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Environmental Contamination and Toxicology

Environmental Contaminants in Eggs of California Least Terns (*Sterna antillarum browni*)

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A severe decline in the coastal breeding population of California least terns (Sterna antillarum browni) in California and Baja California (U.S. Fish Wildl. Serv. 1980) prompted both State and Federal governments to designate it an endangered species in 1970 (Massey 1974). Significant losses of nesting and feeding habitat have contributed greatly to the decline of this subspecies (Massey 1974; Atwood and Minsky 1983). However, environmental contaminants, such as organochlorine compounds and metals, may also have contributed to the decline. California least terns are primarily piscivorous during the nesting period (Massey 1974), feeding predominantly on jack-smelt (Atherinops californiensis), topsmelt (A. affinis), and northern anchovy (Engraulis mordax) (Atwood and Minsky 1983). Topsmelt had the highest levels of DDE (p,p'-DDE) (up to 3 μg/g wet wt) of fish collected from San Diego Bay by Ohlendorf et al. (1985). Eggs of Caspian terns (S. caspia) from that study contained up to 56 μg/g DDE, and DDE was associated with a reduction in eggshell thickness as determined by the thickness index. In addition to shell deficiencies, organochlorines can also cause reduced egg production, aberrant incubation behavior, delayed ovulation, embryotoxicosis, and mortality of chicks and adults (Blus 1982). Mercury (Hg) and selenium (Se) have caused decreased hatchability, altered nesting behavior, and embryotoxicosis in birds in field and laboratory studies (Connors et al. 1975; Ohlendorf et al. 1986; Heinz et al. 1987). Our objective was to evaluate the role of contaminants in the decline of California least terns.

MATERIALS AND METHODS

Unhatched least tern eggs were collected after the nesting season from colonies in San Francisco and San Diego bays, California, during 1981-1987 (Fig. 1). Eggs were obtained from colonies at the Alameda Naval Air Station (NAS) (19 eggs) and the Oakland Airport (13 eggs) in San Francisco Bay and from five colonies in San Diego

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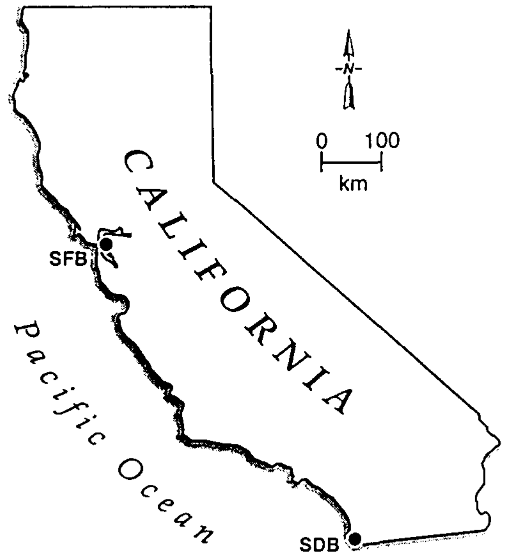

Figure 1. Locations of least tern nesting colonies sampled in California during 1981-1987: SFB - San Francisco Bay; SDB - San Diego Bay.

Bay: Lindberg Field (3 eggs), North Island NAS (20 eggs), Naval Training Center (8 eggs), and D Street/Chula Vista (8 eggs). Three eggs collected at FAA Island in nearby Mission Bay were considered part of San Diego Bay.

Individual eggs were wrapped in aluminum foil and frozen until they could be processed. Each egg was measured and its volume estimated based on its measurements. Each egg was weighed, and the embryo was examined to determine fertility and approximate stage of development. Egg contents were then frozen in chemically clean jars until they were analyzed for contaminants. Eggs from some sites were combined and analyzed as composite samples to increase sample volume, reducing the total number of samples from 74 to 35. Some samples were still insufficient for both organochlorine and metal analyses. Therefore, not all samples analyzed for organochlorines were analyzed for Se or Hg. Thus, 26 samples were analyzed for Hg, 29 for Se, and 31 for organochlorines.

Samples analyzed for Hg and Se were homogenized and freeze-dried for moisture determination. Samples were analyzed for Hg using cold vapor atomic absorption spectrophotometry, with a lower limit of detection (LOD) of 0.01 μg/g (dry wt), and for Se using hydride generation, with an LOD of 0.05 μg/g (dry wt). The accuracy of these results, as measured by spike recovery and reference material analysis, and the precision, as measured by duplicate sample analysis, were acceptable for both analytes. Unless specified otherwise, all Hg and Se residues are expressed on a dry-weight basis to avoid errors in interpretation associated with varying moisture levels in eggs (Adrian and Stevens 1979) and to facilitate comparisons with other recent field studies.

Eggs analyzed for hexachlorobenzene, α-, β-, γ-, and δ-BHC, heptachlor epoxide, oxychlordane, α- and γ-chlordane, trans-nonachlor, cis-nonachlor, endrin, toxaphene, o,p'-DDE, p,p'-DDE, o,p'-DDD, p,p'-DDD, o,p'-DDT, p,p'-DDT, mirex, dieldrin, and total PCBs were individually homogenized, thoroughly mixed with anhydrous sodium sulfate, and Soxhlet-extracted with hexane. The extract was concentrated to dryness for lipid determination. The weighed lipid sample was dissolved in petroleum ether and extracted four times with acetonitrile saturated with petroleum ether. Residues were then partitioned into petroleum ether and fractionated on a Florisil chromatographic column with diethyl and petroleum ethers. A silicic acid chromatographic column was used to separate total PCBs from other organochlorines in one fraction. Fractions were concentrated, and organochlorine pesticides and metabolites and total PCBs were quantified by packed or megabore column, electron capture gas chromatography. Residues in a portion of the samples were confirmed by gas chromatography-mass spectrometry. The precision, as measured by duplicate sample analyses, and the accuracy, as measured by spike recovery, were acceptable for all analytes. The LOD was 0.01 μg/g (wet wt) for organochlorine pesticides and metabolites and 0.05 μg/g (wet wt) for toxaphene and total PCBs. All wet weights were adjusted to fresh wet weights (Stickel et al. 1973), and all residues for organochlorines are reported on that basis.

Means were calculated after transforming concentrations to common logarithms to improve homogeneity of variances. Means were calculated when a contaminant was detected in $\geq$ 50% of the samples, with a value equal to 50% of the LOD assigned to any not-detected values prior to logarithmic transformation.

Eggs are useful indicators of contaminant exposure (Fox and Weseloh 1987), but to reduce bias, eggs should be fresh and collected at random. During our study, eggs were collected because they were abandoned during incubation, died during pipping, or were infertile or addled. In addition, criteria for selection of these eggs were neither temporally nor spatially uniform. Eggs that might have failed as a result of environmental contamination were mixed with those failing for other reasons. Consequently, because of potential bias associated with the collection methods, differences were not compared statistically.

RESULTS AND DISCUSSION

Residues of Hg and Se were detected in all samples, but means and extremes of both elements were higher in eggs from San Francisco Bay (Table 1). Within San Francisco Bay, the mean Hg concentration in eggs from the Oakland Airport seemed to be higher than those from the Alameda NAS, but Se was similar at the two sites (Table 2). Within San Diego Bay, mean Hg concentrations ranged from a low of 0.829 μg/g in eggs from the Naval Training Center to 1.39 μg/g in eggs from D Street/Chula Vista. The lowest concentrations of both Hg (0.56 μg/g) and Se (1.6 μg/g) were found in eggs from North Island (Table 2). Mean Se concentrations within San Diego Bay were relatively uniform.

Table 1. Geometric mean concentrations of mercury (Hg) and selenium (Se) (μg/g dry wt) and organochlorines (μg/g fresh wet wt) in eggs of least terns from San Francisco Bay and San Diego Bay, California, 1981-1987.

	San Francisco Bay			San Diego Bay		
Contaminant	N[1]	Mean	Min/Max	N	Mean	Min/Max
Hg	11	1.88	1.3-3.2	15	1.07	0.56-2.8
Se	12	2.67	2.5-3.1	17	2.41	1.6-2.9
Oxychlordane	13	0.013	ND[2]-0.039	18	0.014	ND-0.092
trans-Nonachlor	13	0.148	0.094-0.32	18	0.097	0.031-0.21
Total PCBs	13	3.66	2.1-5.2	18	1.22	0.71-3.1
p,p'-DDE	13	1.02	0.55-1.9	18	0.936	0.031-1.7
Dieldrin	13	0.095	0.053-0.19	18	0.011	ND-0.038
cis-Nonachlor	13	0.022	ND-0.17	18	NC[3]	ND-0.10

[1]N = sample size; [2]ND = not detected, below the LOD; [3]NC = not calculated, <50% of samples with detected analyte.

The mean Hg concentration in unhatched California least tern eggs collected from Seal Beach National Wildlife Refuge (NWR)(about 145 km north of San Diego Bay) in 1991-1993 was 0.82 μg/g dry wt (M. Rivera, U.S. Fish and Wildlife Service, pers. commun.), similar to the mean we observed in San Diego Bay. Mean Hg concentrations (wet wt) in eggs of Forster's terns (S. forsteri) (0.40 μg/g), Caspian terns (0.20 μg/g), and least terns (0.34 μg/g) from the Texas Gulf Coast were not related to hatching success (King et al. 1991). Reproductive success of common terns (S. hirundo) from Lake Ontario was not reduced at 1.0 μg Hg/g wet wt (Connors et al. 1975), and hatching and fledging success were not reduced in common tern eggs from Germany with 6.7 μg Hg/g wet wt (Becker et al. 1993). Using an average moisture content of 67%, our wet wt values (SFB: 0.629 μg/g and SDB: 0.357 μg/g) are slightly higher than those for terns from Texas but are lower than those in terns from Lake Ontario and Germany. Mercury probably did not impair reproduction of the California least terns in this study.

Mean Se concentrations reported for least, Caspian, and Forster's terns from the Texas Coast were considered to be at background levels (≤ 1 μg/g wet wt) (King et al. 1991). With the exception of one egg from San Francisco Bay (3.1 μg/g dry wt), Se concentrations in all eggs from our study were within the "no effect" range (0.4-0.8 μg/g wet wt or 1-3 μg/g dry wt) suggested by Ohlendorf (1989).

Of the organochlorines listed in Table 1, PCBs, p,p'-DDE, and trans-nonachlor were detected in all samples. Cis-nonachlor was the least common, detected in only 42%

Table 2. Geometric mean concentrations (sample size) and extreme values of mercury (Hg) and selenium (Se) (μg/g dry wt) and organochlorines (μg/g fresh wet wt) in least tern eggs collected from nesting colonies in San Diego Bay and San Francisco Bay, California, 1981-1987.

	San Diego Bay					San Francisco Bay	
Contaminant	NINAS	NTC	LF	D/ChV	FAA	ANAS	OAP
Hg	0.913	0.829	NC[1]	1.39	NC	1.75	2.05
	(8)	(2)	(1)	(3)	(1)	(6)	(5)
	0.56-2.8	0.74-0.92	1.1	0.78-2.3	2.8	1.3-2.4	1.6-3.2
Se	2.21	2.90	NC	2.66	NC	2.64	2.71
	(10)	(2)	(1)	(3)	(1)	(7)	(5)
	1.6-2.9	2.9-2.9	2.6	2.5-2.8	2.8	2.5-2.9	2.5-3.1
Total PCBs	1.459	0.984	1.66	NC	0.776	3.69	3.61
	(9)	(4)	(2)	(1)	(2)	(8)	(5)
	0.95-3.1	0.74-1.4	1.6-1.7	0.77	0.71-0.85	2.1-5.2	2.6-4.7
p,p'-DDE	1.10	0.712	0.0691	NC	0.174	1.04	1.03
	(9)	(4)	(2)	(1)	(2)	(8)	(5)
	0.74-1.7	0.54-1.1	0.056-0.086	0.031	0.16-0.18	0.73-1.9	0.55-1.5
Dieldrin	0.011	0.009	0.008	NC	0.036	0.091	0.103
	(9)	(4)	(2)	(1)	(2)	(8)	(5)
	0.006-0.033	ND-0.038	0.007-0.008	0.008	0.035-0.036	0.053-0.17	0.065-0.19

[1]San Diego Bay: North Island Naval Air Station (NINAS), Naval Training Center (NTC), Lindberg Field (LF), D Street/Chula Vista (D/ChV), and FAA Island (FAA); San Francisco Bay: Alameda Naval Air Station (ANAS) and Oakland Airport (OAP); [2]NC = not calculated, analyte detected in <50% of samples; [3]ND = not detected, below the LOD.

of the analyzed samples. Although cis-nonachlor, trans-nonachlor, and oxychlordane were frequently detected, they were excluded from Table 2 because of their low concentrations. Other organochlorines included in the analyses (See Methods) were detected infrequently and were not included in the tables.

In general, means and extremes of detected organochlorines were similar within (Table 2) and between the bays (Table 1). Means of PCBs in eggs from San Francisco Bay, however, were three times higher than those in eggs from San Diego Bay (Table 1). Ohlendorf et al. (1988) observed this pattern for Caspian tern eggs from San Francisco Bay in 1982 (4.85 μg/g) and San Diego Bay in 1981 (1.70 μg/g). Forster's tern eggs from San Francisco Bay had higher levels of PCBs (5.65 μg/g) in 1982 than did eggs of elegant terns (S. elegans) from San Diego Bay (1.55 μg/g) in 1981 (Ohlendorf et al. 1988). These differences between the bays may be related to local contamination instead of contaminant acquisition on the wintering grounds or along the migratory route (Gilbertson 1974; Custer et al. 1983, 1985) because California least terns forage mostly within 3.2 km of their nest sites during the incubation and chick-feeding stages (Atwood and Minsky 1983; Massey et al. 1992).

PCB concentrations in eggs of California least terns were within the range found in Caspian tern eggs from Texas (1.0-5.4 μg/g) (King et al. 1991), but were generally higher than those in least tern eggs from South Carolina (0.25 - 1.9 μg/g) (Blus and Prouty 1979). Although Caspian tern eggs from the Great Lakes contained mean concentrations of PCBs ranging from 18.5 to 39.3 μg/g, no adverse reproductive effects were noted (Struger and Weseloh 1985). The mean for total PCBs in least terns eggs from Seal Beach NWR was 1.11 μg/g (M. Rivera, U.S. Fish and Wildlife Service, pers. commun.), which was most similar to the mean observed in San Diego Bay.

Mean DDE concentrations seemed similar in the two bays, with similar trends over time. DDE appeared to decline at North Island between 1982-1985 (1.324 μg/g, n = 5) and 1986-1987 (0.864 μg/g, n = 4). There seemed to be a similar decline for DDE in San Francisco Bay from 1981-1985 (1.20 μg/g, n = 7) to 1986-1987 (0.849 μg/g, n = 6). By contrast, least tern eggs from Seal Beach NWR had higher mean concentrations of DDE in both 1991 and 1993, with an apparent increase from 1.96 to 5.19 μg/g over the two years (M. Rivera, U.S. Fish and Wildlife Service, pers. commun.).

The percentage of eggs with detectable cis-nonachlor also appeared to decline with time. In San Diego Bay, detection of cis-nonachlor decreased from 60% of the samples during 1982-1985 to 15% during 1986-1987. In San Francisco Bay, cis-nonachlor was detected in 100% of the eggs in 1981-1985 but in only 17% in 1986-1987.

Both the mean DDE concentration (9.99 μg/g) and the extremes (4.5-23. μg/g) in 16 California least tern eggs collected from Venice Beach (about 45 km north of Seal Beach) during 1981-1985 (Boardman 1988) were higher than those observed in our study. Higher mean concentrations than we found have also been reported for eggs of other tern species from both San Diego Bay (Caspian tern: 9.30 μg/g; elegant tern: 3.79 μg/g) and San Francisco Bay (Caspian tern: 6.93 μg/g; Forster's tern: 1.92 μg/g) (Ohlendorf et al. 1988). However, Blus and Prouty (1979) found concentrations in least terns (0.19-1.22 μg/g) from South Carolina that were not thought to pose any threat to reproduction. Similar values have also been reported not to adversely affect reproductive success in common and Forster's terns (Custer et al. 1983; King et al. 1991).

Few comparable data are available for other organochlorines detected in least terns, but comparisons with concentrations in snowy egrets (Egretta thula), black-crowned night-herons (Nycticorax nycticorax), Caspian terns, and Forster's terns from San Francisco Bay in 1982 (Ohlendorf et al. 1988) indicate that concentrations in these least tern eggs were similar to, or lower than, those not found to impair reproduction.

Contaminants may be among several interactive effects contributing to the recent decline of the California least tern. In conjunction with the decline of available nesting and feeding habitats, elevated levels of contaminants may be detrimental to population growth and stability. In this study, the only contaminant that seemed to be elevated was total PCBs in San Francisco Bay. However, more information is needed to determine isolated and combined effects of contaminant concentrations, for both the short term and long term, in the endangered California least tern population.

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