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Organochlorines and selenium in fishes and colonial waterbirds from the Salton Sea

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INTRODUCTION

Colonial waterbirds nesting at the Salton Sea, California, have been on the decline since 1986 (USFWS 1991, unpubl.; N. Hogg 1992, unpubl.). Species including great blue heron (Ardea herodias), cattle egret (Bubulcus ibis), snowy egret (Egretta thula), and great egret (Casmerodius albus) have had declines in the number of active nests, while double-crested cormorant (Phalacrocorax auritus) and white pelican (Pelecanus occidentalis) no longer regularly nest at the sea. Other colonial waterbirds, such as the black skimmer (Rynchops niger) and gull-billed tern

(Sterna nilotica) also nest at the sea. While the number of active nests of these species has not declined, there has been low reproductive success reported for the past three years (USFWS 1992, 1994 unpubl.).

In 1986, the U.S. Department of the Interior's Irrigation Drainwater Program initiated a number of investigations at the Salton Sea. These investigations were preempted by concerns of potential adverse biological effects of irrigation drainwater contaminants. The purpose of these investigations were to determine if contaminants in irrigation drainage from U.S. Department of the Interior-sponsored irrigation projects have caused or have the potential to cause substantial harmful effects to humans, fish, or wildlife (Setmire et al. 1990). These investigations and earlier studies documented elevated selenium and organochlorine pesticides in fish and waterbirds in the closed basin of the Salton Sea (Platter 1976, unpubl.; Ohlendorf and Miller 1984; Setmire et al. 1990; Ohlendorf and Marois 1990; Rasmussen and Blethrow 1990; Saiki 1990; Setmire et al. 1993). The bioaccumulation of selenium and the DDT metabolite p,p'-DDE was found in greatest concentrations in piscivorous birds. This data raised concerns regarding the recent increase in endangered California brown pelicans utilizing the sea as a post breeding dispersal area and feeding area during El Niño events.

DDT metabolites remain elevated in fish throughout the Salton Sea drainage (Rasmussen and Blethrow 1990, SWRCB 1992). Other organochlorines and newer generation pesticides including chlorpyrifos, dacathal, HCH, HCB, and ethyl parathion have been found in fish tissue at the highest levels in the state (Rasmussen and Blethrow 1990) in the Salton Sea area. During the mid 1980s, total DDT levels exceeded FDA action levels in fish collected from the Alamo River near Calipatria, California (Rasmussen and Blethrow 1990). Setmire et al. (1990) reported sediment concentrations of DDE throughout the Salton Sea drainage near levels found more than 10 years previous. Organochlorines are known to cause adverse effects, including impaired reproduction or mortality in colonial waterbirds with piscivorous diets (Ohlendorf et al. 1978, 1981; Custer et al. 1983; Henny et al. 1984). These effects are associated with the DDT metabolite p,p'-DDE, often causing eggshell thinning or egg breakage (Anderson and Hickey 1972). Crushed, dented, and thin shelled eggs have presently been reported at the Salton Sea in black-crowned nightheron (Nycticorax nycticorax), snowy egret, and great egret (USFWS 1991, 1992 unpubl.; N. Hogg, pers comm.).

Selenium can cause impaired reproduction in birds by embryo mortality, developmental abnormalities, or adult mortality (Ohlendorf et al. 1986a, 1986b, 1988). Selenium levels in Salton Sea fish continue to be the highest in state of California biomonitoring programs (Rasmussen and Blethrow 1990). These high levels led to the California Department of Health Services issuing a health advisory for fish consumption in the Salton Sea. Current and proposed water practices in the Salton Sea area may bring about continued high or increased levels of biologically available selenium through evapoconcentration of selenium associated with drainwater.

As part of the continuing U.S. Department of the Interior Drainwater Investigation, a monitoring program has been proposed to determine current and future effects of selenium and DDE to fish and wildlife (commonly referred to as Phase IV). Reproductive and nesting success studies of colonial waterbirds were proposed as part of later phases of the Salton Sea Drainwater Investigation. Due to declining numbers, nesting attempts, and poor reproductive success, selection of species for monitoring efforts was considered limited. In 1991, the USFWS initiated an effort to document all nesting colonies in the Salton Sea drainage, collect baseline contaminant data on food items and as many colonial waterbird species as possible, in preparation for a large scale biomonitoring program of the sea. The goal of the investigation was to select one or two species of colonial waterbird for use in a long term monitoring program meeting the following requirements - 1) widely distributed throughout the Salton Sea, 2) evidence of drainwater contaminant impacts, and 3) general life history of species well documented. This paper presents the results of this initial investigation.

STUDY AREA AND METHODS

The Salton Sea is located in Imperial and Riverside Counties in the southeastern desert area of California. It was created in 1905, when uncontrolled flooding diverted the Colorado River from its former course into the Gulf of California to the closed basin that is now known as the Salton Sea. The Salton Sea presently acts as a sump, receiving water through a system of controlled drains that are influenced by irrigation practices in the Imperial and Coachella Valleys. Water enters these valleys from the Colorado River via the All American Canal. A more detailed description of the site can be found in Setmire et al. (1990) and Setmire et al. (1993).

Colonial nesting bird surveys were conducted by refuge personnel between April 2 and August 24, 1991 to identify all nesting colonies of waterbirds. Data regarding historical and recent nesting colonies, including Platter (1976, unpubl.), Ohlendorf and Marois (1990), N. Hogg (pers. comm.), R. McKernan (pers. comm.), USFWS (unpubl. reports), California Department of Fish and Game (pers. comm.) were investigated. Surveys were conducted by ground and aerial surveys. The entire Salton Sea shoreline (approx. 177 km) and associated wetlands were surveyed. Colonies identified by aerial surveys were ground-truthed by Salton Sea NWR and/or Carlsbad Field Office staff to verify species and approximate number of nesting birds (by species) in the colony. Access to the colonies varied and was completed by one or several methods including foot, vehicle, kayak, or small boat with outboard motor. Most nests were on snags, dikes or islands surrounded by 0.1 to 2.0 meters of water.

In general, freshly laid eggs were collected from nests randomly during the nesting season from colonies throughout the Sea. The colonies occur typically at the north and south ends of the sea and when possible, eggs from an individual species would be collected from both north and south locations to determine if differences existed between locations. Due to the nearly unsuccessful nesting of gull-billed terns, only addled eggs were collected. All colonies were assigned names based on nearby waterways or roads. Some species such as black-crowned nightheron and snowy egret had at least two clutches a year. Attempts were made to collect only first clutch eggs for all species.

Eggs were collected, placed in an egg carton for safe transport, and placed on ice until returned to the Salton Sea National Wildlife Refuge for processing. Eggs were then refrigerated (< 2 d) before they were weighed and measured. Egg contents was then put into 2 oz. chemically cleaned jars and frozen in preparation for shipment to the analytical laboratory. Data presented has not been adjusted for volume or moisture content.

Eggshells were thoroughly cleaned then stored for > 3 months at room temperature at the Carlsbad Field Office to allow complete drying before eggshell thickness measurements were taken. Shell thickness was measured with a modified Starrett micrometer to the nearest 0.01 mm. Four measurements were made at the equator of the egg to include the shell and shell membrane. A mean shell thickness for each egg was calculated from these measurements.

In general, birds were collected by shotgun with steel shot, occasionally a rifle was used. Upon collection, birds were put on ice and soon after dissected at the Salton Sea National Wildlife Refuge Headquarters. Livers were removed and were placed in a 4 or 8 oz. chemically cleaned jar and frozen for later selenium analysis. Stomach contents were removed and identification of all items possible was made by NWR and/or Carlsbad Field Office staff. Carcasses were prepared as described in King et al. (1987), Boellstorff (1985), and White et al. (1985), then wrapped in aluminum foil, placed in plastic bag, then frozen and stored for later organochlorine pesticide analysis.

Fish were collected from two general habitats - the Salton Sea and major drains entering the sea. Within these general habitats, species of fish were collected from north and south locations of the sea. Bairdiella (Bairdiella icistia) were collected from the sea with the use of experimental size gill nets. Experimental gill nets were used to collect various size of bairdiella eaten by numerous colonial waterbirds at the sea. Sailfin mollies (Poecilia latipinna) and mosquitofish (Gambusia affinis) were collected with the use of baited minnow traps and/or 6 ft seine net from major drains at the north and south ends of the sea. Locations selected were near major nesting colonies or observed feeding areas of waterbirds.

Whole fish samples were collected for chemical analysis. Bairdiella were individually weighed and measured, wrapped in aluminum foil, placed in a plastic bag, then frozen until shipped to the analytical laboratory. Sailfin mollies and mosquitofish were placed in 4 oz chemically clean jars as composites of >10 g for each location and frozen. All fish samples would be analyzed for selenium, organochlorine pesticides, and PCBs.

All samples were chemically analyzed within the quality assurance and control guidelines established by the U.S. Fish and Wildlife Service's Patuxent Analytical Control Facility (USFWS 1990). Selenium analysis was conducted by Environmental Trace Substance Research Center and organochlorine and PCBs analysis was conducted by Hazleton Laboratories America, Inc. Larger samples, such as whole fish and bird carcasses, were first run through a meat grinder. All samples were thereafter homogenized with Spec Industries Inc. Model 800 mixer/mill.

For selenium analysis, a nitric-perchloric digestion was used with approximately 0.5 g of sample homogenate. Selenium determinations were made with the Perkin-Elmer Model 603 or 3030 atomic absorption spectrophotometer with a mounted Varion VGA-76 hydride generation accessory. Standardization was checked every 8-15 samples and approximately 10% of the samples were checked by method of additions to monitor matrix effects. The lower limit of selenium was .03 ug/g wet weight. Duplicate analysis was within 0-17.72%. Spike recoveries ranged from 80.48-118.38%.

Up to 20 g of sample were homogenized and analyzed for organochlorines (including metabolites) and PCBs by gas-liquid chromatography. Homogenates were blended with anhydrous sodium sulfate and extracted for 16 to 20 hrs with methylene chloride. Lipid cleanup of extracts was by gel permeation chromatography. The lower limit of detection for organochlorines was 0.01 ug/g and 0.1 ug/g wet weight for PCBs. No PCBs were detected in any samples. The presence of p,p'-DDE was confirmed in 13% of the samples by gas chromatography-mass spectrometry. Seven percent of the p,p'-DDE samples had duplicate samples run and were within 0-25.53% and spike recoveries ranged from 100-126.33%.

Statistical Analysis. Several data sets were used in the analysis for this study, carcass, liver, eggs, and fish. Due to low sample size and data which were not normally distributed nonparametric statistics were used in all analyses. The p value for rejection of HO was set at $P<.05$ for all analysis, unless otherwise stated. All statistical tests were run using STATGRAPHICS v7.0.

The carcass and liver data consisted of North and South data for Double-crested cormorant, great blue heron, black-crowned night heron and white pelican. Carcass data consisted of analysis for DDE and liver data consisted of analysis for selenium. Kruskal-Wallis was used to test the difference between north and south data within a species for both carcass and liver data. The difference between species was also tested using the Kruskal-Wallis for both carcass and liver data.

Egg data consisted of eggshell thickness measurements, DDE concentration and selenium concentration. A total of seven different bird species eggs were collected including black skimmer, cattle egret, snowy egret, great blue heron, great egret, gull-billed tern and black crowned night heron. The difference between north and south DDE and selenium concentrations in great egrets was tested using the Kruskal-Wallis. Black skimmer data included eggs from three different areas north, south, and Bombay Beach, the difference in DDE and selenium concentrations between these three sites was tested using the Kruskal-Wallis. Gull-billed tern eggs came from Bombay Beach and south. Selenium and DDE concentrations were tested between the two sites using the Kruskal-Wallis. A regression was performed in order to determine the relationship between DDE and eggshell thickness in black skimmer and great egret.

Fish data consisted of analysis on Bairdiella (whole body), Sailfin molly (composite), mosquito fish (composite). Within each of the three species the DDE and selenium concentrations were tested between north and south using the Kruskal-Wallis.

RESULTS AND DISCUSSION

Nesting surveys. Sixteen locations (colonies) were identified as currently or historically having at least one species of nesting colonial waterbird (see Table 1). In 1991, 13 of these colonies were active with a total of 1132 nests. Only 5 of these sites had >10 active nests present. The two largest and most diverse colonies were found at the north end of the sea near the Whitewater River delta. These colonies had 5 and 6 species nesting in each colony. These were the only colonies at the north end of the sea. The 11 active colonies at the south end typically contained <3 species per colony and <120 nests per colony. The Morton Bay colony had 3 species, including one pair of Forster's tern (Sterna forsteri).

Species found at both the north and south ends included great blue heron, great egret, and black skimmer. Gull-billed tern was found nesting only at the south end. Snowy egret, cattle egret, and black-crowned nightheron nested only at the north end. Double-crested cormorant and white pelican were present in both areas, but no nesting attempts were documented.

Cattle egret, snowy egret, and black-crowned nightheron, great egret, and great blue heron were observed nesting at Finney Lake just south of the Salton Sea. This large colony is found in a freshwater impoundment system managed by California Department of Fish and Game and was outside the study area.

The species that are most widely distributed are great blue heron, great egret, and black skimmer. The great blue heron was located at 9 distinct colonies averaging 6.8 birds/colony. Great egrets and black skimmers were found at fewer colonies in greater numbers. Great egrets were at 4 colonies averaging 92.8 birds per colony and skimmers were found at 3 colonies averaging 36.7 birds per colony. With larger numbers at fewer colonies comparisons could be made between colonies. Based on the need to monitor reproductive performance as it relates to contaminant impacts in a statistically valid manner, the great egret and black skimmer are the best candidates for future monitoring.

The Finney Lake population might be considered as part of the Salton Sea colonial waterbird population, but it is unknown if these birds frequently feed at the sea or if they primarily feed in canals and drains throughout Imperial Valley. The colony includes the only nesting cattle egrets, snowy egrets, and black-crowned nightheron in the southern area of the Salton Sea drainage. However, these species are known to double clutch and the nesting initiation is somewhat unpredictable, making these species undesirable for long-term monitoring.

Egg contaminant data and eggshell thickness. Contaminant concentrations of p,p'-DDE and selenium and eggshell thickness measurements are reported in Table 2. Eggs from seven species of birds were analyzed. Waterbirds with more non-piscivorous diets (cattle egret and gull-billed tern) had the lowest DDE concentrations and egrets (great and snowy) had the highest levels. Great egret, snowy egret, and black skimmer all had egg DDE concentrations >10 ppm (wet wt). Black-crowned nightheron had relatively low DDE levels, unlike that found in 1985 as reported in Ohlendorf and Marois (1990). In 1985, black-crowned nightheron DDE concentrations were approximately higher than those associated with reduced reproductive success (Ohlendorf and Marois 1990). The 1985 samples were collected at a colony near the New River which was not active in 1991. The 1991 collection was made at the north end of Salton Sea. This data suggests differences in DDE concentrations of black-crowned nightherons may exist within the Salton Sea. The 1991 great egret egg data compared to 1985 (Ohlendorf and Marois 1990) are lower, but within the same relative range for DDE concentrations. There was no significant difference between concentrations measured at the north and south end colonies. Both sample years are considered elevated and most likely represent levels associated with some level of reduced reproductive success.

No relationship between eggshell thickness and DDE concentrations was observed in great egrets. However, eggshell thickness was negatively correlated with DDE in the black skimmer eggs ($r=-0.58$, $N=12$, $p<0.001$).

Egg Selenium. There was no significant difference between selenium concentrations of eggs collected from the north and south end colonies for great egrets and gull-billed terns. However, black skimmers were significantly different between the south end and the north and Bombay Beach colonies ($p=0.025$).

Liver and carcass data. Selenium concentrations in waterbird livers and p,p'-DDE concentrations in carcasses are reported in Table 3. Two nesting and two non-nesting species were analyzed.

There were no significant differences between north and south end collected birds of any species for liver (selenium) or carcass (p,p'-DDE) samples. Selenium levels tended to be higher in birds feeding on bairdiella (Salton Sea) and DDE levels were higher in birds feeding on mollies and mosquitofish (drains).

All 4 species had selenium concentrations above accepted background levels (Skorupa and Ohlendorf 1991). Double-crested cormorant, white pelican, and black-crowned nightheron had mean selenium values in livers between 10 and 30 ppm (dry wt) which Skorupa et al. (1996) recommends conclusive interpretation is not possible without associated studies of reproductive performance. All selenium values for the non-nesting cormorant and pelican were >10 ppm, representing the highest values found in the sampling area. Cormorant and great blue heron mean selenium levels are comparable to mean concentrations for these species from the Salton Sea in 1986-87 (Setmire et al. 1990). While the data sets are extremely small, this may suggest that contaminants alone are not responsible for continuing decline in Salton Sea colonial waterbird populations. All species sampled represent good indicators to monitor selenium contamination.

All 4 species collected had elevated levels of p,p'-DDE, including values >10 ppm indicating a substantial exposure. The lower mean concentration found in great blue heron were lower than levels associated with mortality or reproductive problems in Ardeids (Blus et al. 1980). The great blue heron is not considered a sensitive species to DDE (Fitzner et al. 1988). Henny (1972) found great blue herons had no serious problems associated with DDE during the height of DDT use. The elevated levels found in black-crowned nightheron support previous data for the species at Salton Sea reported by Ohlendorf and Marois (1990) which found DDE at concentrations higher than those associated with reduced reproductive success of nightherons (Custer et al. 1983, Henny et al. 1984). Cormorant DDE concentrations were higher than previous reported carcass data for the species collected from the polluted Houston Ship Channel (King et al. 1987). However, the mean DDE residue in cormorant and pelican carcasses was well below levels associated with chronic poisoning and reproductive problems in fish eating birds (Blus 1982).

Long term monitoring will focus on reproductive performance that eliminates consideration of cormorants and pelicans. However, the elevated concentrations of selenium and DDE in these species may relate to the lack of nesting attempts at the sea. This aspect of reproductive failure needs to be evaluated in some other effort such as investigating estrogen levels to determine the influence of feminization on the cormorant and pelican population. Based on liver and carcass data as well as previous data, black-crowned nightheron would serve as an excellent indicator for future monitoring efforts.

<u>Fish data</u>. Selenium and p,p'-DDE concentrations in fish collected and analyzed from the Salton Sea area are reported in Table 4. No significant differences were found between north and south ends of the Sea for selenium or DDE in any of the fish species. Bairdiella had the highest mean selenium concentration of the three species sampled, while mean values for sailfin mollies and mosquitofish were comparable. The species collected from the drains had higher DDE

concentrations than bairdiella collected from the Salton Sea. This trend has also been reported in Setmire et al. (1990, 1993).

Bairdiella selenium concentrations were similar to those reported for the Salton Sea in Saiki (1990) and Setmire et al. (1993). Individual bairdiella samples were found to be above the 12 ppm (dry wt) reproductive threshold for sensitive fish reported by Lemly and Smith (1987). However, the toxic threshold for selenium in tissues of saltwater fish such as bairdiella, are unknown (White et al. 1987). Matsui (1989) has documented population declines and deformities of larval stage bairdiella at the Salton Sea. Such malformations have been reported following exposure to a variety of contaminants including pesticides and metals.

High selenium concentrations in freshwater fish from San Felipe Creek, Trifolium Drain, Alamo and New Rivers, were correlated with elevated selenium in water samples (Setmire et al. 1990). This may explain the variability in selenium and possibly DDE in mosquitofish and mollies. All freshwater fish samples collected were below reproductive threshold reported by Lemly and Smith (1987). DDE concentrations in all fish were below guidelines reported in (NAS-NAE 1973).

While selenium and DDE concentrations may not be reflective of adverse effects to fish, they serve as a contaminated pathway to colonial waterbirds. Higher concentrations of DDE are found throughout the trophic system of the canals and drains, while higher selenium concentrations are found throughout trophic systems more closely associated with the Salton Sea.

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Table 1. Summary of 1991 active colonial waterbird nests at Salton Sea.
(GTBH - great blue heron, CAEG - cattle egret, SNEG - snowy egret, GREG - great egret, DCCO - double crested cormorant, BCNH - black crowned night heron, GBTE - gull-billed tern, BLSK - black skimmer)

	GTBH	CAEG	SNEG	GREG	DCCO	BCNH	GBTE	BLSK	TOTAL
Trifolium Drain	0	0	0	0	0	0	0	0	0
Bruchard Bay	0	0	0	0	0	0	0	0	0
New River Delta	3	0	0	0	0	0	0	0	3
Vail Ranch	0	0	0	0	0	0	0	0	0
Lindsey/ Lack	2	0	0	36	0	0	0	0	38
Obsidian Butte	1	0	0	0	0	0	0	0	1
Red Hill/ HQ	5	0	0	0	0	0	0	0	5
Hazard Lakes	0	0	0	0	0	0	0	0	0
Johnson Drain	25	60	60	80	0	100	0	40	365
Bombay Beach	7	0	0	0	0	0	0	0	7
W. Whitewater	10	65	75	250	0	100	0	0	500
Barth Road	0	0	0	0	0	0	50	30	80
W. Poe Road	0	0	0	1	0	0	0	0	1
Morton Bay	0	0	0	0	0	0	80	40	120
Alamo River Delta	4	0	0	0	0	0	0	0	4
Wister WMA Shoreline	4	0	0	4	0	0	0	0	8
Salton Sea Total	61	125	135	371	0	200	130	110	1132

Table 2. Selenium (ppm, dry wt.) and p,p'-DDE concentrations (ppm, wet wt.) found in waterbird eggs at Salton Sea, 1991.

Species	N	Selenium Geometric mean (range)	p-p'-DDE geometric mean (range)
black skimmer	12	4.65 (2.2-8.2)	4.9 (1.8-16.4)
great-blue heron	4	3.86 (2.8-5)	5.78 (2.6-10)
black-crowned night heron	3	5.27 (4.6-6.5)	2.34 (1.7-3.6)
cattle egret	3	3.6 (2.7-5.4)	2.81 (1.6-4.8)
snowy egret	3	3.93 (3.9-4)	15.7 (5-31)
great egret	9	4.77 (3.5-7.1)	8.36 (0.86-31)
gull-billed tern	6	4.10 (3.4-5.3)	1.32 (0.54-2.8)

Table 3. Selenium concentrations (ppm, dry wt.) found in waterbird livers and p,p'-DDE concentrations (ppm, wet wt.) found in waterbird carcasses at Salton Sea, 1991.

Species	N	Selenium Geometric mean (range)	p-p'-DDE geometric mean (range)
double-crested cormorant	6	21.96 (17-29)	2.47 (0.49-11)
great-blue heron	10	9.57 (3.5-17)	1.89 (0.28-25)
black-crowned night heron	4	12.24 (4.8-20)	14.02 (7.8-33)
white pelican	6	14.79 (11-22)	5.43 (1.3-35)

Table 4. Selenium (ppm, dry wt.) and p,p'-DDE concentrations in fish collected from Salton Sea, 1991.

Species	N	Selenium Geometric mean (range)	p-p'-DDE geometric mean (range)
Bairdiella	10	11.12 (8.2-24)	0.08 (0.03-0.39)
sailfin molly	8	4.98 (2.7-6.1)	0.16 (0.08-0.51)
mosquito fish	8	4.87 (2.2-11)	0.24 (0.09-0.96)