CONCENTRATIONS OF SELECTED CHLORINATED HYDROCARBON INSECTICIDES IN BOBWHITE QUAIL IN SOUTH CAROLINA¹

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Chlorinated hydrocarbon insecticides have been the subject of considerable controversy. DDT, the most controversial insecticide, and other chlorinated hydrocarbons have been used extensively until recently when their uses were altered by state and federal legislative and judicial actions. The use of these materials has declined but the problem of environmental pollution still exists because of the persistence and ubiquity of these "hard" pesticides.

Insecticides have been implicated as being the causative factor in the decline of some predatory bird populations. The concern herein is not with a species at the top of a food chain but with the bobwhite quail (*Colinus virginianus*. L.), a favorite game bird which consumes primarily plant material. As a farm game species, the bobwhite is compatible with agriculture and in many instances exists in high populations on farmed areas. Consequently, during all stages of its life history this bird and its food are subjected to direct applications of pesticides of many kinds.

The bobwhite quail may serve as an indicator of the presence of certain persistent insecticides in the areas tested. A decision was made to determine residues of DDT in six tissues and crop contents of bobwhite quail collected from eight areas which had been exposed to various degrees of insecticide application.

METHODS

One hundred seven bobwhite quail from eight areas within South Carolina were collected by shooting (Figure 1). Four areas were primarily agricultural lands (Goat Island, a Hampton County farm, a Holly Hill farm, and Tupelo farm). Major crops produced on these lands were cotton (Gossypium hirsutum L.) and soybean (Glycine max (L.) Merr.). Four additional areas were included because of the limitations on the use of pesticides. Pesticides had never been used on Hobcaw Plantation. Francis Marion National Forest was treated with mirex in 1969 for control of the imported fire ant (Solenopsis saevissima richteri Forel) and BHC had been used for control of the Southern pine beetle (Dendroctonus frontalis Zimmerman). Pesticides had not been used on Groton Plantation (Goosepond and Jointstock collection sites) for 10 years prior to quail collection during the 1969-70 South Carolina hunting season.

Individual specimans were frozen intact in plastic bags as soon after collection as possible. Specimens were transported to the laboratory on ice and placed in a freezer at 0° C until they were removed for thawing and excising of various tissues. Prior to insecticide analysis, crop contents were analyzed to determine volume and frequency of each food item represented therein.

Six tissues-fat, liver, kidney, brain, breast, and gonads—and crop contents were removed from each bird, weighed, freeze dried, weighed again, and ground with mortar and pestle prior to extraction of insecticide. Samples were extracted directly with hexane with the exception of fat samples which were extracted with

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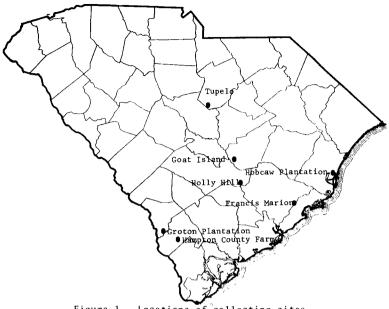


Figure 1. Locations of collecting sites

petroleum ether. Extracts were passed through Florosil (R) columns to remove impurities. The eluants were evaporated to dryness under vacuum and adjusted to 5 ml with nanograde hexane and 5 ul aliquots were injected into the gas chromatograph for analysis.

A ⁶³Ni electron capture detector was utilized to determine insecticide residues. Separations and confirmations were made on 6 ft. X $\frac{1}{4}$ inch U-shaped glass columns packed with the following materials: 10% DC-200 on Gas Chrom Q (80/100), 10% U.C. W-98 on Chromosorb W (60/80), 2% OV-101 and 3% QF-1 on Gas Chrom Q (80/100), and 5% OV-210 on Gas Chrom Q (80/100).

(Goosepond and Jointstock collection sites) for 10 years prior to quail collection

Operating parameters of the gas ch	romatograph were:
Injection port temperature:	220°C
Column temperature:	180° C
Detector Temperature:	275°C
Carrier gas:	High purity nitrogen
Carrier flow:	100 cc/min.
Detector operating conditions:	DC mode
Electrometer:	Pulsed mode 60/6
	Input attenuator 10 ²
	Output attenuator 8, 16, 32

A profile was performed periodically to allow adjustment of the detector to operate at its most efficient voltage output.

Standards utilized in this study were prepared from analytical reagents. Levels of p,p'-DDT, p,p'-DDD, and p,p'-DDE less than 0.01 ppm were recorded as a trace. Recovery tests were performed for each tissue but values given herein were not corrected for the recovery rate. Recovery rates by tissue for p,p'-DDE are as follows: fat, 79%; brain, 80%; breast, 84%; gonad, 80%; and kidney, 86%. All concentrations were recorded on a dry weight basis. The conversion factors for determining wet weight concentrations are as follows: fat, 0.77; brain, 0.24; liver, 0.30; breast, 0.29; gonad, 0.31; and kidney, 0.27. No conversion factor was calculated for crop contents since there was so much variation in food items between individual samples.

RESULTS AND DISCUSSION

Importance of food items based on volume appeared to be a function of availability. In primarily agricultural areas (Table 1) agricultural crops inevitably occurred in the diet of quail. Quail collected from Tupelo, Holly Hill, and Hampton County areas showed a strong dependence upon agricultural crops, mainly soybean and corn)*Zea mays* L.). These areas were also prime examples of "clean farming" wherein all but low areas or forested areas were tilled. Sweetgum (*Liquidambar styraciflua* L.) and pine (*Pinus* spp.). mast were also used as food on these areas. The Goat Island area had more cotton than other areas and birds collected near cotton fields did not have as many agricultural crops upon which to feed. Additionally, an attempt was made on the Goat Island area to leave edges for quail and to burn woodlands for increased quail production. Although the Goat Island area was strictly an agricultural area the quail were not as dependent upon planted fields for food as they were on the other study areas.

The relationship between high percentages of food items by volume and apparent availability was also seen in the food habit data of birds collected form the non-agricultural areas studied (Table 2). Birds from Goose Pond and Jointstock divisions of Groton Plantation showed a preference for native legumes and cultivated crops. As the plantation was managed intensively for bobwhite quail, these food items would be expected to be utilized by the quail. Birds collected on Francis Marion National Forest and Hobcaw Plantation consumed more mast crops since cultivated crops were not available.

	total volum	ie		
Food Types	GI	Т	нн	HC
Mast	66	13	2	38
Native Legumes	18	3	1	2
Cultivated Crops	8	80	96	59
Others	8	4	1	1

 Table 1. Crop contents of bobwhite quail collected from four agricultural areas in South Carolina.

GI - Goat Island; T - Tupelo Farm; HH - Holly Hill Farm; HC - Hampton County Farm.

	percentage of	percentage of total volume			
Food types	GP	J	FM	Н	
Mast	9	3	47	17	
Native Legumes	42	58	47	77	
Cultivated Crops	46	30	0	0	
Others	3	9	6	6	

Table 2.Crop contents of bobwhite quail collected from four non-agricultural
areas in South Carolina.

Legumes consistently occurred in high volumes and frequencies. Beggar lice (Desmodium spp.), partridge pea (Cassia fasciculata Michx), milk pea (Galactia spp.), and bicolor lespedeza (Lespedeza bicolor Turez) appeared to be the important foods common to all eight study areas. Mast apparently was utilized to a great extent when available or when other foods were not available. Comparison of utilization of mast crops between agricultural areas (Goat Island, Hampton County, Hooly Hill, and Tupelo), forested areas (Francis Marion and Hobcaw), and Groton Plantation was of interest. Mast crops were least utilized on the Groton areas where a wide range of foods, both cultivated and native, was available to the birds. From these data, legumes and mast from sweetgum, oak (Quercus spp.), and pine may be assumed to be the staple winter foods on the eight study areas. Where available, cultivated crops were heavily utilized.

From the data collected very little can be said of the relationship between availability and consumption of animal material. The greatest occurrence of animals in the diet was observed in quail collected from Groton and Hobcaw Plantations, the areas of greates and least habitat diversity respectively.

Gas chromatographic detection of p,p'-DDT, p,p'-DDD, and p,p'-DDE was accomplished. Mirex was detected in fat, liver, brain and breast of quail from Francis Marion National Forest which was the only area studied that was included in the 1969 fire ant eradication program utilizing mirex as a toxicant. Several compounds which were suspected to be present in tissues of quail but were not verified were BHC, heptachlor, heptachlor epoxide, aldrin, and dieldrin. Discussion of resultst therefore will be concerned only with residues of p,p'-DDT, p,p'-DDD, p,p'-DDE, and mirex.

DDT was found in all tissues and crop contents from all areas studied with the exception of brains of birds collected from Hobcaw Plantation (Table 3). Quail from agricultural areas had higher concentrations of total DDT (the sum of residues of p,p'-DDT, p,p'-DDD, and p,p'-DDE) than those from areas upon which insecticides had not been used. The most commonly occurring and abundant DDT metabolite was p,p'-DDE.

The mean concentration of total DDT in all tissues from the Hampton County area was significantly greater (P=0.05) than from all other study areas (Table 4-A). The mean concentration in quail tissues from Tupelo and Goat Island was significantly greater (P=0.05) than from all others except the Hampton County farm. Residues in tissues from Goose Pond, Joinstock, Holly Hill, Francis Marion and Hobcaw were not significantly different.

No significant difference in mean concentrations of p,p'-DDT (Table 4-B) or p,p'-DDD (Table 4-C) were found to exist among quail from the study areas. However, there were significant differences among concentrations of p,p'-DDE in quail from the study areas (Table 4-D). Birds from the Hampton County area had significantly greater concentrations of p,p'-DDE than those from the other areas and birds from Tupelo and Goat Island had significantly greater concentrations than those from Goose Pond, Holly Hill, Jointstock, Hobcaw, and Francis Marion National Forest.

The fact that there were significantly higher insecticide concentrations in birds from agricultural areas where pesticides had been used was of interest. Birds collected from the Hampton County farm contained consistently higher concentrations of total DDT and were more dependent upon agricultural crops than those from any other areas. Because of a limited food supply bobwhite quail possibly may have consumed more insecticides by being forced to feed on waste grain and seed that were lying on contaminated soils. Quail on the Goat Island area, for example, were exposed to aerial applications of pesticides used on cotton and soybeans but because other foods and cover were available, they were not as dependent upon the treated fields as were birds from the Hampton County farm.

Hobcaw Plantation and Francis Marion National Forest were relatively free of total DDT contamination as compared to the other areas. Both areas were quite remote and, having had no history of DDT usage, demonstrated the ubiquitous nature of that environmental contaminant. Although chlorinated hydrocarbons have not been used on Groton Plantation since 1960, DDT contamination was found in tissues of quail from Goose Pond and Jointstock divisions. The county in which Groton Plantation is located was heavily farmed and this fact may account for some of the contamination.

A significant difference (P=0.05) in mean concentrations of mirex residues in quail collected from the Francis Marion National Forest was determined by the Duncan's Multiple Range Test (Table 4-E). A mean residue concentration of 0.79 ppm was significantly different from the trace concentration in birds from the Hampton County area. No mirex was detected in tissues of quail from any of the other areas. The Francis Marion National Forest was included in the 1969 imported fire ant eradication program in which mirex was utilized as a toxicant. The Hampton County farm was not included in the eradication program but was close enough so that contaminated birds could have moved into the area.

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Table 3.

			Ŵ	Mean concentration (ppm	ttion(ppm)		
Location	Fat	Liver	Gonads	Kidneys	Brains	BreastCroj	BreastCrop Contents
Agricultural Areas	13.72	4.76	2.82	2.01	1.61	1.22	2.65
Goat Island	14.29	3.15	1.80	0.98	0.45	0.09	2.82
Hampton County	10.71	11.98	8.97	7.82	8.57	7.20	8.74
Holly Hill	2.52	2.91	0.11	0.59	0.14	0.02	0.06
Tupelo	19.73	3.96	1.68	0.95	0.14	0.11	0.36
Non-agricultural Areas	1.15	1.16	0.12	0.25	0.06	0.03	0.30
Francis Marion	0.22	0.25	0.02	0.03	0.01	0.02	0.58
Goose Pond	3.41	2.68	0.47	0.52	0.21	0.06	0.14
Hobcaw	0.25	0.48	TR	0.07	0.00	TR	0.11
Jointstock	1.69	1.79	0.11	0.45	0.06	0.03	0.20
MEAN	6.41	2.64	1.21	0.97	0.75	0.52	1.28

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Location	Mean Residues ((ppm))
A. Total DDT.			
Hampton County Farm**	9.14	a*	
Tupelo**	3.93		b
Goat Island**	3.37		b
Goose Pond	1.08		C
Holly Hill**	0.95		C
Jointstock Francis Marion	0.62		C
Hobcaw	0.16 0.13		(
3. p,p'-DDT.			
	0.27		
Goat Island** Francis Marion	0.27 0.05	a a	
Holly Hill**	0.03	a	
Tupelo**	0.03	a	
Hampton County Farm**	0.02	a	
Jointstock	0.02	a	
Goose Pond	0.01	а	
Hobcaw	0.01	а	
C. p,p'-DDD.			
Tupelo**	0.40	а	
Goat Island**	0.08	а	
Francis Marion	0.02	а	
Hampton County Farm**	TR	а	
Jointstock	TR	а	
Hobcaw	TR	a	
Goose Pond Holly Hill**	TR TR	a a	
D. p,p'-DDE.			
Hammton County Form**	9.11	0	
Hampton County Farm** Tupelo**	3.50	а	b
Goat Island**	3.02		b
Goose Pond	1.07		U
Holly Hill**	0.91		
Jointstock	0.60		
Hobcaw	0.12		
Francis Marion	0.09		
E. Mirex.			
Francis Marion	0.79	а	
Hampton County Farm**	TR		b
Tupelo**	0.00		b
Goat Island**	0.00		b
Goose Pond	0.00		b

Table 4.Significant differences (P=0.05) of insecticide residues in bobwhite
quail collected from eight locations in South Carolina.

Location	Mean Residues (ppm)			
Hobcaw	0.00	Ъ		
Holly Hill**	0.00	b		
Jointstock	0.00	b		

*Means followed by different lower case letters are significantly different (P=0.05).

**Denotes agricultural areas.

Significantly higher concentrations (6.41 ppm) of total DDT were found in the fat tissues with residues in liver (2.64 ppm) also being significantly greater than concentrations in crop contents, gonads, kidney, grain, and breast tissues (Table 5-A). Significantly greater concentrations of p,p'-DDT were found in crop contents than in the tissues although only a comparatively small amount (0.47 ppm) was located in the crop contents. Fat contained 0.01 ppm p,p'-DDT but this concentration was found not to be statistically different from trace concentrations in gonads, breast, and brain, or from no detectable residues of p,p'-DDT in liver and kidney (Table 5-B).

Concentrations of p,p'-DDD (Table 5-C) were detected only in fat (0.33ppm), crop contents (0.15 ppm), and gonads (trace concentrations).

Fat and livers of bobwhite quail analyzed contained greater concentrations of p,p'-DDE than any of the other tissues (Table 5-D). However, all tissues contained at least some p,p'-DDE with the lowest mean concentration of 0.52 ppm in breast tissue and highest mean concentration of 6.07 ppm in fat.

Crop contents contained significantly greater concentrations of p,p'-DDT than the tissues and also contained p,p'-DDD. Although a certain amount of post-mortem breakdown of DDT has been demonstrated (Jefferies and Walker, 1966), the evidence herein indicated the metabolism of DDT to its more stable metabolite p,p'-DDE. The liver, the suspected site of DDT metabolism (O'Brien, 1967), contained no detectable concentrations o p,p'-DDD or p,p'-DDT but contained significantly greater concentrations of p,p'-DDE than any of the other tissues analyzed with the exception of fat. All other tissues, except the kidney, contained at least trace concentrations of p,p'-DDT, but only the crop contents and the highly lipoidal tissues, gonads and fat, contained detectable concentrations of p,p'-DDD (Table 5).

Table	5.	Significant	differences	(P=0.05)	of	insecticide	residues	ın	S1X
		tissues and cr	op contents o	of bobwhit	e qu	ail collected	in South	Car	olina.

Tissue	Tissue Mean	(ppm)		
A. Total DDT.				
Fat	6.41	a*		
Liver	2.64		b	
Crop contents	1.28			C
Gonad	1.21			C
Kidney	0.97			c
Brain	0.75			C
Breast	0.52			Ģ

Tissue	Tissue Mean (ppm)				
B. p,p'-DDT.					
Crop Contents Fat Gonad Breast Brain Liver Kidney	0.47 a 0.01 0.01 TR TR 0.00 0.00	Ե Ե Ե Ե Ե			
C. p,p'-DDD.					
Fat Crop Contents Gonads Liver Kidney Breast Brain	0.33 a 0.15 a TR a 0.00 a 0.00 a 0.00 a 0.00 a				
D. p,p'-DDE.					
Fat Liver Gonad Kidney Brain Crop contents Breast	6.07 a 2.64 1.20 0.97 0.75 0.66 0.52	b	с с с с с с		
E. Mirex					
Fat Liver Breast Brain Kidney Crop contents Gonad	0.87 a 0.20 0.07 TR 0.00 0.00 0.00	Ե Ե Ե Ե			

*Means followed by the different lower case letters are significantly different (P=0.05).

Fat contained significantly higher mean concentrations of mirex than other tissues (Table 5-E). Concentrations in liver, breast, and brain tissues were detected but were not significant.

This study was conducted in only one season of the year. There is a probability of seasonal variation in dosage of agricultural insecticides in nature. Barrier (1970) found seasonal differences in DDT residue levels in tissues as well as in rumen contents of white-tailed deer (*Odocoileus virginianus* L.) collected in Calhoun County, South Carolina. There was no reason to suspect that the same may not be true of the bobwhite quail found in agricultural areas but this could be substantiated only by seasonal collections and residue analyses. The levels of total DDT and mirex residues in the fat of some of the quail studied approached or surpassed the acceptable limits (7.0 and 1.0 ppm., respectively) for poultry set by the USDA (1969). Thus, it is fortuante that DDT usage has since been curtailed. Since these levels are not excessive, any inherent danger in human consumption of these birds is considered doubtful. In most instances, the meat of these birds is considered a delicacy and would not approach the intake of poultry products, a staple source of protein in diets of many humans. The breast muscle is the most commonly eaten portion of quail and had consistently lower concentrations of the insecticides than any of the other tissues studied. Consumption of the other organs and tissues that have higher concentrations of residues is usually avoided. Additionally, the concentrations listed by the FDA are wet weight concentrations and residues in this study were recorded as dry weight concentrations.

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ABSTRACT

A study was conducted involving the analysis for insecticides of selected tissues of 107 bobwhite quail (*Colinus virginianus* L.) collected from eight locations in the Coastal Plain of South Carolina. Six tissues—brain, fat, breast muscle, liver, kidney, and gonad—and crop contents were analyzed by gas chromatographic techniques for p,p'-DDT, p,p'-DDD, p,p'-DDE, and mirex. Total DDT residues were found to differ significantly among areas and tissues studied with highest concentrations being in fat and liver of quail from agricultural areas. Mirex was detected in birds collected from only two of the eight areas studied. Most mirex was found in birds from an area in which mirex had been applied in the 1969 imported fire ant (*Solenopsis saevissima richteri* Forel) eradication program. Mirex was detected in fat, liver, breast, and brain tissues with significantly higher concentrations in the fat samples.

A food habit study was also performed to determine if principal winter foods of quail were those normally affected by application of chlorinated hydrocarbons. Legumes, cultivated crops, and mast of oaks (*Quercus* spp.) and sweetgum (*Liquidambar styraciflua* L.) were among the most important food items. In agricultural areas, crops such as corn (*Zea mays* L.) and soybean (*Glycine max* (L.) Merr.) were utilized heavily as food. Food availability was an important factor. In areas where a large degree of diversification of food and cover was maintained, a large number of plant species occurred in the quail diet.

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