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Organochlorine and Heavy Metal Residues in Falconiforme and Ciconiforme Eggs (Spain)

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Over the last three decades the biosphere has been contaminated almost universally by persistent pollutants of agricultural and industrial origin. Organochlorine pesticides and polychlorinated biphenyls have been documented to have adverse effects of the reproduction and survival of wild birds. Also, environmental contamination from heavy metals has been shown to be a threat to the survival and reproduction of certain birds.

The hazard to Falconiforme and Ciconiforme birds is high because a) they are sensitive to eggshell thinning b) they are high in the food chain (Peakall 1975). The present paper reports the levels of organochlorine compounds and heavy metals in 69 eggs of five species of the avian Falconiforme order and two species of the avian Ciconiforme order collected at Doñana National Park and Castile Plateau (Spain).

Objectives of this study are (1) to determine the levels of organochlorine pollutants and heavy metals in eggs of Falconiforme and Ciconiforme birds of Spain; (2) to evaluate the impact of the contaminants detected on reproductive potential; (3) to evaluate regional patterns of residues.

MATERIALS AND METHODS

Sixty-nine infertile bird eggs were gathered at Doñana National Park and Castile Plateau in 1985 and 1986 breeding season. The Doñana National Park, an area of 50,720 ha, is located in the south-southwest of the Iberian Peninsula. The Doñana National Park, which is also a Scientific Reservation of the Biosphere of UNESCO, and area included in the International Agreement of Ramsar for the protection of wet areas, is of the utmost importance for sedentary birds living there. It is also important for birds migrating between Europe and Africa. The Castile Plateau, located in the centre of the Iberian Peninsula, constitute one important breeding habitat of birds in Spain (Figure 1).

All eggs were collected after they failed to hatch and identified

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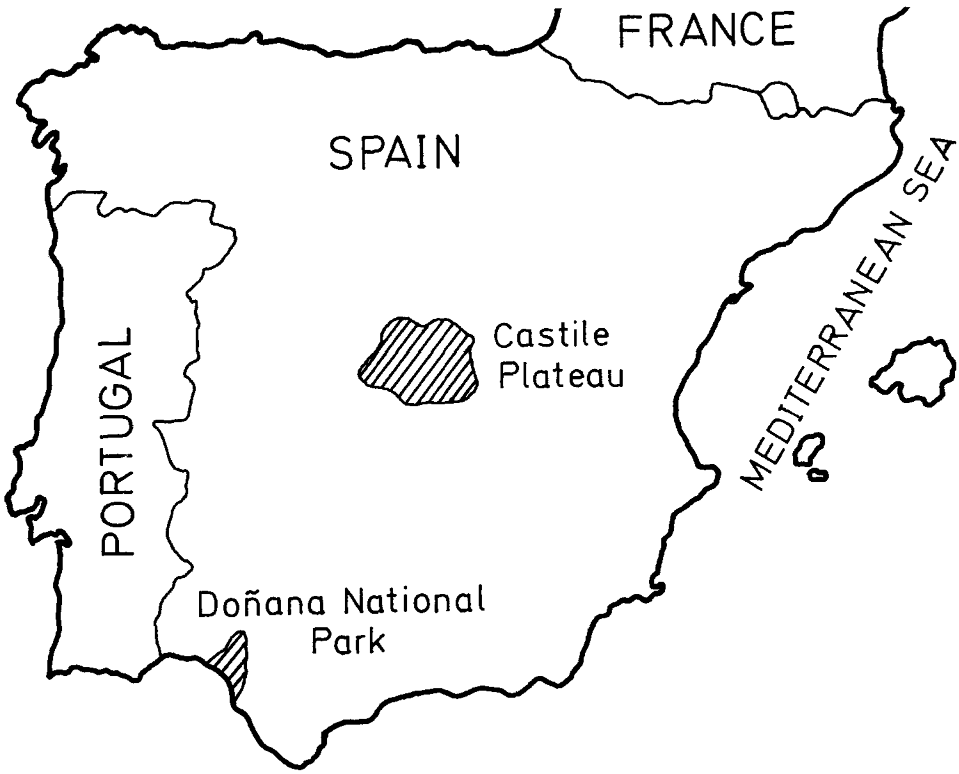

Figure 1. Study areas and sampling locations.

Table 1. Avian species, location and number of eggs collected.

Species	Location	No. of eggs
Spoonbill (*Platalea leucorodia*)	Doñana	9
White Stork (*Ciconia ciconia*)	Doñana	5
Black Kite (*Milvus migrans*)	Doñana	19
Booted Eagle (*Hieraetus pennatus*)	Doñana	3
Buzzard (*Buteo buteo*)	Doñana	2
White Stork (*Ciconia ciconia*)	Castile	16
Black Kite (*Milvus migrans*)	Castile	4
Booted Eagle (*Hieraetus pennatus*)	Castile	3
Buzzard (*Buteo buteo*)	Castile	2
Peregrine Falcon (*Falco peregrinus*)	Castile	3
Royal Eagle (*Aquila chrysaetos*)	Castile	3

as shown in Table 1. They were kept frozen until preanalytical treatment was performed and analyzed individually for residues of organochlorine pesticides, PCBs and heavy metals.

For organochlorines, three grams of the sample were homogenized, mixed with anhydrous sodium sulphate, and extracted with hexane in a Soxhlet apparatus for 8 hours. The extract was cleaned on a partially deactivated Florisil column. The pesticides and

PCBs were separated into four fractions (Cromartie et al. 1975, Kaiser et al. 1980), but fraction I was eliminated since it was not analyzed for HCB or mirex. Due to a lack of Silicar, silica gel chromatography with the same eluting solvents was substituted in the analysis of the eggs contents. Analysis was conducted by gas-liquid chromatography using an electron affinity detector following methods already described in a previous paper (Hernández et al. 1985). Extracts were analyzed for α-HCH, γ-HCH, aldrin, dieldrin, heptachlor, heptachlor epoxide, p,p'-DDE, p,p'-TDE, p,p'-DDT, dichlorobenzophenone and PCBs. The lowest limit of reportable residues was 0.01 ppm (mg/kg) for insecticides and 0.08 ppm for PCBs. Recoveries of insecticides and PCBs from fortified chicken eggs ranged from 89% to 106%, but the residue data in the tables were not adjusted on the basis of these recoveries. All residues are expressed as ppm, wet weight. Geometric means were used to express residue levels. For samples in which no residues could be detected, a value of one-half the reportable limit was assigned for statistical analysis.

The mercury concentration was determined by the flameless atomic absorption method of Hatch and Ott (1968). Lead, cadmium, copper and zinc were analyzed by flame atomic absorption according to the method of Haseltine et al. (1981). The limit of detection were 0.01 ppm for mercury, 0.02 ppm for lead, 0.001 ppm for cadmium, 0.002 ppm for copper, and 0.06 ppm for zinc. Recoveries of mercury, lead, cadmium, copper, and zinc from fortified chicken eggs ranged from 87% to 96%, but the residue data in the tables were not adjusted on the basis of these recoveries. All residues are expressed as ppm wet weight. Geometric means were used to express the residue levels.

RESULTS AND DISCUSSION

Residues of DDE were found in each of the 69 eggs in amounts ranging from 0.06 to 65.86 ppm (Tables 2 and 3). DDT was detected in 60 eggs (87%, range from 0.01 to 2.39 ppm), TDE was detected in 6 eggs (8.7%, range from 0.08 to 0.37 ppm), γ-HCH was detected in 42 eggs (60.8%, range from 0.01 to 1.87 ppm), and α-HCH was detected in 4 eggs (5.8%, range from 0.02 to 0.34 ppm). PCBs ocurred in all 69 eggs in amounts ranging from 0.14 to 18.75 ppm. Residues values detected but not shown in Tables 2 and 3 were: TDE at 0.08, 0.09, 0.12, and 0.17 ppm in Black Kite (Doñana), and 0.18 and 0.37 ppm in Peregrine Falcon (Castile), and α-HCH at 0.34 ppm in Booted Eagle (Doñana), and 0.02, 0.03, and 0.10 ppm in White Stork (Castile). Levels of the other chlorinated pesticides (aldrin, dieldrin, heptachlor, heptachlor epoxide, and dichlorobenzophenone) included in the analytical survey were below their limits of detection. The mean total organochlorine concentrations was 3.4 ppm in Falconiformes and 0.9 in Ciconiformes; the differences in residue levels seem related to differences in feeding habits. DDE constituted about 41.1% of the total residues in Falconiformes and 33.1% in Ciconiformes, PCBs 53.6 and 59.1%, and other compounds combined made up the remaining 5.2 and 7.7%, respectively.

Table 2. Geometric mean and range of organochlorine pollutants and heavy metals in Ciconiforme and Falconiforme eggs from Doñana.

Species	n	γ-HCH	DDE	DDT	PCBs
S.	9	0.006 N.D.-0.03	0.434 0.17-1.23	0.057 0.02-0.12	0.573 0.35-1.29
W.S.	5	0.021 0.01-0.04	0.415 0.27-0.71	0.076 0.02-0.26	0.814 0.23-2.70
B.K.	19	0.023 N.D.-0.09	0.691 0.23-3.23	0.220 N.D.-2.39	2.882 0.54-18.7
B.E.	3	0.012 N.D.-1.87	2.237 0.80-7.20	0.116 0.05-0.28	1.536 0.45-8.43
B.	2	0.034 0.03-0.04	0.293 0.27-0.32	0.028 0.01-0.08	1.649 1.60-1.70

Species	n	Hg	Cd	Pb	Cu	Zn
S.	9	0.17 0.11-0.29	0.09 0.05-0.17	1.09 0.37-3.44	1.17 0.79-1.94	9.61 5.88-17.7
W.S.	5	0.21 0.12-0.31	0.08 0.06-0.15	1.12 0.96-1.41	1.54 1.39-1.63	10.19 8.43-11.8
B.K.	19	0.09 0.03-0.47	0.06 0.04-0.28	0.76 0.35-3.54	0.97 0.67-1.42	9.58 6.35-16.0
B.E.	3	0.18 0.09-0.33	0.04 0.03-0.05	0.67 0.50-1.09	1.16 1.00-1.31	8.42 7.69-9.30
B.	2	0.12 0.10-0.15	0.08 0.06-0.10	0.73 0.70-0.76	1.22 1.17-1.28	9.50 9.19-9.84

S. = Spoonbill, W.S. = White Stork, B.K. = Black Kite, B.E. = Booted Eagle, B. = Buzzard; n = Number of Eggs; N.D. = Not Detected.

It has been proved that DDE is the main pesticide responsible for decreasing the thickness of the eggshell but DDE's incidence is subjected to interspecific variations. Furthermore, the hazard to Falconiforme birds must be imputed to their position in the food chain and their sensitivity to DDE induced eggshell thinning. Keith and Grouchy (1972) reported that 12 ppm of DDE was associated with 20% of the thinning for the Prairie Falcon (*Falco mexicanus*), whereas Peakall et al. (1975) calculated comparable figures of 20 ppm for the Peregrine Falcon (*Falco peregrinus*), and Blus et al. (1972) estimated 8 ppm for the Brown Pelican (*Pelecanus occidentalis*). Kiff et al. (1979) confirmed that 5 ppm of DDE was correlated with 20% of the thinning for the California Condor (*Gymnogyps californianus*). Wiemeyer et al. (1984) reported that

Table 3. Geometric mean and range of organochlorine pollutants and heavy metals in Ciconiforme and Falconiforme eggs from Castile.

Species	n	-HCH	DDE	DDT	PCBs
W.S.	16	0.027 0.01-0.23	0.193 0.06-0.44	0.040 N.D.-0.16	0.434 0.25-1.16
B.K.	4	0.005 N.D.-0.02	0.167 0.12-0.22	0.063 0.01-0.27	0.303 0.20-0.40
B.E.	3	0.025 0.01-0.04	1.168 0.34-8.20	0.156 0.07-0.60	0.754 0.24-3.00
B.	2	0.010 0.01-0.01	0.200 0.13-0.31	0.025 0.02-0.03	0.614 0.54-0.70
P.F.	3	0.021 N.D.-0.14	9.752 2.60-65.8	0.172 0.10-0.35	1.247 0.82-2.11
R.E.	3	N.D.	0.258 0.06-0.68	0.064 0.05-0.08	0.206 0.14-0.32

Species	n	Hg	Cd	Pb	Cu	Zn
W.S.	16	0.11 0.09-0.26	0.07 0.04-0.16	0.93 0.39-2.80	1.41 0.72-4.26	9.30 6.21-19.2
B.K.	4	0.03 0.01-0.10	0.07 0.07-0.08	1.29 1.20-1.45	1.40 0.90-1.88	15.64 7.19-29.4
B.E.	3	0.13 0.10-0.18	0.07 0.04-0.13	1.04 0.32-1.95	1.55 0.84-2.88	10.37 8.29-13.0
B.	2	0.06 0.04-0.10	0.04 0.02-0.08	0.88 0.84-0.93	1.38 1.19-1.61	13.97 13.8-14.1
P.F.	3	0.25 0.09-0.48	0.06 0.05-0.07	1.23 0.78-1.62	1.11 0.89-1.42	12.50 8.76-16.7
R.E.	3	0.05 0.01-0.12	0.09 0.07-0.14	1.40 1.21-1.53	0.90 0.47-2.07	8.42 5.52-11.9

W.S. = White Stork, B.K. = Black Kite, B.E. = Booted eagle, B. = Buzzard, P.F. = Peregrine Falcon, R.E. = Royal Eagle; n = Number of Eggs; N.D. = Not Detected.

5 ppm of DDE was associated with 10% of the shell thinning for the Bald Eagle (*Haliaeetus leucocephalus*) and mean 5-yr production was near normal for breeding areas where eggs contained less than 3 ppm DDE. In 10.2% of the instances the levels reported in this study in Falconiformes are higher than those reported

by Kiff et al. (1979) and Wiemeyer et al. (1984); about 5.1% exceed those shown by Blus et al. (1972) and 2.5% are higher than those assumed by Keith and Grouchy (1972) and Peakall et al. (1975).

All of the 39 Falconiforme eggs analyzed contained residues of PCBs. Unexpected high levels of PCBs were present in one egg of the Booted Eagle (8.4 ppm) collected in Castile and in two eggs of the Black Kite (8.5 and 18.7 ppm) collected in Doñana. It is difficult to separate the effects of PCBs from those of DDE, therefore PCBs and DDE levels in eggs are commonly evaluated collectively; it seems that the PCBs effects are analogous to those produced by DDE on the reproductive mechanisms (Risebrough et al. 1968). DDE and PCBs appeared to interact in reducing breeding success of American Kestrels (Lincer 1972). Residues of other organochlorine pollutants generally ocurred much less frequently, or they were found at much lower levels, or they were below their limits of detection.

The effects of organochlorine pollutants on reproductive success of Ciconiforme birds have not been fully assessed, but in general Ciconiforme birds seem to be less sensitive to DDE than are Falconiforme birds. Henny et al. (1984) found that Black Crowned Night Heron clutch size and productivity decreased and the incidence of cracked eggs increased when DDE levels in eggs were above 8 ppm. Custer et al. (1983) calculated that 10 ppm DDE would cause a 50% decreased in hatching success for this species. All DDE levels reported in this paper in Ciconiformes are lower than those quoted by Henny et al. (1984) and Custer et al. (1983). Low nesting success was reported on Lake Ontario where DDE averaged 4.5-12.4 ppm and PCBs averaged 9.8-63 ppm (Price 1977); in all instances the DDE and PCBs levels reported in this study are considerably lower.

A "frequency index" (FI) was computed by Ohlendorf et al. (1979) by dividing the number of organochlorines detected in all eggs by the total possible ocurrences (number of compounds X number of eggs) to reflect the frequency of organochlorine residue ocurrence in bird eggs. The FI for the Spain eggs was 0.329 (250 detections/759 possible). This was higher than the reported of 0.202 in eggs from Colorado and Wyoming (WcEwen et al. 1984), 0.204 (Southern Atlantic), 0.180 (Inland), and 0.155 (Gulf Coast) (Ohlendorf et al. 1979) and lower than the reported index of 0.492 in eggs from the Great Lakes, and 0.374 (Northern Atlantic) (Ohlendorf et al. 1979).

The eggs of the White Stork, Black Kite, Booted Eagle, and Buzzard from Castile contain lower concentrations of total DDT (DDE + TDE + DDT) and PCBs than those of the same species collected in Doñana. These difference probably reflected the greater contamination of habitat since, although insecticide use is not permited in the Park, some of the agricultural practices in the surrounding areas require the use of insecticides. Likewise, the Guadalquivir River flows through a major urban and industrial centers before it enters the Park. Migrations do not seem to represent an important contribution to the levels of residues found in migratory Falconiformes, as the Black Kite or the Booted Eagle, given that similar levels occur in sedentary species, as the Buzzard, the Peregrine Falcon or the Royal Eagle.

All of the 69 egg contents analyzed contained residues of Hg, Cd, Pb, Cu, and Zn (Tables 2 and 3). In all cases Cd showed the lowest levels with a range from 0.02 (Buzzard, Castile) to 0.16 ppm (White Stork, Castile) followed by Hg with a range from 0.01 (Black Kite, Castile and Royal Eagle, Castile) to 0.48 ppm (Peregrine Falcon, Castile); Pb, range 0.32 (Booted Eagle, Castile) to 3.54 ppm (Black Kite, Doñana); Cu, range 0.47 (Royal Eagle, Castile) to 4.26 ppm (White Stork, Castile); and Zn, range 5.52 (Royal Eagle, Castile) to 29.4 ppm (Black Kite, Castile).

Mercury residues were generally below levels associated with decreased hatchability and nesting survival of other species of birds (Fimreite 1971, Vermeer et al. 1973, Heinz 1976). The eggs of the White Stork, Black Kite, Booted Eagle and Buzzard from Castile Plateau contain lower concentrations of mercury residues than those of the same species collected in the Doñana National Park. Most of the other metals (Cd, Pb, Cu, and Zn) were apparently present at background levels in our egg samples and interpretation of their biological significance is not possible. More integration of field and laboratory research will help determine whether these metals can pose a serious problem to the bird fauna.

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