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Maternal transfer of contaminants in birds: Mercury and selenium concentrations in parents and their eggs

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ABSTRACT

We conducted a detailed assessment of the maternal transfer of mercury and selenium to eggs in three bird species ($n = 107$ parents and $n = 339$ eggs), and developed predictive equations linking contaminant concentrations in eggs to those in six tissues of the mother (blood, muscle, liver, kidney, breast feathers, and head feathers). Mercury concentrations in eggs were positively correlated with mercury concentrations in each of the mother's internal tissues ($R^2 \geq 0.95$), but generally not with feathers. For each species, the proportion of mercury transferred to eggs decreased as mercury concentrations in the mother increased. At the same maternal mercury concentration, the proportion of mercury transferred to eggs differed among species, such that Forster's tern (*Sterna forsteri*) and black-necked stilt (*Himantopus mexicanus*) females transferred more methylmercury to their eggs than American avocet (*Recurvirostra americana*) females. Selenium concentrations in eggs also were correlated with selenium concentrations in the mother's liver ($R^2 = 0.87$). Furthermore, mercury and selenium concentrations in tern eggs were positively correlated with those in the father ($R^2 = 0.84$). Incubating male terns had 21% higher mercury concentrations in blood compared to incubating females at the same egg mercury concentration. We provide equations to predict contaminant concentrations in eggs from each of the commonly sampled bird tissues.

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1. Introduction

Methylmercury contamination of the environment is widespread globally (Driscoll et al., 2013), and methylmercury biomagnifies primarily through aquatic food chains (Wiener et al., 2003) where environmental conditions tend to be conducive to methylmercury production (Ullrich et al., 2001). Birds often are vulnerable to mercury contamination because they are top predators in many aquatic habitats (Scheuhammer et al., 2007), and mercury has long been a primary contaminant of concern for birds (Bond et al., 2015; Monteiro et al., 1997). Because reproduction is among the most sensitive end points for mercury toxicity in birds (Scheuhammer et al., 2007; Wiener et al., 2003), eggs are considered an ideal sampling tissue for evaluating mercury exposure and potential toxicity to birds (Hartman et al., 2013). However, many different bird tissues are commonly sampled to assess mercury exposure (Scheuhammer et al., 2007; Wiener et al., 2003), and often it is difficult to compare mercury concentrations in adult bird tissues to the more highly developed toxicity benchmarks established for eggs (Braune et al., 2012; Heinz et al., 2009b; Kenow et al., 2011). Thus, quantifying the relationship between mercury concentrations, and other contaminants, in parental tissues and their eggs would allow conversion of contaminant concentrations in female tissues to those expected to be in her eggs.

Several prior studies have examined the relationship between mercury concentrations in mothers and her eggs. Lewis et al. (1993) found weak correlations between mercury concentrations in the first egg laid by herring gulls (*Larus argentatus*) and those in the incubating female's liver ($R^2 = 0.18$) and muscle ($R^2 = 0.12$), but not those in the ovary ($R^2 = 0.06$) or feathers ($R^2 < 0.01$). Evers et al. (2003) observed a stronger relationship ($R^2 = 0.79$) between mercury concentrations in nonviable, abandoned eggs and those in the blood of female common loons (*Gavia immer*) that were captured within the same territory in either the specific year or in a different year than when the eggs were collected. Kenow et al. (2015) found an even stronger correlation ($R^2 = 0.97$) between both the first and second laid egg's mercury concentration and those in the blood of

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female common loons that were captured on the nest within a few days of when the eggs were laid. Brasso et al. (2010) also documented a correlation ($R^2 = 0.87$) between the mean mercury concentration in a clutch and those in the blood of female tree swallows (*Tachycineta bicolor*) that were captured at the nest soon after the full clutch was laid. Heinz et al. (2010) fed captive mallards (*Anas platyrhynchos*) a diet containing methylmercury chloride and established a strong relationship between mercury concentrations in a single egg and blood of the female that was bled either the same day the egg was laid ($R^2 = 0.88$) or bled 16–27 days after the egg was laid ($R^2 = 0.67$). Ou et al. (2015) fed captive zebra finches (*Taeniopygia guttata*) a diet dosed with methylmercury cysteine and also observed a strong relationship between mean mercury concentrations in a clutch and those in the female's blood ($R^2 = 0.93$). These results indicate that there likely is a strong relationship between mercury concentrations in females and her eggs, however each study focused on a single species, used a limited number of parental tissues, and had various methodological differences which make it difficult to broadly apply these relationships to birds.

We conducted a detailed assessment of the maternal transfer of mercury to eggs in three species of birds. Because selenium can influence mercury dynamics in birds (Eagles-Smith et al., 2009b; Henny et al., 2002), we also evaluated maternal transfer of selenium to eggs. We selected species that were known to have relatively high (Forster's terns, *Sterna forsteri*; hereafter referred to as terns), moderate (black-necked stilts, *Himantopus mexicanus*; hereafter referred to as stilts), and low (American avocets, *Recurvirostra americana*; hereafter referred to as avocets) exposure to mercury (Ackerman et al., 2013b; Eagles-Smith et al., 2009a) which allowed us to examine maternal transfer of contaminants over a wide range of exposure levels. These species also represented different foraging guilds, with tern diets consisting mainly of fish (McNicholl et al., 2001) and avocet and stilt diets consisting mainly of aquatic invertebrates (Ackerman et al., 2013a; Robinson et al., 1999). We developed predictive equations to link mercury and selenium concentrations in eggs to concentrations in six tissues of the mother (blood, muscle, liver, kidney, breast feathers, and head feathers). We also related mercury and selenium concentrations in eggs to concentrations in the father's blood and liver, respectively, to assess whether mates had similar contamination levels.

2. Material and methods

2.1. Sample collection

During 2005 and 2006, we collected breeding avocets, stilts, and terns at several locations throughout San Francisco Bay, California (37.8°N, 122.3°W). Capture methods are described in detail elsewhere (Eagles-Smith et al., 2008), as these bird collections were used to investigate several topics related to contaminants in birds (Ackerman and Eagles-Smith, 2009; Eagles-Smith et al., 2009a, 2009b, 2008; Herring et al., 2010; Hoffman et al., 2011). Briefly, we collected avocets, stilts, and terns on their nests during early incubation (Eagles-Smith et al., 2008), after we had confirmed during our routine nest monitoring procedures (Ackerman et al., 2014) that their nests contained a full clutch of eggs and were being incubated normally. Overall, 87% of birds were collected before the mid-point of the incubation period as determined by egg flotation (Ackerman and Eagles-Smith, 2010). Nesting birds were collected using self-triggered treadle traps, remotely activated bow nets (Northwoods, Rainer, Washington, USA), or net launchers (Coda Enterprises, Mesa, Arizona, USA). Once the parent was captured, we collected all eggs in the clutch and stored them in a refrigerator until egg dissection. Because both sexes incubate eggs in these three species, we waited until we observed the female return to the target nest before setting the nest trap. We used plumage coloration to identify female stilts (Robinson et al., 1999) and bill shape to identify female avocets (Ackerman et al., 2013a). The sex of Forster's terns cannot be easily determined in the field (Bluso et al., 2006), therefore we collected the incubating parent (male or female) and determined sex during necropsy and genetic analysis (Zoogen Services, Davis, California, USA).

2.2. Egg dissection and adult necropsy

Prior to egg dissection, we allowed refrigerated eggs to come to room temperature and then we 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 a digital balance (Ohaus Adventurer Pro, Ohaus Corporation, Pine Brook, New Jersey, USA). Using clean, stainless steel instruments, we cut a ~15 mm diameter hole in the wide end of each egg and removed the entire egg contents into a tared, sterile 30 or 60 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 and mercury and selenium determination.

Following bird collection, we sampled whole blood from each bird using sodium-heparinized 23–25 gauge needles attached to polypropylene syringes, and then transferred the blood to polypropylene cryovials. Whole blood was collected either via the brachial vein prior to, or from the heart during, necropsy (Eagles-Smith et al., 2008). We also collected fully grown breast feathers from the anterior area of the keel and head feathers from the crown. We then conducted necropsies on each bird using clean, acid-rinsed, and stainless-steel instruments to excise the liver, kidneys, and a portion of breast muscle. We placed tissue samples in I-CHEM glass vials (Chase Scientific Glass, Rockwood, Tennesse, USA) or Whirl-Paks® (Nasco, Modesto, California, USA). All tissue samples were stored on dry ice in the field and at −20 °C in the laboratory until processing and mercury determination.

2.3. Sample processing

We thawed egg samples at room temperature, and then dried the entire egg contents at 50 °C for 48 h or until completely dried. To determine moisture content, we reweighed dried egg contents with a digital balance to the nearest 0.0001 g (Ohaus Adventurer Balance, model AR064; Ohaus, Pine Brook, New Jersey, USA). We then ground the dried egg contents to a powder using a spice grinder with stainless steel blades, followed by further grinding by hand in a mortar and pestle. We thawed liver, kidney, and muscle samples at room temperature, rinsed them in deionized water, and blotted them dry with Kimwipes® (Kimberly–Clark, Roswell, Georgia, USA). We measured each tissue's wet mass and then oven-dried them at 50 °C for 48 h or until they reached a constant mass (0.0001 g; Ohaus Adventurer Balance, model AR064; Ohaus, Pine Brook, New Jersey, USA). We then homogenized each tissue sample separately using a porcelain mortar and pestle. We washed and manually scrubbed feathers in a 1% Alconox solution (Alconox, White Plains, New York, USA) to remove surface debris, and then oven-dried them at 50 °C for 24 h. We stored the processed and dried tissue samples in a desiccator until mercury determination. We thawed blood to room temperature, inverted the cryovials several times, and thoroughly mixed the blood by stirring with a clean pipette tip to ensure sample homogeneity before mercury determination. Some blood samples that were high in mercury were further diluted with deionized water before mercury determination (Ackerman et al., 2008; Eagles-Smith et al., 2008).