



# Season, molt, and body size influence mercury concentrations in grebes<sup>☆</sup>



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## ABSTRACT

We studied seasonal and physiological influences on mercury concentrations in western grebes (*Aechmophorus occidentalis*) and Clark's grebes (*A. occidentalis*) across 29 lakes and reservoirs in California, USA. Additionally, at three of these lakes, we conducted a time series study, in which we repeatedly sampled grebe blood mercury concentrations during the spring, summer, and early fall. Grebe blood mercury concentrations were higher among males ( $0.61 \pm 0.12 \mu\text{g/g ww}$ ) than females ( $0.52 \pm 0.10 \mu\text{g/g ww}$ ), higher among Clark's grebes ( $0.58 \pm 0.12 \mu\text{g/g ww}$ ) than western grebes ( $0.51 \pm 0.10 \mu\text{g/g ww}$ ), and exhibited a strong seasonal pattern (decreasing by 60% from spring to fall). Grebe blood THg concentrations exhibited a shallow, inverse U-shaped pattern with body size, and was lowest among the smallest and largest grebes. Further, the relationship between grebe blood mercury concentrations and wing primary feather molt exhibited a shallow U-shaped pattern, where mercury concentrations were highest among birds that had not yet begun molting, decreased approximately 24% between pre-molt and late molt, and increased approximately 19% from late molt to post-molt. Because grebes did not begin molting until mid-summer, lower grebe blood mercury concentrations observed in late summer and early fall were consistent with the onset of primary feather molt. However, because sampling date was a much stronger predictor of grebe mercury concentrations than molt, other seasonally changing environmental factors likely played a larger role than molt in the seasonal variation in grebe mercury concentrations. In the time series study, we found that seasonal trends in grebe mercury concentrations were not consistent among lakes, indicating that lake-specific variation in mercury dynamics influence the overall seasonal decline in grebe blood mercury concentrations. These results highlight the importance of accounting for sampling date, as well as ecological processes that may influence mercury concentrations, when developing monitoring programs to assess site-specific exposure risk of mercury to wildlife.

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## 1. Introduction

Mercury is highly toxic to fish and wildlife, with documented negative effects on behavior, physiology, survival, and reproduction (Ackerman et al., 2016b; Scheuhammer et al., 2007). Because aquatic habitats tend to support conditions that promote the methylation of inorganic mercury into the more toxic methylmercury (Hall et al., 2008; Ullrich et al., 2001), and because methylmercury biomagnifies up food chains (Wiener et al., 2003), upper

trophic level wetland-dependent organisms, such as piscivorous waterbirds, are particularly at risk to mercury exposure (Ackerman et al., 2016b; Evers et al., 2005). It is for these reasons that avian piscivores are often used as indicators for site-specific exposure risk of mercury contamination on environmental health (Becker, 2003).

Blood is commonly regarded as an ideal tissue for evaluating site-specific mercury exposure risk in birds. Blood mercury concentrations are highly correlated with concentrations of internal tissues (Eagles-Smith et al., 2008), and females' blood mercury concentrations at the time of egg laying often are highly correlated with the mercury concentrations of their eggs (Ackerman et al., 2016a; Evers et al., 2003; Heinz et al., 2010), thereby providing a link to potential effects on reproduction. Furthermore, blood mercury concentrations often reflect recent mercury exposure

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(Monteiro and Furness, 2001), such that sampling blood from individuals at a specific location can reveal mercury exposure risk to reproduction for that location (but see Hartman et al., 2013). However, blood mercury concentrations can change rapidly through time, due to changing environmental factors including bird foraging areas or diet, methylmercury bioaccumulation in prey, and rate of methylmercury production (Eagles-Smith et al., 2009). For example, Rimmer et al. (2010) suggested that blood mercury concentrations of Bicknell's thrush (*Catharus bicknelli*) decreased over the summer as birds switched from more carnivorous detrital-based arthropods (higher mercury) to more herbivorous foliage-based arthropods (lower mercury).

In addition to changing environmental factors influencing seasonal changes in blood mercury concentrations, physiological factors within birds themselves may also contribute to seasonal variation in their mercury concentrations. Transfer of mercury into growing feathers is a major pathway for mercury excretion in birds, and during feather growth the body burden of mercury can decrease (Bearhop et al., 2000; Braune and Gaskin, 1987; Monteiro and Furness, 2001). Many birds undergo a prebasic molt of their flight feathers in late summer or early fall following the breeding season, and a more limited prealternate molt in the winter or early spring (Pyle, 2008). Thus, all else being equal, blood mercury concentration may be expected to decline during the periods of molt and feather growth. In addition, some avian piscivores, such as grebes, undergo breast muscle atrophy and may even become flightless at summer breeding or staging lakes (Jehl, 1997; LaPorte et al., 2013), followed by breast muscle hypertrophy, where breast muscle is regained. Changes in mass associated with breast muscle atrophy may allow for mercury concentrations to increase through mercury mobilization of catabolized tissue (Seewagen et al., 2016) whereas breast muscle hypertrophy may allow for mercury concentrations to decrease through mass dilution (Ackerman et al., 2011). Finally, female birds can reduce their body burden of mercury through transfer of mercury into eggs, potentially resulting in a decrease in blood mercury concentrations of females during the egg-laying period (Ackerman et al., 2016a; Lewis et al., 1993).

Given the potential for mercury concentrations to vary seasonally due to various environmental and physiological factors, sampling date may greatly influence estimation of site-specific mercury exposure risk to birds. Sampling bird mercury concentrations at specific locations throughout the year may allow for identification of periods of high and low mercury exposure, potential causes for these high and low periods, and a more comprehensive evaluation of risk to wildlife. Western grebes (*Aechmophorus occidentalis*) and Clark's grebes (*A. clarkia*) are two closely-related piscivorous waterbirds that breed within freshwater lakes throughout much of western North America, and migrate to, and overwinter along the Pacific coast from southern Alaska to Mexico (LaPorte et al., 2013). Smaller numbers overwinter, and may be resident, at large inland freshwater lakes, particularly in California (LaPorte et al., 2013; Wilson et al., 2013). Previously, Ackerman et al. (2015b) reported a strong relationship between western and Clark's grebe blood mercury concentrations and prey fish mercury concentrations among 25 California lakes. In this study, we evaluated seasonal and physiological influences on blood mercury concentrations of western grebes and Clark's grebes across multiple lakes and reservoirs in California. Western and Clark's grebes are useful species for evaluating physiological effects on mercury concentrations. They exhibit synchronous wing feather molt, whereby all flight feathers are shed and regrown at the same time, and they undergo breast muscle atrophy after arriving at lakes during the breeding season (LaPorte et al., 2013). As a result, western and Clark's grebes experience two dramatic physiological processes (mercury transfer to growing feathers and mass change) that can alter blood mercury

concentrations. Additionally, we repeatedly sampled grebes using a time series design at three sites during the spring, summer, and early fall to gauge how mercury concentrations and grebe morphometrics (e.g., mass, molt) varied over time at specific sites. Our results highlight the influence of sampling date and changes in grebe physiology on interpretation of site-specific exposure risk to mercury, and why these factors are important to incorporate into mercury biomonitoring programs.

## 2. Materials and methods

### 2.1. Study lakes and time series

We captured western grebes and Clark's grebes (hereafter grebes) at 29 lakes and reservoirs (hereafter lakes) throughout California during 2012 and 2013 (see map in Hartman et al., 2016). We sampled grebes at 12 lakes in 2012, 14 lakes in 2013, and at 3 lakes in both years. Grebes used in this study include those sampled at the 25 lakes reported by Ackerman et al. (2015b), where relationships between grebe blood THg concentrations and prey fish THg concentrations were examined, as well as grebes sampled at 3 of these lakes during additional visits not included in Ackerman et al. (2015b), and grebes sampled at four additional lakes. We confirmed that grebes bred at 7 of the 29 lakes in one or both years of the study. However, because most lakes were visited a single time, over 2–3 days, and often before nesting had begun, we could not confirm breeding status at all of the 22 remaining lakes. See Ackerman et al. (2015a) for additional information on the sampled lakes.

At 26 of the 29 lakes, we sampled grebes only once, over a few consecutive days, between April 30<sup>th</sup> and October 6<sup>th</sup>. At the 3 other lakes (Clear Lake, Lake Berryessa, and Lake San Antonio) we carried out a time series study by sampling grebes repeatedly during monthly visits throughout the spring, summer, and early fall. In both 2012 and 2013, we sampled grebes at these three lakes during May 14<sup>th</sup>–June 2<sup>nd</sup> (May), June 17<sup>th</sup>–July 11<sup>th</sup> (June/July), and August 3<sup>rd</sup>–August 29<sup>th</sup> (August). Additionally, in 2012, we sampled grebes at these three lakes during September 7<sup>th</sup>–September 24<sup>th</sup> (September) and, in 2013, we sampled grebes during April 1<sup>st</sup>–April 11<sup>th</sup> (April) at Clear Lake and Lake Berryessa. This yielded a total of five sampling time periods at Clear Lake and Lake Berryessa, and four sampling time periods at Lake San Antonio. We selected the three time series lakes because they each have large summer grebe populations, fish from these lakes had relatively high mercury concentrations (Ackerman et al., 2015b; Davis et al., 2010), and, in the case of Clear Lake, supports a large grebe breeding population.

### 2.2. Grebe sampling

We captured grebes from boats using night-lighting techniques (King et al., 1994; Whitworth et al., 1997). From boats, we directed a spotlight at individual grebes to disorient them, and captured them with a long-handled net. We weighed grebes with a digital bench scale ( $\pm 2$  g, Ohaus ES6R) or spring scale ( $\pm 20$  g, Pesola spring scales) and recorded body measurements, including head-to-bill length, nares-to-bill length, exposed culmen length, bill depth, short tarsus length, and flattened wing length. We measured all morphometrics to the nearest 0.1 mm with digital calipers, except for wing length, which we measured to the nearest 1 mm with a wing ruler. We distinguished western grebes from Clark's grebes by plumage and bill color (LaPorte et al., 2013), and used DNA sex determination (Zoogen Services, Inc., Davis, Ca) or discriminant function analysis based on morphometrics (Hartman et al., 2016) to identify the sex of individuals. We described wing molt by classifying each of the

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