities and reproductive behavior (reviewed in Wolfe et al., 1998; Shore et al., 2011; Whitney and Cristol, 2017). Brain Hg concentrations are also important to note as they may represent a longer term signature of Hg exposure than tissues that turnover at a faster rate.

In this study, we examined Hg concentrations in blood, muscle and especially brain tissues of apparently healthy birds from the northern and eastern coasts of Canada, both areas of Hg concern, to provide a baseline for areas considered to have high Hg in marine food webs. We used this broad sample of species to determine the relationships in concentrations between these tissues (blood, brain, muscle, including examining variation in tissue concentrations within individuals) to convert values among tissues at environmentally-relevant levels in future studies. We also compared our data to that from other studies and modeled the relationship between brain and muscle Hg across multiple coastal and pelagic bird species. Given earlier work by Fort et al. (2014) suggesting that birds wintering along eastern North America are exposed to high Hg, we expected Hg concentrations in tissues of Canadian birds to be relatively higher than found in other regions for which data were available. We also expected the relative values of Hg to be blood > muscle > brain based on the few published values for Hg in these tissues across avian species (e.g., Shore et al., 2011).

#### 2. Methods

We gathered blood, brain and muscle tissue from waterfowl and seabirds collected at various locations across northern and eastern Canada (Fig. 1), both areas of Hg concern. Most tissues came from specimens stored at the National Wildlife Research Centre in Ottawa, or from tissues of hunter-shot birds, all of which were collected as part of other studies (e.g., Holland et al., 2016; Provencher et al., 2016; Mallory and Braune, In press). Other samples were provided as salvage specimens from wildlife management programs (e.g., Seif et al., 2018), including one northern gannet (Morus bassanus) that was found soon after death on the east coast of Nova Scotia. Thus, we did not kill any birds specifically for this project. For the specimens that were obtained, the vast majority were shot by shotgun using steel shot, were healthy in the sense that birds were not emaciated, had no obvious illnesses, and were collected among other members of their species in natural environments. The carcasses were kept cool and then frozen as soon as possible. Afterwards the head, some blood and usually neck muscle tissue were obtained from stored specimens. The skull was cut open and brain tissue was sampled, and similarly samples were taken from neck muscle; for species where neck tissue was not available, breast muscle was taken. Blood was available from some carcasses at the time of capture, or otherwise was extracted from clots in veins or arteries. We took multiple samples of brain and muscle from each bird (n = 2-4; samples were too small to do the same for blood) from consistent locations across individuals; i.e. the same region [or part] of the brain or muscle group. Samples of each tissue were taken (generally  $\sim 1-2$  g),

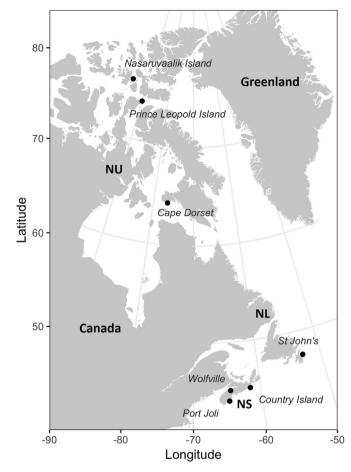


Fig. 1. Map of collection locations for coastal birds used in this study. Sampling sites were all in Canada, and included Nunavut (NU), Newfoundland and Labrador (NL), and Nova Scotia (NS).

freeze-dried, homogenized to powder using a clean mortar and pestle, and transferred to 1.5 mL Eppendorf tubes and frozen until analysis. Note that for some specimens, we could not extract enough blood, so only muscle and brain tissues were available (hence sample sizes differ for comparisons).

We analysed for total mercury (THg) at the Center for Analytical Research on the Environment (CARE) at Acadia University. Samples were analysed on a Nippon Instruments MA-2000 Mercury Analysis System using thermal pyrolysis and gold amalgamation atomic absorption. Samples were reagent blank corrected (average reagent blank  $0.55 \pm 0.29 \,\text{ng/g}, n = 102$ ) with the method detection limit (MDL) calculated as  $3 \times$  the standard deviation of the reagent blanks (MDL = 0.87 ng/g; n = 102). All samples were well above detection limits. Internal quality control included analytical blanks and certified reference material (DORM-4, National Research Council of Canada). The mean recovery for the certified reference material (n = 106) was 120.7% for THg, so we recovery-corrected all THg values. For 88 individuals we had 2-4 samples of muscle from the same birds, and for 30 individuals we had 2-3 samples of brain tissue. We analysed all of these and calculated intra-individual variation in THg concentrations using absolute range of differences within the same individual and tissue, as well as coefficient of variation (CoV). In calculations across species, we used the highest value per tissue per individual, as we felt that this gave the best index of possible toxicological risk levels.

We tested distributions of data using Kolmogorov-Smirnov (K-S) tests. Raw data for all the THg values in all tissue types were somewhat skewed, so we ln-transformed those data. We used general linear models (GLM) to compare ln-transformed THg among tissues and bird

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