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Patterns of Mercury Accumulation in Tissues of Western Sandpipers (*Calidris mauri*) Wintering on the Coast of Sinaloa, Mexico

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Abstract.—Mercury concentration was quantified in liver, muscle, and primary feathers of pooled samples of Western Sandpiper (*Calidris mauri*) collected in December-January 2010 (wintering) and March-April 2011 (premigration) in Ensenada Pabellones and Bahía Santa María, Sinaloa, Mexico, to test for differences between sites, stages, tissues, and age-sex groups. During the winter, mercury concentrations were greater in individuals from Ensenada Pabellones than those from Bahía Santa María (0.81 ± 0.05 vs. 0.57 ± 0.05 mg/kg dry weight, respectively), and greater in liver than in muscle (0.89 ± 0.05 vs. 0.49 ± 0.05mg/kg dry weight, respectively). In the comparison between the stages, wintering birds had greater concentrations of mercury than those of premigration period (0.62 ± 0.05 vs. 0.48 ± 0.05 mg/kg dry weight, respectively). These mercury concentrations were lower than those found in other wintering sites in North America (mean = 4.4 mg/kg dry weight). Although these concentrations suggest that Western Sandpipers wintering on the coast of Sinaloa, Mexico, were being exposed to low mercury concentrations and had not accumulated mercury, it appears that mercury in their tissues decreased during the nonbreeding season. However, these conclusions are based on low numbers in some categories and require further work with individual, rather than pooled, samples. *Received 27 February 2018, accepted 31 May 2018.*

Key words.—bioaccumulation, *Calidris mauri*, coastal wetlands, liver, muscle, nonbreeding, shorebirds.

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Environmental mercury (Hg) contamination is a significant global issue (Eagles-Smith *et al.* 2016). Mercury is most available and harmful to birds in the form of methylmercury, which tends to bioaccumulate and biomagnify through the food chains (Seewagen 2010). Mercury can have detrimental effects on the physiology, behavior, and reproductive success of birds, even at sublethal concentrations (Ackerman *et al.* 2016). In particular, shorebirds feeding in coastal wetlands might be exposed to elevated mercury concentrations because levels of mercury are often elevated in the sediment and its invertebrate fauna (Eagles-Smith *et al.* 2016). Most information on mercury concentrations in shorebirds comes from breeding and stopover sites in the Arctic and temperate zones of the Northern Hemisphere (Ackerman *et al.* 2016). In contrast, there are few studies from tropical and subtropical wintering populations, which are typically in developing countries where mercury contamination may be increased (Seewagen 2010).

Mercury concentrations in specific targeted tissues within bird species reflect different time scales of mercury accumulation. Mercury in feathers reflects blood concentration during the period of feather growth, and feathers are important areas of mercury depuration and sequestration (Eagles-Smith *et al.* 2008; Hartman *et al.* 2017). The liver has higher mercury concentrations, among internal tissues, because it is the primary organ of detoxification (Eagles-Smith *et al.* 2008). Other physiological and biological processes, such as sex, age, and migration, may influence mercury concentration in birds (Eagles-Smith *et al.* 2008; St. Clair *et al.* 2015; Hartman *et al.* 2017). Females can depurate mercury during the breeding season into the eggs they produce (Heinz *et al.* 2010), and adult birds tend to have higher concentrations than immature birds because they have been exposed for a longer time (St. Clair *et al.* 2015). In migratory birds, there are seasonal changes in body mass, with birds preparing for and during migration tending to weigh more than wintering birds (Colwell 2010). Thus, body mass increases can dilute mercury concentration in internal tissues (Hartman *et al.* 2017).

The Western Sandpiper (*Calidris mauri*) is one of the most common shorebirds of the Western Hemisphere. It breeds primarily in the western subarctic of Alaska and winters mostly along the Pacific Coast from California to Peru (Franks *et al.* 2014). Adult (after-hatch-year) birds undergo flight-feather molt immediately after southward migration and arrival on the nonbreeding grounds, whereas immature (hatch-year) birds do not molt their first set of flight feathers, grown on the breeding grounds, at this time (O'Hara *et al.* 2002; Galindo-Espinosa *et al.* 2013). Most Western Sandpipers undergo a prealternate body molt and gain weight during the premigratory period (Fernández *et al.* 2004). Western Sandpipers frequent intertidal mudflats at coastal wetlands and forage on biofilm, and on meiofaunal and macrofaunal invertebrates (Franks *et al.* 2014). Thus, it is possible that both the habitat and diet of Western Sandpipers place them at risk of toxic exposure to mercury.

The coastal wetlands of Sinaloa, Mexico, maintain > 20% of the Western Sandpiper world population during the nonbreeding season (Engilis *et al.* 1998). However, these coastal wetlands are subject to increased inputs of contamination through the waste effluents from intensive agriculture, shrimp aquaculture, and urban sewage from near towns and cities (Ruelas-Inzunza *et al.* 2007; Páez-Osuna and Osuna-Martínez 2015). Thus, wintering shorebirds are exposed to non-essential elements such as mercury. In this study, our objective was to determine baseline Hg concentrations in primary feathers, liver, and muscle, by sex and age class, of Western Sandpipers sampled at Bahía Santa María and Ensenada Pabellones on the coast of Sinaloa, Mexico.

Methods

Study Area

Bahía Santa María (25° 02′ N, 108° 18″ W) is the largest coastal wetland of Sinaloa, Mexico (1,350 km^2), and is about 90 km northwest of Culiacán City. Ensenada Pabellones (24° 26′ N, 107° 33″ W) is a coastal wetland encompassing 800 km^2 and is about 45 km southwest of Culiacán City, Sinaloa, Mexico. High concentrations of Hg have been reported in coastal organisms such as oysters (*Crassostrea corteziensis* and *C. palmula*) and birds (Ruelas-Inzunza *et al.* 2007; Páez-Osuna and Osuna-Martínez 2015).

Fieldwork

The Western Sandpipers examined (n = 68) in this study had accidentally drowned during nocturnal mist netting operations during the 2010-2011 nonbreeding season. Based on their body mass, it appears that these individuals were in good condition before they were caught. Wintering birds from Ensenada Pabellones and Bahía Santa María were collected on 19 December 2010 and 21 January 2011, respectively. Birds during the northward premigration period from Bahía Santa María were collected on 20 March and 3 April 2011 (Table 1). Each individual was sexed on the basis of exposed culmen length measurements (female ≥ 24.8 mm, male ≤ 24.2 mm; Page and Fearis 1971) and aged as hatch-year (< 1 year-old) or after-hatch-year on the basis of plumage coloration and wear of primary feathers (O'Hara *et al.* 2002). Carcasses were placed in a cooler with ice until they were transported to the laboratory. During dissection, we collected muscle, liver, and all primary feathers. Because the individual sample did not provide enough volume for analytical methods, we pooled samples by site, date, sex, and age class (Table 1).

Determination of Hg Concentrations

Feathers were washed in deionized water and acetone and then dried in an oven at 50 °C for 24 hr. Muscle and liver samples were lyophilized for 72 hr (133×10^{-3} mBar and −49 °C). All samples were ground in a ceramic mortar and digested to determine Hg content. The moisture content of each sample was calculated by subtracting the final weight after lyophilization from the initial fresh weight, using an analytical balance. First, 0.25 g of the dried and homogenized content of each sample was digested with 5 mL of 50%HNO_3 in a digestion system (CEM Corporation model $MARS_X$), then a second digestion was performed by addition of 3 mL of H_2O_2 to each vessel (U.S. Environmental Protection Agency 1996) using method 3052. Digested samples were analyzed in an atomic absorption spectrophotometer (PerkinElmer model 1100B). The Hg concentration was determined in a hydride generator (U.S. Environmental Protection Agency 2007) using method 7471B. The Hg concentrations were calculated in mg/kg of dry weight (mg/kg dw). For quality assurance/quality control purposes, a blank solution, a duplicate, and a certified reference material for trace metals (DOLT-4 Dogfish Liver) from the

Table 1. Western Sandpipers sampled at Bahía Santa María and Ensenada Pabellones, Sinaloa, Mexico: number of pooled samples by season (winter and premigration) and sex and age-class; in parenthesis, number of birds per pooled sample.

Sex and Age Class	Ensenada Pabellones – Winter	Bahía Santa María – Winter	Bahía Santa María – Premigration
Hatch year female (HY-F)	1 (4)	1 (3)	1 (1)
After-hatch-year female (AHY-F)	1 (3)	1 (4)	2 (5, 3)
Hatch year male (HY-M)	1 (1)	1 (5)	1 (3)
After-hatch-year male (AHY-M)	3 (5, 5, 4)	1 (6)	3 (6, 4, 6)

National Research Council of Canada were digested with every batch of 10 samples and analyzed with the rest of the samples. The detection limit was 0.0002 mg/kg, the average recovery was 99.8%, and the maximum relative difference was 10.4%.

Data Analysis

The Hg data obtained in this study is limited by the small number of pooled samples, and the variable number of individuals within each pooled sample. For the winter Hg data, we performed a main effects ANOVA to test the effect of the wintering site (Bahía Santa María and Ensenada Pabellones), the tissue type (liver and muscle), and the sex and age class, taking into account number of individuals within each pooled sample. Given the small sample size, we used a variable age-sex group (hatch-year female (HY-F), after-hatch-year female (AHY-F), hatch-year male (HY-M), and after-hatch-year male (AHY-M)) rather than age class and sex individually. Also, we analyzed the Hg concentration by tissue type (primary feather, liver, and muscle) and wintering site for adult birds only (pooled sexes), given that they molt primary feathers at the study sites. We performed a main effects ANOVA to test the effect of season (winter and premigration), tissue type (liver and muscle), and the sex and age class at Bahía Santa María. We report means (± SE). Results were considered significant at $P < 0.05$. All statistical analyses used Statistica Software (StatSoft, Inc. 2005).

RESULTS

Hg concentrations of wintering Western Sandpipers differed by wintering site ($F_{1, 74} = 10.08$; $P < 0.05$) and tissue type ($F_{1, 74} = 35.31$; $P < 0.05$), but they did not differ by age-sex group ($F_{4, 74} = 2.55$; $P = 0.06$). Overall, Hg concentrations were greater in individuals from Ensenada Pabellones than those from Bahía Santa María, and greater in liver than in muscle (Table 2). In the adults only analysis, we obtained similar results: Hg concentrations were greater in birds from Ensenada Pabellones than those from Bahía Santa María (1.27 ± 0.04 vs. 0.93 ± 0.06 mg/kg dw, respectively; $F_{1, 75} = 19.18$; $P < 0.05$), and greater in primary feathers than in liver or muscle (1.98 ± 0.06 vs. 0.83 ± 0.06 vs. 0.50 ± 0.06 mg/kg dw, respectively; $F_{2, 75} = 137.90$; $P < 0.05$).

In the comparison between the winter and premigration stages, Hg concentrations of individuals differed by stage ($F_{1, 86} = 4.17$; $P = 0.04$), tissue type ($F_{1, 86} = 38.93$; $P < 0.05$), and age-sex group ($F_{3, 86} = 4.60$; $P < 0.05$). Wintering individuals had greater concentrations of Hg than those of premigration individuals, and as observed in the wintering site analysis, liver had greater concentrations of Hg than those of muscle (Table 2). Also, Hg concentrations were greater in hatch-year females than in after-hatch-year females, hatch-year males, and after-hatch-year males (0.85 ± 0.10 vs. 0.39 ± 0.06, 0.51 ± 0.07 and 0.46 ± 0.04 mg/kg dw, respectively).

Table 2. Mercury concentrations (means ± SE; mg/kg of dry weight) of Western Sandpipers by wintering site and season at Bahía Santa María and Ensenada Pabellones, Sinaloa, Mexico. Results based on main effects ANOVA to test the effect of the wintering site (Bahía Santa María and Ensenada Pabellones) or season (winter and premigration), and the tissue type (liver and muscle).

		Site		Tissue	
Analysis	Season	Ensenada Pabellones	Bahía Santa María	Liver	Muscle
Between sites		0.81 ± 0.05	0.57 ± 0.05	0.89 ± 0.05	0.49 ± 0.05
Between stages (only Bahía Santa María)	Winter		0.62 ± 0.05	0.80 ± 0.07	0.39 ± 0.07
	Premigration		0.48 ± 0.05	0.58 ± 0.05	0.21 ± 0.05

Discussion

This study of the Hg concentrations in tissues of Western Sandpipers wintering on the coast of Sinaloa is the first analysis from a subtropical wintering population. Although we worked with pooled samples and some caution must be applied, these results could establish a baseline for migratory shorebirds on the Sinaloa coast. Hg concentrations of wintering adult Western Sandpipers were highest in primary feathers, followed by liver, and then muscle. These patterns are consistent with other studies; feather molt represents an important excretory outlet for Hg (Eagles-Smith *et al.* 2008; Ackerman *et al.* 2016), while the liver is involved in Hg detoxification and stores relatively high concentrations of Hg (Eagles-Smith *et al.* 2008). Hg concentrations for liver and muscle for both age classes closely reflect Hg contamination at the Sinaloa coastal wetlands during the nonbreeding season.

The Hg concentrations in wintering birds were higher at Ensenada Pabellones than those at Bahía Santa María, suggesting higher Hg availability. Hg concentrations in the liver (0.9 mg/kg dw) were similar to those found in the liver of wintering American Avocet (*Recurvirostra americana*, 1.0 mg/kg dw) sampled at Ensenada Pabellones (Ruelas-Inzunza *et al.* 2007). However, they were lower than those reported for wintering Western Sandpiper in San Francisco Bay, California, USA (4.4–4.7 mg/kg dw) with known point-source Hg pollution (Hui *et al.* 2001). Hg concentrations above 3.0 mg/kg dw in the liver have been reported to impair reproduction in other avian species (Eagles-Smith *et al.* 2009), and a review of data on North American birds (Ackerman *et al.* 2016) found the lowest concentration at which an adverse effect had been observed (oxidative stress response) to be 1.6 mg/kg dw in the liver. Overall, our results suggest that Western Sandpipers wintering on the coast of Sinaloa have Hg concentrations below this range. However, as mentioned above, due to the pooled nature of our samples, we were unable to determine if individual birds may be above the range.

The wintering samples showed higher Hg concentrations than those from premigration samples in the internal tissues; there was a 27.6% decrease of Hg concentrations in liver and a 46.8% decrease in muscle between stages. Such decreases in Hg concentrations from winter to premigration was expected due to transfer of Hg into the growing feathers during the prealternate body molt and through body mass dilution. In Western Sandpipers, the premigratory period for their northward migration starts in late February, when birds begin the prealternate body molt and progressively gain weight (Fernández *et al.* 2004). The decrease of Hg in internal tissues before departure to the breeding grounds has been found in some other bird species (Hartman *et al.* 2017).

We only found age-sex group differences in the comparison between the wintering and premigration stages. Hatch-year females had greater Hg concentrations that the other age-sex groups. Although Western Sandpipers may have specific sex- and age-related local spatial distribution and site fidelity during the winter (Fernández and Lank 2006), it is unlikely that a differential group exposition occurs during the nonbreeding season. Unfortunately, the pooled nature of our samples limited the power to detect patterns and interactions among factors. Further work with individual, rather than pooled, samples is needed for meaningful comparisons between age-sex groups, between study sites, and between stages.

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