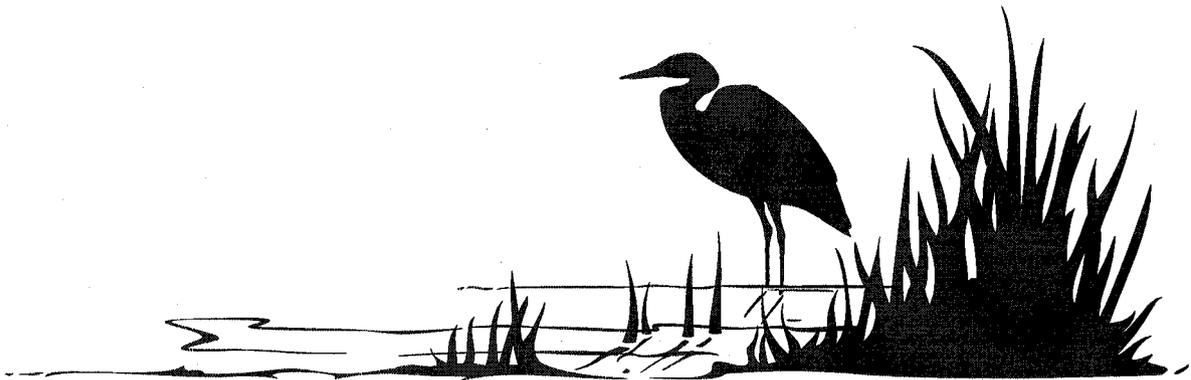U.S. Fish and Wildlife Service
Region 2
Contaminants Program

CONTAMINANTS IN WATERBIRDS, GRACKLES, AND SWALLOWS NESTING ON THE LOWER COLORADO RIVER, ARIZONA, 2000 - 2001

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ABSTRACT

Levels and potential effects of organochlorine compounds and metals were assessed in 106 eggs representing nine avian species nesting at four lower Colorado River National Wildlife Refuges.   Geometric mean DDE residues were highest in the eggs of white-faced ibis *(Plegadis chihi)* (2.34 µg/g wet weight) and double-crested cormorant *(Phalacrocorax auritus)* (0.75 µg/g wet weight).   White-faced ibises fledged < 1.0 young per pair and reproductive success was limited by DDE-induced eggshell thinning and by potentially embryotoxic (>4.0 µg/g) DDE residues detected in 48% of the eggs.   Mean shell thickness of 23 ibis eggs was 0.264 mm, 15% thinner than shells of eggs collected before the widespread use of DDT ($P <0.0001$).   DDE was negatively correlated ($r = -0.51$, $P <0.0001$) with eggshell thickness.   DDT, the parent compound, was detected in 9 of 23 ibis eggs.

DDE in 4 of 22 cormorant eggs (18%) approached or exceeded the lowest observed adverse effect level. Eggshell thickness averaged 0.412 mm, 4.2% thinner than pre-DDT museum eggs (0.43 mm).   DDE residues in cormorant eggs were not correlated ($P = 0.3628$) with shell thickness.   Almost one-half (6/13) the cormorant eggs collected in 2000 were addled. Overall cormorant reproductive success was <1.0 young fledged per pair.

Low DDE residues (≤ 0.70 µg/g wet weight) were detected in the eggs of Clark's grebe *(Aechmophorus clarkii)*, least bittern *(Ixobrychus exilis)*, and great-tailed grackle *(Quiscalus mexicanus)*.   A single great blue heron *(Ardea herodias)* egg contained 4.10 µg/g DDE, a level above the threshold where embryotoxic effects might be expected.

Mercury was present at background levels in all fish-eating bird eggs and in all but one ibis egg.   In the eggs of omnivorous bird species, American coots *(Fulica americana)* and great-tailed grackles, mercury was detected in $\leq 50\%$ of the eggs.   Eggs and nestlings of the primarily insectivorous cliff swallow *(Petrochelidon pyrrhonota)* did not contain detectable mercury concentrations.   Mercury does not appear to be a contaminant of concern for birds nesting along the lower Colorado River.

Of 106 eggs collected from nine species, more than one-third (39/106) contained potentially embryotoxic (> 6.0 µg/g) concentrations of selenium.   The frequency of occurrence of elevated levels of selenium was greatest in eggs of western *(Aechmophorus occidentalis)* (87%) and Clark's grebe (76%).   Generally, eggs of fish-eating birds contained the highest concentrations of selenium, followed by eggs of omnivorous species.   None of the eggs of the insectivorous cliff swallow contained selenium above the 6.0 µg/g toxic threshold.   Selenium concentrations were higher in swallow nestlings than in eggs suggesting that selenium bioaccumulation occurred during the first 10-days of life.

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Numerous studies documented elevated environmental contaminant concentrations in clams, crayfish, and fish collected from the lower Colorado River (Radtke et al. 1988, Schmitt and Brumbaugh 1990, King et al. 1993, Lusk 1993, Ruiz 1994, Villegas 1997, Prieto 1998). However, the effects of contamination on top-of-the-food-chain predators such as fish-eating birds has only been marginally investigated. Chemical analysis of the few waterbird samples collected in past studies indicated potentially toxic concentrations of selenium had accumulated in liver tissues and eggs. Selenium in the livers of three rail species collected in the 1980s ranged from 13.1 to 26.0 µg/g dry weight (Rusk 1991), which was well within the 10 to 30 µg/g dry weight toxic range as suggested by Ohlendorf (1993). In a later study of waterbirds nesting on the lower Colorado River's Imperial National Wildlife Refuge (NWR), 81% contained liver selenium concentrations within the 10 to 30 µg/g dry weight range associated with embryo toxicity and reproductive impairment (Martinez 1994). Selenium concentrations in western (*Aechmophorus occidentalis*) and Clark's grebe *(A. clarkii)* eggs were 2- to 4-times higher than threshold levels associated with embryotoxic effects and teratogenesis (Andrews et al. 1997). Many of the above investigations concluded that selenium concentrations were high enough to be embryotoxic and that future studies were needed to document reproductive success of waterbirds and the incidence of abnormalities in young.

The objectives of our study were to: 1) determine the extent of organochlorine and metal bioaccumulation in waterbirds collected from four refuges on the lower Colorado River, 2) assess the reproductive success of two waterbird species including hatching success, fledging success, and incidence of anomalous young, and 3) document potential adverse effects of contaminants on avian reproductive success.

STUDY AREAS

Field studies focused on four National Wildlife Refuges located on the lower Colorado River. From upstream to downstream, these included Havasu, Bill Williams River, Cibola, and Imperial NWRs. Specific study sites within each refuge were dictated by the location of nesting waterbirds. At Havasu NWR, most samples were collected from Topock Marsh with fewer samples taken from Topock Gorge. At Bill Williams River NWR, almost all waterbird nesting occurred in the cattail *(Typha sp.)* marsh located at the confluence of the Bill Williams River and the Colorado River. Waterbirds at Cibola NWR nested almost exclusively in Cibola Lake. We surveyed Imperial NWR intensively during both years of study, but we were unable to locate an active waterbird colony.

METHODS

Observation period and species selection– Field work extended throughout the 2000 and 2001 nesting seasons.   Observations of waterbird reproductive success focused on a colony of white-faced ibis *(Plegadis chihi)* nesting at Cibola Lake (Cibola NWR) and on a colony of double-crested cormorants *(Phalacrocorax auritus)* nesting in Topock Marsh (Havasu NWR).   Reproductive success of ibises and cormorants was monitored in 2000 only.   In 2001, we redirected field investigation efforts to locate nesting waterbirds at Bill Williams River and Imperial NWRs.   A small sample of cormorant eggs collected by Havasu NWR personnel in 1999 also was analyzed for organochlorine compounds and metals.

In addition to ibis and cormorant eggs, we also collected samples of western grebe, Clark's grebe, least bittern *(Ixobrychus exilis)*, American coot *(Fulica americana)*, great blue heron *(Ardea herodias)*, great-tailed grackle *(Quiscalus mexicanus)*, and cliff swallow *(Petrochelidon pyrrhonota)* eggs on an opportunistic basis.   A small sample of cliff swallow nestlings also was collected.   No attempt was made to document the reproductive success of the grebes, bitterns, herons, grackles, or swallows.

Sample collection and preparation– Ibis and cormorant eggs were collected using the "sample egg method" (Blus 1984) in which one egg was collected per nest and the contaminants in each egg correlated with the success of eggs remaining in the nest.   One egg was collected at random from 2- to 4-egg clutches.   Generally, we waited until clutches were complete before collecting eggs.   In addition to the "sample egg", we also collected any addled eggs or dead young in or below nests.   Eggs were marked with the corresponding nest number using a waterproof felt-point pen. Hatching success of the remaining eggs was monitored on a 4- to 7-day basis.   Nestlings were individually examined shortly after hatching to document the presence or absence of abnormalities.   For bird species other than ibises and cormorants, one egg was collected from each nest on an opportunistic basis whenever new nests were encountered.   Eggs were placed in commercial egg cartons and stored on wet ice in coolers until they could be transferred to a commercial freezer.   We weighed and measured eggs, cut each egg around the girth, and placed its contents in chemically-cleaned jars. Embryo development was assessed using methods described by Taber (1969) based on a 20-day incubation period for the white-faced ibis (Ryder and Manry 1994) and an average 26-day incubation period for cormorants (Hatch and Weseloh 1999).   Jars with egg contents were frozen until analysis.   Eggshells were gently washed and allowed to dry for several weeks. Thickness (shell plus shell membrane) was measured to the nearest 0.01 mm at 3 points around the equator using a Starrett Model 1010M dial micrometer, and a mean thickness was calculated for each egg.   Current eggshell thickness was compared with that of museum eggs collected before the widespread agricultural use of DDT (pre-1947).

One 10-day-old ibis nestling found dead was salvaged for residue analysis. We also collected a small sample of cliff swallow nestlings. Ibis and cliff swallow carcasses were plucked, and bill, feet, wingtips, and gastrointestinal tract were removed and discarded.   Liver tissues were removed and saved for metals analysis. The body remainder was analyzed for organochlorine compounds.

Food items– Whenever possible, regurgitated food items were collected from nestling cormorants. Because of the low number of regurgitated samples available, we also sampled fish of appropriate size seined from areas where cormorants were observed feeding. Regurgitated samples were analyzed individually, and seined fish samples were analyzed by species. No white-faced ibis regurgitated food items were found and no attempt was made to collect representative food items from agricultural fields near the colony where ibis commonly feed.

Chemical analysis– All samples were analyzed for organochlorine compounds including o,p'- and p,p'-DDE; o,p'- and p,p'-DDD; o,p'- and p,p'-DDT; dieldrin; heptachlor epoxide; hexachlorobenzene (HCB); alpha, beta, delta, and gamma BHC; alpha and gamma chlordane; oxychlordane; *trans*-nonachlor; *cis*-nonachlor; endrin; toxaphene; mirex; and total polychlorinated biphenyls (PCB) at Mississippi State Chemical Laboratory, Mississippi State, Mississippi, following methods described by Cromartie et al. (1975) and Kaiser et al. (1980). We report total chlordane which is the sum of the individual chlordane isomers including alpha and gamma chlordane, oxychlordane, *trans*-nonachlor, and *cis*-nonachlor. The lower limit of quantification was 0.01 µg/g (parts per million) for most organochlorine pesticides and 0.05 µg/g for toxaphene and PCBs.   Recovery in spiked samples ranged from 84.5 to 101%. Organochlorine compounds are expressed in µg/g wet weight to facilitate comparing residue levels with those reported in other studies.   Contaminant concentrations in eggs, food items, and bird tissues were tabulated and compared to effect-level thresholds established for each species in laboratory and field studies whenever possible.   With the exception of DDE in white-faced ibis eggs, organochlorine residues in eggs of all species collected in 2000 were below known adverse affect thresholds; therefore, eggs collected in 2001 were not analyzed for organochlorines.

Samples also were analyzed for aluminum, arsenic, barium, beryllium, boron, cadmium, chromium, copper, lead, mercury, magnesium, manganese, molybdenum, nickel, selenium, strontium, vanadium, and zinc at Research Triangle Institute, Research Triangle Park, North Carolina.   Arsenic and selenium concentrations were determined by graphite furnace atomic absorption spectrophotometry (USEPA 1984).   Mercury was quantified by cold vapor atomic absorption (USEPA 1984).   All other elements were analyzed by inductively coupled plasma atomic emission spectroscopy (Dahlquist and Knoll 1978, USEPA 1987).   Blanks, duplicates, and spiked samples were used to maintain laboratory quality assurance and quality control (QA/QC).   QA/QC was monitored by the Service's Patuxent Analytical Control Facility (PACF).   Analytical methodology and reports met or exceeded Service QA/QC standards. Trace element concentrations are reported in µg/g (parts per million) dry weight to facilitate

comparison of results with those of other studies and to avoid errors in interpretation associated with varying moisture levels in eggs and tissues (Stickel et al. 1973). The lower limits of analytical quantification varied by element and by sample and are listed in each respective appendix. Percent moisture also is presented to permit wet weight to dry weight conversions. Dry weight values can be converted to wet weight equivalents by subtracting the percent moisture (as a decimal) from 1.0 and multiplying the resulting number by the dry weight value.

We recognize that not all the elements listed in this report are "heavy metals" or even true metals. But for the sake of convenience, and to avoid often ambiguous terms such as "trace elements, metalloids, and heavy metals," we refer to all elements simply as metals.

Statistical analysis– Organochlorine and metal analytical data were log-transformed to common logarithms to improve homogeneity of variances, and geometric means were calculated when residues were detected in 50% or more of the samples. When means were calculated, a value equal to one-half the lower limit of detection was assigned to any non-detected value prior to log-transformation. Data were then analyzed using analysis of variance to determine statistical differences in residue levels among species. Retransformed geometric means are presented in the text and tables. Eggshell thickness means are arithmetic. Other comparisons of data were made with Student's *t*-test, correlation analyses, and Chi-square tests. A statistical level of $P \leq 0.05$ was considered significant.

RESULTS

A colony of white-faced ibises was located at Cibola NWR in 2000 and double-crested cormorants nested at Havasu NWR in both 2000 and 2001. We also found scattered nests of western and Clark's grebes, least bitterns, American coots, and one great blue heron. In addition to these waterbird species, we also located nesting great-tailed grackles and cliff swallows. The number and location of samples collected is given in Table 1.

White-faced ibis: When the Cibola Lake ibis colony was first discovered on June 21, 2000, only eight active nests with eggs were located, and the colony appeared to be in the egg laying and early incubation period. No nestlings were observed during our first visit to the colony, and we estimated the adult population to be about 50 pairs. Near the end of the incubation period, the total nesting population was estimated by visual counts and aerial photography at approximately 75 pairs (Zaun et al. 2003).

Nineteen nests were marked of which 18 were monitored through hatching. Assuming an average incubation period of 20 days (Ryder and Manry 1994), egg laying in the first nest occurred about June 15 and laying was initiated in the last nest about June 29. Eggs were collected from the date of the first visit until July 20, at which time the incubation period was mostly completed. Twenty-three eggs were collected, 16 from marked nests and seven eggs

found abandoned.   Some of the abandoned eggs were from marked nests, some were not. Since 13 of 30 eggs hatched, some of those that did not hatch were considered abandoned, others were simply lost (fate unknown).   Embryos in 13 of 16 eggs from marked nests were less than one-half developed.   Clutch size in completed nests ranged from 2 to 3 eggs and average clutch size in all 19 nests was 2.5 (Table 2).   Thirteen eggs hatched in 18 nests monitored through hatching for an average of 43% (Table 3).   The fate of the nineteenth marked nest was unknown.   No abnormal nestlings were observed.   Nestling ibis often leave the nest and hide when approached by investigators which makes assessments of fledging success difficult.   However, it can be assumed that fledging success, as defined by the number of young fledged per nesting pair of adults, was <1 since only 13 eggs hatched in 18 nests.

Table 1.   Number and type of samples collected at National Wildlife Refuges on the lower Colorado River, Arizona, 2000 - 2001

| | | Year, sample type, and number collected | | | | | |
| | | 2 0 0 0 | | | 2 0 0 1 | | |
Refuge	Species	Eggs	Whole body	Regurgi-tate	Eggs	Whole Body	Regurgi-tate
Havasu	Double-crested cormorant[1]	22	0	4	0	0	0
	Clark's grebe	6	0	0	1	0	0
	Western grebe	0	1	0	8	0	0
	Least bittern	0	0	0	1	0	0
	American coot	0	0	0	2	0	0
	Great-tailed grackle	0	0	0	10	0	0
	Cliff swallow	0	0	0	4	3	0
	Largemouth bass	0	3	0	0	0	0
	Red shiner[2]	0	2	0	0	0	0
	Threadfin shad[3]	0	1	0	0	0	0
Bill Williams River	Clark's grebe	0	0	0	10	0	0
	Western grebe	0	1	0	8	0	0
Cibola	White-faced ibis	23	1	0	0	0	0
	Least bittern	2	0	0	0	0	0
	Great blue heron	1	0	0	0	0	0
	Great-tailed grackle	6	0	0	10	0	0
Imperial	No samples collected	0	0	0	0	0	0

[1]Nine of 22 cormorant eggs were collected by a cooperator in 1999.

[2]Two composite samples of 30 individuals each.

[3]One composite sample of 28 individuals.

Table 2. White-faced ibis clutch size at Cibola Lake, Cibola National Wildlife Refuge, Arizona, 2000.

	Eggs per nest			Total nests	Total eggs	Mean clutch size
	1	2	3			
No. of nests with eggs	2	5	12	19	48	2.5

Table 3. White-faced ibis nest history and hatching success, Cibola Lake, Cibola National Wildlife Refuge, Arizona, 2000.

Total nests marked	Total eggs laid	# eggs collected for analysis	Fate unknown nests (eggs)		Remaining nests (eggs)		# nests that hatched 1 or more eggs	# eggs hatched	Hatching success eggs hatched ÷ eggs laid
19	48	16	1	(2)	18	(30)	8	13	13 ÷ 30 = 0.43

Organochlorine residues– Residues of seven organochlorine insecticides and PCBs were detected in ibis eggs (Appendix 1). DDE was recovered in all eggs. The geometric mean DDE residue was 2.34 µg/g wet weight (Table 4) and residues in individual eggs ranged from 0.13 to 14 µg/g wet weight. There was no difference ($P = 0.8527$) in geometric mean DDE residues in eggs collected from marked nests (2.14 µg/g) and eggs found abandoned (2.46 µg/g). None of the remaining eggs hatched in nests where the sample egg contained >4.0 µg/g DDE, i.e., the "adverse effect zone" for DDE (Henny and Herron 1989). The geometric mean total chlordane residue in eggs from marked nests (0.01 µg/g) was identical to that in abandoned eggs. DDT was detected in 9 of 23 (39%) eggs. DDT residues generally were correlated with eggs that contained higher DDE residues ($r = 0.60$, $P = 0.0090$); DDT was recovered in only 2 of 12 (17%) eggs that contained <4.0 µg/g DDE, but DDT was present in 7 of 11 (64%) eggs with >4.0 µg/g DDE. Low levels of PCBs, DDD, dieldrin, heptachlor epoxide, and HCB were present in 5 or fewer eggs. The geometric mean DDE residue in ibis eggs was statistically similar ($P > 0.05$) to that of double-crested cormorants (Table 4), but higher than residues detected in the eggs of great-tailed grackles and Clark's grebes. DDE was the only organochlorine compound recovered in the carcass of the dead nestling (0.26 µg/g wet weight).

Eggshell thickness– Two of 16 eggs collected from marked nests were extremely thin-shelled, with a flexible shell that was easily indented with slight finger pressure. These two thin-shelled eggs measured 26% and 29% thinner than normal (pre-DDT). One of 7 abandoned eggs contained an embryo that appeared to have died as a result of the eggshell collapsing during pipping. The sample egg collected from the same nest 16 days earlier was 1 of the 2 visibly thin-shelled eggs. We did not, however, observe any other collapsed or

cracked eggs in or below nests. Mean shell thickness of eggs collected from marked nests (0.264 mm) was identical to thickness of shells of abandoned eggs. Therefore, for additional statistical tests, we combined the egg data from marked nests with data from eggs found abandoned. Overall mean shell thickness of all 23 eggs was 0.264 mm, which was 15% thinner (P <0.0001) than shells of eggs collected before the widespread use of DDT (Table 5). Individual eggshell thinning varied from 1 to 29%. Both log-DDE (r = -0.51, P <0.0001) and log-chlordane (r = -0.2028, P = 0.0310) were negatively correlated with eggshell thickness. Chlordane is not a primary shell thinning agent; therefore, the correlation between total chlordane and shell thinning may have been spurious because eggs with elevated residues of DDE also contained high residues of total chlordane (r = 0.24, P = 0.0172). Because fewer than one-half of the eggs contained residues of other organochlorine compounds, we did not attempt to assess the statistical relationship of these compounds with shell thickness.

Table 4. Geometric mean DDE and PCB residues (µg/g wet weight) in waterbird and great-tailed grackle eggs collected from National Wildlife Refuges on the Colorado River, 1999 - 2000

Species	Year	Refuge	Number of eggs	Geometric mean (n)[1] range	
				DDE	PCB
White-faced ibis	2000	Cibola	23	2.34 (23) 0.13 - 14.0 A[2]	----- (5) ND[3] - 0.18
Double-crested cormorant[4]	1999-00	Havasu	22	0.75 (22) 0.05 - 4.30 AB	0.07 (11) ND - 0.47
Great-tailed grackle	2000	Cibola	6	0.33 (6) 0.13 - 0.70 BC	------ (1) ND - 0.15
Clark's grebe	2000	Havasu	6	0.11 (6) 0.07 - 0.28 C	------ (0)
Least bittern[5]	2000	Cibola	2	----- (2) 0.30 - 0.30	------ (0)
Great blue heron[5]	2000	Cibola	1	----- (1) 4.10	------ (1) 0.24

[1](n) = number of samples with detectible residues.
[2]Means sharing the same letter are statistically similar (P > 0.05).
[3]ND = Not detected.
[4]Nine eggs collected in 1999 and 13 eggs collected in 2000.
[5]Least bittern and great blue heron data were not included in ANOVA calculations due to sample size <3.

Table 5. Comparison of shell thickness measurements of white-faced ibis eggs collected at Cibola Lake, Arizona, 2000, with those from museum collections (pre-DDT)

Year	Number of eggs	Shell thickness (mm)		Percent difference
		mean ± SE	range	
Before 1945[a]	18	0.312 ± 0.006	0.26 - 0.37	-----
2000	23	0.264[b] ± 0.006	0.22 - 0.31	-15

[a]Data from King et al. (1980).
[b]Mean eggshell thickness was significantly different between time periods ($P < 0.0001$, t-test).

Metals– Twelve metals were detected in ibis eggs (Appendix 2). Of the elements present in eggs, mercury and selenium were the most likely to be embryotoxic. Mercury and selenium were recovered in all eggs (Table 6). The geometric mean mercury concentration in eggs collected from marked nests (0.42 µg/g dry weight) was similar ($P = 0.7830$) to that in abandoned eggs (0.38 µg/g). Concentrations of mercury in individual eggs ranged up to 2.72 µg/g. The geometric mean selenium concentration in ibis eggs from marked nests (3.17 µg/g) was similar ($P = 0.3033$) to selenium in abandoned eggs (3.38 µg/g). Selenium in individual eggs ranged from 2.44 to 4.16 µg/g. Both mercury and selenium were detected in the liver of the dead nestling at 0.11 and 4.17 µg/g dry weight, respectively.

Double-crested cormorant: A double-crested cormorant colony was located in emergent dead tree stumps in Topock Marsh (Havasu NWR) southwest of the town of Golden Shores. The cormorant population consisted of 12 pairs in 2000 and 7 pairs in 2001. Reproductive success of cormorants was assessed from April 4 to June 6, 2000. Because of the small number of nesting pairs in 2001, and the disturbance associated with intensive nest monitoring, we did not attempt to assess reproductive success in 2001. Since the colony was relatively small, we were able to monitor all active nests. Six of 12 nests contained eggs and recently hatched young when first observed indicating that the timing of our first visit was close to the hatching date of the first eggs. Average clutch size was 3.0 eggs per nest (Table 7). Hatching success, as defined as the number of eggs hatched divided by the number of eggs laid, was 0.78 (Table 8). The last egg hatched about May 10. No abnormal cormorant nestlings were observed. Fourteen eggs hatched (Table 8) and 11 young survived at least 24 days. Assuming these 11 nestlings fledged, the fledging rate would be 0.78 young fledged per pair of adults.

Table 6. Mercury and selenium concentrations in waterbird, great-tailed grackle, and cliff swallow eggs collected from National Wildlife Refuges on the lower Colorado River, 1999 - 2001

Species	Refuge	Year	Number of eggs	Geometric mean, ppm dry weight, (n)[1] range	
				Mercury	Selenium
Double-crested cormorant	Havasu	1999-00	22	0.41 (22) 0.18 - 1.00 A[2]	4.86 (22) 1.78 - 8.29 AB[2]
White-faced ibis	Cibola	2000	23	0.39 (23) 0.16 - 2.72 AB	3.23 (23) 2.44 - 4.16 C
Western grebe	BWR[3]	2001	8	0.17 (8) 0.12 - 0.28 B	6.61 (8) 4.77 - 8.33 AD
Clark's grebe	Havasu	2000	6	0.05 (6) 0.04 - 0.09 C	5.57 (6) 4.62 - 6.55 ABD
Clark's grebe	BWR[4]	2001	11	0.18 (11) 0.13 - 0.31 B	7.34 (11) 6.20 - 8.40 D
Least bittern	Havasu	2000-01	3	0.37 (3) 0.29 - 0.42 AB	6.05 (3) 5.34 - 7.14 ABD
Great blue heron[5]	Cibola	2000	1	----- (1) 1.33	----- (1) 5.08
American coot[5]	Havasu	2001	2	----- (1) ND[6] - 0.15	----- (2) 6.70 - 7.55
Great-tailed grackle	Cibola	2000-01	26	0.05 (13) ND - 0.46 C	5.36 (26) 3.42 - 10.7 A
Cliff swallow	Havasu	2001	4	----- (0)	3.43 (4) 2.97 - 3.70 BC

[1](n) = number of samples with detectible residues.

[2]Means sharing the same letter are statistically similar. Mercury and selenium concentrations were significantly different among species ($P < 0.0001$).

[3] BWR = Bill Williams River NWR.

[4]In 2001, 10 eggs were collected from Bill Williams River NWR and one egg from Havasu NWR.

[5]Great blue heron and American coot data were not included in ANOVA calculations due to small sample size.

[6]ND = Not detected.

Table 7. Double-crested cormorant clutch size, Topock Marsh, Havasu National Wildlife Refuge, Arizona, 2000.

	Eggs per nest				Total nests	Total eggs	Clutch size
	2	3	4	5			
No. nests with eggs	2	6	3	1	12	36	3.0

Table 8. Double-crested cormorant nest history and hatching success, Topock Marsh, Havasu National Wildlife Refuge, Arizona, 2000.

Total nests marked	Total eggs laid	# eggs collected for analysis	Fate unknown		Remaining		# nests that hatched 1 or more eggs	# eggs hatched	Hatching success eggs hatched ÷ eggs laid
			nests	(eggs)	nests	(eggs)			
12	36	13	2	5	10	18	10	14	14 ÷ 18 = 0.78

A high proportion (33%) of the eggs collected during our first visit to the colony had no embryo development and were addled. Overall, almost one-half (6/13) of the cormorant eggs collected in 2000 were addled. In contrast, none of the 9 eggs collected by the refuge biologist in 1999 were addled.

Organochlorine residues– DDE and PCB were the only organochlorine compounds detected (Table 4, Appendix 3). DDE was present in all eggs and the geometric mean residue was 0.75 µg/g wet weight (Table 4). The geometric mean DDE residue in cormorant eggs was similar to that of white-faced ibises and great-tailed grackles, but higher than residues detected in the eggs of Clark's grebes.

PCBs were recovered at low levels (≤ 0.47 µg/g wet weight) in 11 of 22 eggs. The geometric mean PCB residue for eggs collected in 1999 (0.06 µg/g wet weight) was similar ($P = 0.6290$) to that in eggs collected in 2000 (0.08 µg/g); therefore, the data were combined for additional statistical tests. The geometric mean PCB residue for eggs collected during both years was 0.07 µg/g. No other organochlorine compounds were detected in the cormorant eggs.

Eggshell thickness– Thickness of 22 cormorant eggshells averaged 0.412 mm which was 4.2% thinner than the mean shell thickness of eggs (Anderson and Hickey 1972) collected before the widespread use of DDT (Table 9). Log-DDE was not correlated with shell thickness ($P = 0.3628$). In 2000, the year reproductive success was monitored, eggs hatched in 10 of 12 marked nests. We had hoped to correlate contaminant residues with hatching success, but our sample size from failed nests was too small to facilitate a valid statistical comparison.

Table 9. Comparison of shell thickness measurements of double-crested cormorant eggs collected from Topock Marsh in 1999 - 2000 with museum eggs collected before 1947 (pre-DDT)

| Area | Year | Number of eggs | Shell thickness (mm) | | Percent difference |
			Mean ± SD	Range	
Interior North America[a]	Before 1947	350	0.430 ± 0.003	NA	-----
Topock Marsh	1999 - 2000	22	0.412[b] ± 0.026	0.393 - 0.440	4.2

[a]Data from Anderson and Hickey (1972).
[b]Mean eggshell thickness was not significantly different between years ($P = 0.4717$, t-test).

Metal concentrations– Thirteen elements were detected in cormorant eggs (Appendix 4). Mercury was recovered in all eggs and the geometric mean concentration was 0.41 µg/g dry weight (Table 6). Mercury concentrations in individual eggs ranged from 0.18 to 1.00 µg/g. Selenium also was present in all eggs (geometric mean = 4.86 µg/g, range = 1.78 - 8.29 µg/g).

Nestling cormorants occasionally regurgitated when approached by investigators. We collected regurgitated food samples from four nestlings (Table 1). In addition, we collected potential cormorant food items (fish) by seine in areas where cormorants were seen feeding. Seined samples included three fingerling largemouth bass *(Micropterus salmoides)*, two composite samples of red shiner *(Cyprinella lutrensis)*, and a single composite sample of threadfin shad *(Dorosoma petenense)*. No organochlorine compounds were detected in regurgitated or seined fish samples. Mercury (geometric mean 0.06 µg/g dry weight) was recovered in all four regurgitated samples, but mercury was not present in the fish samples seined from Topock Marsh (Table 10, Appendix 5). All regurgitated and seined samples contained selenium. The geometric mean selenium concentration was higher in the seined largemouth bass than in the regurgitated samples ($P = 0.0056$). However, the composite sample of threadfin shad contained the highest single concentration of selenium (7.93 µg/g dry weight).

Western grebe: Western grebes were observed at all refuges during both years of the study; however, we were able to locate nests only in 2001. Eight eggs were collected from Bill Williams River NWR between July 11 and July 25, 2001. Young grebes leave the nest shortly after hatching negating the possibility of reproductive studies. Western grebe eggs were not analyzed for organochlorine compounds.

Mercury and selenium– Concentrations of 12 metals were recovered in western grebe eggs (Appendix 6). Mercury was detected in all eggs and the geometric mean concentration was 0.17 µg/g dry weight (range = 0.12 - 0.28) (Table 6). Selenium also was present in all eggs (geometric mean = 6.61 µg/g, range = 4.77 - 8.33 µg/g).

Table 10. Geometric mean mercury and selenium concentrations, µg/g dry weight, in double-crested cormorant regurgitate samples and in fish collected from Topock Marsh, Havasu NWR

| Sample | N[1] | Geometric mean (n)[2] and range | |
		mercury	selenium[3]
Regurgitate	4	0.06 (4) 0.04 - 0.10	2.32 (4) 1.88 - 2.93
Largemouth bass	3	----- (0)	4.97 (4) 3.89 - 5.78
Red shiner[4]	2	----- (0)	----- (2) 3.91 - 4.99
Threadfin shad[5]	1	----- (0)	----- (1) 7.93

[1]N = number of samples analyzed.
[2](n) = number of samples with detectible residues.
[3]Selenium concentrations in regurgitate samples were significantly ($P = 0.0056$) lower than those in largemouth bass.
[4]Two composite samples of 30 individuals each.
[5]One composite sample of 28 individuals.

Two adult western grebes found dead by refuge personnel apparently died of starvation as evidenced by the emaciated condition of the carcass.   One specimen was recovered from Bill Williams River NWR and the other from Havasu NWR.   Both carcasses contained low residues of DDE (0.17 and 0.08 µg/g wet weight), DDD (0.02 and 0.04 µg/g) and total PCB (0.10 and 0.07 µg/g) (Table 11).   The carcass recovered from Bill Williams River also had 0.05 µg/g wet weight *trans*-nonachlor.   No other organochlorine compounds were detected in the western grebe carcass tissues.

Relatively low levels of mercury and selenium were recovered in the western grebe liver tissue (Appendix 7).   The sample from Bill Williams River NWR contained 7.75 µg/g dry weight mercury and the liver from the Havasu grebe contained 2.31 µg/g.   These tissues also contained 4.20 and 7.38 µg/g dry weight selenium, respectively.

Clark's grebe: Eggs were collected from May 17 to May 31, 2000 (n = 6) and from June 6 to July 11, 2001 (n = 11).   No adult Clark's grebes were recovered for chemical analysis.

Organochlorine residues– DDE was the only organochlorine compound detected in Clark's grebe eggs (Table 4, Appendix 8) and was recovered in all eggs.   DDE residues in Clark's grebe eggs (geometric mean = 0.11 µg/g wet weight) were significantly ($P<0.0001$) lower than in those of double-crested cormorant and white-faced ibis, but similar to residues in grackles (Table 4).

Table 11.   Organochlorine compound concentrations in carcasses of western grebes found dead and in cliff swallow nestlings collected on Havasu and Bill Williams River National Wildlife Refuges, Arizona, 2000

| Species | Refuge | Concentration, µg/g wet weight | | | Percent moisture | Percent lipid |
		p,p'-DDD	p,p'-DDE	Total PCB		
Western grebe[1]	Bill Williams	0.02	0.17	0.10	72.4	1.31
Western grebe[2]	Havasu	0.04	0.08	0.07	73.1	1.18
Cliff swallow	Havasu	<0.01	0.04	<0.05	67.9	10.2
Cliff swallow	Havasu	<0.01	0.04	<0.05	70.7	9.31
Cliff swallow	Havasu	<0.01	0.02	<0.05	71.2	7.32

[1]Adult western grebe found dead 11/02/98.   This sample also contained 0.05 µg/g *trans*-nonachlor.   No other organochlorine compounds were detected.

[2]Adult western grebe found dead 10/26/2000.

Mercury and selenium– Concentrations of 12 metals were recovered in Clark's grebe eggs (Appendix 6).   The geometric mean mercury level in eggs collected in 2000 (0.05 µg/g dry weight) was significantly ($P < 0.0001$) lower than in eggs collected in 2001 (0.18 µg/g dry weight).   Mercury in individual eggs ranged from 0.04 to 0.31 µg/g (Table 6).   Mercury in Clark's grebe eggs collected in 2000 (0.05 µg/g) was lower than that detected in most other fish-eating birds and identical to levels in the omnivorous great-tailed grackle (0.05 µg/g) (Table 6).   The geometric mean mercury level in Clark's grebe eggs collected in 2001 was significantly lower than that in double-crested cormorant eggs, but similar to the levels reported in the eggs of most other fish-eating birds.   Selenium also was detected in all Clark's grebe eggs and the geometric mean concentration of eggs collected in 2000 (5.57 µg/g) was similar ($P > 0.05$) to the geometric mean of eggs collected in 2001 (7.34 µg/g).   Selenium in individual eggs ranged up to 8.40 µg/g.

Least bittern: Two least bittern nests were located in a small cattail island in Cibola Lake, Cibola NWR, in 2000.   One egg was taken from each nest.   We also collected one additional egg from a nest in the Blankenship Bend area of Havasu NWR in 2001.   Only the two eggs collected in 2000 were analyzed for organochlorines.   DDE was the only organochlorine compound detected.   Each egg contained 0.30 µg/g wet weight DDE (Table 4).   Traces of 12 metals were recovered in bittern eggs (Appendix 9).   We combined the 2000 and 2001 data to calculate a geometric mean and facilitate comparison of least bittern data with those of other waterbirds.   All bittern eggs contained mercury (geometric mean = 0.37, range = 0.29 - 0.42 µg/g) and selenium (geometric mean = 6.05, range = 5.34 - 7.14 µg/g).   The geometric mean mercury concentration in least bittern eggs was higher than that in Clark's grebe eggs

(collected in 2000) and mercury in great-tailed grackle eggs, but the geometric mean mercury concentration was similar to geometric means of all other species.   The geometric mean selenium concentration (6.05 μg/g dry weight) was statistically similar ($P > 0.05$) to that of all other species except the white-faced ibis (3.23 μg/g) ($P < 0.0001$).

Great blue heron: We were able to locate only one great blue heron nest on NWR lands.   A single egg was collected from a heron nest at Cibola NWR's Hart Mine Marsh.   DDE (4.10 μg/g wet weight), PCB (0.24 μg/g), and total chlordane (0.31 μg/g) were the only organochlorine compounds recovered (Table 4, Appendix 8).   Concentrations of 12 metals were detected in the heron egg including a relatively high level of mercury (1.33 μg/g dry weight) and a moderate level of selenium (5.08 μg/g) (Table 6, Appendix 9).   A colony of great blue herons that had been active on Imperial NWR in past years did not nest there during the 2000 - 2001 study period.

American coot: While coots were frequently observed at all refuges during both years of study, we were able to collect eggs only in 2001.   Eggs collected in 2001 were not analyzed for organochlorines.   Concentrations of 11 metals were detected in coot eggs (Appendix 9).   Mercury was recovered only in one egg at 0.15 μg/g dry weight and selenium was present in both eggs (6.70 - 7.55 μg/g) (Table 6).   Because only two eggs were collected, we did not statistically compare mercury and selenium concentrations in coot eggs with those in other species.

Great-tailed grackle: Great-tailed grackle eggs were collected from Cibola NWR in 2000 (n=6) and from both Cibola (n=10) and Havasu (n=10) NWRs in 2001 (Table 1).   No attempt was made to monitor reproductive success at either refuge.

Organochlorine residues– DDE and PCBs were the only organochlorine compounds recovered in grackle eggs (Table 4, Appendix 8).   The geometric mean DDE residue (0.33 μg/g wet weight) was lower than the level in white-faced ibis eggs, but statistically similar ($P > 0.05$) to residues in double-crested cormorants and Clark's grebes (Table 4).   The maximum DDE residue in grackle eggs was 0.70 μg/g.   PCBs were detected only in 1 of 6 eggs at 0.15 μg/g wet weight.

Metals– Concentrations of 12 metals were detected in grackle eggs (Appendix 10).   Mercury was present in 13 of 26 eggs and the geometric mean, 0.05 μg/g, was lower ($P < 0.0001$) than concentrations in all other species except Clark's grebe eggs collected in 2000 (Table 6).   Selenium was recovered in all grackle eggs.   The geometric mean selenium level was lower than that detected in Clark's grebe eggs collected in 2001, higher than concentrations in ibis and cliff swallows, and similar to concentrations in all remaining species (Table 6).

Cliff swallow: An accessible cliff swallow colony was located in Topock Gorge (Havasu NWR) in 2001.   Four eggs and three nestling carcasses were collected for metals analyses. None of the cliff swallow samples were analyzed for organochlorines.   Mercury was not detected in swallow eggs (Table 6, Appendix 9) or liver tissues (Appendix 7).   However, selenium was recovered in all eggs (geometric mean = 3.43 µg/g dry weight) and carcass samples (geometric mean = 6.06 µg/g dry weight).   Other metals detected in cliff swallow egg and carcass tissues are listed in Appendix 7 and 9.

DISCUSSION

White-faced ibis: White-faced ibis populations declined throughout their range in the 1950s and 1960s (Ryder 1967), probably as a result of reproductive failure due to eggshell thinning and loss.   The Great Basin ibis population may have increased since the mid-1970s, (Ryder and Manry 1994), but the Texas Coast population has continued to decline (U.S. Fish and Wildlife Service world wide web site, www.TexasCoastalProgram.fws.gov).   The discovery of the Cibola Lake ibis colony represents the first documented nesting of white-faced ibises in Arizona (Zaun et al. 2003).   Average clutch size (2.5) was lower than that reported in most other studies.   Clutch size for ibises in Utah ranged from 2.8 to 3.5 (Kaneko 1972, Capen 1977, Steele 1984).   Mean clutch sizes for ibises nesting in Nevada were 2.9 to 3.6 (Henny and Herron 1989), and 2.6 to 3.1 for ibises nesting in Texas (King et al. 1980, Custer and Mitchell 1989).

The relatively small clutch size of the Cibola Lake colony may be related to natural latitude variation or to the late timing of the nesting effort.   For species that nest over a broad geographic area, average clutch size usually is greater in populations nesting at more northern latitudes than in those nesting at southern latitudes (Lack 1954, Custer et al. 1983). Throughout most of North America, ibis egg laying and incubation extends from mid-April to late-May (Ryder and Manry 1994).   However, at more southern latitudes such as Texas and Louisiana, egg laying occurred from mid-April through early-July.   Egg laying and incubation at the Cibola Lake colony occurred during the latter portion of the time frame for other colonies located at similar southern latitudes.   Late clutches of white-faced ibises are usually smaller than clutches initiated earlier in the season (Henny and Herron 1989).

Hatching success was strongly associated with the timing of nest initiation.   Eggs hatched in six of seven nests (86%) initiated on or before June 21, but eggs hatched in only 2 of 11 nests (18%) started after June 21 (χ^2 = 5.4029, P = 0.0201).

Organochlorine compounds– DDE in ibis eggs ranged from 0.13 to 14.0 µg/g wet weight. As the concentration of DDE in ibis eggs increases to >4.0 µg/g wet weight, and especially >8.0 µg/g, productivity decreases and the incidence of cracked eggs increases (Henny 1997). In our sample, 11 of 23 eggs (48%) exceeded 4.0 µg/g DDE, and 6 of 23 eggs (26%) exceeded the 8.0 µg/g threshold.   None of the remaining eggs hatched in nests where the

sample egg contained >4.0 µg/g DDE, i.e., the "adverse effect zone" for DDE (Henny and Herron 1989). PCBs were infrequently (5/23) recovered in ibis eggs. The maximum concentration was 0.18 µg/g wet weight. In laboratory studies that assessed the reproductive effects of PCBs on five species of birds, the lowest egg concentration associated reduced reproduction was 15 µg/g wet weight as reviewed by Rice and O'Keefe (1995). In contrast to laboratory studies, assessing the effects of PCBs on wild birds is extremely difficult because PCBs usually occur in combination with one or more other potential toxicants. Reviews of several studies that assessed PCB toxicity to wild birds suggest that the lower range for affecting breeding success is between 3 and 5 µg/g wet weight (Peakall 1986). However, in a later review, (Peakall 1990) suggested that reproductive effects could be expected at egg levels higher than 40 µg/g. By any standard, egg PCB concentrations found in our study were far below any previously reported adverse effect thresholds.

Eggshell thinning– What are the biological consequences of 15% shell thinning as observed in ibises nesting in Arizona? In most waterbird species, eggshell thinning of less than 10% seldom causes egg breakage (Hickey and Anderson 1968). Egg loss usually becomes evident with decreases of 10-15% and serious breakage, usually accompanied by population decline, occurs when average thinning exceeds 15% (Hickey and Anderson 1968; Risebrough et al. 1970; Anderson and Hickey 1972). It is now generally accepted that a long-term average of 18% eggshell thinning in most waterbird species will result in a population decline (Blus 1996). In contrast to the above generalizations, average annual shell thinning of 4 to 14% in white-faced ibis eggs was associated with frequent eggshell breakage and reproductive failure (Capen 1977, King et al. 1980, Henny and Herron 1989). Based on whole egg analyses, limited data suggest that the white-faced ibis and the prairie falcon *(Falco mexicanus)* are the two species most sensitive to DDE-induced eggshell thinning (Blus 1996).

Sources of DDT/DDE– It is unlikely that ibises accumulated high DDT/DDE residues while feeding near the Cibola Lake colony. Studies conducted in that area (Cibola NWR and other lower Colorado River valley sites) documented relatively low residues of organochlorine compounds in resident fish and wildlife (Radtke et al. 1988, Schmitt et al. 1990, King et al. 2000). However, one potential source of high DDT/DDE residues is the lower Gila River, and associated irrigated farmland located approximately 180 - 200 km east of the Cibola Lake colony. The lower Gila River riparian corridor, a natural flight path for waterbirds, links the irrigated farmlands with the Colorado River. Common carp *(Cyprinus carpio)* collected in 1994 from the Gila River contained the highest concentrations of DDE in the nation associated with agriculturally-applied DDT (King et al. 1997). European starlings *(Sturnus vulgaris)* collected near the lower Gila River during a 1982 nationwide survey of 129 sites contained the highest (8.4 µg/g wet weight) DDE concentration in the United States (Bunck et al. 1987). Mallards *(Anas platyrhynchos)* taken in the same general area had the second-highest DDT residue in the nation (Cain 1981). Earth Technology Corporation (1993) concluded, "Based on TCLP analysis, fish and turtles (from the Painted Rock portion of the Gila River) could be considered a hazardous waste and would require treatment and disposal." White-faced ibises are regularly seen feeding in the lower Gila River riparian habitat and in

agricultural fields irrigated with water from the lower Gila River.   Ibises also may be accumulating high DDE on wintering grounds in Mexico (Ryder and Manry 1994), or perhaps in the Imperial Valley of California near the Salton Sea where birds also contain high DDE residues (Ohlendorf and Miller 1984, Mora et al. 1987).

Mercury and selenium– Mercury is an extremely potent embryo toxicant (USDI 1998).   Egg concentrations more closely reflect mercury from recent dietary uptake than from accumulated body burdens (USDI 1998).   Mercury concentrations in wild bird eggs of ≤0.5 µg/g wet weight (~ 2.5 µg/g dry weight) appear to have little detrimental effect on reproduction (Thompson 1996).   Egg concentrations of about 4.0 µg/g dry weight mercury or more, have been associated with decreased reproductive success in some species (Fimreite 1974, Ohlendorf 1993) and also have been associated with altered behavior patterns (Heinz 1979). Only 1 of 23 ibis eggs contained mercury at concentrations higher than the ~2.5 µg/g dry weight background threshold as defined by Thompson (1996).

Selenium is an essential trace element in animal diets, but it is toxic at concentrations only slightly above required dietary levels.   For avian species, the embryo is the life stage most sensitive to selenium poisoning (Heinz 1996).   Selenium in the eggs, rather than in the parent bird, causes developmental abnormalities and death; therefore, egg selenium levels give the most sensitive measure for evaluating hazards to birds.   Background concentrations of selenium in waterbird eggs are generally <3.0 µg/g dry weight (Skorupa and Ohlendorf 1991). From 3 to 6 µg/g dry weight in the eggs is considered a "level of concern" (USDI 1998). Concentrations in this range rarely produce discernible adverse effects but are elevated above typical background concentrations.   The toxicity threshold is >6.0 µg/g selenium in eggs (Skorupa 1998).   Selenium in 74% of the ibis eggs (17/23) exceeded the 3.0 µg/g background concentration, but none exceeded the 6.0 µg/g toxic threshold.   No abnormal ibis young were observed, but that might be expected given the maximum concentration recorded was 4.16 µg/g and the 1% effect concentration threshold for embryo teratogenesis is 14 µg/g for black-necked stilts *(Himantopus mexicanus)* one of the more selenium-sensitive species. There appeared to be little potential for adverse biological effects from either mercury or selenium on the ibis population that nested on the lower Colorado River in 2000.

The 0.11 µg/g dry weight mercury detected in the liver of the dead ibis nestling was well within the normal or background range.   In liver tissues, there is an overlap in mercury concentrations considered "background" and those considered toxic.   Background concentrations of mercury in the liver vary from <1 to 10 µg/g dry weight, but concentrations as low as 6 µg/g can be toxic to some species (Ohlendorf 1993).   The 4.17 µg/g dry weight selenium recovered in the dead nestling also was well within the normal or background range (3 - 10 µg/g dry weight) previously reported for liver tissues of birds (Ohlendorf 1993).

Double-crested cormorant: Double-crested cormorant populations declined drastically throughout United States and Canada from the 1950s to the early-1970s (Anderson and Hickey 1972, Weseloh et al. 1995).   Associated with this decline was an increase in environmental contaminant levels, particularly the insecticide DDT, its metabolite DDE, and the industrial organochlorine compound, PCB.   Cormorants were one of the first species in which population declines were associated with DDE-induced eggshell thinning (Anderson and Hickey 1972).   DDT was suspended as an agricultural insecticide in 1972, and by the 1980s, with declining contaminant levels, most cormorant populations, particularly those in the Great Lakes region, showed signs of recovery (Weseloh et al. 1995, Custer et al. 1999). Cormorant populations nesting on the lower Colorado River have never been thoroughly censused.   The Topock Marsh population has been monitored irregularly since 1968 (Aimee Haskew, pers. comm.).   Although surveys were not completed every year, refuge narrative reports indicate that nesting populations ranged from 8 pairs (1970 and 1987) to 67 pairs (1976).   During the years of our study, 2000 - 2001, we recorded only 12 and 7 nesting pairs, respectively.   There may have been a long-term decline in numbers, but additional study is needed to further assess cormorant population trends throughout the lower Colorado River.

During the 2000 nesting season, 13 double-crested cormorant eggs were collected for residue analysis and shell thickness measurements.   A high proportion (33%) of the eggs collected during our first visit to the colony were addled.   There is always a concern when working in waterbird colonies located in extremely hot climates, that investigator disturbance might result in mortality of a significant number of embryos.   The fact that one-third of the eggs collected on our first visit to the colony were addled suggests that investigator disturbance was not a factor.   Overall, almost one-half (6/13) of the eggs collected in 2000 were addled.

Average clutch size was close to normal.   Clutch size observations in northern United States and Canada suggest that the "modal clutch" is 4 (Hatch and Weseloh 1999).   Palmer (1962) states that the usual clutch is 3-4.   The modal clutch in our study was 3, and average clutch size, 3.0, was probably close to normal for this latitude.   Of 14 eggs that hatched in 12 nests, 11 nestlings survived to about 24 days-of-age.   If all these young fledged, the fledging rate would be slightly less than one young per pair (0.78).

Organochlorine compounds– In studies of cormorants nesting in the Great Lakes region, investigators reported that the lowest observed adverse effect level (LOAEL) for DDE in eggs was 3.5 µg/g wet weight (Tillitt et al. 1992, Yamashita et al. 1993).   These studies and a similar investigation by Custer et al. (1999) indicated that reduced hatching success was related to toxic concentrations of DDE in the egg rather than DDE-induced eggshell thinning. In our study, 4 of 22 cormorant eggs approached or exceeded the 3.5 µg/g adverse effect level.

DDE can have numerous adverse impacts on bird physiology and reproduction.   In extensive experimental trials using ringed turtle doves (*Streptopelia risoria*), overall productivity was reduced by 23% in pairs that had been fed DDE-treated food from 3 to 6 weeks prior to the

reproductive cycle (Keith and Mitchell 1993). The adverse effects of DDE on reproduction can be especially severe during periods of stress, such as food restriction. A 10% food restriction for birds that had been pre-exposed to a DDE-treated diet resulted in an 87% decrease in productivity. Although minor shell thinning was noted, reproductive failure was due primarily to low levels of hormones necessary to develop and maintain active gonads, adequate courtship and brooding behavior, and functional crop glands. Reproduction was depressed by both DDE and by food restriction, and the effects were synergistic (Keith and Mitchell 1993).

DDT, DDE, and chlordane are known endocrine-disrupting compounds and, exposure to endocrine-disrupting chemicals has been associated with abnormal thyroid function (Moccia et al. 1986) , decreased fertility (Shugart 1980), decreased hatching success (Kubiak et al. 1989), altered immune function (Erdman 1998), and feminization of male gull embryos (Fry and Toone 1987). These compounds are stored in body fat and are mobilized during egg laying (Colborn et al. 1993). In the lower Colorado River area, the adverse effects of endocrine-disrupting compounds have been documented in common carp (Bevens et al. 1996, Goodbred et al. 1997), and razorback sucker *(Xyrauchen texanus)* (Tuttle and Orsak 2002) and it is likely that these compounds also are present in fish-eating birds in that area.

PCBs were detected at low levels (≤ 0.47 µg/g) in 11 of 22 cormorant eggs. PCBs at this low frequency of occurrence and concentration are not considered a potential hazard to avian reproduction as the lowest egg concentrations associated with reduced hatchability of five bird species in experimental trials was 15 µg/g wet weight (Rice and O'Keefe 1995). These data are consistent with other studies of contaminants in wildlife of the lower Colorado River which report consistently low levels of PCBs in almost all ecological receptors including invertebrates, fish, and birds (Radtke et al. 1988, Andrews et al. 1997, Tadayon et al. 1997).

Eggshell thinning– Cormorant eggshell thickness averaged 0.412 mm which was 4.2% thinner than that of museum eggs collected before the widespread use of DDT. We found no evidence of cracked or crushed eggs in the nests and no abnormal young were observed. Although overall cormorant reproductive success was relatively poor with less than 1.0 young fledged per pair, there was no evidence to suggest that eggshell thinning was a factor in reproductive failure.

Mercury and selenium– Relatively low mercury levels (0.18 - 1.00 µg/g dry weight) were detected in all cormorant eggs and all concentrations fell below the ~2.5 µg/g background threshold as defined by Thompson (1996). There appeared to be little potential for adverse biological effects from mercury on the cormorant population nesting in Topock Marsh in 1999 - 2000. However, 14 of 22 cormorant eggs (64%) contained selenium concentrations that fell within the "level of concern" (3 to 6 µg/g) (USDI 1998). Seven eggs (32%) contained a potentially toxic (>6.0 µg/g) concentration of selenium.

Regurgitate and fish samples– No organochlorine compounds were detected in regurgitated or seined fish samples.   The absence of organochlorine residues in local fish samples suggests that organochlorine compounds recovered in cormorant eggs may have originated in feeding areas other than Topock Marsh.

Mercury (geometric mean 0.06 µg/g dry weight) was recovered in all four regurgitate samples, but mercury was not present in the fish seined from Topock Marsh (Table 10, Appendix 5).   This also implies that cormorants may have been feeding in areas other than Topock Marsh.

For selenium, the dietary reproductive impairment threshold for nesting aquatic birds is 3 to 8 µg/g dry weight in food items (USDI 1998).   All regurgitate samples collected from nestling cormorants contained < 3.0 µg/g dry weight selenium.   However, largemouth bass, red shiner, and threadfin shad seined from Topock Marsh contained 3.89 to 7.93 µg/g dry weight selenium.   Aquatic birds feeding on fish from Topock Marsh may be ingesting potentially toxic concentrations of selenium.

Western and Clark's grebes: Western and Clark's grebes were considered color phases of a single species, the western grebe, until 1985 (Storer and Nuechterlein 1992).   The breeding range of the two species overlaps and extends from north-central and western Canada south to northern Baja and the Mexican Plateau.   In Arizona, both grebe species nest in substantial numbers on Lake Havasu and other open waters of the lower Colorado River.   These species often nest in loose colonies or as individual pairs.   Both species are precocial with adults and young leaving the nest shortly after hatching.

Both grebe species are primarily fish eaters although some crustaceans and polychaete worms are occasionally reported in the diet (Storer and Nuechterlein 1992).   Both grebes are opportunists when it comes to species of fish taken.   Clark's grebes may forage farther from shore and in deeper water than western grebes (Nuechterlein and Buitron 1989).

Western grebe eggs were not analyzed for organochlorine compounds. The maximum mercury concentration in 8 western grebe eggs was 0.28 µg/g dry weight (Table 6), well below the background threshold of ~2.5 µg/g.   Of eight western grebe eggs collected, one contained above background (>3 µg/g dry weight), but below toxic (>6.0 µg/g), levels of selenium.   Seven eggs contained potentially embryotoxic (6.18 - 8.33 µg/g dry weight) selenium concentrations.

Carcasses of two western grebes found dead contained relatively low residues of organochlorine compounds including ≤0.17 µg/g wet weight DDE and ≤0.35 µg/g PCB. These levels are far below concentrations associated with adverse effects in adult birds.   The grebe from Bill Williams River NWR contained 7.75 µg/g dry weight mercury in the liver and the sample from Havasu NWR contained 2.31 µg/g.   Background concentrations of mercury in the liver are usually ≤10 µg/g dry weight (Ohlendorf 1993); therefore, mercury

concentrations in the livers of adult grebes found dead were within the normal or background range.   Grebe liver tissues also contained 4.20 and 7.38 µg/g dry weight selenium, respectively, and were within the background range of ≤10 µg/g dry weight (Ohlendorf 1993).   Organochlorine compounds, mercury, and selenium were not implicated in the death of the two adult western grebes.

All Clark's grebe eggs contained relatively low (≤0.28 µg/g wet weight) levels of DDE (Table 4) and the maximum residue was below the 3.0 - 4.0 µg/g level associated with reproductive problems in sensitive bird species.   Mercury concentrations in Clark's grebe eggs collected in 2000 (geometric mean = 0.05 µg/g dry weight) were significantly (P <0.0001) lower than those in eggs collected in 2001 (geometric mean = 0.18 µg/g).   The maximum mercury concentration in Clark's grebe eggs was 0.31 µg/g (Table 6) which was well below the background threshold (~2.5 µg/g).   Selenium concentrations in Clark's grebe eggs were similar (P >0.05) between years.   Selenium in all Clark's grebe eggs exceeded the 3.0 µg/g threshold for background levels, and 13 of 17 eggs exceeded the toxic threshold of 6.0 µg/g (Appendix 6).

Least bittern: The least bittern is listed as a year-round resident of the lower Colorado River area (Gibbs et al. 1992); however, no formal censuses have been taken of nesting least bittern numbers.   The dominant food of this species is small fish, primarily topwater species, and invertebrates (Gibbs et al. 1992).   Howell (1932) documented the stomach contents of 93 bitterns collected in Florida and reported that 40% contained small fish, 21% had dragonfly parts, 12% contained aquatic invertebrates and 10% had crustaceans.   Other food items included snakes, frogs, tadpoles, salamanders, leeches, crayfish, and small mammals (Gibbs et al. 1992).

DDE was recovered at low (0.30 µg/g wet weight) levels in the two bittern eggs collected in 2000 (Table 4) and residues were well below the 3.0 - 4.0 µg/g wet weight threshold associated with shell thinning and reproductive problems in sensitive waterbird species.   The maximum mercury concentration recovered in three bittern eggs was 0.42 µg/g dry weight (Table 6) which is within the background range of <2.5 µg/g dry weight.   Selenium was recovered in all three bittern eggs and concentrations ranged from 5.34 - 7.14 µg/g dry weight.   Selenium in two of three bittern eggs was above the background level (3.0 µg/g) and concentrations in one egg (7.14 µg/g) exceeded the 6.0 µg/g toxic threshold.

Great blue heron: The great blue heron nests throughout Arizona.   We observed great blue herons on all refuges but were able to access only one nest.   The great blue heron is primarily a fish eater; however, invertebrates, amphibians, reptiles, and small mammals also have been recorded in dietary observations (Palmer 1962, Kushlan 1978).

We collected one great blue heron egg.   The 4.10 µg/g wet weight DDE residue in the egg was above the 3.0 - 4.0 µg/g threshold where embryotoxic effects might be expected.   The egg also contained 1.33 µg/g dry weight mercury and 5.08 µg/g selenium.   Concentrations of both elements were below toxic thresholds.

American coot: The American coot is one of the most widespread aquatic birds in North America.   We observed coots at all refuges, but we were able to find nests only in 2001. Adult coots are primarily herbivorous throughout the year; however, consumption of animal matter including aquatic insects and mollusks increases during the summer breeding season (Jones 1940).

The two American coot eggs were not analyzed for organochlorine compounds.   The maximum concentration of mercury in coot eggs was 0.15 µg/g dry weight (Table 6), well below the background threshold of ~2.5 µg/g dry weight. Selenium concentrations of 6.7 and 7.55 µg/g dry weight, however, were above the 6.0 µg/g toxic threshold.

Great-tailed grackle: The great-tailed grackle is a year-round resident throughout Arizona. We observed grackles at all refuges and located nesting colonies at both Havasu and Cibola NWRs.   The food habits of this grackle species varies with their breeding cycle; during the breeding season, animal material, primarily arthropods and small vertebrates make up much of the diet along with plant material (Johnson and Peer 2001).   In the nonbreeding season, plant material becomes the dominant food source.   However, absolute volume of plant material does not change throughout the year.   For the purposes of this study, the great-tailed grackle represents an omnivorous species.

DDE residues in six grackle eggs collected in 2000, ranged from 0.13 to 0.70 µg/g wet weight (Table 4) and were below potentially toxic levels.   The frequency of occurrence of eggs with detectible concentrations of mercury was lower in samples from Havasu NWR (3/10) than Cibola NWR (6/10).   Mercury was recovered in one-half (13/26) of the eggs collected during both years of study (Appendix 10) and the maximum concentration, 0.46 µg/g dry weight, was within the background range of <2.5 µg/g. All 26 grackle eggs contained above background concentrations of selenium (Appendix 6).   Nine of 26 eggs (35%) contained potentially embryotoxic (>6.0 µg/g) concentrations (Appendix 10).   The highest concentration of selenium documented in this study, 10.7 µg/g, occurred in a grackle egg collected from Cibola NWR.

Cliff swallow: Cliff swallows were originally birds of the western mountains.   This species has expanded its range across the great plains to eastern North America coincident with the widespread construction of highway culverts used as nesting sites (Brown and Brown 1995). The cliff swallow is found in most northern and eastern portions of Arizona, but in southwestern Arizona, it is confined to the lower Colorado River riparian area.   The cliff swallow feeds almost exclusively on flying insects.   In our study area, most swallow feeding probably occurred over the river and backwater lakes where flying insects are more numerous than in surrounding desert habitats.   The cliff swallow therefore, is an excellent representative of a primarily aquatic insectivorous species.

Cliff swallow eggs and nestlings were collected only in 2001 and the samples were not analyzed for organochlorine compounds. Mercury was not detected in cliff swallow eggs or nestlings suggesting little potential for contamination from local sources. Selenium, however, was present in all eggs (2.97 - 3.70 µg/g dry weight) (Table 6) and nestlings (5.45 - 6.36 µg/g) (Appendix 7). Note that selenium concentrations were higher in nestlings than in eggs suggesting that selenium accumulation occurred during the first 10-days of life. Unfortunately, eggs and nestlings were not taken from the same nest; therefore, the precise bioaccumulation rate from eggs to 10-day old nestlings cannot be determined.

CONCLUSIONS

Reproductive success of white-faced ibises was seriously limited by DDE-induced eggshell thinning and/or by potentially embryotoxic residues detected in 48% of the eggs. Potentially embryotoxic DDE concentrations were detected in 18% of the double-crested cormorant eggs and in one great blue heron egg. The source of the DDE was most likely not of local origin as regurgitate and food items collected from a lower Colorado River backwater area contained low residues of organochlorine compounds. Our data concur with those of other contaminant investigations conducted in the lower Colorado River area that indicate that organochlorine compound residues in resident wildlife are below adverse effect thresholds. Migratory species such as the ibises, cormorants, and the great blue heron most likely bioaccumulated high DDE residues on migration routes or on wintering grounds.

Mercury was present at background levels in all fish-eating birds eggs and in all but one ibis egg. In the eggs of the omnivorous bird species, American coots and great-tailed grackles, mercury was detected in ≤50% of the eggs. Eggs and nestlings of the primarily insectivorous cliff swallow did not contain mercury. Mercury does not appear to be a contaminant of concern for birds nesting along the lower Colorado River.

Above background concentrations of selenium were detected in 94% (100/106) of the eggs of all species and potentially toxic concentrations were present in 8% (8/106). Although selenium accumulated to highest levels in top-of-the-food-chain predators such as the fish-eating birds, high concentrations also were present in the eggs of the omnivorous great-tailed grackle and American coot. Ten-day old cliff swallow nestlings contained higher concentrations of selenium than eggs collected from the same colony suggesting that a significant degree of bioaccumulation occurred during the first few days of life. No evidence of selenium-related reproductive failure was observed.

RECOMMENDATIONS

Our study documented continued high levels of DDE and biologically significant eggshell thinning in the white-faced ibis almost three decades after the national suspension of DDT. Further studies are needed to document areas where contaminant bioaccumulation is most likely. These studies should include assessments of both summer and winter range as well as migration stopover points. Also, there is a strong need to better understand the dynamics of pesticides in wetland and agricultural ecosystems.

High selenium concentrations were not restricted to top level predators and one unexpected result of this study was the occurrence of relatively high selenium levels in the omnivorous great-tailed grackle. Other omnivorous species such as the endangered Yuma clapper rail (*Rallus longirostris yumanensis*) also may be accumulating potentially toxic concentrations of selenium. Addled Yuma clapper rail eggs should be analyzed whenever encountered. A study should be initiated to assess selenium levels and potential adverse effects in a Yuma clapper rail surrogate species such as the Virginia rail *(Rallus limicola)*.

Additional study is needed to assess double-crested cormorant population trends throughout the lower Colorado River. The cormorant population nesting in Topock Marsh at Havasu NWR varied from 8 to 67 pairs between 1970 and 1987. During the years of our study, 2000 - 2001, we recorded only 12 and 7 nesting pairs, respectively. There may be an ongoing long-term decline in numbers. Structured annual censuses are needed to assess nesting populations throughout the region.

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Appendix 1. Organochlorine compound concentrations in white-faced ibis eggs collected at Cibola Lake, Cibola National Wildlife Refuge, Arizona, July 2000, ranked by declining order of DDE residues

Collection condition	Concentration (µg/g wet weight)				Moisture (%)	Lipid (%)
	p,p'-DDE	p'p'-DDT	Total PCB	Total Chlor		
Random[1,2]	14.0	0.09	<0.05	0.03	82.3	5.07
Abandoned[3]	14.0	0.07	0.06	0.04	58.0	9.02
Abandoned	9.00	<0.01	<0.05	0.04	79.3	7.36
Random[2]	9.00	0.20	<0.05	0.01	82.4	4.66
Random[3,4]	8.80	0.07	0.18	0.03	81.8	5.86
Random	8.30	0.05	<0.05	<0.01	83.1	5.94
Random[2,4]	5.80	<0.01	0.06	0.03	84.0	4.07
Abandoned	5.00	<0.01	0.05	0.01	77.2	6.66
Abandoned	5.00	<0.01	<0.05	0.02	82.8	6.33
Random	4.90	0.03	<0.05	0.02	84.6	5.06
Random	4.50	0.02	<0.05	0.02	82.5	5.80
Random	3.50	<0.01	0.06	0.01	82.8	5.09
Random	3.40	<0.01	<0.05	0.01	82.3	5.00
Random[5]	3.00	0.04	<0.05	<0.01	82.0	4.94
Abandoned	2.90	<0.01	<0.05	<0.01	82.1	3.27
Random	2.80	<0.01	<0.05	<0.01	82.7	4.56
Random[6]	2.00	<0.01	<0.05	0.02	82.9	4.66
Abandoned	0.33	<0.01	<0.05	<0.01	80.9	5.91
Random	0.31	<0.01	<0.05	<0.01	83.5	5.42
Random[1,6]	0.26	0.01	<0.05	0.05	82.3	6.29
Random	0.19	<0.01	<0.05	<0.01	83.0	5.62
Abandoned	0.18	<0.01	<0.05	<0.01	82.2	5.87
Random	0.13	<0.01	<0.05	<0.01	80.0	5.14

[1]Dieldrin was detected in one egg from nest 10 (0.55 µg/g) and in one egg from nest 2 (0.09 µg/g).

[2]p,p'-DDD detected in four eggs: 1 (0.47 µg/g), 5 (0.06 µg/g), 10 (0.07 µg/g), and 17 (0.14 µg/g).

[3] Pipped egg. Embryo dead in shell.

[4]Visibly thin-shelled eggs found in nests these nests.

[5]Egg from nest 12 also contained 0.06 µg/g HCB.

[6]Heptachlor epoxide present in egg from these nests at 0.12 µg/g and 0.06 µg/g.

Appendix 2.   Metals in white-faced ibis eggs collected from the Cibola National Wildlife Refuge, Arizona, 2000

| Contaminant concentration, µg/g dry weight[1] | | | | | | | | | | | | Moist (%) |
Al Zn	Ba	Cr	Cu	Fe	Hg	Mg	Mn	Mo	Se	Sr		
6.96	0.86	<0.54	3.62	64	0.21	497	2.83	<0.54	2.71	11.2	58.4	84.0
<5.15	1.88	<0.52	3.46	67	0.17	467	1.53	<0.52	3.67	16.7	61.0	82.9
<5.15	1.88	<0.52	3.46	67	0.17	467	1.26	<0.53	3.37	13.3	51.8	82.8
<5.31	0.63	<0.53	4.26	84	0.17	503	1.07	<0.53	4.16	11.3	41.5	80.7
<5.31	<0.53	<0.53	4.28	75	0.50	530	2.27	<0.53	3.06	15.3	51.7	82.4
<5.26	0.71	<0.53	4.04	81	0.48	577	21.3	<0.51	3.69	36.2	56.8	81.9
14.6	1.75	1.99	5.98	140	0.52	742	1.43	<0.51	3.32	14.3	59.4	82.3
<5.12	1.34	<0.51	3.86	94	0.29	443	1.17	<0.52	3.51	10.1	45.7	81.2
<5.21	0.53	<0.52	4.48	66	0.33	423	2.66	0.88	3.44	12.0	59.5	82.8
<5.53	<0.55	<0.55	2.92	84	0.22	395	2.65	<0.52	2.94	11.0	50.5	82.6
<5.17	<0.52	<0.52	3.93	84	0.80	522	1.34	<0.53	2.44	18.2	66.5	82.5
<5.29	0.79	<0.53	4.39	119	0.28	650	1.59	<0.52	3.25	21.7	78.5	78.7
<5.22	1.57	<0.52	4.41	109	0.23	664	9.36	<0.53	2.70	22.9	71.3	82.4
12.5	0.70	4.62	6.05	165	0.30	875	1.31	<0.52	3.20	15.2	54.7	79.4
<5.17	1.64	1.00	4.09	127	0.95	410	2.72	<0.53	3.31	12.1	63.5	56.2
9.01	<0.53	<0.53	4.50	90	0.21	797	1.77	<0.54	3.51	15.8	49.5	82.3
<5.38	0.81	<0.54	3.34	97	0.16	424	1.53	<0.52	3.16	12.8	53.5	82.8
<5.20	0.92	<0.52	3.82	85	1.40	573	1.33	<0.54	3.34	16.8	54.9	82.2
<5.36	1.12	<0.54	4.22	106	1.93	472	1.66	<0.50	4.10	6.78	49.9	81.2
<5.02	<0.50	<0.50	3.16	97	0.31	566	1.79	<0.48	3.38	17.0	56.3	82.7
<4.80	1.02	<0.48	3.61	103	2.72	647	2.33	<0.53	2.59	15.1	59.7	83.1
<5.31	0.68	<0.53	3.05	112	0.89	498	2.75	<0.50	3.22	10.5	54.4	82.3
<5.01	0.57	<0.50	3.19	81	0.18	595	2.04	<0.51	2.85	14.6	57.3	82.5

[1]Arsenic, boron, beryllium, cadmium, and vanadium were not detected in any samples.

Appendix 3. Organochlorine compound residues, ppm wet weight, in double-crested cormorant eggs collected from Topock Marsh, Havasu National Wildlife Refuge, Arizona, 1999 - 2000[a,] ranked in order of declining DDE concentrations

Year collected	p,p'-DDE	Total PCB	Percent Moisture	Percent Lipid
1999[b]	4.30	<0.05	81.1	5.85
1999	4.20	0.36	84.4	3.06
1999	3.60	0.35	81.5	5.84
2000	3.40	<0.05	82.7	5.70
1999	2.50	0.06	79.6	5.66
1999	2.20	<0.05	78.5	8.07
2000	2.10	0.47	84.9	2.81
1999	1.40	<0.05	79.1	5.32
2000	1.20	0.40	83.8	4.90
2000	1.00	0.22	82.5	4.05
1999	0.80	<0.05	81.2	5.79
1999	0.79	0.23	81.0	6.47
1999	0.69	<0.05	83.4	3.47
2000	0.67	0.18	83.7	5.48
2000	0.40	<0.05	84.3	3.68
2000	0.39	<0.05	80.6	5.68
2000	0.25	0.28	79.4	5.57
2000	0.21	0.20	81.3	5.18
2000	0.17	<0.05	84.8	2.71
2000	0.15	<0.05	83.4	5.11
2000	0.14	0.08	80.2	4.99
2000	0.05	<0.05	83.4	4.74

[a]No other organochlorine compounds were detected.
[b]Samples collected in 1999 were collected by Havasu NWR Refuge Biologist.

Appendix 4.   Metals in double-crested cormorant eggs collected from Topock Marsh, Havasu National Wildlife Refuge, 1999 -2000

Year	Element concentration, µg/g dry weight[1]											Moist (%)
	Ba	Cr	Cu	Fe	Hg	Mg	Mn	Mo	Se	Sr	Zn	
1999	<0.53	<0.53	7.70	120	0.86	510	0.82	1.40	4.37	9.04	51.6	81.6
1999	0.75[2]	2.04	7.61	177	0.31	677	3.47	1.11	4.15	17.6	78.9	81.1
1999	0.71	<0.53	6.29	140	0.31	550	1.33	0.84	4.62	9.63	51.4	79.5
1999	0.73	<0.48	7.29	188	0.78	695	1.62	1.56	3.95	16.4	68.9	81.5
1999	<0.51	0.92	8.32	134	0.71	793	3.31	0.75	6.22	11.9	67.8	83.0
1999	<0.54	1.24	7.72	119	0.39	727	2.48	0.84	4.65	14.1	57.6	84.0
1999	<0.55	<0.55	7.51	180	0.41	615	3.29	1.17	6.22	8.21	86.6	79.3
1999	0.95	<0.52	7.69	198	0.65	726	1.77	1.69	3.80	16.9	76.2	81.8
1999	<0.51	<0.51	7.63	108	1.00	563	0.67	1.28	3.95	8.69	50.3	79.4
2000	<0.54	<0.54	5.78	118	0.48	621	1.63	1.21	5.10	7.72	45.8	81.9
2000	<0.54	0.99	6.27	139	0.28	544	1.76	1.11	5.40	7.82	52.8	79.7
2000	<0.54	<0.54	7.04	103	0.68	466	2.44	1.10	4.51	6.18	54.6	82.8
2000	<0.52	0.87	7.51	123	0.37	587	1.62	0.97	7.06	6.53	48.2	79.0
2000	<0.54	<0.54	6.85	110	0.41	595	1.48	1.06	4.04	6.63	65.5	76.1
2000	<0.52	0.54	6.26	122	0.18	619	1.11	0.83	7.46	7.50	43.5	81.1
2000	<0.53	0.67	6.65	122	0.52	586	1.49	0.92	3.97	3.95	55.8	85.0
2000	<0.52	0.63	6.42	130	0.35	593	2.39	1.27	5.05	5.97	49.4	80.4
2000	<0.53	<0.53	6.65	129	0.29	400	1.82	1.40	6.39	6.21	53.5	81.7
2000	<0.51	<0.51	6.92	120	0.36	438	1.90	1.57	6.07	4.93	53.0	82.3
2000	0.51	<0.48	6.07	158	0.32	588	2.96	1.22	1.78	6.74	58.6	82.3
2000	<0.53	1.06	9.23	152	0.24	897	2.28	0.77	8.29	9.97	65.7	83.7
2000	<0.48	<0.48	6.47	116	0.21	527	2.31	1.08	4.62	5.79	50.0	81.8

[1]Arsenic, barium, boron, beryllium, cadmium, and vanadium were not detected in any samples.

[2]Aluminum and nickel were present in this egg at 3.44 and 0.16 µg/g.

Appendix 5. Metals in double-crested cormorant regurgitate and in whole fish collected from Topock Marsh, Havasu NWR, 2000

Sample	N	Element concentration, μg/g dry weight[1]													Moist (%)
		Al	As	Ba	Cr	Cu	Fe	Hg	Mg	Mn	Se	Sr	V	Zn	
Regurgitate[2]	1	7.08	<0.42	3.33	1.85	2.71	58.0	0.08	1834	7.43	2.93	241	<0.52	106	77.7
Regurgitate	1	13.2	0.49	8.29	1.73	5.11	132.	0.04	1512	48.1	1.88	113	0.68	105	73.7
Regurgitate	1	8.63	<0.42	4.62	3.01	5.22	103.	0.04	1371	39.6	2.00	78.2	<0.52	64.8	72.0
Regurgitate[2]	1	<5.38	<0.43	1.80	1.70	2.60	59.4	0.10	1856	3.99	2.64	226	<0.54	94.5	75.7
Largemouth bass	1	51.5	<0.80	11.8	1.22	2.66	85.5	<0.04	2550	20.8	5.47	283	<1.00	153	76.3
Largemouth bass	1	<10.0	<0.80	5.42	1.76	2.06	38.4	<0.04	2183	5.40	3.89	185	<1.00	99.8	75.0
Largemouth bass	1	79.7	<0.78	4.98	1.82	1.93	91.5	<0.04	2161	9.34	5.78	154	<0.98	115	74.9
Red shiner	30	23.3	<0.75	10.4	1.61	3.20	75.0	<0.04	1790	14.3	3.91	173	<0.94	235	72.7
Red shiner	30	40.0	<0.99	12.7	<0.99	4.29	61.7	<0.04	1675	11.5	4.99	198	<0.99	207	74.6
Threadfin shad[3]	28	1057	2.71	26.4	4.13	5.37	1567	<0.04	2009	38.2	7.93	97.5	3.57	82.6	76.0

[1]Boron, beryllium, cadmium, and molybdenum were not detected in any samples.
[2]Portions of a bass (species unknown) were recognizable.
[3]Lead and nickel were detected in the threadfin shad sample at 1.44 and 1.55 μg/g dry weight.

Appendix 6.   Metals in Clark's and western grebe eggs collected from Havasu and Bill Williams River National Wildlife Refuges, 2000 - 2001

| Species | Year | Refuge | Element concentration μg/g dry weight[1] | | | | | | | | | | | | Moist (%) |
			Al	B	Ba	Cr	Cu	Fe	Hg	Mg	Mn	Se	Sr	Zn	
Clark's grebe	2000	Havasu	<5.81	<2.33	2.66	<0.58	3.36	127	0.09	378	1.92	4.62	11.41	57.6	80.8
Clark's grebe	2000	Havasu	<5.40	<2.16	<0.54	<0.54	2.65	153	0.06	366	2.79	6.27	4.64	48.8	78.5
Clark's grebe	2000	Havasu	<5.40	<2.16	<0.54	<0.54	2.50	105	0.05	388	1.84	6.55	5.81	45.5	80.3
Clark's grebe	2000	Havasu	<5.23	<2.09	<0.52	<0.52	1.88	126	0.05	413	2.11	5.37	8.07	42.5	77.8
Clark's grebe	2000	Havasu	<5.17	<2.07	<0.52	<0.52	2.66	114	0.04	386	2.60	5.42	6.78	51.9	77.8
Clark's grebe	2000	Havasu	<4.91	<2.16	0.55	<0.49	2.75	115	0.05	380	2.06	5.42	9.65	60.6	78.1
Clark's grebe	2001	Havasu	2.60	<0.50	<0.99	<0.10	2.00	134	0.29	326	1.07	6.70	7.22	45.5	77.9
Clark's grebe	2001	BWR	2.65	<0.50	<0.99	<0.10	2.73	124.	0.17	385.	2.11	8.29	4.17	46.4	79.9
Clark's grebe	2001	BWR	2.10	<0.50	<0.99	<0.10	2.30	94.9	0.16	498.	3.23	8.40	7.32	51.1	78.6
Clark's grebe	2001	BWR	2.49	1.12	<0.99	<0.10	2.06	100.	0.15	350.	1.44	7.11	4.23	44.0	78.3
Clark's grebe	2001	BWR	3.77	0.67	<0.99	2.45	2.22	94.4	0.14	301.	2.74	6.80	2.29	36.8	76.9
Clark's grebe	2001	BWR	1.70	<0.50	<0.99	<0.10	1.83	92.1	0.16	297.	3.38	7.10	2.33	37.0	78.1
Clark's grebe	2001	BWR	1.79	<0.50	<0.99	<0.10	2.95	141.	0.18	416.	2.33	7.93	4.45	53.7	79.3
Clark's grebe	2001	BWR	2.14	0.80	<0.99	<0.10	2.57	93.4	0.18	369.	3.82	6.20	4.26	37.2	77.9
Clark's grebe	2001	BWR	1.59	<0.50	<0.99	<0.10	2.24	96.2	0.15	378.	1.99	7.83	4.65	50.3	78.4
Clark's grebe	2001	BWR	1.26	<0.50	<0.99	<0.10	2.27	121.	0.13	360.	2.64	7.27	3.48	47.8	78.0
Clark's grebe	2001	BWR	1.36	<0.50	<0.99	<0.10	2.61	101.	0.31	415.	2.11	7.40	6.32	45.2	77.2
West. grebe	2001	BWR	1.76	<0.50	<0.99	<0.10	2.76	134.	0.14	335.	2.30	4.77	4.82	46.6	79.5
West. grebe	2001	BWR	1.69	<0.50	<0.99	<0.10	2.69	115.	0.14	359.	2.03	7.21	4.45	48.8	76.9
West. grebe	2001	BWR	2.28	<0.50	<0.99	<0.10	2.55	123.	0.15	377.	2.22	6.60	3.01	43.1	77.9
West. grebe	2001	BWR	1.55	<0.50	<0.99	<0.10	2.44	93.6	0.21	459.	1.66	8.33	2.72	34.7	77.3
West. grebe	2001	BWR	2.64	<0.50	<0.99	<0.10	2.51	112.	0.17	446.	2.45	7.38	4.15	48.7	76.8
West. grebe	2001	BWR	4.69	<0.50	<0.99	<0.10	2.78	154.	0.16	417.	2.25	6.38	7.25	45.3	77.2
West. grebe	2001	BWR	3.07	1.04	<0.99	<0.10	3.56	113.	0.12	400.	2.64	6.66	4.98	41.8	81.0
West. grebe	2001	BWR	1.49	<0.50	<0.99	<0.10	2.23	111.	0.28	346.	3.32	6.18	5.17	46.7	78.4

[1]Arsenic, beryllium, cadmium, lead, molybdenum, nickel, and vanadium were not detected in any samples.

Appendix 7.   Metals in liver tissues of white-faced ibis and western grebes found dead and in cliff swallow nestling carcasses from Cibola, Bill Williams River, and Havasu National Wildlife Refuges, Arizona, 2000 - 2001

Species	Refuge	Contaminant concentration, µg/g dry weight[1]														Moist (%)
		Al	B	Cd	Cr	Cu	Fe	Hg	Mg	Mn	Mo	Pb	Se	Sr	Zn	
W-f ibis[2]	Cibola	<10.4	<4.17	<0.21	58.8	58.3	1113	0.11	790	18.9	1.23	4.28	4.17	2.59	<1.04	73.2
W. grebe[3]	Bill Wm.	1.55	3.90	1.08	0.11	48.9	8728	7.75	637.	12.5	1.11	<0.05	4.20	0.25	75.9	73.4
W. grebe[3]	Havasu	3.03	3.55	1.20	<0.10	63.6	9990	2.31	661.	15.4	1.20	<0.05	7.38	0.46	64.6	75.1
C. swallow	Havasu	6.89	5.31	<0.19	<0.19	14.7	1216	<0.10	773.	6.26	<1.95	0.10	6.51	10.9	26.8	71.0
C. swallow	Havasu	4.45	4.94	<0.28	<0.28	18.5	943	<0.14	747.	5.76	<2.79	0.53	5.45	8.12	30.1	70.9
C. swallow	Havasu	5.72	3.02	<0.21	<0.21	15.4	1387	<0.11	780.	7.34	<2.13	0.22	6.36	5.65	34.8	68.9

[1]Arsenic, barium, beryllium, and vanadium were not detected in any samples.

[2]Nestling ibis found dead. This sample also contained 4.28 µg/g nickel.

[3]Adult grebes found dead by refuge personnel.

Appendix 8.   Organochlorine compound concentrations, µg/g wet weight, in Clark's grebe, great-tailed grackle, least bittern, and great blue heron eggs collected on Havasu and Cibola National Wildlife Refuges, Arizona, July 2000

Species	Refuge	p,p'-DDE	Total PCB	Moisture (%)	Lipid (%)
Clark's grebe	Havasu	0.10	<0.05	77.9	9.23
Clark's grebe	Havasu	0.11	<0.05	81.4	3.62
Clark's grebe	Havasu	0.10	<0.05	78.6	8.85
Clark's grebe	Havasu	0.07	<0.05	80.5	7.54
Clark's grebe	Havasu	0.28	<0.05	78.4	8.11
Clark's grebe	Havasu	0.08	<0.05	78.3	4.11
Great-tailed grackle	Cibola	0.13	<0.05	84.2	3.61
Great-tailed grackle	Cibola	0.15	<0.05	83.7	3.99
Great-tailed grackle	Cibola	0.62	0.15	64.7	8.86
Great-tailed grackle	Cibola	0.70	<0.05	83.2	3.35
Great-tailed grackle	Cibola	0.45	<0.05	78.9	5.73
Great-tailed grackle	Cibola	0.33	<0.05	84.9	4.82
Least bittern	Cibola	0.30	<0.05	78.8	6.38
Least bittern	Cibola	0.30	<0.05	82.0	5.85
Great blue heron[1]	Cibola	4.10	0.24	83.2	4.69

[1]This sample also contained 0.31 µg/g total chlordane.

Appendix 9.   Metals in American coot, least bittern, cliff swallow, and great blue heron eggs collected from Havasu and Cibola National Wildlife Refuges, Arizona 2000 - 2001

Species	Contaminant concentration, μg/g dry weight[1]														Moist (%)
	Al	As	B	Ba	Cr	Cu	Fe	Hg	Mg	Mn	Mo	Se	Sr	Zn	
Least bittern	6.04	<0.43	<2.13	0.76	<0.53	6.44	129	0.41	506	2.87	<0.53	5.34	4.81	67.1	78.8
Least bittern	<5.36	0.64	<2.15	1.00	<0.54	8.26	136	0.42	600	3.32	<0.54	5.83	8.42	68.6	81.5
Least bittern	2.79	<0.50	1.05	<1.00	<0.10	5.20	98.2	0.29	481	2.12	<1.00	7.14	3.79	63.8	82.9
Cliff swallow	7.79	<0.67	1.22	16.9	0.21	2.13	115.	<0.05	293.	2.95	<1.35	3.20	37.0	61.2	77.8
Cliff swallow	18.4	<0.71	1.27	12.9	<0.14	2.04	122.	<0.05	315.	4.22	<1.42	2.97	41.5	60.8	76.6
Cliff swallow	16.3	<0.98	1.24	3.16	0.35	2.18	93.0	<0.05	358.	2.39	<1.95	3.64	18.5	73.2	83.7
Cliff swallow	10.4	<0.98	6.17	2.46	0.26	1.86	92.5	<0.05	319.	2.49	<1.95	3.70	18.0	64.4	80.4
Amer. coot	1.89	<0.50	2.17	1.26	<0.10	1.85	83.6	<0.05	377.	1.12	<0.99	6.70	7.22	45.5	75.1
Amer. coot	2.02	<0.50	0.88	3.06	<0.10	3.22	108.	0.15	415.	2.28	<0.99	7.55	12.8	46.0	75.8
Gr. blue heron	9.18	<0.45	<2.24	1.21	1.63	5.33	157	1.33	787	7.74	1.10	5.08	15.0	67.5	82.6

[1]Beryllium, cadmium, lead, nickel, and vanadium were not detected in any samples.

Appendix 10. Metals in great-tailed grackle eggs collected from Cibola and Havasu National Wildlife Refuges, Arizona 2000 - 2001

Year	Refuge	Contaminant concentration, μg/g dry weight[1]												Moist
		Al	B	Ba	Cr	Cu	Fe	Hg	Mg	Mn	Se	Sr	Zn	(%)
2000	Cibola	<5.14	<2.06	0.86	<0.51	3.77	115	0.09	484	4.86	4.51	14.7	65.1	84.0
2000	Cibola	<8.14	<3.26	<0.81	<0.81	2.29	146	0.07	483	4.20	3.84	11.4	56.0	83.0
2000	Cibola	<5.25	<2.10	0.98	<0.52	2.75	130	<0.02	411	1.82	4.05	5.99	79.8	69.9
2000	Cibola	<5.36	<2.15	0.90	<0.54	2.56	121	0.03	516	1.85	4.21	6.55	77.2	84.0
2000	Cibola	<22.5	<9.01	5.44	<2.25	3.00	140	<0.09	470	2.68	4.20	24.0	73.4	93.4
2000	Cibola	18.0	<2.19	1.36	<.548	1.71	139	0.11	427	3.11	4.99	13.5	56.1	81.0
2001	Cibola	1.65	1.65	4.23	<0.10	3.24	172.	0.13	368.	1.10	8.17	23.6	79.4	81.7
2001	Cibola	1.29	1.13	2.07	0.11	1.97	211.	<0.05	364.	3.37	4.59	10.1	66.7	76.0
2001	Cibola	1.83	0.60	1.53	<0.10	2.43	116.	<0.05	369.	1.28	7.54	12.8	60.2	72.9
2001	Cibola	1.23	0.50	1.24	<0.10	2.70	115.	<0.05	423.	3.28	3.98	8.00	76.7	81.4
2001	Cibola	1.98	<0.50	1.44	<0.10	3.40	150.	0.46	403.	2.12	8.89	39.7	57.6	74.1
2001	Cibola	2.02	1.64	3.75	<0.10	3.52	182.	0.44	581.	1.66	7.82	53.8	73.6	76.0
2001	Cibola	2.60	0.91	1.19	<0.10	2.80	96.8	0.05	342.	0.63	6.91	11.8	38.8	80.0
2001	Cibola	2.16	0.67	<1.00	0.15	1.80	121.	<0.05	416.	2.01	3.74	8.51	51.3	56.0
2001	Cibola	2.03	0.72	3.70	<0.10	1.91	153.	0.08	411.	2.27	6.54	18.4	61.0	79.9
2001	Cibola	2.08	<0.50	<0.99	<0.10	3.25	154.	0.12	399.	1.63	10.7	34.8	55.3	81.3
2001	Havasu	2.30	1.22	4.81	<0.10	1.88	117.	<0.05	321.	1.96	5.34	19.2	57.5	93.4
2001	Havasu	2.75	0.62	4.30	0.19	1.76	175.	<0.05	311.	1.93	4.48	11.5	63.5	81.5
2001	Havasu	4.84	<0.50	6.31	0.14	2.13	137.	0.05	549.	3.02	6.95	55.5	62.6	83.2
2001	Havasu	3.02	<0.50	2.80	1.64	2.15	153.	<0.05	396.	2.04	8.72	18.7	62.7	83.8
2001	Havasu	4.10	2.54	5.59	0.18	2.50	188.	<0.05	375.	2.53	4.49	11.8	69.6	77.9
2001	Havasu	2.25	0.94	3.36	<0.10	1.83	185.	<0.05	303.	2.51	4.22	12.2	75.7	79.2
2001	Havasu	1.74	1.41	<0.98	<0.10	1.69	100.	0.09	333.	2.42	5.53	10.3	52.5	82.3
2001	Havasu	3.93	1.02	1.84	0.11	3.09	162	0.24	290.	2.90	5.53	13.9	58.6	82.7
2001	Havasu	8.73	2.31	1.29	<0.10	2.27	89.5	<0.05	397.	1.33	3.42	19.3	50.5	84.4
2001	Havasu	2.35	1.36	1.31	<0.10	1.81	138.	<0.05	357.	2.66	3.68	13.5	64.0	82.7

[1]Arsenic, beryllium, cadmium, lead, molybdenum, nickel, and vanadium were not detected in any samples.