Effects of DDT on Bobwhite Quail (*Colinus virginianus*) Adrenal Gland

by

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INTRODUCTION

There has been considerable research to determine some of the lethal effects of chlorinated hydrocarbon pesticides (SCHAFER 1972, TUCKER and HAEGELE 1970), but knowledge of the sublethal effects are also of importance. Potential subtle endocrine influences of these pesticides could alter the population dynamics of the species affected (FRIEND AND TRAINER 1970).

Chlorinated hydrocarbon pesticide residues occur in almost all birds analyzed in many countries (STICKEL 1968). Several studies have shown that sublethal levels of pesticide ingestion can reduce reproductive efficiency in avian species (AZEVEDO et al. 1965, COOKE 1973, DEWITT and GEORGE 1960, GENELLY and RUDD 1956, HEATH et al. 1969, WURSTER 1969).

Other reports indicate that chlorinated hydrocarbons may have a definite effect on the endocrine system of avian species. White leghorn cockerels (Gallus sp.) fed DDT at 100ppm for 32 weeks showed marked reduction in both comb and testicular development (ECOBICHON and SASCHENBRECKER 1968). Ovulation time of Bengalese finches (Lonchura striata) was delayed progressively in relation to magnitude of DDT (11 to 54ppm) dosage (JEFFERIES 1967). Ringdoves (Streptopelia risoria) also showed that chlorinated hydrocarbons cause significant delay in ovulation time (PEAKALL 1970). Pigeons (Columba livia) fed a diet containing p,p-DDE had increased adrenal weights (JEFFERIES and FRENCH 1972). Adult male mallard ducks fed diets

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containing 2.5 to 250ppm DDT did not demonstrate any significant changes in adrenal weights or a change in the percentage of cortex (NAUMAN 1969). A diet of 265ppm DDT fed to 6-week-old male and female turkeys (Meleagris sp.) for 7 and 15 weeks, did not cause any discernible gross histological alterations of the testes, ovary or thyroid (SIMPSON et al. 1972). The functional state of the adrenals in birds may change without subsequent change in adrenal weight (DE ROOS 1963). The organisms response to pollutants is species specific, and is related to a great number of environmental variables as well as the physiological state of the individual at the time of exposure (GISH and CHURA 1970, TUCKER and HAEGELE 1970).

The objective of this research was to determine if feeding sublethal levels of DDT to bobwhite quail, would cause any detectable histological alterations in their adrenal glands.

PROCEDURES

Data discussed in this paper is a portion of an investigation to determine the metabolic effects of DDT on bobwhite quail (LUSTICK et al. 1973, VOSS 1971).

Quail for this research were obtained from a game farm at Urbana, Ohio. Each treatment group of 7 birds was maintained outdoors in separate wire enclosures (915cm x 1070cm x 1425cm).

The quail were 28 days old (30 June) when placed on a diet containing 0, 10, 50, 150ppm technical grade DDT (Diamond Shamrock Chemical Co., Houston, Texas, 77015)® The technical grade DDT was dissolved in Mazola® corn oil and mixed 2:98 with Purina® game bird startena (medicated) SQ. Food and water were given ad libitum.

The birds were sacrificed after being fed DDT for 242 days (27 February). There were no fatalities during this period. The quail remains were placed in plastic bags and stored in a freezer at -20°F for a short period of time before dissection. Later, tissues required for gas-liquid chromatography and histological examination were excised. An electron capture gas-liquid chromatography technique was used to determine the DDT residues in the brain tissues (BARRY et al. 1965, VOSS 1971).

The adrenals from each bird were placed in Bouin-Dubosco fixative (HUMASON 1967) for 24 hours, and then stored in 70 percent ethyl alcohol until all the adrenal tissues were embedded for sectioning. Among 20 species of birds (exclusive of chicken, pigeon, and duck) there was no sex difference in the size of the adrenals (HARTMAN and BROWNELL 1949). In domestic fowl there was no significant difference in the size of the right and left adrenals and cortical and medullary tissue are intimately mixed together with about the same proportion in all parts of the gland (SAUER and LATIMER 1931). There is no significant difference in the cortical medullary ratio between adrenals from the same bird (FLICKINGER 1961). Therefore, only one adrenal from each quail was selected for sectioning.

The center longitudal portion of the adrenals were sectioned 6 to 8μ thick and stained with Masson trichrome stain (HUMASON 1967). Some modification in staining times were made: for Delafield's hematoxylin 1/2 to 1 minute, and for Masson A 4 minutes. The lighter staining cells with a deep staining nuclei were identified as cortical (interrenal) tissue, medullary (chromaffin) tissue was darker staining (BRADLEY 1950, JONES et al. 1958, STURKIE 1965).

A 10 x 10mm square grid micrometer was inserted in a 10X ocular lens of a binocular compound microscope (American Optical Corp. Buffalo, New York 14215). Using a 43X objective lens (430X magnification) the type of tissue (cortex, medullary, or other) at the intersecting points of the grid was recorded (CHAMOT and MASON 1938, SWINGLE and REMINGTON 1944).

Four sections of an adrenal from each quail were counted and readings were taken from the top, middle, and bottom of each section, giving a total of 12 readings for each quail adrenal (1200 intersecting points).

RESULTS AND DISCUSSION

The DDT metabolite most commonly found in the brain tissues by gas-liquid chromatography was p,p'-DDE, except for the birds fed 150ppm DDT; in these, about one-third of the total residue was p,p'-DDT (Table 1). There was a gradual increase of DDE residues in the brain tissue as the DDT diet level increased. The LD_{50} of technical DDT for bobwhite quail ranges from 1170 to 1610ppm (HILL et al. 1971, VOSS 1971).

Table 1. The effects of sublethal DDT diet on brain residues, adrenal/body weight ratio, and cortical/medullary ratios of bobwhite quail.

DDT in diet (ppm)	Sex	Mean DDT* residues in brain tissues. ($\bar{X} \pm$ S.D. ppm)	Adrenal/body wt. % (gms) X 100 (paired adrenals)	Cortical/medullary $\bar{X} \pm$ S.D. (4 sections/bird)
0	M	0	0.3	1.15 ± 0.47
0	M		0.6	1.39 ± 0.44
0	F		0.8	1.36 ± 0.87
0	F		0.8	1.13 ± 0.55
10	M	0.33 ± 0.20	0.6	1.76 ± 1.11
10	M		1.0	0.54 ± 1.84
10	F		0.9	1.16 ± 0.60
10	F		0.1	1.67 ± 0.58
50	M	7.50 ± 2.39	0.2	3.11 ± 1.25
50	M		0.8	1.44 ± 0.64
50	M		1.2	1.66 ± 0.68
50	F		0.3	1.44 ± 0.33
50	F		0.9	2.86 ± 1.07
50	F		0.3	2.08 ± 1.39
50	F		0.8	1.69 ± 0.58
150	M	13.95 ± 3.76	0.1	2.72 ± 1.47
150	M		1.0	2.38 ± 1.35
150	F		0.7	2.27 ± 1.62
150	F		0.7	2.55 ± 1.22

* All brain residues were p,p'-DDE except the 150ppm group, which was 10.88ppm p,p'-DDE and 3.07ppm p,p'-DDT.

Paired adrenal weight expressed as a percentage of total body weight was calculated for each bird (Table 1.). A least squares linear regression and analysis of variance was performed on the male and female data separately to compare the percentage of paired adrenal weight to the DDT diet. No significant difference was found ($P > 0.99$).

The histological examination of the adrenal sections indicated that as the DDT diet level increased there was a subsequent increase in the cortical/medullary ratio (C/M, Table 1). A least squares linear regression and analysis of variance was performed on the male and female cortical medullary ratios separately. The values for the male data were: $Y=1.322 + 0.009X$; $r^2= 0.440$, $P < 0.10$. The values for the female data were: $Y=1.435 + 0.007X$; $r^2= 0.481$, $P < 0.05$. The male and female cortical/medullary ratios were not combined because in chickens there is approximately equal amounts of cortical and medullary tissue in males but slightly more cortical tissue in females (OAKBERG 1951).

The increase in adrenal cortical tissue without any apparent increase in adrenal weight may indicate hypotrophy of the medullary tissue, but supporting evidence is lacking. The action of certain toxic substances can result in adrenal cortical hypertrophy with increased secretion of cortical hormones. These corticosteroids have an effect on the gonads, as shown by the masculinizing effects of cortical tumors of the adrenal, reported in hens (HÖHN 1961). Administration of adrenal corticosteroids (cortisone) to laying birds decreases shell deposition (URIST and DEUTSCH 1960). In young cockerels treated with technical DDT, there was reduced concentrations of corticosteroids in adrenals and in plasma (SREBOCARN et al. 1971).

Daily intramuscular injections of adrenaline into house sparrows (Passer domesticus) of both sexes, suppressed the development of gonads, accessory and secondary sex characters (bill color in male) of the normal breeding cycle (PERRY 1941). Similar results were also found in male domestic fowl (WHEELER et al. 1942).

SUMMARY AND CONCLUSIONS

A wide range of responses to sublethal levels of DDT exist, many of which are species specific and vary within each species depending upon age, sex, and

physiological state. Sublethal levels of DDT do cause an increase in the adrenal cortical tissue of bobwhite quail, which may cause increased secretion of corticosteroids, and in turn affect reproduction. A delicate homeostatic balance exists within the avian endocrine system which may be disturbed by feeding sublethal levels of chlorinated hydrocarbon pesticides. This adverse effect on the endocrine system may cause subtle reproductive failures which go unnoticed until the population is greatly reduced.

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