

PESTICIDE RESIDUES IN MEAT AND MILK

Storage of Dieldrin in Tissues and Its Excretion in Milk of Dairy Cows Fed Dieldrin in Their Diets

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Dieldrin, fed in the diet of dairy cows at levels of 0.1, 0.25, 0.75, and 2.25 p.p.m., appeared in the milk at the end of 6 to 12 weeks at average levels of 0.02, 0.06, 0.11, and 0.28 p.p.m., respectively. Tissue analyses at the end of the feeding period indicated that dieldrin was present in fats at concentrations varying from 0.24 p.p.m. at the lowest level of feeding to 5.48 at the highest. The amounts of dieldrin found in other tissues was proportionate to their fat content.

AS THE CHLORINATED HYDROCARBON INSECTICIDES have long been known to be stored in animal fat to varying degrees, the extent of the usage of these insecticides on forage crops depends greatly on their propensity for storage in fat and/or their excretion in milk. A few insecticides have been assigned tolerances in the fat of certain animals, but to date regulatory agencies have tended to maintain that milk is not to be contaminated by pesticides or other foreign substances.

Some research has been done by various workers (7-3, 5) which indicated that dieldrin was excreted in milk at fairly high levels, when cows were sprayed with it or fed at fairly high dosage rates.

The present study was initiated to determine whether or not there was a level of feeding below which contamination of fat, and especially milk, of dairy cows would not occur.

Methods

With the exception of one milking Shorthorn, the animals selected for testing were all grade Guernsey or Holstein

cows. They were deliberately selected at random to avoid any possible bias attributable to the selection of highly bred uniform animals and to reflect the degree of variance to be found in average farm herds.

Rations for the cows consisted of No. one grade, first cutting alfalfa and a concentrate mixture of 1/3 oats and 2/3 corn ground in a hammer mill. The land on which the rations were grown and the crops themselves had never been treated with an insecticide. Each cow was stanchioned and fed the oats-corn concentrate from the same individual feed box, while during hay-feeding periods each cow was tied with a rope halter and fed from the same feed rack throughout the experiment. Before beginning the test, efforts were made to estimate the approximate feed intake for each cow. During the experiment the amount of feed provided for each cow was slightly less than the average amount the individual cows had been found to consume. This procedure reduced to a minimum the amount of unconsumed feed. Immediately prior to each feeding, dieldrin, formulated in acetone at various con-

centrations, was pipetted onto the rations (hay and grain) so that 1 ml. of the solution was sufficient to add the desired amount of dieldrin, in terms of parts per million, to 1 pound of feed.

Both the rations and milk samples were analyzed before the cows were put on dieldrin intake to ensure that they contained no dieldrin residues.

On days when milk samples were to be collected, the entire production of each cow was placed in a separate, sterile, galvanized can. The milk in each container was thoroughly mixed and representative samples were placed in milk cartons lined with Pliofilm bags. Samples were then labeled and placed in a cold room at 0° F. until analysis.

When cows were slaughtered 12 and 18 weeks after the start of the experiment, appropriate tissues were collected, placed in labeled Pliofilm bags, and stored at 0° F. until analysis.

Daily records were kept on milk production, feed provided, feed consumed, and feed not consumed. Monthly weighing of the cows established weight gains or losses.

Groups of four cows each were fed at

Table I. Dieldrin Levels in Milk of Dairy Cows Sampled during 12 Weeks of Dieldrin Intake and Additional 6-Week Feed-off

Dieldrin in Diet, P.P.M.	Dieldrin, P.P.M. ^a										
	Dieldrin Intake Period, ^b Weeks							Feed-off Period, ^c Weeks			
	0	3/7	1	2	4	8	12	0 ^d	4	6	
0	<0.01	...	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
0.1	<0.01	...	<0.01	<0.01	<0.01	0.01	0.02	0.04	0.03	<0.01	
0.25	<0.01	...	0.02	0.02	0.02	0.03	0.06	0.07	0.02	0.01	
0.75	<0.01	0.04	0.04	0.06	0.07	0.11	0.11	0.16	0.15	0.04	
2.25	<0.01	0.06	0.16	0.17	0.16	0.18	0.28	0.24	0.21	0.04	

^a Corrected for check values and to 4% butterfat.

^b Milk from four cows, dosage analyzed through 12 weeks, then two slaughtered for tissue analysis.

^c Milk from remaining two cows per dosage analyzed through 6-week feed-off period.

^d Same as 12-week, but average of two cows not sacrificed.

Table II. Ratios of Dieldrin in Fat to Dieldrin in Milk at Ends of Intake and Feed-off Periods

Dieldrin in Diet, P.P.M.	12 Weeks on Dieldrin			6 Weeks Feed-off Period			Decline in 6 Weeks, %	
	P.P.M.		Ratio fat/milk	P.P.M.		Ratio, fat/milk	Fat	Milk
	Fat ^a	Milk ^b		Fat ^a	Milk ^b			
0.1	0.24	0.014	16.8	0.33	<0.01	>33.4	0	>30.1
0.25	0.92	0.076	12.1	0.47	0.015	31.0	49.2	80.1
0.75	1.34	0.112	12.0	0.90	0.036	25.1	32.6	67.7
2.25	5.48	0.305	18.0	3.81	0.040	96.4	30.4	87.0
Av.			15.7			>54.9	30.8	>80.1

^a Av. renal and body fats. ^b Av. of two cows sacrificed at this interval.

levels of 0, 0.1, 0.25, 0.75, and 2.25 p.p.m. of dieldrin in the diet for 12 weeks. At that time, two cows from each group were slaughtered and samples of tissues were collected. To determine the amount of dieldrin which would be lost from the tissues during a 6-week feed-off period, the remaining cows were fed on dieldrin-free diets for this period, at which time they were slaughtered.

The specific colorimetric method of O'Donnell *et al.* (7), as modified in the Shell method series (8), was used to determine microgram quantities of dieldrin in milk and tissues. Samples of whole milk, 600 grams in size, and 50-gram ground tissue samples were saponified directly with ethanolic potassium hydroxide on a steam bath, extracted with Skellysolve B, washed with distilled water, dried with anhydrous sodium sulfate, and chromatographed on activated magnesium oxide-Celite (2 to 1), followed by the development of color specific for dieldrin. For the amounts used, sensitivity of the method for milk was 0.01 p.p.m. and for tissues was 0.1 p.p.m. Recoveries, added to check samples, generally ranged between 80 and 120%.

Milk samples were analyzed for their butterfat content by the Dairy Science Department of the University of Illinois.

Results and Discussion

Levels of dieldrin appearing in milk of dairy cows during the 12-week dieldrin-intake period followed by a 6-week period on dieldrin-free rations are shown in Table I. At the end of the 12-week period, dieldrin was present in the milk in measurable quantities at all levels of feeding. While individual cows on the 0.1 p.p.m. feeding level showed concentrations in their milk greater than 0.01 p.p.m. (the sensitivity of the method) as early as the second week of feeding, the average concentration for the four cows at that dosage did not exceed 0.01 until the eighth week. The two higher dosages produced measurable amounts of dieldrin in milk at the first sampling 3 days after the start of dieldrin feeding. The amounts of dieldrin appearing in both milk and fat appeared to be roughly proportional to the amounts in the diet.

In evaluating the data obtained at the

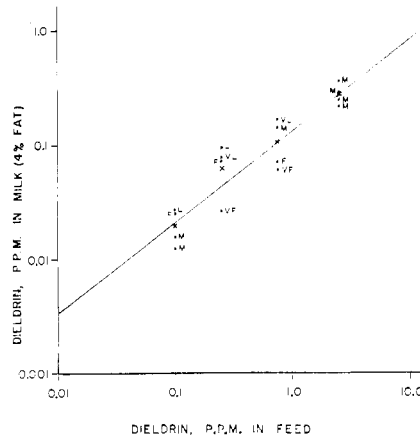


Figure 1. Dieldrin in milk, cows on continuous dieldrin intake for 12 weeks at rates of 0.1, 0.25, 0.75, and 2.25 p.p.m. in the total feed intake

Ratings for fatness of animals as judged by examination of dressed carcass
 M. Medium VF. Very fat
 F. Fat VL. Very lean
 L. Lean

end of the 12-week feeding period, when dieldrin intake is plotted against the average dieldrin content of milk on log-log graph paper, a straight-line relationship is strongly suggested. The trend is apparent despite the fact that the data show considerable variability and the obvious effect of variations in the fatness of cows on the dieldrin content of their milk (Figure I). The ratings as to fatness indicated in Figure I are only relative and not precise values. Nevertheless, it seems that there is a definite relationship between the fatness of the animal and the amount of dieldrin appearing in its milk. The same can be said for the amounts of dieldrin stored in fat.

From the data presented, it would appear also that the amount of dieldrin appearing in milk on any given day is dependent upon two factors—the daily intake and the existing concentration in the fat of the animal. This is demonstrated rather strongly in Table II. Some have theorized that the concentration of pesticide in milk of 4% butterfat content should be 1/25 the concentration appearing in the fatty tissues of the animal, but this does not appear to be the case. In this experiment, at the end

Table III. Dieldrin in Cow Tissues after 12 Weeks of Dieldrin Intake Followed by 6 Weeks of Feed-off

Dieldrin in Diet, P.P.M.	Average Total Intake, Mg./Kg.	Dieldrin, F.P.M. ^a																				
		Brain		Heart		Liver		Kidney		Renal Fat		Body Fat		Omentum Fat		Heart Fat		Rump Fat		Udder Fat		
		12	18	12	18	12	18	12	18	12	18	12	18	12	18	12	18	12	18	12	18	
0	0	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
0.1	0.293	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
0.25	0.750	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
0.75	2.169	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
2.25	6.556	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1

^a Corrected for check values. Two cows per dosage slaughtered at 12 weeks, two remaining feed-off and slaughtered at 18 weeks.

of the 12-week period during which the cows were on dieldrin intake, ratios of dieldrin in fat to dieldrin in milk ranged between 12 and 18 to 1 and averaged 15.7 to 1 rather than 25 to 1. This phenomenon has also been reported by Kiigemagi *et al.* (6) who used endrin. When the dieldrin intake was discontinued, however, and the amount of dieldrin appearing in milk became dependent upon only one source—the existing concentration in the fat of the animal—the ratios became greater than the accepted figure of 25 to 1. The per cent decline in fat residues after the animals had been off dieldrin for 6 weeks was only about 30%, while the decline in dieldrin in milk was greater than 80%. This is borne out also in related work with other insecticides (4).

Table III gives dieldrin residues in various cow tissues at the end of 12 weeks

of dieldrin intake and after an additional 6-week feed-off period. Concentrations in the various tissues are roughly proportional to the fat content of the tissues.

Steaks and roasts from cows which had been fed high dosages of dieldrin were cooked and compared for dieldrin content with similar raw cuts. No significant losses of dieldrin due to cooking were evident.

Weight gains of treated cows were comparable to those of checks. During the experiment and at slaughter, animals showed no evidence of pathology. Milk production throughout was consistent with that which could be expected.

Literature Cited

- (1) Claborn, H. V., Wells, R. W., *Agr. Chem.* 7(10), 28-9 (1952).
- (2) Claborn, H. V., U. S. Dept. of Agr.,

Bull. **ARS-33-25**, 1-30 (1956).

- (3) Ely, R. E., Moore, L. A., Carter, R. H., Hubanks, P. E., Poos, F. W., *J. Dairy Sci.* 37, 1461-5 (1954).
- (4) Gannon, N., Link, R. P., Decker, G. C., unpublished data.
- (5) Harris, J. R., Stoddard, G. E., Bateman, G. Q., Shupe, J. L., Greenwood, D. A., Harris, L. E., Bahler, T. L., Lieberman, F. V., *J. Agr. Food Chem.* 4, 694-6 (1956).
- (6) Kiigemagi, U., Sprowls, R. G., Terriere, L. C., *Ibid.*, 6, 518-21 (1958).
- (7) O'Donnell, A. E., Johnson, H. W., Jr., Weiss, F. T., *Ibid.*, 3, 757-62 (1955).
- (8) Shell Method Series, 638/56 (1956).

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PESTICIDE RESIDUES IN MEAT

Storage of Dieldrin in Tissues of Steers, Hogs, Lambs, and Poultry Fed Dieldrin in Their Diets

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Dieldrin was fed in the diet to various animals for 12 weeks at levels of 0.1, 0.25, 0.75, 2.25 p.p.m. Samples of tissues were then analyzed for dieldrin. It was detectable in the fat of all animals fed on the various intake levels. When detectable in other tissues, residues were proportional to the fat content of the tissues. For each species the amount of dieldrin stored appeared to be proportional to the rate of intake. Laying hens stored the greatest quantity of all, but their eggs contained very little dieldrin. Steers stored more dieldrin in their tissues in terms of parts per million than did hogs, while lambs stored less than hogs.

DIELDRIN, AN INSECTICIDAL MATERIAL consisting of not less than 85% 1,2,3,4,10,10 - hexachloro - 6,7 - epoxy-1,4,4a,5,6,7,8,8a - octahydro - 1,4-endo - exo - 5,8 - dimethanonaphthalene (HEOD), for several years has been a very effective insecticide on a large variety of insects attacking many different crops. It has consequently been registered and approved for use on many of these crops. As a chlorinated hydrocarbon, it is known to be stored in animal fat as a result of ingestion or contact (1-3). Therefore, the extent of its usage on forage to be used as animal feed will to a considerable degree be determined by its propensity for storage in animal tissues. Studies reported in this paper were initiated to develop data considered a prerequisite to the possible establishment of tolerances for dieldrin on forage and in animal fat.

Methods

Animals under test were fairly uniform in age, breed, and weight. They consisted of Black Angus steers, Yorkshire hogs, Shropshire lambs, White Rock fryers, and Leghorn laying hens.

Rations for the various breeds were as follows—steers: ground shelled corn and alfalfa hay; hogs: ground shelled corn, dehydrated alfalfa meal, soybean

oil meal, ground limestone, and steamed bone meal; lambs: shelled corn, linseed meal, and alfalfa hay; fryers: growing mash; and hens: laying mash. All rations were obtained from the University of Illinois feed storage. Prior to the start of feeding, they were analyzed to ensure that they bore no dieldrin residues.

Before the experiment was started, efforts were made to estimate the ap-

Table I. Dosage Rates and Number of Animals on Test

Dieldrin in Diet, P.P.M.	No. of Animals per Dosage				
	Steers	Hogs	Lambs	Fryers	Hens
0	2	2	2	6	4
0.1	3	3	3	6	5
0.25	3	3	3	6	5
0.75	3	2	2	6	5
2.25	2	2	2