Bull. Environ. Contam. Toxicol. (1990) 45:523–530

Bulletin of Environmental Contamination and Toxicology

Organochlorine Insecticide Residues in Birds and Bird Eggs in the Coastal Plain of Israel

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A large number of reports have appeared in the literature regarding insecticide residues in birds and bird eggs and their ecological impact on bird populations (see Edwards, 1975; Moriarty, 1975; Brown, 1978, for references). Generally, much larger quantities of insecticide residues were found in raptorial and fish-eating birds than in herbivorous and insectivorous species.

Residues of organochlorine insecticides and their metabolites in bird eggs have been implicated in the thinning of egg shells with a consequence of egg breakage or reproductive failure (Koeman et al, 1967; Ratcliffe, 1967; Wurster,1969; Peakall, 1970; Bitman, 1970; Cope, 1971; Jeffries and French, 1971; Moriarty, 1983, to mention only a few). In Israel (Mendelssohn, 1972; Mendelssohn and Paz, 1977; Mendelssohn et al. 1979) as in many other countries, certain insectivorous and omnivorous bird species have declined in numbers as a result of feeding on pesticide-contaminated food in heavily sprayed agricultural areas. Heavy losses were also recorded among several raptorial species due to secondary poisoning caused by frequent rodenticide applications to control the Levant vole *Microtus guentheri* (Mendelssohn, 1972).

The lack of sufficient information on the insecticide residues in birds and their eggs in our area prompted us to monitor several bird species for insecticide residues, particularly DDT and its metabolites, in an intensive agricultural area of the Coastal Plain. Polychlorinated biphenyls (PCBs), a group of industrial chemicals, were also analyzed in some samples because of their ubiquitous presence in the environment and their similarity to DDT in physical and chemical properties. Earlier accounts (prior to 1966) of the presence of large quantities of chlorinated hydrocarbon insecticides in birds may have been inaccurate, possibly due to the presence of PCBs which interfered with the analysis of these compounds (Jensen, 1966).

It was hoped that the information obtained would enable us to: (1) select key species that could be used effectively to monitor

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the insecticide contamination of birds, (2) determine whether residue levels were sufficiently high to warrant special investigation, and (3) use the data as a baseline for comparison in the assessment of insecticide residues in birds in future monitoring work.

MATERIALS AND METHODS

Eggs were collected for three seasons between 1975 and 1978 from an agricultural area along the coast and were brought to the laboratory the same day. Whenever possible, the eggs were processed the same day of arrival. Otherwise, the eggs were kept frozen until use. Birds were captured by gunshot and mist nets in the same areas where the eggs were collected and were brought to the laboratory the same day. The tissues intended for analysis were excised and extracted immediately or kept frozen until use.
The egg content was weighed and homogenized in a blender. Small eggs weighing 5 grams or less were used in their entirety. Five gram aliquots were taken from larger eggs. The homogenate was placed in a mortar together with sufficient anhydrous sodium sulfate (granular, residue brand) and ground to a dry flowing powder. The latter was transferred to a thimble and was extracted with a 1:4 mixture of acetone:hexane for 2 hrs in a Soxhlet apparatus. At the end of this period the solvent was decanted and concentrated to 1-2 mL in preparation for cleanup. Bird tissues were extracted and prepared for cleanup in the same manner as eggs

Cleanup of residues was made by the method described in FDA Manual (1971) using activated Florisil and elution with 6% diethyl ether in petroleum ether. PCBs were separated from organochlorine insecticides on silicic acid columns by the method of EPA (1980). Perchlorination of PCBs with $SbCl_5$ was made according to Berg et al. (1972) and Armour (1973) .The resultant decachlorobiphenyl was quantitated against Aroclor 1254 which was treated in the same manner, or against commercial decachlorobiphenyl (both obtained from Analabs, North Haven, CT, USA). $SbCl_5$ was purchased from Merck-Schuchardt, Munich, W. Germany. All other chemicals and solvents were of reagent grade and were obtained locally.

Statistical analysis was made by the Nested-Anova method (Sokal and Rohlf, 1969)

REEULTS AND DISCUSSION

The quantities of DDE found in bird eggs and in tissues of birds are given in Tables 1 and 2, respectively. Occasionally, small amounts of DDT were detected but these values are not reported. PCBs were found in practically all samples, but due to the qualitative nature of the analysis at this stage, these values are not reported. Without exception, DDE was the predominant compound found in all samples. A total of 459 eggs comprising sixteen bird species were analyzed. In addition, tissues of 75 birds of eleven species were examined for residues. The tissues included liver, heart, brain, muscle, skin and feathers.

Table 1. Residues of DDE in eggs of sixteen bird species

Species	No. of clutches	No. of eggs	DDE (ppm)[a] Range	Avg±SD	Food pref
Chukar (Alectoris chukar)	5	45	0.09-0.47	0.20±0.14	H
Feral pigeon (Columba livia)	4	8	0.10-1.15	0.57±0.44	G
Greenfinch (Carduelis chloris)	9	41	0.11-0.71	0.35±0.21	G
Turtle dove (Streptopelia turtur)	19	36	0.00-0.60	0.23±0.17	G
Palm dove (S. senegalensis)	26	48	0.00-0.25	0.04±0.06	G
Cattle egret (Bubulucus ibis)	11	36	0.31-1.30	0.62±0.38	O
Hooded crow (Corvus corone)	6	18	0.40-0.76	0.61±0.14	O
House sparrow (Passer domesticus)	18	79	0.00-0.85	0.28±0.22	O
Crested lark (Galerida cristata)	5	17	0.21-0.61	0.36±0.15	I/G
Yellow vent bulbul Pycnonotus xanthopygos	17	38	0.27-1.85	0.57±0.43	I/F
Blackbird (Turdus merula)	13	28	0.11-1.43	0.66±0.46	I/F
Graceful warbler (Prinia gracilis)	9	25	0.00-3.13	0.96±1.05	I
Kestrel (Falco tinnunculus)	3	11	0.30-0.67	0.46±0.15	C
Common tern (Sterna hirundo)	7	16	0.20-1.90	0.88±0.60	C
Little egret (Egretta alba)	10	28	0.17-6.26	1.61±1.43	P
Night heron Nycticorax nycticorax)	12	32	0.10-5.20	1.62±1.41	P

[a] DDE was calculated as Average±SD of the total No. of clutches.

Food preference: H--Herbivore; G--Granivore; O--Omnivore; I--Insectivore; I/G--Insectivore/Granivore; I/F--Insectivore/ Fructivore; C--Carnivore; P--Piscivore.

As Table 1 shows, the highest amounts of DDE were found in the Kestrel and Common tern (carnivores), and in the fish-eating birds Little egret and Night heron. This is to be expected since these species are high up in the food chain where bioaccumulation of pesticide residues is of common occurrence. The Graceful warbler, a tiny bird with a voracious appetite for insects also had a high level of DDE. The lowest amounts of DDE were found in eggs of Chukar, a herbivore, and in those of Palm dove and Turtle dove , both granivores.

Table 2. Statistical analysis of the variation in DDE content of eggs among species, clutches within species, and eggs within a clutch of the same species

Variance Component	Level	SS	DF	MS	FS	P
Among species	2	79.058	15	5.270	5.582	<0.001
Clutches within species	1	122.748	157 140.7*	0.781 0.944*	4.899	<0.001
Eggs within clutch	0	63.314	332	0.190		

*Corrected for unequal sample size (Sokal and Rohlf, 1969).

Analysis of variance components

Variance component	Level	Variance	% Variation
Among species	2	0.139	25.97
Clutches within species	1	0.207	38.59
Eggs within clutch	0	0.190	35.43

Statistical analysis of the results (Table 2) showed highly significant differences in DDE content among the different species and among egg clutches of the same species. Analysis of variance components (Table 2) indicated that 35% of the total variation was due to differences among eggs of the same clutch; 39% was due to differences among clutches within a species; and 26% was due to differences among the different species.

The analysis of bird tissues (Table 3) showed that on the average, the heart contained the highest amounts of DDE, followed by liver, brain and muscle. Actually , skin and feathers contained higher amounts of DDE but the number of samples analyzed was small in comparison. This is not surprising since heart tissue and skin contain large amounts of fat where organochlorine insecticides are easily stored. In some instances, PCBs were also found in heart and liver tissues but these were analyzed only qualitatively, hence, these data are not reported.
Most consistent results were found in liver which is the site of detoxification of many lipid-soluble organic compounds. As

expected , muscle tissue contained the lowest amounts of residues and this is consistent with the low fat content of this tissue. It must be emphasized that these quantities of DDE are in no way harmful to the birds since birds which survived a DDT-treatment in the laboratory contained much larger amounts of DDT, DDE and DDD in their tissues (Stickel et al, 1966; Hill et al, 1971; Stickel, 1973). However, one must be cognizant of the fact that even low levels of DDE over a long period of exposure can affect eggshell thickness and cause egg breakage or reproductive failure.

Table 3. DDE residues in various tissues of eleven bird species collected in an agricultural area of the Coastal Plain.

SPECIES	n	LIVER	HEART	BRAIN	MUSCLE	SKIN and FEATHERS
Cattle egret	5	.47±.14	.48±.06	.39±.18	.48±.18	.56±.11
Crested lark	12	.45±.07	.68±.24	.38±.17	.13±.02	.45±.18
Yellow vent bulbul	11	.36±.07	-	.41±.26	.11±.03	1.40±1.10
Greenfinch	5	.39±.12	-	.11±.04	.09±.07	-
Hooded crow	7	.21±.08	.41±.04	.25±.05	.12±.04	-
Blackbird	5	.30±.09	.35±.18	.18±.05	.17±.04	-
House sparrow	10	.34±.10	.55±.19	.29±.07	.17±.06	-
Palm dove	4	.40±.20	.25±.10	.18±.06	.03±.02	-
Turtle dove	6	.30±.10	.39±.18	.37±.15	.03±.02	.32±.15
Feral pigeon	5	.24±.05	-	.06±.06	.04±.04	-
Graceful warbler	5	whole body .15±.06				

More recently , we analyzed eggs of two species of fish-eating birds, i.e., the Night heron and Little egret. The results are given in Table 1 for DDE and in Table 4 for PCBs. It is evident that larger quantities of DDE and PCBs were found in these species since their food, consisting mainly of carp and Tilapia fish, contains substantial amounts of these compounds (Perry et al.1983)

The intensive agricultural practices of the relatively small arable land area of Israel favor the outbreak of numerous pests and, consequently, the use of increasing amounts of pesticides. Contamination of the local environment by pesticides over the past 3-4 decades has had some subtle effects on bird populations and on other wildlife .

Table 4. Residues of PCBs in eggs of two fish-eating bird species

Species	No. of clutches	No. of eggs	PCBs (ppm)	
			Range	Avg±SD
Little egret (<u>Egretta alba</u>)	6	8	0.11-0.98	0.54±0.28
Night heron <u>Nycticorax</u> <u>nycticorax</u>)	7	10	0.12-2.84	0.77±0.64

The detrimental effects of pesticides on certain bird population densities in Israel have been well documented (Mendelssohn, 1972; Mendelssohn and Paz, 1978). On the other hand, following long periods of widespread insecticide applications, several bird species such as the Bulbul, the Blackbird, and the House sparrow displayed a considerable increase in numbers to the point of their becoming pests of agriculture. In particular, due to secondary poisoning by repeated use of the rodenticide thallium sulfate, the disappearance of the Sparrow hawk (<u>Accipiter nisus</u>), a raptorial species which feeds mainly on other birds and on field mice, resulted in an abundance of blackbirds and bulbuls in many areas as a consequence of reduced predation (Mendelssohn, 1972). There also exists the possibility that the above three species have become tolerant or resistant to insecticides as they continue to live, feed and breed in agricultural areas heavily treated with insecticides.

Biochemical investigations on the above three species (Yawetz et al. 1978a, 1978b, 1979) revealed the presence in their livers of substantial amounts of cytochrome P-450, an enzyme involved in the metabolism of various lipophilic insecticides. This adds support to the possibility of these species becoming more tolerant to insecticides other than DDT.

The results of this monitoring project show that DDT, DDE, and PCBs are present in avian species in the Israeli environment. The predominant compound was DDE. The presence of PCBs in eggs and tissues of most bird species analyzed (albeit qualitatively in most samples) is of great concern since little is known about the effects of PCBs on the physiology of avian reproduction

This is the first purposeful monitoring project of this kind in Israel taking into account several species of birds of varying food habits. These data can be used as a guideline and a baseline for future monitoring work. We recommend the bulbul as the most suitable species for future monitoring work for the following reasons: The bulbul is a territorial bird, quite stable, has a small number of eggs per clutch (usually 3) which is a desirable parameter in monitoring work, the eggs are large (approx.3.5 g) in proportion to body weight (approx. 35 g), lives a good number of years relative to palearctic birds, the nests are easy to find, and it is widespread throughout the country (Hasson ,1978).

Other suitable species are the Blackbird, the Graceful warbler and the Barn owl. The mixed function oxidase system of the Barn owl (Tyto alba) and its induction by PCBs has been extensively investigated in our laboratory (Rinsky and Perry, 1981, 1983) and we find this species, as a representative of the carnivores, to be most suitable for monitoring work.

Acknowledgments. The present investigation was supported by a grant from the Environmental Protection Service, Ministry of the Interior, Israel.

REFERENCES

Armour JA (1973) A quantitative perchlorination of polychlorinated biphenyls as a method for confirmatory residue measurements and identification. J Assoc Off Anal Chem 56: 987-993.

Berg OW, Diosady PL, Rees GAV (1972) Column chromatography separation of polychlorinated biphenyls from chlorinated hydrocarbon pesticides and their subsequent gas chromatographic quantitation in terms of derivatives. Bull Environ contam Toxicol 7: 338-347.

Bitman J, Cecil HC, Fries GE (1970) DDT-induced inhibition of avian shell gland carbonic anhydrase: A mechanism for thin eggshells. Science 168: 594-596.

Brown AWA (1978) Ecology of pesticides. Wiley-Interscience, John Wiley & Sons, New York.

Cope OB (1971) Interactions between pesticides and wildlife. Ann Rev Entomol 16:325-364.

Edwards CA (1973) Persistent pesticides in the environment, 2nd ed CRC Press Inc, Cleveland, Ohio.

EPA (1980) Manual of analytical methods for the analysis of pesticides in humans and environmental samples. Watts RR ed. U.S. Environmental Protection Agency, Health Effects Research Laboratory, Environmental Toxicology Division, Research Triangle Park, North Carolina, Section 5, pp 1-19.

FDA (1971) Pesticide Analytical Manual Vol 1. Food and Drug Administration, U.S. Dept HEW, Washington, D.C.

Hill EF, Dale EW, Miles JW (1971) DDT intoxication in birds; subchronic effects and brain residues. Toxicol Appl Pharmacol 20: 502-514

Jeffries DJ, French MC (1971) Hyper and hypothyroidism in pigeons fed DDT: an explanation for the thin egg shell phenomenon. Environ Pollut 1:235-242.

Jensen S (1966) Report of new chemical hazard. New Scientist 32:612

Koeman JH, Oudejans RCHM, Huisman EA (1967) Danger of chlorinated hydrocarbon insecticides in bird eggs. Nature 215:1094-1096.

Mendelssohn H (1972) Effect of pesticides on bird life: The impact of pesticides on bird life in Israel. Bull Int'l Council for Bird Preservation.

Mendelssohn H, Paz U. (1977) Mass mortality of birds of prey caused by Azodrin, an organophosphorus insecticide. Biol Conservation 11:163-170.

Mendelssohn H, Schlueter P, Aderet Y (1979) Report on Azodrin poisoning of birds of prey in the Huley Valley in Israel. Int'l Council for Bird Preservation 13:124-129.
Moriarty F (1975) Organochlorine Insecticides: Persistant Organic Pollutants. Academic Press Inc. (London) LTD.
Moriarty F (1983) Ecotoxicology. Academic Press Inc. (London) LTD
Peakall DB (1970) p,p'-DDT: Effect on calcium metabolism and concentration of estradiol in the blood. Science 168:592-594.
Perry AS, Gasith A, Mozel Y (1983) Pesticide residues in fish and aquatic invertebrates. Arch Toxicol Suppl 6:199-204.
Ratcliffe DA (1967) Decrease in eggshell weight in certain birds of prey. Nature (London) 215:208-210.
Sokal RR, Rohlf FJ (1969) Biometry. The principles and practical statistics in biological research. W. H. Freeman, San Francisco.
Stickel LF (1973) Pesticide residues in birds and mammals. In Edwards CA (ed) Environmental Pollution by Pesticides, Plenum Press London, pp 254-312.
Stickel LF, Stickel W, Christensen R (1966) Residues of DDT in brains and bodies of birds that died on dosage and in survivors. Science 151:1549-1551.
Wurster CF (1969) Chlorinated hydrocarbons and avian reproduction. How are they related? In Miller MW, Berg GG (eds) Chemical Fallout, Charles Thomas, pp 368-389.

Received November 1, 1989; accepted February 15, 1990.