

The desorption of ammonia by citrus fruits presents a confused picture. Much of this apparent desorption is probably simple dilution, by fresh air, of the ammonia-air mass surrounding the fruits inside the experimental chambers. Not all the instances of apparent desorption can be accounted for by this simple explanation, however, for some moist or wounded fruits and vegetables unquestionably desorb measurable ammonia after treatment.

The fiberboard composing citrus shipping cartons also sorbs ammonia during the treatment, then desorbs the major part of the contained ammonia into an ammonia-free atmosphere. As with many of the fruits and vegetables tested, the desorption process with fiberboard is essentially complete within a few hours after initiation. The many types of fiberboard and fiberboard components examined in detail exhibited very similar sorption characteristics. Ammonia sorption by fiberboard is not directly related to moisture content.

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INSECTICIDE RESIDUES

The Endrin Content of Eggs and Body Tissue of Poultry Receiving Endrin in Their Daily Diet

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The tissues and eggs of chickens ingesting small amounts of endrin daily were examined for endrin residues using a specific spectrophotometric method of analysis. Results were confirmed with a nonspecific bioassay method sensitive to 0.02 p.p.m. of endrin. At the lowest level of intake, 0.1 p.p.m., endrin could not be detected in the eggs, but was present in the body fat. Both tissues were contaminated with endrin at higher levels of intake.

THE fate of insecticides in the animal body is a subject of considerable importance in modern pest control. Because many contemporary pesticides possess residual properties which make them effective for weeks or months, they can conceivably be distributed rather widely from their original site of application. A material used on forage or grain crops, for example, may be present as minute residues on the harvested crop and eventually become deposited in the tissues of the animal eating these feed-stuffs. The results of a study of the distribution of endrin (1,2,3,4,10,10-

hexachloro - 6,7 - epoxy - 1,4,4a,5,6,7,8,8a - octahydro - 1,4 - endo - endo - 6,8 - dimethanonaphthalene) in poultry tissue are described in this paper.

The fate of insecticides applied to or consumed by poultry has been investigated by several workers. Hoskins (9) and Furman and Bankowski (5) have described the absorption of benzene hexachloride by chickens following treatment of their roosts with various formulations of this compound. Draper and coworkers (3) have found that after 2 years of DDT intake the DDT level in poultry fat is approximately 10 times

that of the diet. When Sherman and Rosenberg (13) fed isodrin at 3.5 mg. per kg. of diet and endrin at 4.3 mg. per kg., they found 0.6 p.p.m. of isodrin and 6.7 p.p.m. of endrin in the fat 24 hours later. Fairchild and Dahm (4) reported that when chickens were given a single dust application of lindane or dieldrin, dieldrin persisted in the body fat for as much as 12 weeks with residue levels higher than those found at one week. In the case of lindane, residues of 11 p.p.m. were found one week after dusting with a steady decrease to about 0.3 p.p.m. at 8 weeks. Marsden

and Bird (7) found that turkeys fed DDT for 7 to 8 weeks stored the insecticide in their fat at concentrations ranging from four to eight times that in the diet.

The purpose of the work described here was to expose laying pullets and broiler chickens to small daily doses of endrin for prescribed periods, during which eggs, body fat, and certain meat cuts were to be analyzed for endrin residues.

Experimental

The feeding experiments were begun in July 1956, when the Delaware X New Hampshire male chicks were approximately 1 month of age and the White Leghorn pullets were 6 months old. Seven birds were used for each level of endrin in the broiler feeding tests and six pullets for each level in the egg deposition studies. The broiler feeding experiments were continued for 6 weeks, at which time all birds were slaughtered. The pullets were kept on the experimental diet for 8 weeks; then the endrin was removed and the hens were continued on a normal diet for 4 additional weeks.

A week's supply of both the broiler and layer rations was prepared once each week, and was fortified with 0.1, 0.25, and 0.75 p.p.m. of endrin. Unfortified control diets were also fed in each experiment. High corn type all-mash rations were employed in both the broiler and layer experiments. Body weight, egg production, feed consumption, and mortality records were kept throughout the experiments.

As a result of certain discrepancies in the analyses, it was decided to repeat the broiler feeding experiment the following season, starting in April 1957. The design of the experiment was essentially the same as that previously conducted, except that male broilers of a New Hampshire X Delaware mating were used and the levels of endrin fortification were increased to 0.25, 0.75, and 2.25 p.p.m. in addition to the unsupplemented rations. To check the effect of a feed-off period on the endrin content of broiler body fat, an additional group was given 2.25 p.p.m. of endrin daily for 6 weeks and then changed to an endrin-free diet for 2 weeks.

Sampling Methods. Egg samples from the laying pullets were taken at 1, 2, 4, and 8 weeks during the feeding of endrin and at 4 weeks after the cessation of endrin feeding. Sample eggs were taken during the fourth, fifth, and sixth days of the week and broken into a Waring Blendor and homogenized. One hundred-gram subsamples were removed and stored at 0° C. pending analysis. At the end of the feed-off period the birds were slaughtered and body fat samples were taken for endrin analysis.

The broilers in the 1956 experiment

Table I. Endrin Content of Eggs and Fat from Treated Pullets

Endrin in ^b Diet, P.P.M.	Endrin Content, P.P.M. ^a											
	During Endrin Feeding						After Endrin Feeding Ceased					
	1 Week		2 Weeks		4 Weeks		8 Weeks		Eggs, 4 Weeks		Fat, 4 Weeks	
	Sp.	Bio.	Sp.	Bio.	Sp.	Bio.	Sp.	Bio.	Sp.	Bio.	Sp.	Bio.
0.00	<0.1	<0.02	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	0.04
0.10	<0.1		<0.1	<0.1		<0.1		<0.1		<0.1		0.21
0.25	<0.1		<0.1	<0.1	0.05	0.2	0.31	<0.1	0.13	0.3	0.59	
0.75	<0.1		<0.1	0.1		0.3	0.36	0.2	0.17	1.1	1.07	

^a Data corrected for apparent endrin content of control samples and rounded to nearest 0.1 p.p.m. for spectrophotometric method and 0.02 p.p.m. for bioassay method. Values below these levels are indicated. Multiple analyses of same sample have been averaged.

^b Six birds used for each feeding level.

Table II. Endrin Content of Broiler Tissue

Endrin in ^b Diet, P.P.M.	Endrin, P.P.M., after 6-Week Endrin Intake ^a							
	1956 Experiments				1957 Experiments			
	Fat		Breast		Drumstick		Fat	
	Sp.	Bio.	Sp.	Bio.	Sp.	Bio.	Sp.	Bio.
0.00	<0.1	<0.02	<0.1	...	<0.1	<0.1	<0.1	<0.1
0.10	0.5	0.66	<0.1	0.04	<0.1
0.25	0.7	1.46	0.1	...	<0.1	0.6	1.0	
0.75	1.6	3.10	0.2	0.24	0.3	3.6	4.3	
2.25						17.0 ^c	18.0 ^c	

^a Data corrected for apparent endrin content of control samples and rounded to nearest 0.1 p.p.m. for spectrophotometric method and 0.02 p.p.m. for bioassay method. Values below these levels are indicated. Multiple analyses of same sample have been averaged.

^b Seven chicks per feeding level.

^c Endrin content of corresponding sample 2.9 p.p.m. after 2 weeks without endrin intake.

were slaughtered at the end of the sixth week of feeding and were sampled for breast meat, drumsticks, and body fat. In the 1957 broiler experiments, only body fat samples were analyzed. In all cases body fat desposits were found to be very meager. Only by carefully gleaning the entire carcass was sufficient fat for analysis obtained.

Analytical Methods. All samples were analyzed by the spectrophotometric method of Bann and coworkers (7), which is specific for endrin and sensitive to 0.1 p.p.m. In addition, approximately half of the samples were analyzed independently, using a mosquito larvae bioassay sensitive to 0.02 p.p.m. of endrin (10).

To validate the two analytical methods, control samples were analyzed frequently with and without fortification with endrin. Recoveries averaged 99% for the specific method and 108% for the bioassay method.

Results and Discussion

Weight gain, egg production, feed consumption, and mortality appeared normal for the particular birds involved and the environmental conditions imposed.

The results of the endrin determinations on eggs and tissue are shown in the accompanying tables. According to the results in Table I, a dietary level of 0.10 p.p.m. of endrin can be con-

sumed for 8 weeks without danger of contaminating the eggs with endrin. At 0.25 p.p.m. and higher, definite deposition of endrin in egg tissue occurs, sometimes after 2 to 4 weeks of intake. This egg contamination does not readily disappear, being evident for at least 1 month after exposure to endrin is stopped.

The results of the body fat and meat tissue analyses are shown in Tables I and II. A definite accumulation of endrin in fat was shown in both experiments, with even the lowest dietary level showing evidence of deposition. Meat tissue showed endrin contamination at both the 0.25-p.p.m. and the 0.75-p.p.m. intake levels. Cooking failed to eliminate the residues.

A surprising development in the 1956 experiment was the lack of agreement between the endrin contents as measured by the two independent methods. In all but one case where samples were analyzed by both methods, the bioassay results were appreciably higher than the spectrophotometric results. As considerable experience had been gained in the use of these two methods in analyzing animal tissues—which up to this point had shown them to agree—it was considered possible that a toxic metabolite of endrin was produced in this case. It was decided to repeat the experiment to check this point. The results of the second series of tests are shown in Table II. Much closer analytical agreement was reached. The explanations for the discrepancies in the first experiments are

Table III. Estimated Endrin Recovery in Broiler Feeding Experiments^a

Endrin Feeding Group, P.P.M.	Total Feed Consumed, G.	Total Endrin Intake, Mg.	Body Wt. at Slaughter, G.	Est. Body Fat, G.	Endrin in Fat, P.P.M.	Est. Total Body Endrin, Mg.	Endrin Recovery, %
1956 Experiments							
0.1	3180	0.32	1606	160	0.6	0.1	31
0.25	2950	0.73	1501	150	1.0	0.15	20
0.75	3180	2.38	1596	160	2.5	0.4	16
1957 Experiments							
0.25	3420	0.85	1721	170	1.0	0.17	20
0.75	3220	2.42	1638	160	4.0	0.6	25
2.25	3400	7.65	1685	170	18.0	3.1	40

^a Calculations based on actual feed intake and body weight records and on assumption that endrin is found only in body fat which was estimated to be 10% of total body weight.

not apparent, but the results of the second feeding experiments seem to eliminate the possibility of a toxic metabolite being produced by poultry. Reasonably good agreement as to levels of endrin in the fat was obtained in the two separate feeding experiments.

In similar studies with other farm livestock (10, 14) it was found that the maximum endrin content of body fat was approximately 1.00 p.p.m. after continuous feeding of endrin dietary levels up to 2.00 p.p.m. In the case of poultry, however, levels several times this concentration are reached. The most obvious explanation for this apparent greater tendency to accumulate endrin in body fat is that much less fat is present in poultry tissue. Literature sources (2, 12) suggest that poultry of the type used in this experiment may contain about 10% of total body fat. On the other hand, hog carcasses run as high as 50% of fat (7), and beef or lamb carcasses may vary from 20 to 50% in fat content (6, 8). This amounts to a concentration factor of 2 to 5 times for the endrin in poultry fat, compared to that in the other livestock mentioned.

In considering the results of the analyses of poultry tissues after a period of endrin ingestion, it must be concluded that the tendency for endrin to be deposited in edible tissue is rather strong. However, levels fed in these experiments are probably much higher than would be encountered in practice. Only a small percentage of the modern poultry diet is derived directly from sources where insecticides such as endrin are at present used. In these experiments the alfalfa meal content of the diet

was 3% or less. Grains, if exposed to insecticides at all, would carry exceedingly low residues. From these considerations it is highly unlikely that the endrin content of a total poultry diet would reach 0.1 p.p.m.

Inasmuch as feed consumption, body weight, and analytical data were available, it was thought worthwhile to attempt to account for the endrin ingested by the broilers in the two experiments. This has been done in Table III. One approximation which had to be made to arrive at these figures was that for total body fat content. The 10% value referred to previously was used. An assumption made was that endrin was present only in the fat tissue. As can be seen in the table, the calculations lead to the conclusion that up to about 40% of the endrin ingested during the experiments had been accounted for.

The endrin recovery calculations in the poultry feeding experiment were of sufficient interest to lead to similar approximations in the case of hogs, lambs, beef, and dairy cattle which had been given endrin in their diet (10, 14). The estimates were made in the same manner as those shown in Table III using detailed records of feed intake, body weight, and milk production, and endrin residue levels of body fat and milk. Total fat estimates needed in the calculations were those referred to previously. As a result of these calculations it was concluded that only a small part of the insecticide had been accounted for. Hogs appear to retain about 3% of the ingested endrin. Approximately 5% of the endrin was recovered in the

case of the lambs, 7% with the dairy cows, and 12% with the beef cattle.

Although the endrin balance calculations are based on several assumptions, it seems safe to conclude that when this insecticide is ingested in small amounts, a large percentage is disposed in some manner other than fat storage. One possibility is that endrin is metabolized and stored in a chemical form not detected by the two analytical methods. Another is that endrin or endrin metabolites are excreted. Further work needs to be done to learn more about the fate of this and similar insecticides when ingested. The nature and disposition of any metabolites produced will lead to a better understanding of the hazards associated with the presence of pesticide residues in animal tissues.

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