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SELENIUM BIOACCUMULATION AND BODY CONDITION IN SHOREBIRDS AND TERNS BREEDING IN SAN FRANCISCO BAY, CALIFORNIA, USA

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Abstract—The present study evaluated Se bioaccumulation in four waterbird species ($n = 206$ birds) that breed within San Francisco Bay, California, USA: American avocets (*Recurvirostra americana*), black-necked stilts (*Himantopus mexicanus*), Forster's terns (*Sterna forsteri*), and Caspian terns (*Hydroprogne caspia*). Selenium concentrations were variable and influenced by several factors, including species, region, reproductive stage, age, and sex. Adult Se concentrations (μg/g dry wt) in livers ranged from 3.07 to 48.70 in avocets (geometric mean ± standard error, 7.92 ± 0.64), 2.28 to 41.10 in stilts (5.29 ± 0.38), 3.73 to 14.50 in Forster's terns (7.13 ± 0.38), and 4.77 to 14.40 in Caspian terns (6.73 ± 0.78). Avocets had higher Se concentrations in the North Bay compared to the South Bay, whereas stilt Se concentrations were similar between these regions and Forster's terns had lower Se concentrations in the North Bay compared to the South Bay. Female avocets had higher Se concentrations than male avocets, but this was not the case for stilts and Forster's terns. Of the factors assessed, reproductive stage had the most consistent effect among species. Prebreeding birds tended to have higher liver Se concentrations than breeding birds, but this trend was statistically significant only for Forster's terns. Forster's tern chicks had lower Se concentrations than Forster's tern adults, whereas avocet and stilt adults and chicks were similar. Additionally, body condition was negatively related to liver Se concentrations in Forster's tern adults but not in avocet, stilt, or Caspian tern adults and chicks. These variable results illustrate the complexity of Se bioaccumulation and highlight the need to sample multiple species and examine several factors to assess the impact of Se on wildlife.

Keywords—Bird Bioaccumulation Body condition San Francisco Bay Selenium

INTRODUCTION

Selenium is an essential micronutrient for vertebrates and is beneficial at low doses [1]. At exposure levels only slightly above beneficial doses, however, Se can be highly toxic to wildlife [1,2]. Diet is the main avian exposure pathway for Se [1,3], and bird tissues and egg Se concentrations respond quickly to changes in dietary Se exposure [4–6]. Selenium toxicity in birds has been detected primarily via teratogenic effects on developing embryos [7,8] and increased embryo mortality [9,10]. Sublethal effects also may occur in adults at elevated concentrations [11], with few overt signs other than emaciation, cracking of keratinaceous structures, and feather loss [12–15].

The San Francisco Bay Estuary (CA, USA) is the largest estuary on the west coast of North America and is recognized as a site of hemispheric importance to shorebirds [16,17]. Nearly 1,000,000 shorebirds and approximately 39% of the diving ducks within the Pacific Flyway use the estuary annually [17,18] (http://www.sfbayjv.org/estuarybook.php). Currently, however, several trace elements, including Se, occur at such elevated concentrations that the estuary is listed as impaired under section 303(d) of the Federal Clean Water Act [19] (http://www.waterboards.ca.gov/water_issues/programs/tmdl/docs/303dlists2006/swrcb/r2_final303dlist.pdf). Previous studies have demonstrated that diving ducks bioaccumulate Se to potentially toxic concentrations while wintering in San Francisco Bay, with geometric mean liver Se concentrations commonly approaching or exceeding 33 μg/g dry weight in surf scoters (*Melanitta perspicillata*) and lesser and greater scaup (*Aythya affinis* and *Aythya marila*, respectively) [11,20–24]. Long-billed dowitchers (*Limnodromus scolopaceus*) and western sandpipers (*Calidris mauri*) wintering in San Francisco Bay also have mean liver Se concentrations exceeding 11 μg/g dry weight [25], a level that has been associated with reproductive impairment in mallards (*Anas platyrhynchos*) [10,26].

Although extensive research has been conducted regarding Se exposure to waterbirds wintering in San Francisco Bay, few studies have assessed Se bioaccumulation in birds that breed locally within the estuary [27,28]. Additionally, Se levels are elevated in fish within San Francisco Bay [29,30], yet little work has been done concerning risk to fish-eating birds. The objective of the present study was to examine Se bioaccumulation in four waterbird species that breed within San Francisco Bay (CA, USA): American avocets (*Recurvirostra americana*, hereafter avocets), black-necked stilts (*Himantopus mexicanus*, hereafter stilts), Forster's terns (*Sterna forsteri*), and Caspian terns (*Hydroprogne caspia*). These species have different foraging habitats and diets that represent several distinct compartments within the San Francisco Bay food web. Avocets and stilts, the two most abundant breeding shorebirds in San Francisco Bay [17,31], forage primarily on aquatic insects and crustaceans [32,33]. Stilts, however, use more vegetated marsh habitats than avocets do, whereas avocets use more tidal flat habitats along the bay's margins [31,34]. Forster's terns and Caspian terns are piscivorous [35,36], but

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Forster's terns mainly forage on small fish (length, <10 cm [36]) within wetlands along the bay's margins [37], whereas Caspian terns mainly forage on larger fish (length, 5–30 cm [35]) within open-bay habitats [38]. In addition, avocets and stilts overwinter in San Francisco Bay and the Central Valley (CA, USA) [17,32,33], whereas Forster's terns and Caspian terns mainly overwinter outside the estuary and arrive in the spring to breed [35,36]. These differences in habitat use and diet, together with effects of reproductive stage, sampling region, and sex, were predicted to result in differential Se bioaccumulation and exposure risk. The relationship between body condition and liver Se concentrations also was examined to assess exposure risk in San Francisco Bay waterbirds.

MATERIALS AND METHODS

Study site

Selenium concentrations in waterbirds were studied throughout the San Francisco Bay Estuary in 2005. Main study regions included the North Bay (San Pablo Bay and Napa–Sonoma Marsh Wildlife Area; salt ponds 1, 1A, and 2 as well as Dutchman Slough, Rush Creek, and West End Duck Club), Central South Bay (Eden Landing Ecological Reserve; salt ponds E4, E6, E8, and E10), and Lower South Bay (Alviso salt pond complex of the Don Edwards San Francisco Bay National Wildlife Refuge; salt ponds A1, A5, A8, A11, A16, and A21 as well as Coyote Creek Marsh, New Chicago Marsh, and Rectangle Marsh). Study area maps and pond locations are available in Ackerman et al. [34,37].

Bird sampling

Adults were collected during the prebreeding season (March 3–April 12 for avocets and stilts and March 23–April 21 for Forster's terns and Caspian terns) and during the breeding season (May 19–July 5 for avocets and stilts and May 26–June 28 for Forster's terns). Collection of Caspian terns was not permitted during the breeding season. Prebreeding birds were collected using shotguns with steel shot or high-powered air rifles. Breeding birds were collected on their nests during midincubation using self-triggered treadle traps or remotely detonated bow nets (Northwoods) and net launchers (Coda Enterprises). Chicks were collected from July 5 to August 4, 2005, near nesting sites using long-handled nets or shotguns with steel shot. Each bird was weighed with a spring scale (Pesola AG), and exposed culmen length and short tarsus (tarsometatarus bone) length were measured with digital calipers (Fowler) and flattened wing length with a wing board. Birds were sexed via morphology and necropsy. All birds were collected under the guidelines of the U.S. Fish and Wildlife Service migratory bird and California Department of Fish and Game scientific collecting permits as well as the U.S. Geological Survey, Western Ecological Research Center, Animal Care and Use Committee.

Selenium determination

Necropsies were conducted in the field on birds using clean, acid-rinsed, stainless-steel scalpels, scissors, and forceps to excise the livers for Se analyses. Liver samples were placed in Whirl-Paks® (Nasco) or I-CHEM glass vials (Chase Scientific Glass) and immediately placed on dry ice until they could be stored frozen at −20°C. For processing, liver samples were thawed at room temperature, rinsed with deionized water, and blotted dry with clean Kimwipes® (Kimberly–Clark), then measured for wet mass to the nearest 0.0001 g on an analytical balance (Ohaus Adventurer Balance, model AR064; Ohaus). Liver samples were oven-dried at 50 to 60°C for 48 h or until they reached a constant mass. Once dried, each liver sample was reweighed to the nearest 0.0001 g, and the samples were then homogenized to a uniform consistency using a Wiley mill (Thomas Scientific) and a porcelain mortar and pestle.

Dried liver samples were stored in glass vials and sent to the Trace Elements Research Laboratory (Texas A&M University, College Station, TX, USA) to determine Se using hydride generation followed by atomic fluorescence. Certified reference materials for Se in tissues (National Institute of Standards and Technology 2976), matrix spikes, duplicate samples, and blanks were analyzed for quality-control purposes. Recoveries for certified reference materials averaged 106.3% ± 3.1% ($n = 11$), matrix spikes averaged 102.0% ± 7.0%; ($n = 11$), and absolute relative percentage difference for duplicates averaged 3.8% ± 1.2% ($n = 11$). Liver Se concentrations were reported on a dry-weight basis to control for differences in moisture content among birds; average moisture content was 67.2% ± 0.3% for avocets, 66.3% ± 0.4% for stilts, 64.9% ± 0.8% for Caspian terns, and 63.7% ± 0.4% for Forster's terns.

Statistical analysis

Using a tiered approach, the present study tested whether Se concentrations in waterbird livers differed among factors with JMP® software (Ver 5.0, SAS Institute). Selenium concentrations were $\log_e$ transformed to meet the assumptions of normality of residuals and homogeneity of variance. Unless otherwise noted, the reported model-based geometric mean ± standard error Se concentrations were based on back-transformed least-square means ± standard errors. The model-based standard errors of the geometric means were calculated by the delta method [39].

In the first stage of analysis, analysis of variance (ANOVA) was used to test whether liver Se concentrations in adults differed among species using only our data for prebreeding birds from the North Bay region. This data partitioning was necessary, because Caspian terns were allowed to be collected only during the prebreeding season and only in the North Bay whereas collection of the other three species was permitted during all time periods and regions. In the next stage of analysis, Caspian tern data were excluded and ANOVA was used to test the expanded global model that included species (avocets, stilts, and Forster's terns), reproductive stage (prebreeding and breeding), sampling region (Lower South Bay, Central South Bay, and North Bay), and sex as main effects and species × reproductive stage, species × region, species × sex, and reproductive stage × sex as two-way interactions. Nonsignificant interactions were dropped from the final model. Significant interactions were found between species and region and between species and sex (see *Results*); therefore, separate ANOVAs were conducted for each species to test the effects of reproductive stage, region, and sex. Tukey honestly significant difference multiple pairwise comparisons with $\alpha < 0.05$ were used to test differences between categories for significant variables. To present data graphically, least-square mean Se concentrations were back-transformed from the global model that included species × reproductive stage, species × region, and species × sex interactions.

For chicks, ANOVA was similarly used to test whether liver Se concentrations differed among species (avocets, stilts, and

Forster's terns), regions (Lower South Bay and Central South Bay), and the species × region interaction. Unlike the adult analyses, neither reproductive stage nor sex was included as a main effect in the model, and the chick sampling did not include the North Bay (because of reduced breeding effort by waterbirds in this region). Similar to adults, the global model indicated a significant species × region interaction (see *Results*). Thus, separate ANOVAs also were conducted for each species to test the effect of sampling region, and separate ANOVAs were conducted for each region to test the effect of species. Tukey honestly significant difference pairwise comparisons with $\alpha < 0.05$ was then used to test differences between categories for significant variables.

Selenium concentrations between chicks and adults could be compared only for the Lower South Bay, where we had sampled both chicks and breeding adults. To do this, ANOVA was used, with species, sex, and age (chicks or adults) as main effects. For this analysis, only the data collected during the breeding time frame for adults were used. To present the chick data graphically, least-square mean ± standard error concentrations were back-transformed from the global model that included species and sampling region as main effects and the species × region interaction.

The last stage of the analyses tested whether body condition was related to liver Se concentrations. Principal components analysis (PCA) of three structural body size measurements (length in mm of flattened wing, short tarsus, and exposed culmen) was used for each species and sex. Only a few male Caspian terns ($n = 1$) and stilts ($n = 3$) were available, so the PCA analysis was not split by sex for these species or for chicks. The first principal component (PC1) was used as an index of structural body size. For adults, analysis of covariance (ANCOVA) was then used to test whether bird mass ($\log_e$ transformed) was related to liver Se concentrations, and bird body size (PC1), sex, and reproductive stage were included to account for other factors that might influence a bird's body condition. As discussed above, neither reproductive stage nor sex was included in the model to test effects on body condition in chicks. Because any size-related variation in body mass was accounted for by including the PC1 of body size as a covariate, the results were interpreted as whether bird body condition was related to Se concentrations. Because Se could have a nonlinear effect, models also were assessed that included a quadratic term for Se concentrations.

RESULTS

Liver Se concentrations were analyzed in 156 adults and 50 chicks of four waterbird species. Across all regions, sexes, and reproductive stages, Se concentrations in livers of adult waterbirds in San Francisco Bay were 7.92 ± 0.64 μg/g dry weight in avocets ($n = 59$; range, 3.07–48.70 μg/g dry wt), 5.29 ± 0.38 μg/g dry weight in stilts ($n = 40$; range, 2.28–41.10 μg/g dry wt), 6.73 ± 0.78 μg/g dry weight in Caspian terns ($n = 10$; range, 4.77–14.40 μg/g dry wt), and 7.13 ± 0.38 μg/g dry weight in Forster's terns ($n = 47$; range, 3.73–14.50 μg/g dry wt). Selenium concentrations in 27% of avocet, 5% of stilt, 20% of Caspian tern, and 15% of Forster's tern livers exceeded 11 μg/g dry weight. For chicks, liver Se concentrations across all regions and sexes were 4.31 ± 0.34 μg/g dry weight in avocets ($n = 17$; range, 2.42–8.05 μg/g dry wt), 4.35 ± 0.39 μg/g dry weight in stilts ($n = 16$; range, 2.11–7.98 μg/g dry wt), and 3.19 ± 0.10 μg/g dry weight in Forster's terns ($n = 17$; range, 2.55–4.39 μg/g dry wt).

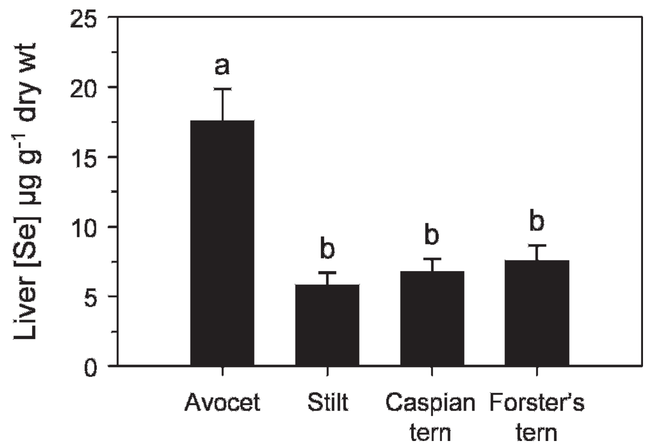

Fig. 1. Selenium concentrations (dry wt) in livers of four waterbird species during the prebreeding season (March 3 to April 21, 2005) in North San Francisco Bay (CA, USA). Different letters above bars indicate statistical difference (Tukey pairwise comparisons, $p < 0.05$).

Adults

Selenium concentrations differed among species using only the data for prebreeding birds in the North Bay (ANOVA: species, $F_{3,36} = 12.75$, $p < 0.0001$; sex, $F_{1,36} = 0.02$, $p = 0.89$). Tukey pairwise comparisons showed that Se concentrations in avocets (17.55 ± 2.31 μg/g dry wt) were higher than those in all other species but did not differ among stilts (5.81 ± 0.88 μg/g dry wt), Caspian terns (6.73 ± 0.97 μg/g dry wt), and Forster's terns (7.55 ± 1.09 μg/g dry wt) (Fig. 1).

The next stage of analysis excluded Caspian tern data and tested whether adult Se concentrations in livers differed among variables using our expanded global model. The final model indicated significant species × region and species × sex interactions (ANOVA: species, $F_{2,133} = 3.58$, $p = 0.03$; reproductive stage, $F_{1,133} = 8.38$, $p = 0.01$; region, $F_{2,133} = 5.11$, $p = 0.01$; sex, $F_{1,133} = 0.29$, $p = 0.59$; species × region, $F_{4,133} = 15.81$, $p < 0.0001$; species × sex, $F_{2,133} = 6.68$, $p = 0.002$). Therefore, three separate ANOVAs for each species were conducted to test for differences among regions and between sexes and reproductive stages.

American avocets

Selenium concentrations in avocets differed among sampling regions and sexes but not between reproductive stages (ANOVA: reproductive stage, $F_{1,54} = 2.09$, $p = 0.15$; region, $F_{2,54} = 18.75$, $p < 0.0001$; sex, $F_{1,54} = 11.08$, $p = 0.002$) (Fig. 2). Avocets in the North Bay had higher Se concentrations (12.59 ± 1.93 μg/g dry wt) than avocets in the other two regions, but the Lower South Bay (4.91 ± 0.46 μg/g dry wt) and Central South Bay (6.55 ± 1.10 μg/g dry wt) regions did not differ (Tukey tests). Female avocets also had higher Se concentrations (9.55 ± 0.81 μg/g dry wt) than male avocets (5.73 ± 0.93 μg/g dry wt).

Black-necked stilts

Selenium concentrations in stilts differed among sampling regions but did not vary greatly between reproductive stages or sexes (ANOVA: reproductive stage, $F_{1,35} = 1.51$, $p = 0.23$; region, $F_{2,35} = 6.65$, $p = 0.01$; sex, $F_{1,35} = 1.43$, $p = 0.24$) (Fig. 2). Stilts in the Central South Bay had lower Se concentrations (3.44 ± 0.68 μg/g dry wt) than birds in either the North Bay (6.01 ± 0.89 μg/g dry wt) or Lower South Bay

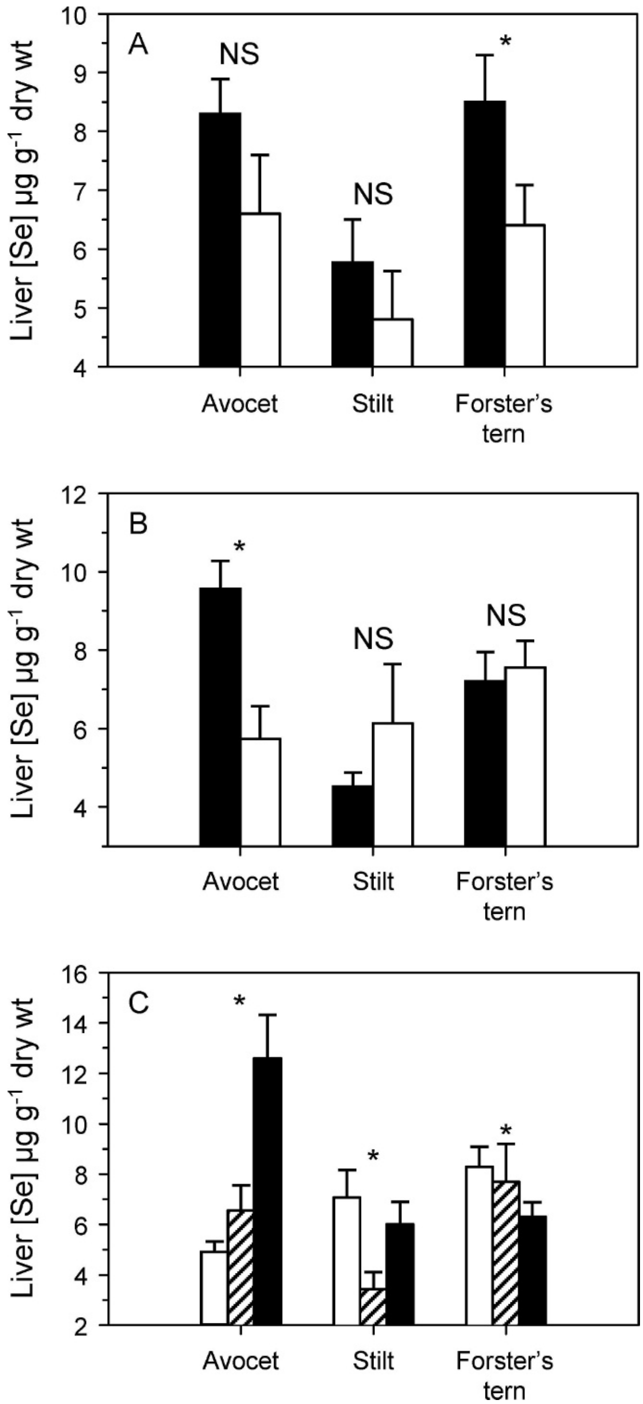

Fig. 2. Selenium concentrations (least-squares mean ± standard error; dry wt) in adult livers for (**A**) prebreeding (■) and breeding (□), (**B**) female (■) and male (□), (**C**) Lower South Bay (□), Central South Bay (▨), and North Bay (■) waterbirds in San Francisco Bay (CA, USA). An asterisk (*) indicates a statistically significant difference for each species ($p < 0.05$). NS = not significant.

(7.07 ± 1.10 µg/g dry wt), but the North Bay and Lower South Bay Se concentrations were similar (Tukey tests).

Forster's terns

Selenium concentrations in Forster's terns differed between reproductive stages and among sampling regions but not between sexes (ANOVA: reproductive stage, $F_{1,42} = 6.37$, $p = 0.02$; region, $F_{2,42} = 3.11$, $p = 0.05$; sex, $F_{1,42} = 0.21$, $p = 0.65$) (Fig. 2). Prebreeding Forster's terns had higher Se concentrations (8.50 ± 0.67 µg/g dry wt) than breeding terns captured on their nests (6.41 ± 0.57 µg/g dry wt). Forster's tern Se concentrations in the Lower South Bay (8.29 ± 0.67 µg/g dry wt) also were higher than those in the North Bay (6.30 ± 0.48 µg/g dry wt) but were no different than those in the Central South Bay (7.69 ± 1.25 µg/g dry wt; Tukey tests).

Adult body condition

The PCA indicated that structural size body measurements (wing, culmen, and tarsus lengths) were correlated as expected, and PC1 accounted for 48% (male) and 45% (female) of the morphological variation in avocet, 63% in stilt, 62% in Caspian tern, and 51% (male) and 65% (female) in Forster's tern adults. Eigenvector weights of PC1 were positive and ranged from 0.49 to 0.80 (male) and 0.40 to 0.87 (female) for avocets, 0.69 to 0.85 for stilts, 0.35 to 0.96 for Caspian terns, and 0.03 to 0.88 (male) and 0.68 to 0.90 (female) for Forster's tern adults.

For Forster's terns, adult body condition was negatively related to liver Se concentrations when controlling for other physical factors that could influence body condition (ANCOVA: liver Se, $F_{1,37} = 5.43$, $p = 0.03$; body size, $F_{1,37} = 2.64$, $p = 0.11$; reproductive stage, $F_{1,37} = 5.17$, $p = 0.03$; sex, $F_{1,37} = 1.77$, $p = 0.19$) (Fig. 3). Body condition was not related to liver Se concentrations in avocets (ANCOVA: liver Se, $F_{1,44} = 0.10$, $p = 0.75$; body size, $F_{1,44} = 9.04$, $p = 0.01$; reproductive stage, $F_{1,44} = 3.85$, $p = 0.06$; sex, $F_{1,44} = 1.97$, $p = 0.17$), stilts (ANCOVA: liver Se, $F_{1,35} = 1.95$, $p = 0.17$; body size, $F_{1,35} = 1.03$, $p = 0.32$; reproductive stage, $F_{1,35} = 18.64$, $p < 0.0001$; sex, $F_{1,35} = 0.05$, $p = 0.83$), or Caspian terns (ANCOVA: liver Se, $F_{1,3} = 0.05$, $p = 0.84$; body size, $F_{1,3} = 0.61$, $p = 0.49$; sex, $F_{1,3} = 4.66$, $p = 0.12$). A quadratic term for Se concentrations also was included in all models; however, the quadratic term was not significant for any species (all $p > 0.15$).

Chicks

Our global model indicated a significant species × region interaction (ANOVA: species, $F_{2,44} = 5.17$, $p = 0.01$; region, $F_{1,44} = 27.06$, $p < 0.0001$; species × region, $F_{2,44} = 8.74$, $p = 0.001$). We therefore conducted separate ANOVAs for each species and region.

Selenium concentrations differed among species within the Lower South Bay (ANOVA: species, $F_{2,30} = 19.75$, $p < 0.0001$) but not in the Central South Bay (ANOVA: species, $F_{2,14} = 0.48$, $p = 0.63$) (Fig. 4). Chick Se concentrations in the Lower South Bay were higher in stilts (5.44 ± 0.38 µg/g dry wt) and avocets (5.01 ± 0.33 µg/g dry wt), which did not differ, than in Forster's terns (3.17 ± 0.20 µg/g dry wt; Tukey tests). Chick Se concentrations in the Central South Bay were 2.97 ± 0.24 µg/g dry wt in stilts, 3.26 ± 0.26 µg/g dry wt in avocets, and 3.29 ± 0.29 µg/g dry wt in Forster's terns.

Selenium concentrations differed among regions for avocets (ANOVA: region, $F_{1,15} = 10.06$, $p = 0.01$) and stilts (ANOVA: region, $F_{1,14} = 28.85$, $p < 0.0001$) but not for Forster's terns (ANOVA: region, $F_{1,15} = 0.26$, $p = 0.62$) (Fig. 4). Chick Se concentrations were higher in the Lower South Bay than in the Central South Bay for both avocets (Lower South Bay, 5.01 ± 0.40 µg/g dry wt; Central South Bay, 3.25 ± 0.36 µg/g dry wt) and stilts (Lower South Bay, 5.42 ± 0.37 µg/g dry wt; Central South Bay, 2.97 ± 0.26 µg/g dry wt), but they were not different for Forster's terns (Lower South Bay, 3.16 ± 0.12 µg/g dry wt; Central South Bay, 3.29 ± 0.20 µg/g dry wt).

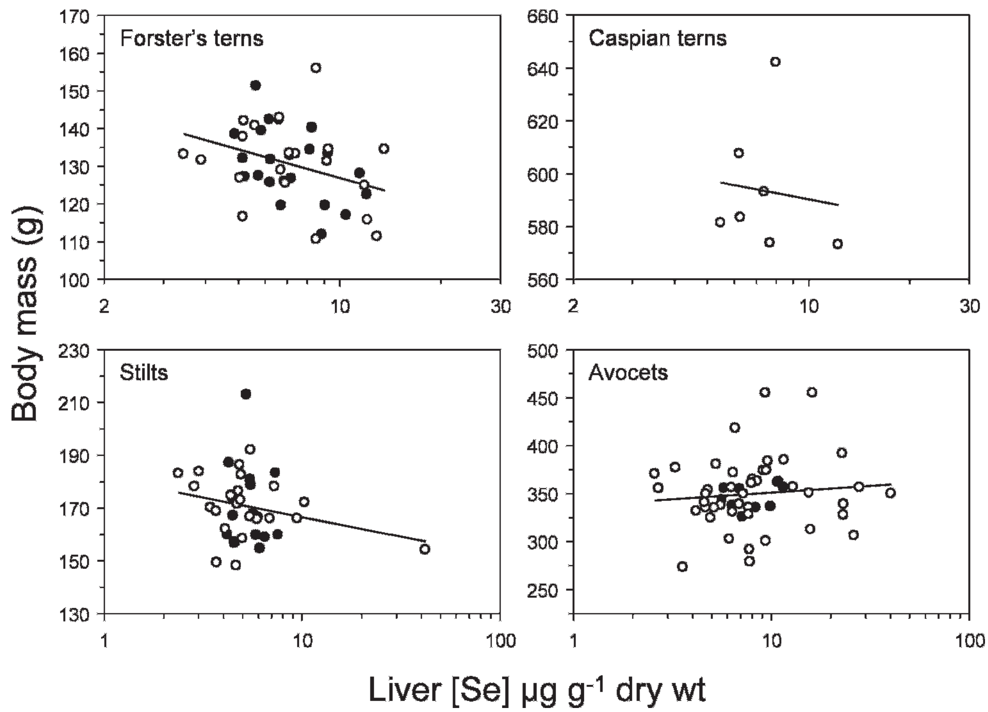

Fig. 3. Partial leverage plots depicting the relationship between adult body mass in grams and liver Se concentrations (dry wt) while accounting for the effects of structural body size, reproductive stage, and sex for four waterbird species during the prebreeding (○) and breeding (●) seasons in San Francisco Bay (CA, USA).

Chicks versus adults

We found significant species × age and species × sex interactions (ANOVA: species, $F_{2,61} = 3.50$, $p = 0.04$; age, $F_{1,61} = 25.41$, $p < 0.0001$; sex, $F_{1,61} = 0.03$, $p = 0.86$; species × age, $F_{2,61} = 19.95$, $p < 0.0001$; species × sex, $F_{2,61} = 3.90$, $p = 0.03$). Separate ANOVAs for each species therefore were conducted to remove the interaction terms. For avocets (ANOVA: age, $F_{1,17} = 0.04$, $p = 0.84$; sex, $F_{1,17} = 2.88$, $p = 0.11$) and stilts (ANOVA: age, $F_{1,19} = 1.22$, $p = 0.28$; sex, $F_{1,19} = 8.45$, $p = 0.01$), Se concentrations did not differ between ages while accounting for sex. For Forster's terns, Se concentrations were higher in breeding adults (7.39 ± 0.49 µg/g dry wt) than in chicks (3.17 ± 0.24 µg/g dry wt) sampled in the same region (ANOVA: age, $F_{1,25} = 69.88$, $p < 0.0001$; sex, $F_{1,25} = 0.32$, $p = 0.57$). Tests were not conducted for Caspian tern chicks, because they were not sampled.

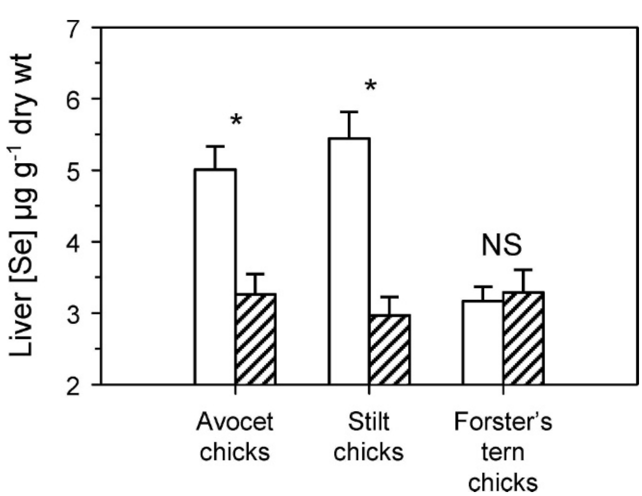

Fig. 4. Selenium concentrations (least-squares mean ± standard error; dry wt) in waterbird chick livers sampled from the Lower South Bay (□) and Central South Bay (▨) of San Francisco Bay (CA, USA). An asterisk (*) indicates a statistically significant difference for each species ($p < 0.05$). NS = not significant.

Chick body condition

The PCA indicated that morphometrics were correlated, and PC1 accounted for 79, 97, and 62% of the morphological variation in avocet, stilt, and Forster's tern chicks, respectively. Eigenvector weights of PC1 were positive and ranged from 0.86 to 0.91 for avocets, 0.96 to 0.99 for stilts, and 0.15 to 0.97 for Forster's tern chicks. Chick body condition was not influenced by liver Se concentrations in avocets (ANOVA: liver Se, $F_{1,8} = 0.42$, $p = 0.54$; body size, $F_{1,8} = 8.38$, $p = 0.02$), stilts (ANOVA: liver Se, $F_{1,13} = 0.04$, $p = 0.85$; body size, $F_{1,13} = 153.12$, $p < 0.0001$), or Forster's terns (ANOVA: liver Se, $F_{1,12} = 1.08$, $p = 0.32$; body size, $F_{1,12} = 0.72$, $p = 0.41$).

DISCUSSION

Selenium concentrations in livers of San Francisco Bay waterbirds were variable, ranged over an order of magnitude, and depended on several factors, including species, region, reproductive stage, age, and sex. Some of these effects were not consistent among species, however, and significant species × region and species × sex interactions were found. For example, avocets had higher Se concentrations in the North Bay compared to the Lower South Bay, whereas stilt Se concentrations did not differ between these regions and Forster's terns had lower Se concentrations in the North Bay compared to the Lower South Bay. Similarly, female avocets had significantly higher Se concentrations than male avocets, but this was not the case for stilts or Forster's terns. Of the factors assessed, reproductive stage had a more consistent effect among species, with prebreeding birds tending to have higher liver Se concentrations than breeding birds. Forster's terns in particular had significantly higher Se concentrations when they arrived in the San Francisco Bay (March–April) than when they nested (May–June), suggesting that Se exposure at overwintering areas along the Pacific Coast of California and Mexico may be higher than that in San Francisco Bay. Large numbers of avocets and stilts overwinter in San Francisco Bay [17], however, and their Se concentrations also tended to decline from prebreeding to breeding, indicating that Se exposure simply may be lower in spring compared to winter in San Francisco Bay. These variable results among species underscore the complexity of Se cycling in wetlands and subsequent bioaccumulation.

In general, geometric mean Se concentrations in the breeding waterbirds studied (avocets, 7.92 μg/g dry wt; Forster's terns, 7.13 μg/g dry wt; Caspian terns, 6.73 μg/g dry wt; and stilts, 5.29 μg/g dry wt) were lower than those in wintering shorebirds [25] and diving ducks [11,20,23,24] previously sampled in the San Francisco Bay Estuary. For example, Se concentrations in livers of greater scaup (19.3 μg/g dry wt) and surf scoters (34.4 μg/g dry wt) from San Francisco Bay in 1982 [20], greater scaup and canvasbacks from San Pablo Bay (scaup, 20.7 μg/g dry wt; canvasbacks, 9.02–11.9 μg/g dry wt), Central Bay (scaup, 32.7 μg/g dry wt), Grizzly Bay (canvasbacks, 5.66 μg/g dry wt), Alviso Slough (canvasbacks, 10.4 μg/g dry wt), and Alameda Flood Control Channel (scaup, 27.4 μg/g dry wt; canvasbacks, 14.2 μg/g dry wt) in 1986 [23], and greater scaup and surf scoters from Tomales Bay (scaup, 13 μg/g dry wt; scoter, 20 μg/g dry wt) and Suisun Bay (scaup, 67 μg/g dry wt; scoter, 119 μg/g dry wt) [11] in 1989 were higher than those species we studied. Liver Se concentrations in long-billed dowitchers (Hayward, 12.1 μg/g dry wt; Newark, 27.2 μg/g dry wt) and western sandpipers (Hayward, 13.4 μg/g dry wt; Newark, 12.2 μg/g dry wt) during the winter of 1991 to 1992 [25] also generally were higher than the birds examined in the present study. Lower Se concentrations in breeding shorebirds and terns in 2005 compared to the diving ducks studied in the 1980s and shorebirds in the early 1990s likely were not caused by a decrease in Se availability over time. For example, Se concentrations in surf scoter livers sampled during the same year of the present study were still at elevated levels (36.3 μg/g dry wt; S.E.W. De La Cruz, U.S. Geological Survey, Vallejo, CA, USA, unpublished data). Instead, higher Se concentrations in diving ducks compared to shorebirds and terns in San Francisco Bay likely is the result of diet and how Se cycles in the estuary. Diving ducks forage mainly on benthic bivalves that bioaccumulate Se to a higher degree than aquatic invertebrates typically consumed by shorebirds, and this difference in Se concentrations has been shown to propagate up the food webs in San Francisco Bay [29,40].

Avocet and stilt liver Se concentrations were much lower in San Francisco Bay than in California's South Grasslands region (avocets, 67.3 μg/g dry wt; stilts, 35.6 μg/g dry wt), Kesterson Reservoir (avocets, 28.4 μg/g dry wt; stilts, 46.4 μg/g dry wt), and North Grasslands region (avocets, 13.3 μg/g dry wt; stilts, 12.7 μg/g dry wt) sampled in 1984 [41]. Selenium levels at Kesterson Reservoir were shown to cause embryo deformity and mortality in several waterbird species [9]. Liver Se concentrations in avocets and stilts in San Francisco Bay were similar to Se concentrations at Volta Wildlife Area in California's Southern Central Valley (avocets, 6.38 μg/g dry wt; stilts, 7.82 μg/g dry wt), which was used as a reference site for the Kesterson Reservoir studies, and no effects of Se on bird reproduction were detected at these levels [41,42]. However, several avocets, especially prebreeding females in the North Bay, had high levels of Se contamination (maximum, 48.7 μg/g dry wt).

Reproductive impairment in mallards has been associated with liver Se concentrations exceeding 11 μg/g dry weight, and physiological impairment has been associated with levels exceeding 33 μg/g dry weight [10,26]. Although species are known to differ in their sensitivity to Se contamination [43], these levels still may provide a useful benchmark for preliminary assessments of potential risk to waterbirds. Average liver Se concentrations for San Francisco Bay waterbirds were well below these toxicity levels, but Se concentrations varied by as much as 16-fold in avocets. We found that 27% of avocets, 20% of Caspian terns, 15% of Forster's terns, and 5% of stilts had liver Se concentrations of 11 μg/g dry weight or greater, but that only 2% of shorebirds and no terns had concentrations of 33 μg/g dry weight or greater. Nonetheless, body condition was negatively related to liver Se concentrations in adult Forster's terns while explicitly accounting for other factors that could influence body condition, such as reproductive stage and sex. The relationship was weak, however, and whether this pattern actually represents cause and effect is unclear, especially because the same negative relationship was not found in the other three species.

At Kesterson Reservoir, ducks were considered to be among the most sensitive to Se, stilts to have average sensitivity to Se, and avocets to be less sensitive to Se [43]. Marine birds, such as terns, also may be relatively tolerant to Se toxicity, because marine environments typically have higher Se contamination than freshwater ecosystems and seabirds may have evolved mechanisms to deal with elevated Se exposure [1]. For example, several sea ducks appear to be unaffected by relatively high levels of Se exposure [6,44]. Thus, if Se was having adverse effects on waterbirds within San Francisco Bay, the authors expected to find effects on body condition in stilts rather than in Forster's terns.

Ohlendorf et al. [12] found that body weight was negatively related to liver Se concentrations for American coots (*Fulica americana*) but not for stilts or avocets at Kesterson Reservoir. Body weights of surf scoters wintering in San Francisco Bay were not related to liver Se concentrations [22]. Takekawa et al. [24] also found that only one of 13 body condition indices for canvasback ducks (*Aythya valisineria*) wintering in San Francisco Bay was negatively related to liver Se concentrations and that none of the body condition indices was related to liver Se concentrations in lesser or greater scaup. Body condition of lesser scaup, ring-necked ducks (*Aythya collaris*), and white-winged scoters (*Melanitta fusca*) also was not negatively related to liver Se concentrations [44,45]. Captive breeding studies have found that body weights of mallard and scaup decline when they are fed elevated dietary levels of Se in the form of selenomethionine (>15 μg/g dry wt) but that body weights return to normal quickly after the contaminated diet is removed [6,13]. Some studies have actually found positive relationships between liver Se concentrations and body condition [44,45]. Thus, there remains limited evidence for a consistent negative effect of Se on waterbird body condition.

Evaluating the deleterious effects of Se on waterbirds also is hampered by its complicated toxicology and interactions with other contaminants. Selenium can induce a hormetic effect in vertebrates, potentially enhancing egg hatchability at low doses and reducing hatchability at high doses [1]. Additionally, Se can be an important component in detoxifying mercury [46,47]. In areas such as San Francisco Bay, where numerous contaminants, especially mercury [48,49], may be impairing waterbirds, the interpretation of deleterious effects because of Se contamination likely is confounded by a combination of its low-dose benefits, its interaction with mercury, and the potentially toxic effects of several other contaminants.

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