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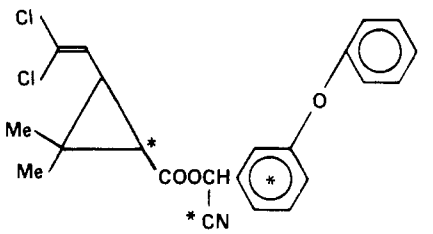
Metabolism of *cis*- and *trans*-Cypermethrin in Rats. Balance and Tissue Retention Study

Maureen J. Crawford, Andrew Croucher, and David H. Hutson*

The fate of the cis and trans isomers of the pyrethroid insecticide cypermethrin (NRDC 149), α-cyano-3-phenoxybenzyl 3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate, has been studied in rats (1–5 mg/kg) by using three forms of radiolabeling (benzyl-^{14}C, cyclopropyl-^{14}C, and cyano-^{14}C). Radioactivity derived from the benzyl-^{14}C and cyclopropyl-^{14}C labeling was rapidly eliminated, mostly in the urine. Tissue residues were generally very low, e.g., 0.01 μg/g in brain, with the exception of fat (~1 μg/g). Residues derived from the cis isomer tended to be higher than those derived from the trans isomer. The rate of depletion of the residues derived from [benzyl-^{14}C]-*cis*-cypermethrin was rapid ($t_{1/2}$ was less than ~1 day) from all tissues except fat, from which radioactivity was eliminated with a half-life of 11–12 days. This residue was largely due to unchanged *cis*-cypermethrin. [*cyano*-^{14}C]Cypermethrin afforded radioelimination and distribution characteristics similar to those reported for the cyanide ion.

Cypermethrin (NRDC 149, I) is one of the pyrethroid α-cyano-3-phenoxybenzyl esters which combine high insecticidal activity with a degree of photostability suitable for use in the field (Elliott, 1976). Part of the successful development of this compound is a study of its mammalian toxicology which includes a knowledge of its biotransformation in experimental animals. It is a complex molecule in terms of stereochemistry, and furthermore, as it is an ester, its fate in biological systems must be studied by using a radiolabel in both the acid and alcohol moieties. Cypermethrin possesses three chiral carbon atoms and is therefore a mixture of eight isomers. It was discovered some years ago (Abernathy and Casida, 1973) that the relative orientation of the ethenyl group and the carboxylic group on the cyclopropane ring of the pyrethroid insecticides has a dominant effect on the rate of enzymatic hydrolysis of these compounds. Hence, the traditional "cis" and "trans" nomenclature is still in common use. In the rat metabolism study reported here, two isomer mixtures were used; these are referred to as the cis and trans isomers throughout. C-1 (cyclopropyl) and C-α (benzyl) were racemic. Three positions of ^{14}C labeling were utilized: the cyclopropyl group, the 3-phenoxybenzyl group, and the cyano group:



I (cypermethrin ; * = ^{14}C–labelling)

MATERIALS AND METHODS

Chemicals. [*benzyl*-^{14}C]-*cis*- and *trans*-cypermethrin [α-cyano-3-phenoxy[^{14}C]benzyl 3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate, I] (34 μCi/mg) were synthesized at the Shell Biosciences Laboratory. [*cyclopropyl*-^{14}C]Cypermethrin (cis and trans mixture) (9.6 μCi/mg), the separate cis and trans isomers (11.1 μCi/mg), and [*cyano*-^{14}C]cypermethrin (7.13 μCi/mg) were obtained from the same source. All compounds were analyzed by thin-layer chromatography (TLC) on Merck silica gel F_{254} plates developed in toluene and were either >99.5% pure or purified by further TLC in toluene before use. The isomeric composition of some of the compounds was

Shell Research Ltd., Shell Toxicology Laboratory, Sittingbourne Research Centre, Sittingbourne, Kent ME9 8AG, U.K.

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measured by radio high-pressure LC as described later. Nonradioactive *cis*-cypermethrin and *trans*-cypermethrin were also obtained from Shell Biosciences Laboratory.

Treatment of Animals. *Balance and Tissue Residue Studies.* Six male and six female rats (Wistar strain, bred in Shell Toxicology Laboratory), 12 weeks of age, with mean weights of approximately 320 and 210 g, respectively, were each given a single oral dose of [*benzyl*-^{14}C]-*cis*-cypermethrin (0.61 mg, 1.7–2.5 mg/kg) in 0.5 mL of corn oil. Each rat was individually housed in a glass metabolism cage (Jencons Ltd., Hemel Hempstead, U.K.) and given free access to food and water. Urine and feces were collected daily until the rats were sacrificed, this being after 24 h for two rats of each sex, 72 h for a further two rats of each sex, and 8 days for the remaining animals. The animals were killed by anaesthetizing with Nembutal, followed by cardiac puncture. The blood was assayed for radioactivity as were samples of the kidney, liver, brain, skin, fat, muscle, gastrointestinal tract, and remaining carcass. All excreta and animal tissues were stored at −15 °C until required for radioassay. [*benzyl*-^{14}C]-*trans*-Cypermethrin (0.615 mg) in 0.8 mL of corn oil was dosed to three rats of each sex weighing approximately 250 g (male) and 200 g (female) (2.5–3.1 mg/kg). Respired air was collected from one male and one female by drawing air through the cages and into 5 N NaOH solution (300 mL). Urine, feces, and tissues were collected as described above. [*cyclopropyl*-^{14}C]Cypermethrin (0.52 mg) in corn oil (0.8 mL) was dosed to three rats of each sex weighing approximately 430 g (male) and 240 g (female) (1.2–2.2 mg/kg). Two more rats weighing 570 g (male) and 280 g (female) were dosed with 0.52 g of [*cyclopropyl*-^{14}C]cypermethrin, and respired air, urine, and feces were collected as described above for 72 h. Two female rats were dosed orally with 1.075 mg of [*cyclopropyl*-^{14}C]-*cis*-cypermethrin in 0.5 mL of corn oil, and a further two female rats were dosed with 0.87 mg of the trans isomer in 0.45 mL of oil. Urine and feces were collected daily for 3 days for radioassay; tissues were not analyzed. [*cyano*-^{14}C]-Cypermethrin (0.81–1.22 mg) was dosed in corn oil (0.37–0.56 mL) to three rats of each sex (4.3 mg/kg). Products were collected and the animals were sacrificed as described above at 72 h. $^{14}CO_2$ was collected as described above from one more rat of each sex dosed with [*cyano*-^{14}C]cypermethrin.

Fat Residue Depletion Study. Eight female rats were weighed, and each was dosed with 0.55 mg of [*benzyl*-^{14}C]-*cis*-cypermethrin (29.4 μCi/mg) in 0.25 mL of corn oil. The animals were maintained in normal holding cages which were cleaned daily for the first 3 days (to prevent recycling of radiochemical by coprophagy) and thereafter at 2–3-day intervals. The rats were killed in groups of two by cervical dislocation at 8, 14, 25, and 42 days after dosing. Fat (peritoneal), liver, and kidney samples were removed for radioassay. They were stored at −20 °C prior to analysis.

Biliary Elimination Study. Two male rats were fitted with biliary cannulae under anaesthesia (Abou-El-Makarem et al., 1967) and then dosed with [*cyclopropyl*-^{14}C]cypermethrin. Bile was collected for 4–5 h while maintaining the anaesthesia (thiopentone). A third rat was dosed orally with [*cyclopropyl*-^{14}C]cypermethrin prior to cannulation. Further details of the experiment are given in the footnotes to Table VIII.

Radioanalysis. Radioassay of urine and NaOH solution was carried out in NE 260 scintillator solution (Nuclear Enterprises Ltd., Edinburgh, Scotland) in a Packard Model 2450 liquid scintillation spectrometer. Blood (100 μL), feces (30–110 mg), and skin, kidney, muscle, brain, liver, intestine, and remaining carcass (30–150 mg) were combusted in duplicate in a Packard 306 sample oxidizer. The $^{14}CO_2$ produced was trapped in Carbosorb (9 mL) and mixed automatically with Permafluor (12 mL) for counting. Samples were analyzed in duplicate. Fat samples (30–50 mg) were combusted in triplicate or quadruplet. Efficiency of combustion (82–92%) was routinely monitored by combusting [^{14}C]glucose standard solution and [*cyclopropyl*-^{14}C]- and [*benzyl*-^{14}C]cypermethrin on cellulose powder. Counting efficiency was measured by the sample channels ratio method.

High-Pressure Liquid Radiochromatography. This was carried out by using simultaneous monitoring of the column effluents with a CE 212 variable-wavelength UV monitor (Cecil Instruments Ltd., Cambridge, U.K.) and a Packard Model 3021 flow analyzer. Cypermethrin isomers were analyzed on a column of 5-μm Hypersil (Shandon Southern Products Ltd., Runcorn, U.K.) (20 cm × 4.5 mm) run in hexane–30% water-saturated dichloromethane (4:1 v/v). R_t values (relative to the first peak eluted) were as follows: 1*R*,cis-*R* and 1*S*,cis-*S* isomers, 1.00; 1*R*,cis-*S* and 1*S*,cis-*R* isomers, 1.13; 1*S*,trans-*S* and 1*R*,-trans-*R* isomers, 1.38; 1*S*,trans-*R* and 1*R*,trans-*S* isomers, 1.52. This procedure was used to show that the [*benzyl*-^{14}C]-*cis*-cypermethrin contained radioactivity in the ratio 53:47 (*RR* + *SS*:*RS* + *SR*); the trans isomer ratio was 57:43. Quantification was obtained with a Model 5680 printing autoscaler (E.S.I. Nuclear Ltd., Reigate, U.K.).

Analysis of Fat Samples for Cypermethrin. Fat samples (2 g) from each of the two rats (depletion study) sacrificed at 8 days (no. 3 and 7) were combined. A similar sample was made from the rats sacrificed 25 days after dosing (no. 2 and 6). The weights and residues in each individual sample were computed to give calculated concentrations of residue in the 8- and 25-day samples of 0.32 and 0.17 μg (as cypermethrin) per g, respectively. The two samples were extracted with acetone–hexane (2:3) and cleaned up for GLC by a method in current use in Shell Toxicology Laboratory (Baldwin and Lad, 1979). The extract was radioassayed before and after cleanup, and the content of *cis*-cypermethrin in the extract was measured by GLC. Conditions were as follows: glass column, 1 m × 3 mm containing 4% (m/m) OV1 on 80–100-mesh Gas-Chrom Q; nitrogen carrier gas at a flow rate of 100 mL/min; column temperature, 260 °C; detector temperature, 315 °C; retention time of *cis*-cypermethrin, 2 min 55 s. Two spiked samples of fat (0.05 μg/g) were taken through the analytical procedure with a mean recovery of 77.5%.

RESULTS

Elimination of Radioactivity. *Benzyl*-^{14}*C Label.* The rates and routes of excretion of radioactivity following the administration of single oral doses of *cis*- and *trans*-cypermethrin are illustrated in Figure 1. Elimination was rapid and was mainly via the urine with the exception of the cis isomer in female rats. The difference between male and female rats in this respect was statistically significant. For example, the 0–24-h mean urinary output of radioactivity for 6 male rats was 53.0% (SEM ± 1.56) and for female rats was 35.4% (SEM ± 4.65). The Student *t* value (3.5872) indicated a significant difference at 0.01%. A sex difference was not observed with the trans isomer, which was 70% eliminated in the urine in 3 days. No $^{14}CO_2$ elimination was observed. Mean recovery for the cis isomer (including tissues, cage washings, etc.) was 98.9% in 8 days and for the trans isomer was 102.2% in 3 days (Table I).

Cyclopropyl-^{14}*C Label.* The radioactivity derived from

Table I. Total Recovery of Radioactivity from Rats following Oral Administration[a] of Benzyl-^{14}C-Labeled Cypermethrins

isomer dosed	sex	urine	feces	cage washings	skins	intestines	carcasses	total
cis	male	57.5	20.7	1.4	4.7	14.6	6.0	104.9
cis	male	48.8	29.1	1.1	2.5	13.3	4.9	99.7
cis	male	63.0	28.7	0.1	3.7	1.6	4.2	101.3
cis	male	66.3	25.9	0.2	0.9	3.0	3.1	99.4
cis	male	62.8	32.6	0.4	0.4	0.9	3.5	100.6
cis	male	60.9	31.0	1.7	0.7	0.7	1.9	96.9
	mean							100.5
cis	female	43.2	30.4	1.6	4.5	6.1	3.9	89.7
cis	female	28.8	48.0	0.6	2.7	16.4	4.4	100.9
cis	female	31.5	71.6	0.08	1.4	0.6	1.4	106.6
cis	female	35.8	51.2	1.0	1.3	0.7	3.0	93.0
cis	female	61.4	34.9	0.08	1.6	0.7	2.5	101.2
cis	female	49.6	40.2	0.4	0.2	0.4	1.3	92.1
	mean							97.3
trans	male	66.8	27.2	1.5	1.0	0.7	0.4	97.6
trans	male	65.5	38.2	0.7	1.9	1.1	0.9	108.3
trans	male	81.9	19.5	0.2	0.4	0.5	0.6	103.0
	mean	71.4	28.3	0.8	1.1	0.8	0.6	103.0
trans	female	74.4	22.2	2.8	0.7	1.4	0.8	102.3
trans	female	74.0	26.6	0.7	0.5	0.6	0.5	102.9
trans	female	74.9	19.4	0.9	2.9	0.6	0.5	99.2
	mean	74.4	22.7	1.5	1.4	0.9	0.6	101.5

[a] Six rats of each sex were given single oral doses of [*benzyl*-^{14}C]-*cis*-cypermethrin (1.7–2.5 mg/kg) in corn oil. Two males and two females were sacrificed 1, 3, and 8 days after dosing. Three rats of each sex were similarly dosed with [*benzyl*-^{14}C]-*trans*-cypermethrin (2.5–3.1 mg/kg) and sacrificed 3 days after dosing. Results are expressed as percentage of given dose.

Table II. Total Recovery of Radioactivity from Rats following Oral Administration[a] of Cyclopropyl-^{14}C-Labeled Cypermethrin (Cis plus Trans Mixture)

sex	urine	feces	cage washings	skins	intestines	carcasses	total
male	55.8	25.3	6.1	1.7	10.7	4.5	104.1
male	57.1	29.8	7.3	0.6	5.7	3.4	103.9
male	54.6	30.9	5.6	1.2	8.9	3.3	104.5
mean	55.8	28.7	6.3	1.2	8.4	3.7	104.2
female	71.6	23.4	2.4	0.8	1.9	1.7	101.8
female	62.3	29.7	5.6	1.0	1.9	2.2	102.7
female	65.7	27.8	5.1	1.5	4.7	1.8	106.6
mean	66.5	27.0	4.4	1.1	2.8	1.9	103.7

[a] Three rats of each sex were given single oral doses of [*cyclopropyl*-^{14}C]cypermethrin (1.2–2.2 mg/kg) and sacrificed 3 days after dosing. The results are expressed as percentage of the given dose.

[*cyclopropyl*-^{14}C]-*cis,trans*-cypermethrin was eliminated mainly in the urine of both male and female rats (Figure 1); the remainder was eliminated in the feces. Less than 0.1% of the dose was eliminated as $^{14}CO_2$, and therefore the total recovery in the balance study (in which $^{14}CO_2$ was not measured) was good (Table II).

Cyano-^{14}C Label. The pattern of elimination of the cyano-^{14}C label was very different from those of the other two labels. This was expected because other α-cyano pyrethroids are metabolized by hydrolysis, releasing cyanide ion which is rapidly converted into thiocyanate ion; the latter, however, is only slowly eliminated from the body (Ohkawa et al., 1979). The fecal elimination profile was very similar to that for the other labels and suggests that much of the fecal radioactivity is in the form of a structure which retains the whole molecular skeleton. The low yield of $^{14}CO_2$ (1–2%) suggests that very little of the cyanide ion entered the one-carbon pool. Total recovery, including the substantial amount in the tissues, is shown in Table III.

Tissue Residues. *Benzyl-^{14}C Label.* Residues derived from *cis*-cypermethrin were measured 1, 3, and 8 days after a single oral dose. The results (Table IV) therefore provide a measure of the rate of depletion of the radioactivity from the various tissues. One day after dosing, residual radioactivity lay in the order fat > liver > kidney > blood >

Table III. Total Recovery of Radioactivity from Rats following Oral Administration[a] of Cyano-^{14}C-Labeled Cypermethrin (Cis plus Trans Mixture)

						intestines							
sex	urine	feces	$^{14}CO_2$[b]	cage washings	skin	stomach	stomach contents	small intestine	small intestine contents	large intestine	large intestine contents	carcass	total
male	12.1	41.3		0.5	6.8	0.5	5.5	0.7	0.9	0.4	1.4	11.7	81.8
male	6.9	51.9		0.2	12.5	0.4	6.4	0.6	0.8	0.4	1.6	8.2	89.9
male	6.0	32.8		0.3	12.8	1.0	7.7	0.7	0.4	0.9	7.2	12.3	82.1
mean	8.3	42.0	1.2	0.3	10.7	0.6	6.5	0.7	0.7	0.6	3.4	10.7	85.7[c]
female	12.6	56.5		0.5	8.8	0.7	6.0	1.1	0.9	0.4	1.4	8.9	97.8
female	7.2	65.9		0.6	10.3	0.8	4.7	0.7	0.3	0.2	0.7	6.1	97.5
female	9.0	49.3		0.4	22.7	1.4	7.9	0.7	1.6	0.3	3.0	6.7	103.0
mean	9.6	57.2	1.5	0.5	13.9	1.0	6.2	0.8	0.9	0.3	1.7	7.2	100.8[c]

[a] Three rats of each sex were given single oral doses of [*cyano*-^{14}C]cypermethrin (4.3 mg/kg) and sacrificed 3 days after dosing. [b] $^{14}CO_2$ measurements were made on one animal of each sex additional to those described above. [c] Includes $^{14}CO_2$.

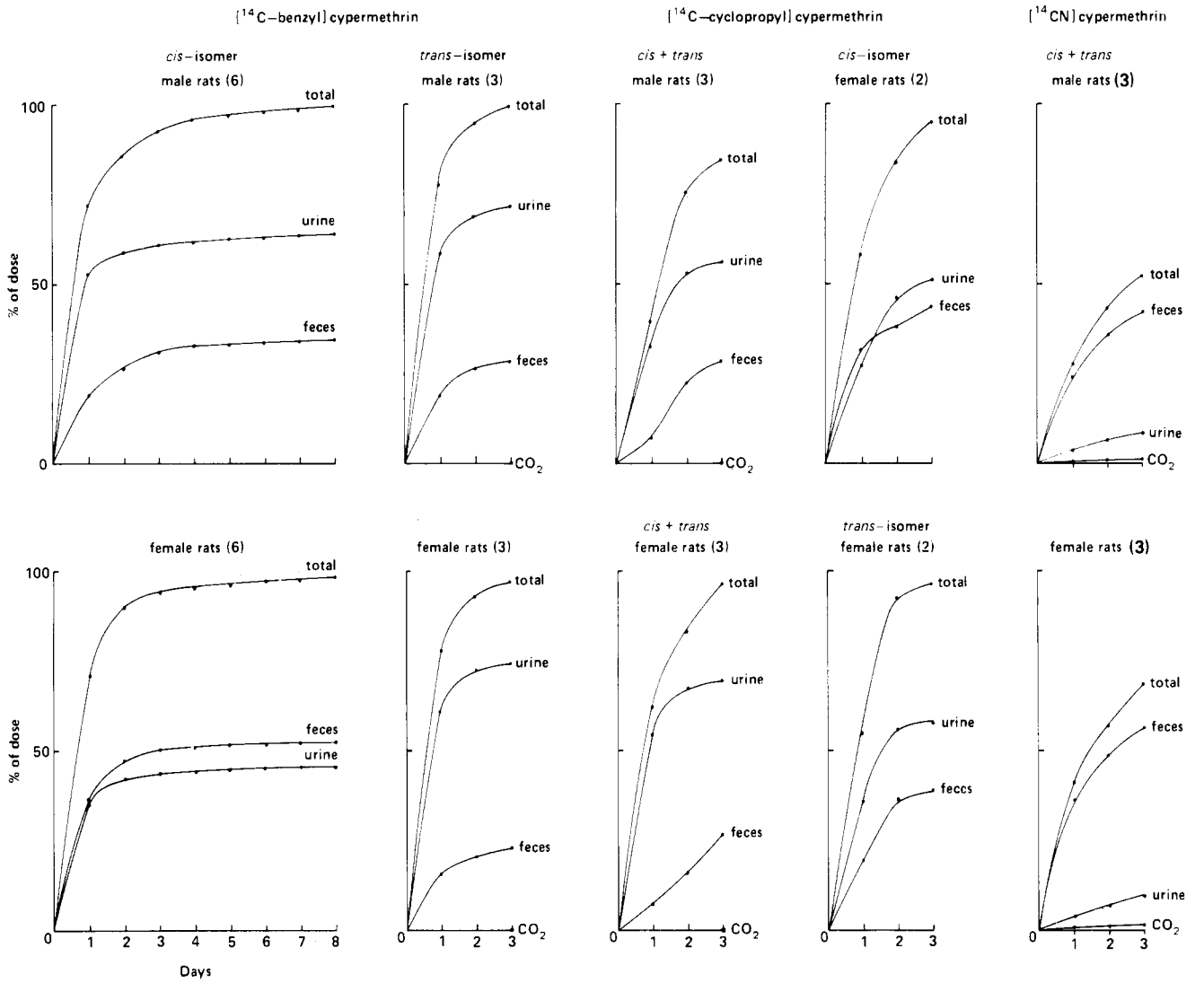

Figure 1. Rates of elimination of radioactivity from rats dosed with variously labeled forms of cypermethrin.

Table IV. Radioactivity[a] Remaining in Rat Tissues 1, 3, and 8 Days after a Single Oral Dose of [*benzyl*-^{14}C]-*cis*-Cypermethrin (1.7–2.5 mg/kg)

	male			female		
tissue	day 1	day 3	day 8	day 1	day 3	day 8
liver	0.42	0.19	0.053	0.41	0.08	0.048
	0.56	0.18	0.062	0.98	0.072	0.034
kidney	0.17	0.052	0.014	0.17	0.027	0.015
	0.16	0.074	0.021	0.30	0.036	0.010
fat	1.08	1.02	0.97	1.34	0.83	0.93
	0.91	0.83	1.33	1.46	1.04	1.07
muscle	0.023	0.006	0.002	0.024	0.004	0.009
	0.022	0.008	0.005	0.053	0.003	0.004
brain	0.005	0.002	0.001	0.008	0.002	0.001
	0.01	0.002	0.001	0.024	0.002	0.001
blood	0.13	0.026	0.01	0.16	0.016	0.007
	0.14	0.030	0.013	0.47	0.019	0.008

[a] Individual values are given, expressed as micrograms of cypermethrin per gram of tissue.

muscle > brain and ranged from 1–2 μg/g in fat to 0.005–0.024 μg/g in brain. The residues depleted rapidly from all tissues except fat. Residues derived from the trans isomer were measured only 3 days after dosing, as were the residues derived from the other labeling modes. These day 3 values are compared with those for [*benzyl*-^{14}C]-*cis*-cypermethrin in Table V. Trans isomer residues follow the same general pattern as that found for the cis isomer with the exception that radioactivity in the fat was much lower (particularly in male rats).

Cyclopropyl-^{14}*C Label.* Residues derived from the cis/trans mixture were measured at day 3 (Table V). They were consistently low in all tissues with the exception of fat, in which they were intermediate between those for the (benzyl-^{14}C-labeled) *cis*- and *trans*-cypermethrin.

Cyano-^{14}*C Label.* These residues, although somewhat higher than those derived from benzyl-^{14}C and cyclopropyl-^{14}C labels, followed the same general pattern (i.e., high only in fat). Much of the residual radioactivity was associated with the stomach (content), intestines, and skin (Table III) and not with the internal organs.

Identity and Kinetics of Depletion of Residue in Fat. The only residue which was significantly higher than the average was that derived from the cis isomer (all labels) in fat. This was studied further with more animals over a longer period (up to 42 days). The results (Table VI) show that radioactivity was eliminated from fat with a half-life of 11.7 days (95% confidence limits 8.6–18.2). The very low residues in liver and kidney were eliminated at a similar rate: $t_{1/2}$ was 8.0 (5.3–16.6) and 15.4 (12.7–19.5), respectively. The residue in fat was found to be mostly unchanged *cis*-cypermethrin (Table VII). It is likely that the liver and kidney residues at any point between 8 and 42 days were due to the metabolic processing of a small amount of cypermethrin released from fat.

Elimination of Radioactivity in Bile. Compared with the urinary route, only a very small proportion of orally

Table V. Radioactivity[a] Remaining in Rat Tissues 3 Days after a Single Oral Dose of Benzyl-^{14}C-, Cyclopropyl-^{14}C-, and Cyano-^{14}C-Labeled Cypermethrin

	liver				kidney				fat				muscle				brain				blood			
	benzyl-^{14}C		cPr-^{14}C[b]	CN-^{14}C[b]	benzyl-^{14}C		cPr-^{14}C	CN-^{14}C	benzyl-^{14}C		cPr-^{14}C	CN-^{14}C	benzyl-^{14}C		cPr-^{14}C	CN-^{14}C	benzyl-^{14}C		cPr-^{14}C	CN-^{14}C	benzyl-^{14}C		cPr-^{14}C	CN-^{14}C
sex	cis	trans			cis	trans			cis	trans			cis	trans			cis	trans			cis	trans		
males	0.19	0.055	0.30	0.92	0.052	0.053	0.10	1.07	1.02	0.018	0.14	0.64	0.006	0.005	0.01	0.39	0.002	0.001	0.01	0.11	0.026	0.015	0.07	2.16
	0.18	0.055	0.37	0.88	0.074	0.041	0.08	1.11	0.83	0.12	0.31	1.16	0.008	0.003	0.01	0.40	0.002	0.001	0.007	0.13	0.030	0.018	0.04	2.36
		0.052	0.45	1.48		0.041	0.10	1.66		0.26	0.43	1.87		0.009	0.02	0.55		0.002	0.008	0.20		0.012	0.04	2.98
means	0.18	0.054	0.37	1.09	0.063	0.045	0.09	1.28	0.92	0.19	0.31	1.22	0.007	0.006	0.01	0.45	0.002	0.001	0.009	0.15	0.028	0.015	0.05	2.50
females	0.08	0.086	0.14	1.04	0.027	0.070	0.06	1.17	0.83	0.65	0.87	0.63	0.004	0.004	0.01	0.36	0.002	0.001	0.006	0.11	0.016	0.031	0.04	2.50
	0.072	0.067	0.10	0.61	0.036	0.057	0.05	0.67	1.04	0.38	0.64	0.28	0.003	0.002	0.007	0.22	0.002	0.001	0.01	0.08	0.019	0.015	0.04	1.41
		0.076	0.12	0.80		0.045	0.06	0.82		0.39	0.64	1.43		0.005	0.008	0.39		0.001	0.006	0.09		0.014	0.04	1.48
means	0.076	0.076	0.12	0.82	0.031	0.057	0.06	0.89	0.93	0.47	0.72	0.78	0.003	0.005	0.009	0.32	0.002	0.001	0.008	0.09	0.017	0.02	0.04	1.80

[a] Individual values are given, expressed as micrograms of cypermethrin per gram of tissue. [b] cPr-^{14}C, cyclopropyl-^{14}C; CN-^{14}C, cyano-^{14}C.

Table VI. Residual Radioactivity in Tissues of Female Rats following the Oral Administration of a Single Dose of [*benzyl*-^{14}C]-*cis*-Cypermethrin (~ 2.5 mg/kg)

rat	wt at dosing, g	sacrificed, days after dosing	wt at death, g	residues, μg/g[a] fat	liver	kidney
3	253	8	277	0.34	0.016	0.008
7	200	8	220	0.31	0.020	0.009
1	217	14	248	0.26	0.008	0.007
5	200	14	230	0.54	0.007	0.007
2	221	25	255	0.195	0.003	0.004
6	205	25	260	0.150	0.005	0.004
4	221	42	297	0.046	<0.001	<0.001
8	234	42	308	0.055	<0.001	<0.001

[a] Expressed as cypermethrin.

Table VII. Identification of the Residue in Fat

fat sample[a]	residue, μg/g,[b] by radioanalysis: tissue combustion	assay of extract	assay of final extract	residue, μg/g, by GLC: final extract	corrected for recovery[c]
rat (day 8)	0.32	0.29	0.20	0.22	0.28
rat (day 25)	0.17	0.15	0.13	0.13	0.17

[a] See Table VI. [b] Expressed as cypermethrin. [c] Two spiked samples of fat (0.05 μg/g) were taken through the analytical procedure, giving a mean recovery of 77.5%.

Table VIII. Elimination of Radioactivity in Bile of Male Rats following Oral Dosing with [*cyclopropyl*-^{14}C]-Cypermethrin

rat	dose,[a] mg	collection time, h	% dose in bile
1[b]	0.52	4	1.6
2[c]	0.53	4	1.5
3[b]	0.26	5	0.95

[a] Rats weighed 250–300 g. [b] Cannulated before dosing. [c] Cannulated after dosing.

administered [*cyclopropyl*-^{14}C]cypermethrin (metabolites) was eliminated in the bile (Table VIII). The use of β-glucuronidase treatment and TLC analysis indicated that the radioactivity was due to the glucuronide conjugates of the *cis*- and *trans*-cyclopropanecarboxylic acid derived from the hydrolysis of cypermethrin. No unchanged cypermethrin was found in the bile, and none would be expected in view of its highly lipophilic character. The conjugates (M_r 385) fall very near the minimum molecular weight required for secretion into rat bile (325 ± 50) (Hirom et al., 1972) and, therefore, would be expected to be excreted mostly in the urine.

DISCUSSION

The results show that cypermethrin is rapidly eliminated from rats with the exception of a small proportion of the cis isomer which, having reached the fat, is eliminated from that tissue with a half-life of 12 days. The characteristics of the elimination are very similar to those of the other cyano-3-phenoxybenzyl pyrethroids fenpropathrin (Crawford and Hutson, 1977) and deltamethrin (Ruzo et al., 1978) and of the primary alcohol ester permethrin (Gaughan et al., 1977). The incorporation of the α-cyano group into the 3-phenoxybenzyl esters introduces a secondary alcohol function into the cyano pyrethroids which increases

their stability to the hepatic carboxylesterases (Soderlund and Casida, 1977). However, it apparently has little effect on metabolism and elimination in vivo.

The difference between the cis and trans isomers of cypermethrin in the amounts retained in the fat is of interest. In terms of lipophilic character, there is little difference between the two isomers, and experiments in mice (Hutson et al., 1981) show that the amount reaching fat is similar with both isomers. The difference lies in the rate of release from this tissue. It is possible that hydrolysis of the ester bond occurs in the fatty tissue, perhaps catalyzed by a lipase. This hydrolysis would be expected to proceed much more rapidly with the trans isomer than with the cis isomer.

The rapid elimination of cypermethrin from rats is due primarily to the efficient cleavage of the ester bond giving rise to polar metabolites which are further oxidized and conjugated before excretion. The structural elucidation of these metabolites and their mechanisms of formation will be described in a subsequent paper.

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Phenthoate Applied to California Citrus Trees: Residue Levels on Foliage and Soil, in Air, and on and in Fruit

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Phenthoate [Cidial, ethyl α-[(dimethoxyphosphinothioyl)thio]benzeneacetate] was field applied to California orange, lemon, and grapefruit trees. Applications included two dilute spray rates and one low-volume rate. Residue levels of phenthoate and its oxygen analogue (oxon) were determined for assisting in setting worker reentry safety intervals and legal fruit tolerances. The half-lives for the dissipation of phenthoate from orange, lemon, and grapefruit rind were 15 ± 1, 20 ± 8, and 20 ± 5 days, respectively. The maximum rind residue of phenthoate oxon was 0.09 ppm in orange rind. No determinable phenthoate residues (>0.03 ppm) were found in the edible portion of the citrus fruits. Dislodgeable phenthoate residues dissipated rapidly from foliage. The half-lives were 3.6 ± 0.3 and 3.1 ± 0.3 days for phenthoate dissipation from orange and lemon foliage, respectively. The maximum oxon level found was 0.07 μg/cm^2 of leaf surface. Phenthoate appeared to persist longer on orange fruit surfaces ($t_{1/2}$ = 10 days) than on the corresponding foliar surfaces ($t_{1/2}$ = 3.9 days). Phenthoate dissipation from soil dust on the grove floor after a 7.5 lb of AI (1500 gal)$^{-1}$ acre^{-1} application had a $t_{1/2}$ value of 29 days. The highest level of phenthoate and its oxon found were 180 and 30 ppm, respectively. Air samples collected from under a tree after a 7.5 lb of AI (100 gal)$^{-1}$ acre^{-1} application contained 10 μg of phenthoate/m^3 at 3-days postapplication and lesser amounts thereafter.

Phenthoate [ethyl α-[(dimethoxyphosphinothioyl)-thio]benzeneacetate; Cidial, Elsan, Papthion, Tanone] is a broad-spectrum scalicide/thripsicide/acaricide that has been used for ~18 years for pest control on citrus in the Mediterranean countries. It is a potentially useful insecticide for the chemical control of the California red scale (*Aonidiella aurantii* Mask) and the woolly whitefly (*Aleurothrixus floccosus*) in California. The woolly whitefly has moved into portions of southern California but has not yet developed into a major pest.

Fruit residue data for phenthoate after application to mature lemon, grapefruit, and orange trees and dislodgeable foliar residue data after application to orange trees were reported by Iwata et al. (1977a). However, the U.S. Environmental Protection Agency (EPA) has requested that additional information be provided prior to the granting of a registration for use of phenthoate on citrus. Since the emphasis on residue data relevant to assessing worker exposure to cholinesterase-inhibiting insecticide residues was not as great in 1973–1974 when the tests were conducted as it currently is, Iwata et al. (1977a) did not provide data on dislodgeable residue levels of phenthoate

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