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journal homepage: www.elsevier.com/locate/envpolMaternal transfer of mercury to songbird eggs[☆]Joshua T. Ackerman^{*}, C. Alex Hartman, Mark P. Herzog

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ABSTRACT

We evaluated the maternal transfer of mercury to eggs in songbirds, determined whether this relationship differed between songbird species, and developed equations for predicting mercury concentrations in eggs from maternal blood. We sampled blood and feathers from 44 house wren (*Troglodytes aedon*) and 34 tree swallow (*Tachycineta bicolor*) mothers and collected their full clutches ($n = 476$ eggs) within 3 days of clutch completion. Additionally, we sampled blood and feathers from 53 tree swallow mothers and randomly collected one egg from their clutches ($n = 53$ eggs) during mid to late incubation (6–10 days incubated) to evaluate whether the relationship varied with the timing of sampling the mother's blood. Mercury concentrations in eggs were positively correlated with mercury concentrations in maternal blood sampled at (1) the time of clutch completion for both house wrens ($R^2 = 0.97$) and tree swallows ($R^2 = 0.97$) and (2) during mid to late incubation for tree swallows ($R^2 = 0.71$). The relationship between mercury concentrations in eggs and maternal blood did not differ with the stage of incubation when maternal blood was sampled. Importantly, the proportion of mercury transferred from mothers to their eggs decreased substantially with increasing blood mercury concentrations in tree swallows, but increased slightly with increasing blood mercury concentrations in house wrens. Additionally, the proportion of mercury transferred to eggs at the same maternal blood mercury concentration differed between species. Specifically, tree swallow mothers transferred 17%–107% more mercury to their eggs than house wren mothers over the observed mercury concentrations in maternal blood (0.15–1.92 $\mu\text{g/g ww}$). In contrast, mercury concentrations in eggs were not correlated with those in maternal feathers and, likewise, mercury concentrations in maternal blood were not correlated with those in feathers (all $R^2 < 0.01$). We provide equations to translate mercury concentrations from maternal blood to eggs (and vice versa), which should facilitate comparisons among studies and help integrate toxicity benchmarks into a common tissue.

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1. Introduction

Mercury pollution is globally pervasive and methylmercury bioaccumulation continues to be an environmental problem (Driscoll et al., 2013; Krabbenhoft and Sunderland, 2013; Eagles-Smith et al., 2016). Birds are often used as bioindicators of environmental mercury contamination and avian reproduction has been shown to be sensitive to mercury toxicity (Scheuhammer et al., 2007; Ackerman et al., 2016b). Mercury monitoring programs often use several different bird tissues to assess contaminant exposure, and this complicates data comparisons among studies (Evers et al., 2011; Ackerman et al., 2016b). Furthermore, avian

toxicity benchmarks have been developed for several tissues, and integrating the effects of mercury toxicity across tissues is difficult (Ackerman et al., 2016b). Quantifying the relationships in mercury concentrations among tissues would facilitate converting mercury concentrations in one tissue into equivalent mercury concentrations in another tissue and provide the ability to translate mercury concentrations across bird tissues. The most commonly recommended tissues for sampling environmental mercury exposure in birds are adult blood and eggs (Ackerman et al., 2016b).

Equations for estimating mercury concentrations in eggs using mercury concentrations in the mother are available for a few species of birds sampled in the wild (Evers et al., 2003; Brasso et al., 2010; Kenow et al., 2015; Ackerman et al., 2016a) or in captivity as part of artificial dosing studies (Heinz et al., 2010; Ou et al., 2015). Only one study has compared the maternal transfer of contaminants among species and found that, at a given maternal blood

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mercury concentration, the proportion of mercury transferred to eggs differed among species (Ackerman et al., 2016a). That study was conducted on three species in the order Charadriiformes (Ackerman et al., 2016a); most of the other maternal transfer studies were on common loons (*Gavia immer*; Evers et al., 2003; Kenow et al., 2015). In contrast, few studies have examined the transfer of mercury from mothers to eggs in songbirds (Brasso et al., 2010). Songbirds may be more sensitive to mercury toxicity than some other avian taxa (Heinz et al., 2009), and the limited data available suggest that songbird mothers may transfer mercury to eggs at a lower rate than other avian taxa (compare Brasso et al., 2010; Ackerman et al., 2016a).

Our objective was to evaluate the maternal transfer of mercury to eggs in songbirds, and determine whether this relationship differed among songbird species. We examined the relationship between mercury concentrations in mothers and eggs in two species of songbirds: house wrens (*Troglodytes aedon*) and tree swallows (*Tachycineta bicolor*). These species are insectivorous and often forage in riparian and aquatic habitats that can be elevated in methylmercury contamination. Because they readily use artificial nest boxes, we were able to locate nests across a diversity of habitats which allowed us to examine the transfer of mercury into eggs across a wide range of maternal mercury concentrations. Additionally, wrens (Family Troglodytidae) and swallows (Family Hirundinidae) have been among the most studied songbirds for mercury contamination in the wild (Gerrard and Louis, 2001; Custer et al., 2007; Brasso and Cristol, 2008; Hothem et al., 2008; Hawley et al., 2009; Hallinger and Cristol, 2011; Hartman et al., 2013) and there are some studies that have suggested potential negative effects of mercury exposure on reproduction in these taxa (Heinz et al., 2009; Jackson et al., 2011). Therefore, providing maternal transfer equations for these songbird species should be useful for many of the studies investigating mercury contamination in songbirds.

2. Material and methods

2.1. Sample collection

During 2013–2015, we sampled songbirds and collected eggs at the Cache Creek Settling Basin (38.7°N, 121.7°W) and Cosumnes River Preserve (38.3°N, 121.4°W) within the Central Valley of California. At these sites, we installed and have maintained nearly 500 artificial nest boxes since 2012. Artificial nest boxes equipped with wigwag traps were constructed following standard guidelines for swallows (<http://golondrinas.cornell.edu/>). We checked nest boxes approximately twice weekly for nesting activity during the breeding season from late March through early June. We evaluated relationships between mercury concentrations in a mother's blood and her eggs during two different incubation stages: (1) house wren and tree swallow mothers that were captured and bled within 3 days of clutch completion and (2) tree swallow mothers that were captured and bled during mid to late incubation (6–10 days during incubation).

2.1.1. Sample collection: at clutch completion

For mothers that were captured within 3 days after clutch completion, we included only nests that were discovered during the egg-laying stage, by floating eggs to confirm that they were still fresh and had not advanced in embryo development (Ackerman and Eagles-Smith, 2010). For these nests, we returned to the nest at the expected clutch completion date. Clutch completion dates were estimated by taking the absolute difference between the initial clutch size and the expected full clutch size (typically 6 eggs for tree swallows and 7 eggs for house wrens) and adding this value

to the date of the initial nest visit with the assumption that females would lay one egg per day on average. We then returned to the nest the following day after the expected clutch completion date to confirm that the clutch had been completed and no new eggs had been added to the clutch. If new eggs had been added to the clutch, then we returned to the nest on the following day and repeated this procedure until no new eggs had been added to the clutch. Once we confirmed clutch completion, we attempted to capture the incubating female using two methods. First, we quietly approached the nest box and placed a net around the nest box entry and exit hole and attempted to flush the bird from the nest box into the net. If this trapping attempt was unsuccessful, we attached a fishing line to the wigwag (small wooden block that can swing to cover the exit hole), moved away from the nest, watched the nest until the female returned, and then triggered the wigwag to cover the entry and exit hole to trap the bird. Once captured, we verified sex of the bird by plumage (for tree swallows) and the presence of a brood patch. We sampled whole blood from the brachial vein using sodium-heparinized capillary tubes that were thereafter sealed with crito-caps and stored in sealed polypropylene cryovials. Blood was stored on ice in the field and at -20°C in the laboratory until mercury determination. We also collected breast feathers and stored them in Whirl-Paks[®] (Nasco, Modesto, California, USA). We then banded birds with a U.S. Geological Survey leg band, measured their external morphology, and released them at the capture site. After the female was captured and bled, we collected all eggs in the clutch and stored them in a refrigerator until egg dissection.

2.1.2. Sample collection: during mid to late incubation

For a separate group of tree swallow nests, we captured mothers during mid to late incubation to test whether the timing of blood sampling influenced the resulting relationship between mercury concentrations in maternal blood and eggs. These nests were monitored during incubation and were targeted for female capture approximately 8 days after clutch completion (range: 6–10 days during incubation). The incubation period of tree swallows is typically 13–14 days (Winkler et al., 2011), therefore we refer to our sampling period as mid-late incubation. After the female was captured and bled, we collected only one egg randomly from her clutch (instead of collecting the full clutch).

2.2. Egg dissection

We allowed refrigerated eggs to come to room temperature and then measured egg length and width to the nearest 0.01 mm using digital calipers (Fowler, Newton, Massachusetts, USA) and total egg weight (with eggshell) to the nearest 0.01 g on an analytical balance (Ohaus Adventurer Pro, Ohaus Corporation, Pine Brook, New Jersey, USA). Using clean, stainless steel instruments, we cut a ~5-mm diameter hole in the wide end of each egg and removed the entire egg contents into a tared, sterile 30-mL jar. We then measured egg content weight (without eggshell) with a digital balance to the nearest 0.01 g, and stored egg contents at -20°C until processing.

2.3. Sample processing

We determined total mercury (THg) concentrations and used them as an index of methylmercury (MeHg) concentrations because nearly all mercury in bird eggs (Ackerman et al., 2013), blood (Rimmer et al., 2005), and feathers (Bond and Diamond, 2009) is in the more toxic methylmercury form. THg concentrations were determined on a dry-weight basis. For eggs, we thawed egg contents at room temperature and then dried the entire egg contents at 50°C for 48 h or until completely dried. To determine

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