

Table IV. Partial Correlations Among Pesticide Concentrations in Ewe Body and Milk Fat^a

Item ^b	1	2	3	4	5	6	7	8	9	10	11	12
	Milk fat											
(1) DDE	1.00	-0.13	0.19	0.86 ^e	-0.45	0.57 ^d	0.74 ^e	0.00	0.15	0.55 ^e	-0.46	-0.06
(2) DDD			-0.29	0.27	0.03	-0.37	-0.14	0.08	-0.38	-0.20	0.01	-0.36
(3) DDT				0.37	-0.54 ^d	-0.44	0.09	-0.21	0.01	0.02	0.30	0.02
(4) Total DDT					-0.56 ^d	0.47	0.59 ^d	-0.04	-0.05	0.62 ^e	-0.28	-0.22
(5) Dieldrin						-0.77 ^e	-0.09	0.19	0.13	0.03	-0.26	-0.04
(6) Ratio ^c							0.31	-0.10	0.33	0.31	-0.04	0.41
	Body fat											
(7) DDE								0.36	0.51 ^d	0.92 ^e	-0.76 ^e	0.07
(8) DDD									0.30	0.56 ^d	-0.63 ^e	0.43
(9) DDT										0.75 ^e	-0.64 ^e	0.64 ^e
(10) Total DDT											-0.86 ^e	0.36
(11) Dieldrin												-0.30
(12) Ratio ^c												1.00

^a Partial correlations coefficients were derived on intragroup variance basis; residual d.f. = 15; ewe parturition weight was included as continuous independent variable. ^b For identification, the item numbers across the top of the table correspond to those traits (listed by number in parentheses) down the left margin. ^c Ratio = total DDT: dieldrin. ^d $p < 0.05$. ^e $p < 0.01$.

Durham, W. F., Cueto, C., Hayes, W. J., *Amer. J. Physiol.* **187**, 373 (1956).

Food and Drug Administration, "Pesticide Analytical Manual," United States Department of Health, Education, and Welfare, Washington, D. C., 1969.

Harter, H. L., *Biometrics* **16**, 671 (1960).

Harvey, W. R., "Least squares analysis of data with unequal subclass numbers," United States Department of Agriculture, Agricultural Research Service, 1960, pp 20-28.

Kunze, F. M., Laug, E. P., *Fed. Proc.* **12**, 339 (1953).

Laben, R. C., Archer, T. E., Crosby, D. G., Peoples, S. A., *J. Dairy Sci.* **48**, 701 (1965).

Laben, R. C., Archer, T. E., Crosby, D. G., Peoples, S. A., *J. Dairy Sci.* **49**, 1488 (1966).

National Research Council, "Nutrient Requirements of Domestic Animals. No. 5 Nutrient Requirements of Sheep," 4th ed, revised, Washington, D. C., 1968.

Street, J. C., *Science* **146**, 1580 (1964).

Street, J. C., Blau, A. D., *Toxicol. Appl. Pharmacol.* **8**, 497 (1966).

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Certain Physiological Factors Affecting Organochlorine Pesticide Metabolism in Young Ovines Contaminated *in utero*

Hugo Varela-Alvarez, John D. Sink,* and Lowell L. Wilson

Thirty crossbred lambs (15 males and 15 females), the progeny of ewes contaminated with DDT and dieldrin, were used to determine the effects of placental transfer, length of contamination, sex, and age on the body storage and elimination of these pesticides. Three lambs of each sex were sacrificed at birth. Similar numbers of both a suckling and an artificially raised group were sacrificed at 5 and 10 weeks of age. Placental

transfer of the pesticides occurred, resulting in contamination of all newborn lambs. Suckling lambs accumulated significantly more DDT, DDT metabolites, and dieldrin because of constant exposure to pesticide through their mother's milk. Of the suckling lambs, females stored significantly more DDT and DDT metabolites than did males, but dieldrin storage was the reverse.

Placental transfer of organochlorine pesticides (Braund *et al.*, 1968; Finnegan *et al.*, 1949; Hathway, 1965) and residue elimination through the milk (Braund *et al.*, 1967; Brown *et al.*, 1966; Crosby *et al.*, 1967; Gannon and Decker, 1960; Laben *et al.*, 1965; Zweig *et al.*, 1961) have been reported. More information is needed regarding the metabolism of pesticides in young ruminants contaminated

in utero and subsequently through their mother's milk and regarding the effect of certain physiological factors on the rate of deposition and elimination of residues. Sex affects deposition rate in rats (Durham *et al.*, 1956), but little or no information is available regarding sex effects on pesticide metabolism in ruminants. The purposes of this study were to ascertain placental transfer, and to determine the effect of length of contamination, sex, and age on the rate of deposition and elimination of DDT, DDT metabolites, and dieldrin in progeny of ovine females contaminated *prepartum*.

* Department of Animal Science, The Pennsylvania State University, University Park, Pennsylvania 16802.

Table I. Least-Squares Means for Newborn Lamb Pesticide Body Fat Concentration and Other Traits by Lamb Sex

Item	Males	Females	Overall mean
Body fat weight, g	86.4	77.0	81.7
Body fat, %	2.1	1.7	1.9
DDE, ppm	41.0	16.2	28.6
DDD, ppm	27.5	4.4	16.0
DDT, ppm	27.1	5.6	16.4
Total DDT, ppm	95.6	26.2	60.9
Dieldrin, ppm	27.2	10.8	19.0
Total DDT:dieldrin ratio	3.52	2.43	2.98
Total body DDT, mg	8.3	2.0	5.1
Total body dieldrin, mg	2.3	0.8	1.6
Body ash (DMB), % ^a	21.8	18.4	20.1
Body moisture, %	78.4	82.5	80.5

^a DMB = dry matter basis.**MATERIALS AND METHODS**

Animals and Treatments. Thirty crossbred lambs (*Ovis aries*) representing both sexes were used in this study. These lambs were the progeny of crossbred 2-year-old, first-lamb ewes. Prior to breeding, the ewes were contaminated with purified DDT and dieldrin (50 mg/kg body weight), as reported previously (Varela-Alvarez *et al.*, 1973). After birth, the lambs were divided into the following groups with equal numbers of each sex in each group: those that were sacrificed immediately (newborn, $n = 6$); those that were to remain with their dams (suckling, $n = 12$); and those that were removed from their dams immediately and raised artificially on a noncontaminated ration (artificially raised, $n = 12$). Six lambs (three males and three females) from each of the latter two groups were sacrificed at 5 weeks of age and six more were sacrificed at 10 weeks of age. The lambs raised artificially were given a commercial milk replacer and free access to grain, hay, water, and mineralized salt, all of which were examined and found free of organochlorine pesticide contaminants.

Sampling and Analyses. All of the lambs were exsanguinated, eviscerated, pelted, and frozen, and the entire carcass was ground. Duplicate samples were taken for chemical and pesticide analyses. Moisture, ash, and ether extract were determined according to AOAC (1970) procedures. The methods used in the pesticide analysis have been described (Varela-Alvarez *et al.*, 1973).

Statistical Analysis. Statistical analyses were by least-squares analyses of variance, which included: lamb age and lamb group (newborn, suckling, and artificially

raised); lamb sex and lamb group as fixed main effects; and their two-way interactions. Partial correlation coefficients were derived on an intragroup variance basis among individual observations within each lamb age. Lamb birth weight, body fat percent, and lamb weight at sampling were included in all analyses as continuous independent variables (Harvey, 1960).

RESULTS AND DISCUSSION

Placental Transfer. Placental transfer of DDT, DDT metabolites, and dieldrin occurred in each ewe, resulting in contamination of all newborn lambs. The least-squares means by lamb sex for newborns' body fat concentrations of DDE, DDD, DDT, total DDT, dieldrin, total DDT:dieldrin ratio, the calculated body content of total DDT and dieldrin, and other traits are presented in Table I.

Concentrations of DDD in newborn lamb body fat averaged 27.5 and 4.4 ppm for male and female lambs, respectively. DDT concentration values were 27.1 and 5.6 ppm for male and female lambs. All other newborn traits studied apparently were not affected by lamb sex. Means for total DDT and dieldrin in the whole lamb body were 5.1 and 1.6 mg, respectively, and mean lamb birth weight was 4.6 kg.

The partial correlations among pesticide concentration in lamb body fat and other traits of the newborn lambs are presented in Table II. Partial correlation coefficients were derived on an intragroup variance basis. To remove from the experimental error the greater amount of fatness associated with the larger lambs, lamb birth weight and body fat percent were included as continuous independent variables.

The body fat concentration (ppm) of DDE, as well as total DDT, were both positively correlated 0.99 with body moisture percent, whereas a negative correlation (-0.95) between percent moisture and dieldrin concentration was noted. On the other hand, DDE and total DDT were negatively related (-0.99 and -0.95, respectively), while dieldrin was positively associated (0.99) with body fat percent. Such opposite relationships of the pesticides with these two body constituents can be expected in view of the well established negative correlation between body moisture and body fat (Reid *et al.*, 1968). However, the dissimilar association between the pesticides and the same body constituent is rather puzzling.

The total body burden of DDT was significantly related (0.96) to body ash content, but the total body burden of dieldrin was not.

Length of Contamination and Lamb Age. Least-squares means for lamb body fat pesticide concentration, pesticide content, and other traits by lamb group and lamb age are presented in Table III.

Table II. Partial Correlations Among Newborn Body Fat Pesticide Concentrations and Other Traits^a

Item ^b	1	2	3	4	5	6	7	8	9	10	11	12
(1) DDE, ppm	1.00	0.81	0.85	0.97 ^c	-0.97 ^c	0.82	-0.88	0.99 ^d	-0.99 ^d	-0.97 ^c	-0.72	-0.98 ^c
(2) DDD, ppm		1.00	0.99 ^d	0.92	-0.67	0.33	-0.99 ^d	0.89	-0.73	-0.65	-0.99 ^d	-0.69
(3) DDT, ppm			1.00	0.94	-0.72	0.39	-0.99 ^d	0.91	-0.77	-0.70	-0.98 ^c	-0.74
(4) Total DDT, ppm				1.00	-0.91	0.67	-0.96 ^c	0.99 ^d	-0.95 ^c	-0.89	-0.86	-0.92
(5) Dieldrin, ppm					1.00	-0.92	0.76	-0.95 ^c	0.99 ^d	-0.99 ^d	0.56	0.99 ^d
(6) Total DDT:dieldrin ratio						1.00	-0.45	0.73	-0.89	-0.93	-0.19	-0.91
(7) Ash, %							1.00	-0.94	0.81	0.74	0.96 ^c	0.77
(8) Moisture, %								1.00	-0.96 ^c	-0.93	-0.81	-0.95 ^c
(9) Body fat, %									1.00	0.99 ^d	0.62	0.99 ^d
(10) Body fat weight, g										1.00	0.53	0.99 ^d
(11) Total body DDT, mg											1.00	0.58
(12) Total body dieldrin, mg												1.00

^a Correlations derived on intragroup variance basis; residual d.f. = 2; lamb birth weight and body fat percent were included as continuous independent variables. ^b For identification, the item numbers across the top of the table correspond to those traits (listed by number in parentheses) down the left margin. ^c $p < 0.05$. ^d $p < 0.01$.

Table III. Least-Squares Means for Pesticide Body Fat Concentrations and Other Traits for Suckling and Artificially Raised Lambs by Lamb Age

Item	Suckling, weeks		Artificially raised, weeks	
	5	10	5	10
	Body fat weight, kg	9.6	15.0	9.8
Body fat, %	38.2	22.6	40.3	22.4
DDE, ppm	10.3	9.5	0.4	0.8
DDD, ppm	10.1	0.5	0.1	0.1
DDT, ppm	7.5	2.1	0.2	0.0
Total DDT, ppm	27.9	12.1	0.7	0.9
Dieldrin, ppm	10.2	1.5	1.0	0.2
Total DDT:dieldrin ratio	2.74	8.07	0.70	4.50
Total body DDT, mg	266.5	181.2	3.9	5.6
Total body dieldrin, mg	97.2	22.1	4.8	1.1

There was no significant difference in total body burden of DDT and dieldrin between newborn lambs and those artificially raised and sacrificed 5 and 10 weeks later. However, there was a significant difference between newborn lambs and artificially raised lambs with respect to individual DDE, DDD, DDT, total DDT, and dieldrin concentrations in the body fat. These results indicate that no significant degradation or elimination of pesticides occurred in the artificially raised lambs, but that the pesticide residue concentration was diluted with increased body fat deposition.

All pesticide concentrations in body fat and total dieldrin body content were significantly affected by group when the artificially raised group was compared with the suckling lamb group. Suckling lambs averaged consistently greater than did the artificially raised group in concentration and total body content of pesticides. This can be explained by realizing the suckling lamb group was exposed to a constant intake of pesticide.

As lambs in the suckling group became older, the concentration of certain pesticides declined. Mean body fat DDD concentration means were 10.1 and 0.5 ppm at 5 and 10 weeks of age, respectively; the same trend was also present for DDT (7.5 vs. 2.1 ppm), total DDT (27.9 vs. 12.1 ppm), and dieldrin (10.2 vs. 1.5 ppm). However, the total DDT:dieldrin ratio increased significantly with age. Mean ratios were 0.70 and 4.50 at 5 and 10 weeks of age, respectively. There was a significant interaction between sex and age in the suckling lamb group on body fat concentration of DDT and dieldrin and on the total DDT:dieldrin ratio.

The significant decline with time/lamb age in body fat concentration of DDD, DDT, dieldrin, total DDT, and content of total DDT and dieldrin in suckling lambs indicates that, despite the continuous intake of pesticide by the suckling animals, there is a definite body metabolism that reduced concentration and total body content of organochlorine pesticides. It also indicates that the elimination rate of DDT, its metabolites, and dieldrin was significantly greater than the rate of pesticide ingestion.

Sex Effects. There was no significant sex effect on pesticide concentration or content in artificially raised lambs (Table IV). Conversely, there was a significant effect of sex in the suckling lamb group on certain pesticide residues. In the latter group, females averaged higher than did males in DDT concentration (6.2 vs. 3.4 ppm) and total DDT concentration (22.2 vs. 17.8 ppm). Durham *et al.* (1956) reported that at any level of intake above 0.05

Table IV. Least-Squares Means for Pesticide Body Fat Concentrations and Other Traits for Suckling and Artificially Raised Lambs by Lamb Sex

Item	Suckling		Artificially raised	
	Males	Females	Males	Females
	Body fat weight, kg	11.2	13.4	10.1
Body fat, %	26.9	33.9	29.3	33.4
DDE, ppm	9.4	10.4	0.3	0.9
DDD, ppm	5.0	5.6	0.1	0.1
DDT, ppm	3.4	6.2	0.1	0.1
Total DDT, ppm	17.8	22.2	0.5	1.1
Total body DDT, mg	111.0	180.6	4.6	12.2
Dieldrin, ppm	6.4	5.3	0.7	0.5
Total body dieldrin, mg	39.7	42.9	6.7	6.6
Total DDT:dieldrin ratio	2.78	4.18	0.71	2.20

mg/kg per day, female rats stored more DDT and derived compounds that did males. In contrast, female lambs averaged less than did males for dieldrin concentration in the present study (5.3 vs. 6.4 ppm).

Total body content of dieldrin was also significantly affected by sex. The means were 42.9 and 39.7 mg for female and male lambs, respectively. These results substantiate an interaction between DDT and dieldrin reported by Street (1964). The change in dieldrin concentration of male and female lambs compared with the change in DDT concentration is probably because total DDT was greater in female lambs, producing a stronger interaction between the two pesticides to degrade dieldrin more rapidly. Ratio of total DDT to dieldrin was greater in female than in male lambs (4.18 vs. 2.78). The results of this study suggest that there is a concentration limit of DDT and dieldrin that initiates the interaction mechanism.

LITERATURE CITED

- Association of Official Analytical Chemists, "Official Methods of Analysis," 11th ed, Washington, D. C., 1970.
- Braund, D. G., Brown, L. D., Huber, J. T., Leeling, N. C., Zabik, M. J., *J. Dairy Sci.* **51**, 116 (1968).
- Braund, D. G., Brown, L. D., Leeling, N. C., Zabik, M. J., Huber, J. T., *J. Dairy Sci.* **50**, 991 (1967).
- Brown, W. H., Witt, J. M., Whiting, F. M., Stull, J. M., *Bull. Environ. Contam. Toxicol.* **1**, 21 (1966).
- Crosby, D. G., Archer, T. E., Laben, R. C., *J. Dairy Sci.* **50**, 40 (1967).
- Durham, W. F., Cueto, C., Hayes, W. J., *Amer. J. Physiol.* **187**, 373 (1956).
- Finnegan, F. K., Haag, H. B., Larson, P. S., *Proc. Soc. Exp. Biol. Med.* **72**, 357 (1949).
- Gannon, N., Decker, G. C., *J. Econ. Entomol.* **53**, 411 (1960).
- Harvey, W. R., "Least squares analysis of data with unequal subclass numbers," United States Department of Agriculture, Agricultural Research Service, 1960, pp 20-28.
- Hathway, D. E., *Arch. Environ. Health* **11**, 380 (1965).
- Laben, R. C., Archer, T. E., Crosby, D. G., Peoples, S. A., *J. Dairy Sci.* **48**, 701 (1965).
- Reid, J. T., Bensadoun, A., Bull, L. S., Burton, J. H., Gleeson, P. A., Han, I. K., Joo, Y. D., Johnson, D. E., McManus, W. R., Paladines, O. L., Stroud, J. W., Tyrell, H. F., VanNierkerk, B. D. H., Wellington, G. H., in "Body Composition in Animals and Man," National Academy of Sciences, Washington, D. C., 1968.
- Street, J. C., *Science* **146**, 1580 (1964).
- Varela-Alvarez, H., Sink, J. D., Wilson, L. L., *J. Agr. Food Chem.* **22**, 407 (1973).
- Zweig, G., Smith, L. M., Peoples, S. A., Cox, R., *J. Agr. Food Chem.* **9**, 481 (1961).

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