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UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF

PREVENTION, PESTICIDES AND TOXIC SUBSTANCES

DATE: June 18, 2002

TXR No. 0050649

MEMORANDUM

SUBJECT: **PERMETHRIN**: Updated DERs and Executive Summaries

FROM: Yung G. Yang, Ph.D. Yung G. Yang 6/20/2002
Toxicology Branch
Health Effects Division (7509C)

THROUGH: Alberto Protzel, Ph.D.
Branch Senior Scientist, Toxicology Branch
Health Effects Division (7509C)

TO: Stacey Milan/Robert McNally
PM 60
Special Review and Reregistration Division (7508W)

DP Barcode:	D269531	**Submission:**	S504352
Chemical:	Permethrin	**Case:**	819432
PC Code:	109701	**CAS No.:**	52645-53-1

Action: Prepare and update the toxicology database of permethrin for a RED.

Response: The toxicology database for permethrin has been prepared and reviewed by the Toxicology Branch. The database has been peer reviewed by the Health Effects Division's Hazard Identification Assessment Review Committee (HIARC) on April 18, 2002. The toxicology RED chapter for permethrin has been prepared under a separate memorandum to support a Reregistration Eligibility Decision (RED).

The updated DERs and executive summaries are as follows and are attached.

[redacted]

13544

054554

Chemical:	**Permethrin, mixed cis,trans**
PC Code:	**109701**
HED File Code	**13000 Tox Reviews**
Memo Date:	**06/18/2002**
File ID:	**TX050649**
Accession Number:	**412-03-0017**

HED Records Reference Center
11/04/2002

DETAILED EXECUTIVE SUMMARY

PERMETHRIN/109701

SALMONELLA/ESCHERICHIA/MAMMALIAN ACTIVATION GENE MUTATION ASSAY [OPPTS 870.5100[1] (§84-2)]
MRID 00126834

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Health Sciences Research Division
Oak Ridge National Laboratory*
Oak Ridge, TN 37831
Task Order No. 02-05

Primary Reviewer: B.L. Whitfield, Ph.D.	Signature:	B L Whitfield
	Date:	MAR 2 6 2002
Secondary Reviewers: Cheryl B. Bast, Ph.D., D.A.B.T.	Signature:	Cheryl B Bast
	Date:	MAR 2 6 2002
Robert H. Ross, Group Leader	Signature:	Robert H. Ross
	Date:	MAR 2 6 2002
Quality Assurance: LeeAnn Wilson, M.A.	Signature:	L. A. Wilson
	Date:	MAR 2 6 2002

Disclaimer

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Oak Ridge National Laboratory, Managed and Operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

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[1] 870.5100 - Reverse mutation *E. coli* WP2 and WP2uvrA
870.5140 - Gene mutation *Aspergillus nidulans*
870.5250 - Gene mutation *Neurospora crassa*

EPA Reviewer: Yung Yang, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/21/2002
Signature: Joycelyn Stewart
Date 6/3/2002

DATA EVALUATION RECORD

TXR#: 0050649

STUDY TYPE: ***In vitro* Bacterial Gene Mutation** *Salmonella typhimurium* / mammalian activation gene mutation assay [OPPTS 870.5100[2] (§84-2)]

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin (96% a.i., 45% cis and 55% trans); Pydrin (95% a.i.); Fenpropathrin (97% a.i.); and Cypermethrin (96.5% a.i., 47% cis and 53% trans)

SYNONYMS: FMC 33297 (for Permethrin)

CITATION: Suzuki, H. (1977) Studies on mutagenicity of some pyrethroids on *Salmonella* strains in the presence of mouse hepatic S9 fractions. Biochemical Toxicology Laboratory, Research Department, Institute for Biological Science, Hyogo, Japan. Laboratory Report No. AT-70-0157. July 1, 1977. MRID 126834. Unpublished.

SPONSOR: Not specified.

EXECUTIVE SUMMARY: In a reverse gene mutation assay in bacteria (MRID 126834), strains TA98, TA100, TA1535, TA1537 and TA1538 of *S. typhimurium* were exposed to Permethrin (96% a.i., Lot # A-4) in DMSO at concentrations of 10, 100 and 1000 µg/plate in the presence and absence of mammalian metabolic activation (S9-mix). Three other pyrethroids were also tested at the same three concentrations: Pydrin (95% a.i., Lot No. 022016), Fenpropathrin (97% a.i., Lot No. 022018) and Cypermethrin (96.5% a.i., Lot No. T-1). The S9-fractions were obtained from Kanechlor 400 induced ICR, CDF1, BALB/c, ddY, BDF1 and C57BL/6 mouse liver.

Permethrin was tested up to 1000 µg/plate but the justification for this upper dose was not given. No significant increases (defined as a two-fold or greater increase) in the average number of revertants per plate over the corresponding solvent control values were seen in any tester strain at any test material concentration with or without S9-mix from any of the mouse strains. The

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[2] 870.5100 - Reverse mutation *E. coli* WP2 and WP2uvrA, *S. typhimurium* TA97, TA98, TA100, TA1535, TA1537
870.5140 - Gene mutation Aspergillus nidulans
870.5250 - Gene mutation Saccharomyces cerevisiae

solvent and positive control values were appropriate. **Under the condition of this study, permethrin was not mutagenic either with or without S9-mix.**

This study is classified as **Unacceptable/Guideline.** It does not satisfy the guideline requirements for Test Guideline OPPTS 870.5100[1]; OECD 471 for *in vitro* mutagenicity (bacterial reverse gene mutation) data. No signed and dated GLP statement was provided, individual plate counts were not reported, the composition of the S9-mixes were not detailed, no historical control values were provided and no justification for the upper concentration of test materials was given. Because of the number of omissions, the age of the study and likely unavailability of original data and the short time required to conduct an Ames Test, the study should be repeated.

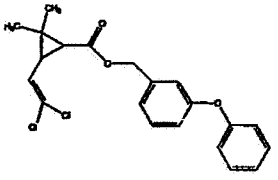
COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1.a. Test material: Permethrin

Description:	Not provided
Lot/Batch #:	A-4
Purity:	96% a.i. (45% cis and 55% trans)
CAS # of TGAI:	Not provided (52645-53-1 from ChemIDplus)
Solvent Used:	DMSO



1.b. Test material: Pydrin

Description:	Not provided
Lot/Batch #:	22016
Purity:	95% a.i.
CAS # of TGAI:	Not provided (51630-58-1 from ChemIDplus)
Structure:	Not provided
Solvent Used:	DMSO

1.c. Test material: Fenpropathrin

Description:	Not provided
Lot/Batch #:	22018
Purity:	97% a.i.
CAS # of TGAI:	Not provided (64257-84-7, 39515-41-8 from ChemIDplus)
Structure:	Not provided
Solvent Used:	DMSO

1.d. Test Material: Cypermethrin

Description:	Not provided
Lot/Batch #:	T-1
Purity:	96.5% a.i. (47% cis and 53% trans)
CAS # of TGAI:	Not provided (52315-07-8 from ChemIDplus)
Structure:	Not provided
Solvent Used:	DMSO

2. Control materials:

Negative: None

Solvent (final conc'n): DMSO / 10 µL/plate

Positive: Nonactivation:

Sodium azide ____ µg/plate
2-Nitrofluorene ___ µg/plate
9-Aminoacridine ____ µg/plate
Other (list):

Activation:

2-Aminoanthracene (2-anthramine) _____ µg/plate
Other (list):
2-Acetylaminofluorene 10 and 100 µg/plate (all strains)
Dimethylnitrosamine 1000 and 2000 µg/plate (all strains)

The two positive controls, both requiring activation, were used with and without S9-mix.

3. Activation: S9 derived from

x	Induced		Aroclor 1254		Rat	x	Liver
	Non-induced		Phenobarbitol	x	Mouse		Lung
			None		Hamster		Other
		x	Other: Kanechlor 400		Other		

The S9-fraction was obtained from ICR, CDF1, BALB/c, ddY, BDF1 and C57Bl/6 mice.

Describe S9 mix composition: Composition of the S9-mix was not provided.

4. **Test organisms: *S. typhimurium* strains**

	TA97	x	TA98	x	TA100		TA102		TA104
x	TA1535	x	TA1537	x	TA1538		list any others:		

Properly maintained? ☐ Yes ☐ No
Checked for appropriate genetic markers (*rfa* mutation, R factor)? ☐ Yes ☐ No

The information required to answer the above two questions was not provided.

5. **Test compound concentrations used:**

Mutagenicity assay:
Nonactivated and activated conditions: 10, 100, 1000 µg/plate of the respective pyrethroid in all strains. Three replicate trials were conducted.

B. **TEST PERFORMANCE:**

1. **Type of *Salmonella* assay:**
x standard plate test
__ pre-incubation (__ minutes)
__ "Prival" modification *(i.e. azo-reduction method)*
__ spot test
__ other

2. **Protocol:** The experimental protocol description was limited to the comment "the bacterial cells (ca. 2 x $10^{8)}$ mixed with S-9 and the chemical dissolved in DMSO were poured onto minimal plate and incubated for 48 hr at 37°C."

3. **Statistical analysis:** No statistical analysis was performed.

4. **Evaluation criteria:** The average number of revertants per plate was reported. No other evaluation criteria were given.

II. **REPORTED RESULTS:**

A. **PRELIMINARY CYTOTOXICITY ASSAY:** No preliminary cytotoxicity assay was reported.

B. **MUTAGENICITY ASSAY:** Three concentrations of Permethrin or one of the other pyrethroids (10, 100 or 1000 µg/plate) were tested without S9-mix and with an S9-mix from each of the six mouse strains. There was no evidence of a mutagenic effect of any of the four pyrethroids in any tester strain at any test material concentration with or without S9-

mix. Results of the mutagenicity assay with Permethrin are summarized in Appendix Table 1 (MRID 126834, pp. 6).

III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** The investigators concluded that neither Permethrin or any of the other pyrethroids tested were mutagenic in any of the tester strains at any concentration up to 1000 µg/plate, with or without S9-mix from any of the six mouse strains.

B. **REVIEWER COMMENTS:** The reviewer agrees with the investigators' conclusion of negative responses based on the information provided; however, the study does not meet some of the current acceptance criteria. Individual plate counts were not provided, no signed and dated GLP statement was provided, no historical control values were provided and the components of the cofactor solution and the percent S9-fraction in the mix were not given. No justification was given for limiting the upper dose to 1000 µg/plate rather than the limit value of 5000 µg/plate.

The study is unacceptable as presented but would be acceptable if the testing laboratory could provide the missing information. Due to the age of the study, the missing data may no longer be available and a repeat assay is more practical.

C. **STUDY DEFICIENCIES:** Study deficiencies were described in the Reviewer Comments section. Most of the deficiencies are likely data presentation deficiencies rather than experimental deficiencies; however, because of the number of omissions, the age of the study and the short time required to conduct another Ames Test, the study should be repeated.

REFERENCES:

Ames, B.N., J. McCann and E. Yamasaki (1975) Methods for detecting carcinogens and mutagens with the *Salmonella*/mammalian microsome mutagenicity test. Mutation Res. 31:347-364.

APPENDIX

(MRID 126834)

THE FOLLOWING ATTACHMENTS ARE NOT AVAILABLE ELECTRONICALLY. SEE THE FILE COPY.

Tox Review 50649

Page 10 is not included in this copy.

Pages _____ through _____ are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

X FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

EXECUTIVE SUMMARY

PERMETHRIN

STUDY TYPE: SUBCHRONIC NEUROTOXICITY- RAT [NONGUIDELINE] MRID NO. 40766807

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order 01-85

Primary Reviewer:
C. B. Bast, Ph.D., D.A.B.T.

Signature: Cheryl B Bast
Date: JAN 0 5 2001

Secondary Reviewers:
S. S. Talmage, Ph.D., D.A.B.T.

Signature: Sylvia S. Talmage
Date: JAN 0 5 2001

Robert H. Ross, M.S., Group Leader

Signature: Robert H. Ross
Date: JAN 0 5 2001

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PERMETHRIN

Subchronic Neurotoxicity Study [Nonguideline]

EPA Reviewer: Yung G. Yang, Ph.D. Date: 10/10/2001
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: J. Stewart, Ph.D. Date: 11/24/2001
Toxicology Branch, HED (7509C)

DATA EVALUATION RECORD

TXR# 0050649

This is an updated executive summary of a DER (MRID 40766807, HED Doc. No. 005946. The final conclusion has not been changed.

STUDY TYPE: Subchronic Neurotoxicity-Rat; Nonguideline

OPPTS Number: Nonguideline
DP BARCODE: D269531
PC CODE: 109701

SUBMISSION CODE: S504352
TOX CHEM NO: 652B

TEST MATERIAL: Permethrin (98%), 40:60 cis/trans

CHEMICAL NAME: Permethrin

CITATION: Snodgrass, H.L. and Cantu, R.M. (1987). Neurotoxicity in rats following subchronic ingestion of Permethrin-treated food. Aberdeen Proving Ground, MD, Study No. 75-51-0351-87. March 6, 1987. MRID 40766807. Unpublished.

SPONSOR: U.S. Army Environmental Hygiene Agency

EXECUTIVE SUMMARY: In a subchronic oral neurotoxicity study (MRID 40766807), Sprague-Dawley rats (10/sex/group) were administered Permethrin (98%, 40:60 cis/trans, Lot No. PL85-216) in acetone at concentrations of 0, 100, 200, or 400 mg/kg/day in the diet for 90 days (main study). Two control groups were included, one was an untreated control group and the other was a vehicle (acetone treated diet) control group. After the 90 days, the rats in the main study were sacrificed by a special procedure designed to allow for fixation of the nervous system *in situ*. The experiment also included a special recovery component that consisted of 10 male and 10 female rats in the 400 mg/kg/day and untreated control groups; these animals were sacrificed 6 weeks after the completion of dosing after being maintained on untreated control diet. Neurological tissues from control and high-dose animals were examined microscopically. Functional observational battery (FOB) and motor activity testing were not performed.

There were no treatment-related deaths. Clinical signs included hyperexcitability, intermittent tremors, and irritability in mid-dose males during the first 3 weeks of treatment and intermittent tremors in mid-dose females during the first week of treatment. High-dose rats exhibited hyperexcitability, intermittent and continuous tremors, twitching, nystagmus (males only) and combativeness (males only) throughout the treatment period. Body weight gain was decreased 6 to 13% in high-dose males from treatment week 11 to post-dosing week 2; and 5 to 9% in high-dose females compared to controls from weeks 3 to 13. No treatment-related food consumption effects were noted. There were no gross lesions associated with treatment and there were no

microscopic observations indicative of a neurotoxic effect.

The systemic LOAEL is 200 mg/kg/day based on tremors and irritability. The systemic NOAEL is 100 mg/kg/day. The NOAEL is > 400 mg/kg/day with respect to morphological and histological changes.

This study is classified **Acceptable/Nonguideline.** The data provide useful information suggesting no morphological or histological effects in rats fed 400 mg/kg/day in the diet for 90 days.

DATA EVALUATION RECORD

PERMETHRIN (PP557)
(S-504352)

STUDY TYPE: MULTIGENERATION REPRODUCTION - RAT (870.3800 [§83-4a])
MRID 92142092, 120271, 92142031

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task No. 02-04

Primary Reviewer:
Mary Lou Daugherty, M.S.

Signature: Robert H. Ross for M L Daugherty
Date: NOV 28 2001

Secondary Reviewers:
Carol Forsyth, Ph.D., D.A.B.T.

Signature: Carol Forsyth
Date: NOV 28 2001

Robert H. Ross, M.S. Group Leader

Signature: Robert H. Ross
Date: NOV 28 2001

Quality Assurance:
Gary Sega, Ph.D.

Signature: Robert H. Ross for Gary Sega
Date: NOV 28 2001

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PERMETHRIN

Reproduction Study [870.3800 (§83-4a)]

EPA Reviewer: Yung G. Yang, Ph.D. Yung G. Yang, Date 6/5/2002
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Joycelyn Stewart, Date 6/5/2002
Toxicology Branch, HED (7509C)

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Multigeneration Reproduction - Rat [OPPTS 870.3800 (§83-4)]

DP BARCODE: D269531
P.C. CODE: 109701

SUBMISSION CODE: S504352
TOX. CHEM. NO.: 652BB

TEST MATERIAL (PURITY): PP557 (Seven batches used: purity ranged from 94.0% to 98.9%; nominal isomeric ratio, 40% *cis*:60% *trans*.)

SYNONYMS: PP557; 3-phenoxybenzyl (±)-*cis*:*trans*-3-(2,2-dichlorovinyl)-2,2-dimethyl-cyclopropane-1-carboxylate.

CITATION: Hodge, M.C.E., Banham, P.B., Glaister, J.R., et al. (1977) Permethrin (PP557): 3-Generation Reproduction Study in Rats, Volumes I and II. ICI Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire SK10 4TJ, UK. CTL Report Number: CTL/P/361. CTL Study Number: RR0015. December 1977. MRID 120271. Unpublished.

Barber, J.E. (1990) Phase 3 reformat of MRID 120271. ICI Americas, Inc., Agricultural Products, Wilmington, Delaware 19897. May 2, 1990. MRID 92142092. Unpublished.

Guttmann, E.M. (1990) Phase 3 Summary of MRID 120271. ICI Americas, Inc., Agricultural Products, Wilmington Delaware 19897. April 26, 1990. MRID: 92142037. Unpublished.

SPONSOR: ICI Americas Inc., Wilmington, Delaware.

EXECUTIVE SUMMARY: In a three generation reproduction study (MRID 92142092, 120271, 92142037), permethrin, PP557, (purity, 94.0-98.8%; batch numbers, P34, P35, P36, P52, P44, BX4, and BX6) was administered to groups of 12 male and 24 female Wistar rats in the diet at concentrations of 0, 500, 1000, or 2500 ppm (0, 25, 50, and 125 mg/kg/day, respectively, using a standard conversion factor of 0.05). Two litters were produced by each generation. F_0, F_1, and F_2 parental animals received test or control diet for 12 weeks post weaning and were then paired for mating to produce the A litters. After various rest periods, the F_0, F_1, and F_2 parental animals were remated to produce the B litters. Test diets were administered during mating, gestation and lactation for three successive generations throughout the study. The F_2 parents were mated for a

third time, using the same breeding pairs as for the B litters, producing the C litters for a developmental toxicity evaluation. Ten males of the F_1 generation were maintained on experimental diets until they were 54-55 weeks old and were submitted for microscopic examination of selected neurological tissues.

No animals of the parental generations died during the study, although a few were killed because of conditions not related to administration of PP557. There were no dose- or treatment-related effects on body weights, body weight gains, food consumption, or food efficiency.

Treatment-related clinical signs in high-dose parental animals were limited to whole body tremors, occurring in all parental generations (exception: tremors were not observed in the F_0 males) during the first few days of the premating period. In the 2500-ppm groups, the incidence rates for the tremors were 20/24 (F_0 females), 11/12 and 24/24 (F_1 males and females, respectively), and 12/12 and 24/24 (F_2 males and females, respectively). Tremors were also observed in pregnant and lactating females exposed to 2500 ppm PP557. There were no tremors at 0 ppm in any generation. The tremors were intermittent and transient. Neuropathy was not observed in a special microscopic examination of selected neurological tissues from F_1 males continued on test for one year.

Gross examination at necropsy did not reveal any dose- or treatment-related findings, nor did microscopic examination of grossly abnormal tissues from all parents surviving to scheduled termination and of reproductive tissues from animals suspected of infertility.

Therefore, the LOAEL for systemic toxicity is 2500 ppm (125 mg/kg/day) based on tremors observed in the F_0 females, and the F_1 and F_2 males and females. The systemic toxicity NOAEL is 1000 ppm (50 mg/kg/day).

Mating performance, fertility, and pup growth and survival were not affected by PP557 treatment in the F_1, F_2, and F_3 generations.

In the F_3C litters, there were no developmental effects associated with the administration of PP557 over three generations. The percentages of male fetuses of the 1000- and 2500-ppm groups (39.0 and 44.7%, respectively) were lower than the control value (53.2%), but the effect was not associated with increased resorptions and was not dose-related. Also, no consistent effect on sex ratios was observed in other litters or generations of the study and the effect is not considered to be treatment-related.

Therefore, the reproductive toxicity NOAEL is ≥2500 ppm (125 mg/kg/day) and the reproductive toxicity LOAEL is not identified.

Microscopic examination of F_3B weanlings revealed dose-related increases in centrilobular hypertrophy of the liver. The incidences of slight and moderate centrilobular hypertrophy were dose-related, ranging from 0 to 80% for the males and from 10 to 100% for the females. The HIARC determined that the hypertrophy of the liver is an adaptive and reversible effect and is not considered as an adverse effect. 153

The NOAEL for offspring toxicity is ≥2500 ppm (125 mg/kg/day). The offspring LOAEL is not established.

The study is classified as **Acceptable/Guideline** and satisfies the requirements for a reproduction study (OPPTS 870.3800 [§83-4a]) in rats. Deficiencies/deviations in OPP guidelines include the following: (1) lack of homogeneity and stability analyses of the test diets, (2) F_0 rats were weanlings instead of 8-10 weeks old at start of study, (3) histopathology of reproductive organs performed only on selected animals, and (4) one male per two females were mated, instead of one male per female. The author of the 1990 reformatted document acknowledged these deficiencies and provided justification for the differences. This reviewer agrees with the 1990 reviewer that the study is acceptable.

COMPLIANCE: Signed and dated GLP and Data Confidentiality statements were provided. Quality Assurance and Flagging statements were not provided. The GLP statement noted that "the study was carried out in 1977 prior to the implementation of GLP and therefore no claim is made to its GLP compliance."

I. MATERIALS AND METHODS

A. MATERIALS

1. Test material: PP557.

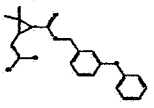
   Description: Not avaliable.

   Lot No.: Seven batches of PP557 used in the study were assigned numbers P34, P35, P36, P52, P44, BX4, and BX6.

   Purity: For the seven batches, purity ranged from 94.0-98.9%; isomeric ratios ranged from 36.2% *cis*:61.0% *trans* to 43.9% *cis*:55.0% *trans* (nominal, 40% *cis*:60% *trans*.)

   Stability of compound: The authors noted that the stability of PP557 in the diet had been verified over a period of four weeks, but did not supply data for this. Dietary analyses (conducted by the investigators) up to 63 days after preparation of the diets used in this study indicated that the compound is stable for at least two months; however, this reviewer could find no information regarding storage conditions prior to these analyses.

   CAS No.: [52645-53-1].
   Structure:



2. Vehicle and/or positive control 1564

The vehicle and negative control were prepared from a stock diet obtained from Oakes Limited, Congleton, Cheshire. The control diet was 77 parts stock diet, 18 parts malt extract, and 5 parts maize oil. No positive control was used in this study.

3. Test animals

Species: Rat
Strain: Wistar
Age and weight at start of study: approximately 4 weeks old; males: 49.8 g; females: 47.8 g
Source: Alderley Park, Cheshire.
Housing: Animals were housed in Wilmslow mobile rat racks. Cages were of 19 gauge standard galvanized wire mesh on three sides and floor with a solid back. Cages were suspended over collecting trays lined with absorbent paper. During gestation and lactation the cages housing females were fitted with solid floors and supplied with autoclaved paper bedding. For the mating period, two females were caged with one male. After confirmation of pregnancy and during lactation, the females were housed individually.
Diet: Stock diet was available *ad libitum*.
Water: Source not identified, available *ad libitum*.
Environmental conditions:
Temperature: 21-25°C
Humidity: 45-60% (with occasional fluctuations.)
Air Changes: Not given.
Photoperiod: 12 hour light/12 hour dark
Acclimation period: 3 days

B. PROCEDURES AND STUDY DESIGN

1. In life dates

Start: November 17, 1975; end: August 11, 1977

2. Mating procedure

The F_0 and F_1 parental animals produced A and B litters while the F_2 parents produced A, B, and C litters. Following a 12-week premating period, procedures were, in general, as follows: two females were placed with one male of the same treatment group, the mating periods were 14 days, pregnancy was determined mainly by the presence of sperm in vaginal smears, the day on which sperm were detected in the vaginal smear was considered to be day 0 of gestation, and sibling matings were avoided. If mating had not occurred after the given time periods, the male was exchanged for another male of proven fertility from the same group. If the second mating was unsuccessful, the female was considered to be infertile.

Variations in these procedures occurred for the various groups and are as follows: the mating period of the F_1 parents for the A litters was reduced to seven days; for all other groups, the mating period was 14 days. For F_0 females (in the production of A and B litters), vaginal smears were examined daily for evidence of mating. The day on which sperm were detected was designated day 0 of pregnancy. After a positive smear, a second smear was taken in the next expected estrous cycle. If no sperm were present, the female was presumed pregnant and smearing ceased. F_0 females were observed 2-3 times per week for abdominal enlargement and when this occurred, the male was removed.

For F_1 parental females, (in the production of the A litters) smearing continued after the presence of sperm was noted to determine if the female was pregnant, pseudo-pregnant, or continued into another estrous cycle. The male was removed when a positive smear was obtained with the female in estrous.

For the F_1 females (in the production of the B litters) and the F_2 females (in the production of the A and B litters), vaginal smearing stopped when blood was detected in the smear (at around day 15). If spotting did not occur and if there was no significant body weight gain, a second mating was arranged.

3. Study schedule

F_0, F_1, and F_2 parental animals received test or control diet for 12 weeks post weaning and were then paired for mating to produce the A litters. After various rest periods (see next paragraph), the F_0, F_1, and F_2 parental animals were remated to produce the B litters. Test diets were administered during mating, gestation and lactation for three succesive generations throughout the study. On day 21 the pups were weaned onto the same diets as their parents. The F_2 parents were mated a third time for the evaluation of developmental toxicity.

Animal sacrifices were as follows: the F_0 and F_2 parents were sacrificed at the end of their respective breeding programs; F_1 parents were maintained on their respective experimental diets until week 54-55 when the males were examined for neuropathology; the F_1A, F_2A, and F_3A pups were sacrificed for necropsy as weanlings; after selection of the F_1 and F_2 parents, the remaining F_1B and F_2B pups were sacrificed for gross necropsy; and the F_3B weanlings were sacrificed for necropsy and for microscopic examination of tissues from selected pups.

Rest periods between weaning of the A litters and remating to produce the B litters for the different generations were as follows: F_0, most remated after 10 days' rest, some rested for up to 28 days; F_1, some remated after 1-10 days' rest, others 1-2 weeks later; and F_2, 5-14 days for most, others 1-2 weeks after that. It appears that the investigators varied the rest periods either to avoid littering and weaning over Christmas or to assure that all animals of the same generation would be mated on the same day.

4. Animal assignment

Animal assignments are listed in Table 1. Future F_1 and F_2 parents were selected from as many of the F_1B and F_2B litters as possible, based on the mean group litter size ±3 at day 10 and representing the mean pup weight for each sex per litter. Where possible, only one pup per sex was selected per litter. If litters were insufficient or single sex, more than one animal of a sex was selected from the same litter (the genealogy of the parents was accurately recorded.) In addition to 12 males and 24 females selected for pairing, six extra females were incorporated into each group. At about nine weeks of age all females were examined for a congenital abnormality, imperforate vagina, and were replaced by one of the six if the condition was present.

TABLE 1. Animal assignment

Dose group	Dietary concentration of PP557 (ppm)	No. of parental animals per group[a]					
		F_0 Generation		F_1 Generation		F_2 Generation	
		Male	Female	Male	Female	Male	Female
Control	0	12	24	12	24	12	24
Low	500	12	24	12	24	12	24
Mid	1000	12	24	12	24	12	24
High	2500	12	24	12	24	12	24

[a]Six extra females were incorporated into each group to replace any female found to have an imperforate vagina.
Data taken from text table p. 15 and text pp. 15-17, MRID 92142092.

5. Dose selection rationale

The authors noted that dose levels were selected from the results of a preliminary investigation in which PP557 was fed at dietary levels of up to 10,000 ppm over a 28-day period. The results of this study were not given in the report and, except for the notation that one investigator had previously shown PP557 to have low acute and sub-acute oral toxicity, no further discussion of dose selection was found.

6. Dietary preparation and analysis

The control diet was prepared by mixing a stock ration (77 parts) with malt extract (18 parts) and maize oil (5 parts); a small amount of tap water was added to aid mixing. Dietary mixtures containing PP557 were prepared weekly during most of the study, and occasionally at two-week intervals during the latter stages. The appropriate quantity of test substance was mixed with a small portion of the maize oil, then mixed with malt and water and added to the stock ration. The amount of test article used for each dietary preparation was adjusted for the purity of that particular batch. The ingredients for each dietary concentration were mixed mechanically for ten minutes, then were extruded into pieces 1 cm in diameter and 3-5 cm in length. The pieces were dried in a vacuum oven at 30 mm mercury and at temperatures not exceeding 40°C, or in open trays in the diet storage area. The PP557 concentration in

the diet was determined at various intervals ranging from 2 to 63 days after preparation of the diets. Three different methods of analysis were employed in this study, the investigtors noted that "the various methods reflect the continuing development of the methodology to gain improvements." The early methods did not separate the *cis* and *trans* isomers.

Results –

Homogeneity analysis: not performed.

Stability analysis: The authors noted that the stability of PP557 in the diet over a period of four weeks had been verified, but these data were not provided. The data from the concentration analyses indicate that PP557 in the diet is stable for up to 63 days after preparation. Mean concentrations of duplicates of the 500, 1000, and 2500 ppm samples analyzed at 63 days were 94, 86, and 102% of nominal, respectively.

Concentration analysis: The mean results of the concentration analyses on duplicate samples ranged from 73 to 125% of nominal values. The actual concentrations of 14 of 23 samples were within 10% of the theoretical value, 6 were within 15% and 2 were 20% more or less than the theoretical value. Isomeric ratios, measured at six of these time points, ranged from 36.2% *cis*:61.0% *trans* to 43.9% *cis*:55.0% *trans* (nominal, 40% *cis*: 60% *trans*).

C. OBSERVATIONS

1. Parental animals - All animals were observed daily for clinical or behavioral abnormalities. Swabs or blood samples were submitted for bacteriological or viral screening if there was clinical evidence of suspected infection. Food consumption per cage and individual body weights were recorded weekly during the 12 weeks post weaning. The body weight of any female with an imperforate vagina was replaced with the body weight of the replacement female (because food consumption was on a per cage basis, those values could not be replaced.) Although body weights were also recorded on days 0, 7, 14, and 21 of gestation, they were only included in the tabulations if the date of mating was reliably established by vaginal smear. Dams were not weighed during lactation.

2. Litter observations - Litter observations were made as shown in Table 2. All litters were examined within twenty hours of parturition for the number of live and still-born pups and for general health and vigor of the pups, based on color, movement, size, presence of milk or dehydration, and abnormalities. Litters were examined daily for gross abnormalities and disease, and were counted, sexed, and weighed on days 0, 4, 10 and 21 post partum. Litters were not culled on lactation day 4.

TABLE 2. Litter observations				
Observation	Lactation day			
	Day 0	Day 4	Day 10	Day 21
Pup survival	Daily			
Clinical signs	Daily			
Gross abnormalities	Daily			
No. pups	x	x	x	x
Pup weight	x	x	x	x
Sex of each pup	x	x	x	x

Data taken from text p. 18, MRID 92142092.

3. Postmortem observations

a. Parental animals: During the production of the "A" litters, any female not pregnant after being paired with two males, or any male suspected of being infertile, was killed and the reproductive tract submitted for histological examination, along with any abnormal tissues. The same tissues were also taken from any female with an imperforate vagina and, in some cases, from any animal suspected of infertility during mating for the "B" litters.

At the end of their breeding program the parents of each generation were killed (with an overdose of halothane vapor) and their major organs were examined grossly. Samples of abnormal tissues were submitted for histological evaluation. Necropsy was usually performed when the parents were about six months old; however, the F_1 parents were maintained on the experimental diets until they were 54-55 weeks old, when the males were examined for neuropathology. Neural tissues examined in the one-year-old males were as follows: cerebrum, anterior spinal cord, anterior dorsal root ganglia, cerebellum, posterior spinal cord, posterior dorsal root ganglia, trigeminal ganglia, gastrocnemius muscle, sciatic nerve, lumbrical muscle, sural nerve, and posterior tibial nerve.

Rats becoming ill, undergoing a difficult parturition, or dying during the course of the study were necropsied and samples of the tissues from the various systems (listed below) were submitted for histological evaluation, along with any tissue that appeared abnormal. Tissues from this group of adults were examined microscopically and are indicated (X) in the following table.

X	DIGESTIVE	X	CARDIOVASC./HEMAT.	X	NEUROLOGIC
	Tongue		Aorta	X	Brain
	Teeth	X	Heart	X	Sciatic nerve
	Oral cavity		Bone marrow	X	Spinal cord
X	Salivary glands	X	Lymph nodes (mesenteric)	X	Pituitary
	Esophagus	X	Spleen		Eyes (optic n.)
X	Stomach	X	Thymus		
X	Duodenum				GLANDULAR
X	Jejunum		UROGENITAL	X	Adrenal gland
X	Ileum	X	Kidneys		Harderian gland
X	Cecum		Ureter		Lacrimal gland
X	Colon	X	Urinary bladder		Mammary gland
	Rectum	X	Testes	X	Thyroid
X	Liver	X	Epididymides		
X	Pancreas	X	Prostate		OTHER
			Vas deferens		Bone
	RESPIRATORY	X	Seminal vesicle	X	Voluntary muscle
X	Trachea	X	Ovaries		Skeletal muscle
X	Lung		Oviduct		Skin
	Nose/nasal cavity	X	Uterus		Tail
	Pharynx		Vagina	X	All gross lesions and masses
	Larynx				

b. Offspring: All pups found dead were preserved and decalcified. Free-hand sectioning and dissection were performed in an attempt to establish cause of death.

Ten males and ten females from each dose group of F_3B weanlings were necropsied and microscopic examinations were conducted on the same tissues listed for the adult animals in the table above.

The remaining offspring from the F_3B generation and from all other generations except F_3C were killed as weanlings and examined at necropsy internally and externally for abnormalities of major tissues, organs, and structures. Samples of most abnormal tissues were submitted for histological examination.

Selected pups from the F_2A generation were also examined histologically to investigate the unusually high mortality rates in these litters. Thirty-nine neonates dying during the first two weeks post partum were examined grossly and the following tissues underwent microscopic examination: nasal cavity, brain, heart, lungs, liver, spleen and kidney.

For a group of randomly selected F_2A weanlings (5/sex/dose) killed at 3 weeks of age, the major thoracic and abdominal viscera were examined grossly and the larynx, trachea, lungs, thymus, heart, liver, kidney, spleen, stomach and small intestine were examined microscopically.

The remainder of the F_2A weanlings, killed at 3 weeks of age were examined grossly. Only macroscopically abnormal tissues were submitted for histological examination.

4. Developmental Toxicity Study

The F_2 parents were mated for a third time, using the same breeding pairs as for the F_3B litters, producing the F_3C litters. At least 11 pregnant females in each dose group were killed by cervical dislocation on day 20, the day before expected parturition. The remaining females were discarded after necropsy.

For each pregnant female in the developmental toxicity study, the intact uterus was examined for numbers of live fetuses, early resorptions and late resorptions. Fetuses were removed and evaluated for viability, weight, sex, and abnormalities. Corpora lutea were counted and uterine resorptions were counted and classified as early or late.

In two-thirds of the fetuses the viscera were examined grossly and the carcass fixed in 70% methanol. The fetuses were stained with Alizarin Red, cleared, and examined for skeletal defects and retardation of ossification. The remaining fetuses were decalcified in Bouin's and examined for soft tissue defects by free-hand sectioning and dissection based on Wilson's technique.

D. DATA ANALYSIS

1. Statistical analyses: The data for each generation were processed by computer and analyzed by statistical tests as noted for each of the various parameters listed below using the control group as a reference point. In addition to the parameters listed below, the following were analyzed by Student's *t* test: fetal and maternal group mean body weights, food consumption, and the mean interval between a positive vaginal smear and parturition.

2. Indices: The following reproductive indices were calculated:

Male fertility index (%): (No. successful males/total no. males mated) x 100
Analyzed by Chi Squared test

Female fertility index (%): (No. of litters born/number of females mated) × 100
Analyzed by Chi Squared test.

Offspring viability indices: The following litter indices were calculated.

Viability index: No. live born/number born
This index was calculated for each litter and transformed by $\sin^{-1}\sqrt{P}$ before analysis by Student's *t* test.

Group mean litter size at parturition and days 4, 10, or 21 post partum using mean X at parturition and mean X and Y subsequently, analyzed by Student's *t* test (the authors note that "mean Y excludes the loss of whole litters often due to maternal neglect which adversely affects mean X values and therefore more emphasis was placed on the former for interpretative purposes.")

Mean X: No. of young/number of litters born
Mean Y: No. of young/number of litters at 4, 10, or 21 days

The mean of Y was used by the current reviewer to calculate postpartum survival.

Lactation index: No. of pups surviving at day 21/number of pups born alive

The index was calculated for each litter, transformed by $\sin^{-1}\sqrt{P}$, and analyzed by Student's *t* test.

3. Developmental Toxicity Study

Mean fetal body weights, the number of implantations, the number of viable fetuses and the number of resorptions were analyzed according to the following:

Analysis of variance was carried out and the group means were compared by a one-sided Student's *t* test using the pooled within-group variance.

Fetal abnormalities and differences in skeletal ossification were analyzed by a 2×2 contingency table based on litter rate.

Preimplantation loss (implantation efficiency) was analyzed as follows:
No. of implantations/no. of corpora lutea

4. Historical control data

Historical control data were not provided.

II. RESULTS

A. PARENTAL ANIMALS

1. Mortality and clinical signs

No parental animals were found dead, but several were killed during the study because of illness or for humane reasons. In the F_0 group, three females were killed because of dystocia. In the F_1 generation, two males were killed, one with hemorrhagic prostatitis and the other with respiratory disease; two F_1 females were killed because of severe weight loss and probable dystocia, respectively; and a third

was killed because of severe weight loss and the presence of large amounts of blood in the vaginal smear halfway through gestation; the cause was uncertain. In the F_2 generation, one female was killed because she had appeared pregnant, but no litter was observed; the investigators suggested that she had possibly eaten her litter. In the F_2 generation, two males were killed, one because of hemorrhage resulting from trauma to the skull and the other because of malocclusion. (The times of sacrifice were not found in the text or tables). None of these deaths were considered to be related to treatment.

Whole body tremors appeared in all parental generations at 2500 ppm. The tremors were transient and intermittant. Table 3 shows that (1) the tremors occurred during weeks 0-1 or 0-3 of the premating period, just after the beginning of treatment (for F_0 females, tremors were first observed on day 0 of premating, but the duration of the observation for the high-dose group was not legible in the report table); (2) 83.3-100% of females in all three generations and 91.7 and 100% of males in the F_1 and F_2 generations, respectively, exhibited slight or moderate tremors, whereas no tremors were observed among males of the F_0 generation. Tremors persisted in the high-dose females throughout gestation and lactation.

Tremors were also observed in two low- and mid-dose F_0 females during the first week of treatment and in one mid-dose F_1 male during week 3. No other low- or mid-dose animals were affected in any generation and the tremors observed in these animals were isolated occurrences.

Conjunctivitis was observed in male and/or female parents of all three generations and all control and treatment groups during the premating period. Ringtail was observed in the F_0 parents at 0, 500, 1000, and 2500 ppm, and in the F_2 parents at 0, 1000, and 2500 ppm, but not at 500 ppm. Neither the conjunctivitis nor the ringtail was dose-related. The incidence of conjunctivitis was low and the ringtail did not persist and was attributed to low humidity.

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TABLE 3. Incidence of whole body tremors in F_0, F_1, and F_2 parents[a] fed PP557 during the premating period

Severity	Observation	Dose group (ppm)			
		0	500	1000	2500
F_0 generation females					
Slight	Number of observations	0	0	2	4
Moderate	Number of observations	0	2	0	23
	Number of affected animals	0/24	2/24 (8.3%)	2/24 (8.3%)	20/24 (83.3%)
	Weeks observed	–	0	0	0-[b]
F_1 generation males					
Slight	Number of observations	0	0	1	12
Moderate	Number of observations	0	0	0	10
	Number of affected animals	0/12	0/12	1/12 (8.3%)	11/12 (91.7%)
	Weeks observed	–	–	3	0-3
F_1 generation females					
Slight	Number of observations	0	0	0	28
Moderate	Number of observations	0	0	0	13
	Number of affected animals	0/24	0/24	0/24	24/24 (100%)
	Weeks observed	–	–	–	0-3
F_2 generation males					
Moderate	Number of observations	0	0	0	12
	Number of affected animals	0/12	0/12	0/12	12/12 (100%)
	Weeks observed	–	–	–	0-1
F_2 generation females					
Moderate	Number of observations	0	0	0	24
	Number of affected animals	0/24	0/24	0/24	24/24 (100%)
	Weeks observed	–	–	–	0-1

[a]Tremors were not observed in males of the F_0 generation.
[b]Unreadable in report table.
Data taken from Tables 3, 4, and 5, pp.36, 37, and 38, respectively, MRID 92142092.

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2. Body weight and food consumption

a. Premating - Selected body weights for the F_0, F_1, and F_2 adults are presented in Tables 4, 5, and 6, respectively. The body weight tables of the original report contained areas of illegible data. For Tables 4, 5, and 6, those data have been omitted in some cases and/or calculated by the reviewer where possible. Generally, absolute body weights and premating weight gains (weeks 0-12) for the males and females of all generations were similar between the treated and control groups throughout the study. The following differences in body weights, compared to control values, were statistically significant: (1) in the F_1 males, weights were increased by 13.3% ($p<0.05$) at 500 ppm, week 4; (2) in the F_1 females, weights were increased by 4.2% ($p<0.01$), 1.6% ($p<0.05$), and 5.8% ($p<0.05$) at 500 ppm, weeks 2, 4, and 6, respectively, and by 5% ($p<0.05$) at 2500 ppm, week 6; (3) in the F_2 males, weights were increased by 13.6% ($p<0.05$) and 8.0% ($p<0.05$) at 1000 ppm, weeks 0, and 2, respectively. These increases in absolute body weights were sporadic, not dose-related, and were not considered to be treatment-related. The investigators did not record parental body weights beyond premating, except for females that were weighed during pregnancy.

Mean total food consumption per cage during the 12-week premating period was comparable for all groups, with a few exceptions. Values were increased in F_1 males at 500 ppm ($p<0.05$) and 2500 ppm ($p<0.01$), in F_1 females at 2500 ppm ($p<0.05$), and in F_2 females at 1000 ppm ($p<0.05$). Mean food intake per gram body weight gained (food utilization) for treated animals was comparable to control values within each sex/generation, but most utilization values were consistently higher for females (6.1-6.6 g) than for males (4.9-5.1g).

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TABLE 4. Mean body weights (g) of the F_0 adults during the premating period				
Week of Study	Treatment Group			
	0 ppm	500 ppm	1000 ppm	2500 ppm
Males				
Week 0	50.0	49.6	50.4	49.4
Week 2	136.3	134.2	132.7	130.3
Week 4	232.4	232.9	230.9	228.8
Week 6	305.2	303.0	304.3	300.9
Week 8	365.8	355.8	355.2	358.8
Week 12 (end of premating)	433.1	419.5	420.0	422.8
Premating weight gain (weeks 0-12)	383.1	369.9	369.6	373.3
Females				
Week 0	48.1	47.6	47.9	47.4
Week 2	117.0	119.0	–[b]	115.8
Week 4	165.4	169.0	164.0	170.3
Week 6	197.8	202.0	195.2	200.1
Week 8	223.0	226.1	228.8[b]	229.8
Week 12 (end of premating)	252.5	255.3	245.6[b]	254.7
Premating weight gain (weeks 0-12)	204.4	207.7[a,b]	197.7 [a,b]	207.3

[a]Calculated by reviewer.
[b]Difficult to read on report table.
Data taken from Tables 9 and 10, pp. 42 and 43, respectively, MRID 92142092.

TABLE 5. Mean body weights (g) of the F_1 adults during the premating period

Week of Study	Treatment Group			
	0 ppm	500 ppm	1000 ppm	2500 ppm
Males				
Week 0	37.3	39.8	36.0	37.5
Week 2	102.6	113.9	107.8	101.4
Week 4	191.3	216.8*	204.0	191.8
Week 6	273.2	281.4	283.6	274.3
Week 8	329.3	353.3	343.8	336.0
Week 12 (end of premating)	404.1	434.3	433.0	424.4
Premating weight gain (weeks 0-12)	366.8	394.5[a,b]	397.0	386.9[a,b]
Females				
Week 0	35.3	37.0	35.3	36.3
Week 2	98.7	102.9**	97.6*	96.3*
Week 4	148.0	150.4*	151.[b]	–[b]
Week 6	186.6	197.4*	191.5	196.0*
Week 8	219.2	221.7	219.0[b]	219.3
Week 12 (end of premating)	249.1	234.2	246.1	252.0
Premating weight gain (weeks 0-12)	213.8[a,b]	197.2[a,b]	210.8[a,b]	215.7[a,b]

[a]Calculated by reviewer.
[b]Difficult to read on report table.
Significantly different from controls, *p<0.05, **p<0.01.
Data taken from Tables 11 and 12, pp. 44 and 45, respectively, MRID 92142092 .

TABLE 6. Mean body weights (g) of the F_2 adults during the premating period

Week of Study	Treatment Group			
	0 ppm	500 ppm	1000 ppm	2500 ppm
Males				
Week 0	38.9	37.8	44.2*	38.0
Week 2	112.1	119.0	121.1*	108.9
Week 4	208.8	214.0	223.1[a]	197.5
Week 6	291.0	300.0	300.2	290.8
Week 8	–[a]	–[a]	344.1	–[a]
Week 12 (end of premating)	442.1	438.3	–[a]	419.9
Premating weight gain (weeks 0-12)	403.2	400.5	–[a]	381.9[a,b]
Females				
Week 0	37.5	36.8	41.2*	34.5
Week 2	99.3	101.1	108.9*	94.9
Week 4	195.4	190.9	141.3	–[a]
Week 6	194.2	194.9	198.1	191.1
Week 8	228.1	222.0	224.3	–[a]
Week 12 (end of premating)	261.4	254.4	255.3[a,b]	246.5[a,b]*
Premating weight gain (weeks 0-12)	223.9	217.6	214.1	212.0

[a] Difficult to read on report table.
[b] Calculated by reviewer.
Significantly different from controls, *p<0.05.
Data taken from Tables 13 and 14, pp. 46 and 47, MRID 92142092.

b. Gestation and lactation - Statistically significant decreases in body weight gains were observed during days 0-21 of pregnancy in the following: F_1 females with B litters at 500 ppm ($p<0.01$) and 1000 ppm ($p<0.01$); and F_2 females with A and B litters at 500 ppm ($p<0.01$) and 1000 ppm ($p<0.05$ or 0.01). These values were sporadic and not dose-related and values for all other groups were comparable to those of controls.

3. Test substance intake

Test substance intake was not calculated by the study author. Using a food factor of 0.05 for the adult rat, doses for the 500, 1000, and 2500 ppm groups were estimated by the reviewer to be 25, 50, and 125 mg/kg/day, respectively.

4. Reproductive function

The investigators did not evaluate estrous cyclicity, sperm characteristics, or sexual maturation.

5. Reproductive performance

The reproductive performances of the F_0, F_1, and F_2 animals are summarized in Tables 7, 8, and 9, respectively. Gestation indices and mean gestation lengths were similar between the treated and control groups of all three generations.

Male and female fertility indices for control and treated groups of most generations were ≥90%, with the following exceptions: the female fertility indices for the F_1 generation during production of the A litters were 83.3, 87.5, and 87.5% at 0, 500, and 1000 ppm, respectively; however, the female fertility index at 2500 ppm was 95.8%.

Male and female mating indices were comparable between controls and treated groups in most cases, with the following exceptions: for the F_0 generation (B litters), male mating indices for the 500- and 1000-ppm groups were 17% lower than control values, but were within 10% of controls at 2500 ppm; for the F_1 generation, the male and female mating indices ranged from 83.3 to 100%, but were higher than, or comparable to, control values. Note that the text and tables of this study do not state the total number of males mated for the B litters of each generation. Therefore, the total number of males mated was estimated in each instance, based on the male fertility indices, the reproductive data for individual animals, and information from pathology records regarding animals that failed to breed. The estimated number was used to calculate the male mating indices. Ovarian follicles, corpora lutea, and implantation sites were evaluated as part of the developmental toxicity study.

TABLE 7. Reproductive performance of F_0 rats fed PP557

Observation	0 ppm	500 ppm	1000 ppm	2500 ppm
A litters				
Number males/females paired	12/24	12/24	12/24	12/24
Number females with evidence of mating (positive vaginal smear)	22	21	23	22
Number females gravid (%)	24 (100.0)	22 (91.7)	24 (100.0)	24 (100.0)
Number of males siring a litter	12	12	12	12
Mean gestation length (days)	22.1	21.8	22.1	21.9
Gestation index (%)[a]	100.0	100.0	100.0	100.0
Male mating index (%)[a]	91.7	100.0	100.0	100.0
Female mating index (%)[a]	91.7	87.5	95.8	91.7
Male fertility index (%)	100.0	100.0	100.0	100.0
Female fertility index (%)	100.0	91.6	100.0[b]	100.0
B litters				
Number males/females paired	12/24	12/22	12/23	12/24
Number females with evidence of mating	21	17	18	21
Number females gravid (%)	23 (95.8)	21 (95.4)	23 (100.0)	24 (100.0)
Number of males siring a litter	12	11	11	12
Mean gestation length (days)	22.2	22.0	22.1	22.0
Gestation index (%)[a]	95.6	95.4	100.0	100.0
Male mating index (%)[a,c]	100.0	83.3	83.3	91.7
Female mating index (%)[a]	87.5	77.3	78.3	87.5
Male fertility index (%)	100.0	91.7	91.7	100.0
Female fertility index (%)	95.8	95.4	100.0[b]	100.0

[a]Calculated by reviewer:
female mating index = (no. females with positive vaginal smears/no. females mated) x 100
male mating index = (no. males with females showing positive vaginal smears/no. males mated) x 100
gestation index = (no. females with live born/no. females pregnant) x 100
[b]Includes one female whose pups were removed by Caesarean section.
[c]Based on at least one female having a positive vaginal smear or being pregnant.
Data taken from Tables 22 and 23, pp. 55 and 57, respectively, and from Appendix E, pp. 332-339, MRID92142092.

TABLE 8. Reproductive performance of F_1 rats fed PP557

Observation	0 ppm	500 ppm	1000 ppm	2500 ppm
A litters				
Number males/females paired	12/24	12/24	12/24	12/24
Number females with evidence of mating (positive vaginal smear)	20	20	22	22
Number females gravid (%)	20 (83.3)	21 (87.5)	21 (87.5)	23 (95.8)
Number of males siring a litter	11	11	11	11
Mean gestation length (days)	21.9	21.9	22.0	22.0
Gestation index (%)[a]	100.0	100.0	100.0	100.0
Male mating index (%)[a]	83.3	91.7	83.3	83.3
Female mating index (%)[a]	83.3	83.3	91.7	91.7
Male fertility index (%)	91.6	91.6	91.6	91.6
Female fertility index (%)	83.3	87.5	87.5	95.8
B litters				
Number males/females paired	10/20	10/21	11/21	11/23
Number females with evidence of mating	20	19	19	23
Number females gravid (%)	20 (100)	19 (90.5)	21[b] (100)	22[c] (95.6)
Number of males siring a litter	9	9	10	10
Mean gestation length (days)	21.9	22.1	21.9	21.7
Gestation index (%)[a]	95.0	100.0	95.2[b]	100.0
Male mating index (%)[a,d]	90.0	90.0	90.9	90.9
Female mating index (%)[a]	100.0	90.5	90.5	100.0
Male fertility index (%)	90.0	90.0[e]	90.9[e]	100.0
Female fertility index (%)	95.0	90.5	100.0	95.6

[a]Calculated by reviewer:
female mating index = (no. females with positive vaginal smears/no. females mated) x 100
male mating index = (no. males with females showing positive vaginal smears/no. males mated) x 100
gestation index = (no. females with live born/no. females pregnant) x 100
[b]Includes one female whose litter was thought to have been resorbed.
[c]Includes one female whose pups were thought to have been cannibalized.
[d]Based on at least one female having a positive vaginal smear or being pregnant.
[e]Unreadable in report table, calculated by reviewer.
Data taken from Tables 24 and 25, pp. 59 and 61, respectively, Appendix E, pp. 340-347, MRID 92142092.

TABLE 9. Reproductive performance of F_2 rats fed PP557

Observation	0 ppm	500 ppm	1000 ppm	2500 ppm
A litters				
Number males/females paired	12/24	12/24	12/24	11/24
Number females with evidence of mating (positive vaginal smear)	20	22	22	24
Number females gravid (%)	23 (95.8)	22 (91.7)	24 (100)	23 (95.8)
Number of males siring a litter	11	12	11	11
Mean gestation length (days)	21.9	22.0	22.1	22.1
Gestation index (%)[a]	100.0	100.0	100.0	100.0
Male mating index (%)[a]	91.7	100.0	91.7	100.0
Female mating index (%)[a]	83.3	91.7	91.7	100.0
Male fertility index (%)	91.7	100.0	91.7	100.0
Female fertility index (%)	95.8	91.4	100.0	95.8[b]
B litters				
Number males/females paired	11/23	11/22	12/24	10/23
Number females with evidence of mating	22	20	23	23
Number females gravid (%)	21 (91.3)	22 (100)	23 (95.8)	23 (100)
Number of males siring a litter	10	11	11	10
Mean gestation length (days)	21.9	22.0	22.0	21.9
Gestation index (%)[a]	100.0	100.0	100.0	100.0
Male mating index (%)[a,c]	90.9	100.0	100.0	100.0
Female mating index (%)[a]	95.6	90.9	95.8	100.0
Male fertility index (%)	90.9	100.0	100.0	100.0
Female fertility index (%)	91.3	100.0	95.8	100.0

[a]Calculated by reviewer:
female mating index = (no. females with positive vaginal smears/no. females mated) x 100
male mating index = (no. males with females showing positive vaginal smears/no. males mated) x 100
gestation index = (no. females with live born/no. females pregnant) x 100
[b]Includes one female whose pups were thought to be cannibalized.
[c]Based on at least one female having a positive vaginal smear or being pregnant.
Data taken from Tables 26 and 27, pp. 63 and 65, respectively, and Appendix E, pp. 348-355, MRID 92142092.

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6. Parental postmortem results

a. Organ weights - Organ weights were not recorded for the parental generations of this study.

b. Pathology

1) Macroscopic pathology - Among the F_0, F_1, and F_2 parents surviving to scheduled termination, gross examination of major tissues revealed no excessive or dose-related pathology. Males and females of all three generations exhibited conjunctivitis and the F_0 generation had ringtail, but neither finding was dose-related or considered treatment-related.

2) Microscopic pathology - Only grossly abnormal tissues were examined microscopically. Examination of tissues from F_0, F_1, and F_2 parents surviving to scheduled termination revealed no excessive or dose-related pathology.

Reproductive tissues from females that were not pregnant with the "A" litter after mating with two males and tissues from some females producing the "A", but not the "B" litter, were examined grossly and some were submitted for histological examination, as were reproductive tissues of males suspected of infertility. The total numbers in each group that were examined grossly were as follows: F_0 - 4 females, 2 males; F_1 - 12 females, 8 males; and F_2 - 6 females, 3 males. Other than three "old hemorrhages" in two of four F_0 females and in one of six F_2 females, microscopic abnormalities were not observed in any reproductive tissues from the six males and twenty females examined.

Males of the F_1 parental generation were maintained on experimental diets until they were 54-55 weeks old, then sacrificed for neuropathological examination. Selected neurological tissues from ten control males and ten 2500-ppm males were examined. Single multiple degenerating nerve fibers were observed in both the controls and the 2500-ppm group. These abnormalities were present only in the anterior dorsal root ganglia (0/10 controls, 1/10 treated), trigeminal galglia (2/10 controls, 3/10 treated), sciatic nerve (5/10 controls, 8/10 treated), sural nerve (3/10 controls, 4/10 treated), and posterior tibial nerve (10/10 controls, 9/10 treated). These lesions were present in controls and treated animals and are not considered to be treatment-related. No microscopic abnormalities were observed in the cerebrum, anterior spinal cord, posterior spinal cord, gastrocnemius muscle, or lumbrical muscle of the controls or the PP557-treated group.

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B. OFFSPRING

1. Viability and clinical signs

Viability data for the F_1, F_2, and F_3 litters during lactation are given in Tables 10, 11, and 12, respectively. The number of viable litters on day 0, mean live litter size on day 0, survival at day 0, post-natal survival, and the viability and lactation indices were comparable between treated and control animals for three generations, two litters each, with a few exceptions: (1) the viability index of the F_1A pups in the 1000-ppm group was 6.0% lower than the control value ($p<0.05$), but the effect was not dose-related; the investigators noted that the viability index for the F_1A controls was unusually high; (2) the mean live litter sizes for F_2A pups were increased by 20.9% ($p<0.05$) at 1000 ppm, and by 18.7% (not statistically significant) at 2500 ppm; (3) survival among the F_2A pups during days 4-21 was decreased by 19.9%, 10.3%. and 18.9% at 500, 1000, and 2500 ppm, respectively, and this is reflected in the lactation indices which were reduced by about 28% (part of the value unreadable in report), 4.6%, and 11.4%, respectively; and (4) the viability indices for the F_2B pups were different from the controls at 1000 ppm and 2500 ppm ($p<0.05$ for both), but the differences were not dose-related and do not appear to be treatment-related. These deviations from control values do not appear to be treatment-related.

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TABLE 10. Viability of F_1A and F_1B pups during lactation

Observation	0 ppm	500 ppm	1000 ppm	2500 ppm
A litters				
Number of viable litters	24	22	24	24
Mean live litter size on day 0	10.0	11.3	11.5	10.9
Survival at day 0 (% per litter)	99.2	98.4	96.2	96.7
Viability index	0.993	0.984[a]	0.933*	0.965[a]
Survival days 0-4 (%)[b]	93.8	94.8	95.3	92.7
Survival days 4-21 (%)[c]	100.0	98.7	98.9	93.4
Lactation index	0.879	0.93[a]	0.917	0.878[a]
Whole litter loss days 0-4 (n)	2	0	1[d]	1
Sex ratio at day 0 (% male)	54.2	49.8	48.3[a]	45.2[a]
B litters				
Number of viable litters	22	21	23[e]	24
Mean live litter size on day 0	10.3	10.4	11.6	11.0
Survival at day 0 (% per litter)	94.0	91.2	96.4	97.2
Viability index	0.914	0.908[a]	0.9[a]	0.9[a]
Survival days 0-4 (%)[b]	97.0	94.8[f]	93.3	98.9[f]
Survival days 4-21 (%)[c]	98.7	96.3	97.2	100.0
Lactation index	0.943	[a]	[a]	0.919[a]
Whole litter loss days 0-4 (n)	1	1[d]	1[d]	1
Sex ratio at day 0 (% male)	55.5	51.2	50.0[a]	[a]

[a] Difficult to read in report table.
[b] Calculated by reviewer: (no. pups alive at day 4/no. pups alive at day 0) x 100
[c] Calculated by reviewer: (no. pups alive at day 21/no. pups alive at day 4) x 100
[d] Difficult parturition. Dam autopsied, pups removed.
[e] Includes one female whose pups were removed by Caesarean section.
[f] Not all offspring detected at day 0.
Data taken from Tables 22 and 23, pp. 55-58, and Appendix E, pp. 332-339, MRID 92142092.

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TABLE 11. Viability of F_2A and F_2B pups during lactation

Observation	0 ppm	500 ppm	1000 ppm	2500 ppm
A litters				
Number of viable litters	20	21	21	23
Mean live litter size on day 0	9.1	10.2	11.0*	10.8
Survival at day 0 (% per litter)	94.3	94.4	98.2	97.3
Viability index	0.946	0.943	0.908[a]	0.973
Survival days 0-4 (%)[b]	91.3	86.1	94.4	92.8
Survival days 4-21 (%)[c]	95.2	76.3	85.4	77.2
Lactation index	0.840[a]	0.6__[a]	0.801[a]	0.744
Whole litter loss days 0-4 (n)	2	0	0	0
Sex ratio at day 0 (% male)	54.7	44.5	44.5	49.2
B litters				
Number of viable litters	19	19	20	22
Mean live litter size on day 0	10.8	11.0	9.8	11.6
Survival at day 0 (% per litter)	90.7	98.6	100.0	99.6
Viability index	0.930[a]	0.9_6[a]	1.000[d]*	0.9__[a]*
Survival days 0-4 (%)[b]	94.7	95.2	98.0	97.3
Survival days 4-21 (%)[c]	94.9	97.0	99.5	96.4
Lactation index	0.8_6[a]	0.900	___[a]	0.94_[a]
Whole litter loss days 0-4 (n)	2[e]	0	1[f]	0
Sex ratio at day 0 (% male)	40.6	54.6	52.6	47.7

[a]Difficult to read in report table.
[b]Calculated by reviewer: (no. pups alive at day 4/no. pups alive at day 0) x 100
[c]Calculated by reviewer: (no. pups alive at day 21/no. pups alive at day 4) x 100
[d]Includes one female whose litter was thought to have been resorbed.
[e]One animal had difficult parturition. Dam autopsied, fetuses removed.
[f]Thought to have been cannibalized.
*Statistically significant at $p<0.05$.
Data taken from Tables 24 and 25, pp. 59-62, and Appendix E, pp. 340-347, MRID 92142092.

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TABLE 12. Viability of F_3A and F_3B pups during lactation

Observation/study time	0 ppm	500 ppm	1000 ppm	2500 ppm
A litters				
Number of viable litters	23	22	24	23[d]
Mean live litter size on day 0	11.1	10.8	10.8	11.4
Survival at day 0 (% per litter)	96.6	99.2	94.6	97.8
Viability index	0.9[a]	0.932[a]	0.9[a]	0.9[a]
Survival days 0-4 (%)[b]	86.3	90.4	96.6	91.4
Survival days 4-21 (%)[c]	93.2	98.6	98.8	98.8
Lactation index	0.763	–[a]	0.947**	–[a]
Whole litter loss days 0-4 (n)	3	0	0	1
Sex ratio at day 0 (% male)	53.4	50.2	48.6	51.2
B litters				
Number of viable litters	21	22	23	23
Mean live litter size on day 0	12.7	11.9	11.4	11.6
Survival at day 0 (% per litter)	99.6	97.0	94.6	96.1
Viability index	0.936	0.9[a]	0.9[a]	–[a]
Survival days 0-4 (%)[b]	94.0	98.8	98.8	95.5
Survival days 4-21 (%)[c]	98.4	98.1	99.6	100.0
Lactation index	[a]	[a]	–[a]	–[a]
Whole litter loss days 0-4 (n)	0	0	1	1
Sex ratio at day 0 (% male)	50.4	52.0	50.4	51.0

[a]Difficult to read in report table.
[b]Calculated by reviewer: (no. pups alive at day 4/no. pups alive at day 0) x 100
[c]Calculated by reviewer: (no. pups alive at day 21/no. pups alive at day 4) x 100
[d]Includes one female whose litter was thought to have been cannibalized.
**Statistically significant at $p<0.01$.
Data taken from Tables 26 and 27, pp. 63-66, and Appendix E, pp. 348-355, MRID 92142092.

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Increased mortality was observed in the F_2A pups. Animals that appeared healthy, as well as those described as "wasting" (characterized by loss of vigor, empty alimentary tracts and thin appearance) were among those that died. The investigators performed additional studies to determine the specific cause of the increased mortalities. These studies suggested that respiratory infection (see section B.4.b. Offspring/Postmortem Results/Pathology) and/or external noise and vibration from building work adjacent to the animal room may have contributed to the deaths. The batch of test diet, tremors in the F_1 female parents, and viral infection were ruled out as possible causes of the deaths.

Tremors were seen occasionally in 3- to 5-week-old F_2A pups of the 2500 ppm group awaiting necropsy and ringtail was seen on pups of the F_1A and F_1B litters. All other pups appeared healthy throughout lactation.

2. Body weight

Body weights of the F_1, F_2, and F_3 pups during lactation are given in Tables 13, 14, and 15, respectively. Statistically significant changes, compared to control values, were observed only in weights of the F_1A males, of the F_2A and F_2B males and females, and of the F_3A males.

In the F_1A males, the mean day-4 body weight was increased by 11.5% at 2500 ppm ($p<0.05$). In the F_2A males, day-4 body weights were decreased by 17.0% at 500 ppm ($p<0.01$), 12.8% at 1000 ($p<0.01$), and 11.7% at 2500 ppm ($p<0.05$), and day-10 body weights were reduced by 21.2% at 500 ppm (not statistically significant), 20.1% at 1000 ($p<0.05$) and assumed to be decreased at 2500 ppm ($p<0.05$) (numbers in the report were illegible and the percentage reduction at 2500 ppm could not be calculated). In the F_2A females, day-4 body weights were decreased by 12.6% at 500 ppm, by 10.3% at 1000 ppm ($p<0.05$ for both), and by 9.2% at 2500 ppm (not statistically significant). Day-10 body weights of the F_2A females were reduced by 18.0 and 22.1% at 1000 and 2500 ppm, respectively, but these reductions were not statistically significant. In the F_2B pups, statistically significant increases of 12.3 to 16.7% ($p<0.05$ or $p<0.01$) were observed in the body weights of male and female pups at 1000 ppm, days 4, 10, and 21, but not at any other dose. In F_3A male pups, day-4 body weights were increased by 9.9% ($p<0.05$) at 1000 ppm, but at no other dose.

TABLE 13. Mean body weights (g) of F_1A and F_1B pups during lactation					
Day of lactation		0 ppm	500 ppm	1000 ppm	2500 ppm
F_1A pups					
Day 0	Male	5.8	5.8	5.7	5.7
	Female	5.3	5.5	5.4	5._[a]
Day 4	Male	7.8	8.2	8.8	8.7*
	Female	7.6	7.7	8.3	8.1
Day 10	Male	15.6	16.5	14.9	14.0
	Female	15.3	15.8	14.1	15.4
Day 21	Male	33.2	34.0	33.8[a]	29.8[a]
	Female	32.9	33.3	31.0	32.6
F_1b Pups					
Day 0	Male	6.1	6.9	5.8	_[a]
	Female	5.7	5.5[a]	5.4	5.5
Day 4	Male	8.7	8.7	8.8	9.0[a]
	Female	8.4	8.2	8.2	
Day 10	Male	17.3	17.3	16.8[a]	17.0[a]
	Female	16.9	16.5	16.0	
Day 21	Male	38.0[a]	38.0	36.2	36.9[a]
	Female	36.7	37.1	33.9	33.7

[a]Difficult to read in report table.
Data taken from Tables 28 and 29, pp. 67 and 68, respectively, MRID 92142092.
Significantly different from control: *p<0.05.

TABLE 14. Mean body weights (g) of F_2A and F_2B pups during lactation					
Day of lactation		0 ppm	500 ppm	1000 ppm	2500 ppm
F_2A pups					
Day 0	Male	5.8	5.6	5.6	5.5
	Female	5.5	5.2	5.2	5.2
Day 4	Male	9.4	7.8**	8.2**	8.3*
	Female	8.7	7.6*	7.8*	7.9
Day 10	Male	18.4	14.5[a]	14.7[a]*	–[a]*
	Female	17.2	16.9	14.1	13.4[a]
Day 21	Male	38.5	39.[a]_	37.3[a]	37.9
	Female	38.1	38.3	–[a]	34.3[a]
F_2 B Pups					
Day 0	Male	5.9	6.0	6.8	6.0
	Female	5.6	5.7	5.6	5.7
Day 4	Male	8.3	8.8	9.7**	8.9
	Female	8.1	8.5	9.1*	8.4
Day 10	Male	16.9	18.0	19.6**	17.6
	Female	16.2	17.4	18.9[a] **	16.7
Day 21	Male	37.3	38.2[a]	42.7**	31.9[a]
	Female	36.1	37.3	40.9**	31.5

[a]Difficult to read in report table.
Data taken from Table 30 and 31, pp. 69 and 70, respectively, MRID 92142092.
Significantly different from control: *p<0.05; **p<0.01

TABLE 15. Mean body weights (g) of F_3A and F_3B pups during lactation

Day of lactation		0 ppm	500 ppm	1000 ppm	2500 ppm
F_3A pups					
Day 0	Male	5.8	5.8	5.7	5.7
	Female	5.5	5.5	5.3	5.4
Day 4	Male	8.1	8.6	8.9*	8.8[a]
	Female	8.1	8.4	8.5	8.5
Day 10	Male	16.1	16.4[a]	16.8	16.6[a]
	Female	16.1	15.7	16.8	15.9
Day 21	Male	37.4	37.8[a]	36.6[a]	36.1
	Female	37.0	37.0	35.1[a]	35.0[a]
F_3b Pups					
Day 0	Male	6.1	6.0	6.0	6.1
	Female	5.7	5.7	5.7	5.7
Day 4	Male	8.7	8.6	9.0	8.9
	Female	8.2	8.1	8.6	8.5
Day 10	Male	17.[a]	17.2	17.8	17.4
	Female	16.8	16.4	17.2	16.7
Day 21	Male	38.2	36.0	37.5	[a]
	Female	36.5	35.4	34.1	34.7

[a]Difficult to read in report table.
Data taken from Table 32 and 33, pp. 71 and 72, respectively, MRID 92142092.

3. Offspring developmental milestones

Pup developmental landmarks, such as auditory canal opening, pinna unfolding, and eye opening, were not monitored in this study.

4. Offspring postmortem results

a. Organ weights - Organ weights of offspring were not recorded in this study.

b. Pathology

1) Macroscopic pathology - Gross findings in all generations of pups that died from day 0 to day 21 postpartum were sporadic and not related to treatment. Many of the F_2A pups found dead exhibited dilatation of the trachea and/or bronchioles. Incidences of this observation at 0, 500, 1000, and 2500 ppm were 3, 5, 6, and 3 (30%, 26%, 43%, and 21%), respectively, for the males, and 1, 2, 3, and 5 (25%, 20%, 38%, and 31%), respectively, for the females. However, the effect was neither dose- nor treatment-related.

2) Microscopic pathology

To determine the cause of increased pup mortalities in the F_2A litters, the following histological examinations were performed on the following groups: (1) 39 pups that died during the first two weeks of life, selected in about equal numbers from each sex and dose group - nasal cavity, brain, heart, lungs, liver, spleen and kidney were examined. The main findings among these animals were collapse and congestion of the lungs and inflammation of the nasal cavity, especially of the olfactory mucosa. The investigators noted that it was difficult to determine if the lung congestion/collapse was an antemortem or postmortem change and that autolysis hampered histological evaluation.

(2) a group of randomly selected weanlings (5 males and 5 females per group, killed at three weeks of age) - larynx, trachea, lungs, thymus, heart, liver, kidney, spleen, stomach and small intestine were examined. Evidence of respiratory disease was observed in the tissues from two females and three males; all other tissues were normal.

(3) the remainder of the weanlings killed at three weeks of age - only grossly abnormal tissues from these animals (7 males and 1 female) were submitted for histological examination. Three rats had bronchiolitis with or without alveolitis, three had pupillary membrane of the eye, and one had a dilated renal pelvis. One rat had no significant histological abnormalities. The investigators found no evidence that PP557-induced toxicity caused the deaths and speculated that respiratory infection or environmental factors could have been responsible.

Microscopic examination of ten F_3B weanlings of the control group and ten of the 2500 ppm group revealed increases in centrilobular hypertrophy of the liver that were somewhat dose-related. Liver tissues from ten animals each of all four dose groups were then examined and the results are shown in Table 16. When combined, the incidences of slight and moderate centrilobular hypertrophy were dose-related, ranging from 0 to 80% for the males and from 10 to 100% for the females. Findings in the treated weanlings of all generations included low incidences of cystitis, pyelonephritis, and persistent pupillary membrane. None were dose-related.

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TABLE 16. Centrilobular hypertrophy in F_3B weanlings								
	Males				Females			
	Dose (ppm)				Dose (ppm)			
	0	500	1000	2500	0	500	1000	2500
Total animals examined	10	10	10	10	10	10	10	10
Slight hypertrophy	0	4	3	5	1	2	5	4
Moderate hypertrophy	0	0	3	3	0	0	0	6
Total (%)	0 (0%)	4 (40%)	6 (60%)	8 (80%)	1 (10%)	2 (20%)	5 (50%)	10 (100%)

Data taken from Table 41, p. 92, MRID 92142092.

5. Developmental toxicity study (F_3C)

The results of the developmental toxicity study are presented in Tables 17 (litter data), 18 (results of visceral examination), and 19 (significant findings in skeletal examination.) Table 17 shows that there were no statistically significant changes in number of viable fetuses and mean litter weight. At 2500-ppm, statistically significant increases in implantations per litter (10.2%, $p<0.05$) and mean number of fetuses per litter (18.6%, $p<0.05$) were observed, in comparison to control values, along with a significant decrease in mean fetal weights (6.1%, $p<0.05$). The number of viable fetuses in this group was also increased by 29.8%, but the increase was not statistically significant. Early resorptions were decreased and late resorptions were increased at 2500 ppm, but when both types of resorptions were combined, the total resorptions were similar for controls and treated animals. Table 17 shows that the mean number of corpora lutea per pregnancy and implantation efficiency were similar for all dose groups, but the percent of male fetuses varied from 39.0 to 56.8%.

External abnormalities were limited to the following: one 500-ppm fetus with gastroschisis; one 1000-ppm fetus with interscapular hemorrhage, probably resulting from mechanical injury; and one 2500-ppm fetus with a suspected ruptured spleen, but internal examination revealed no abnormality.

Table 18 shows minimal increases in the numbers of fetuses with slight pelvic dilatation, but these were neither dose-related nor statistically significant. Litter incidences were not given. Table 19 shows only skeletal variations that were increased or decreased. Statistically significant differences from controls include: increases in wide fontanale at 500 and 2500 ppm (10.8% and 8.6%, respectively, $p<0.01$ for both); increased incidences of vertebrae that were not ossified, ranging from 4.5% to 10.8% ($p<0.05$ or 0.01); and increases in short or extra ribs, ranging from 8.6% to 20.3% ($p<0.05$). None of these effects were dose-related.

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TABLE 17. Cesarean section observations				
Observation	Dose			
	0 ppm	500 ppm	1000 ppm	2500 ppm
No. Litters	11	12	11	12
Corpora Lutea/Pregnancy (mean ± s.d.)	13.1 ± 2.3	13.4 ± 1.3	12.7 ± 1.9	14.7 ± 1.9
Mean Implantations/Litter	11.8	12.8	12.8	13.0*
Implantafion efficiency[a] (mean ± s.d.)	0.90 ± 0.14	0.94 ± 0.09	1.02 ± 0.10	0.94 ± 0.08
Total Live Fetuses	124	148	136	161
Live Fetuses/Litter	11.3	12.3	12.4	13.4*
Mean Fetal Weight (g)	4.9	4.7	4.7	4.6*
Sex Ratio (% Male)	53.2	56.8	39.0	44.7
Resorptions				
Early resorptions (%)	6 (4.6)	5 (3.3)	4 (2.8)	3 (1.8)
Late resorptions (%)	0 (0.0)	0 (0.0)	1 (0.7)	2 (1.2)

[a]Implantation efficiency = no. implants/no. corpora lutea
Significantly different from control, *p<0.05
Data taken fromTable 53, pp. 106-107, MRID 92142092.

TABLE 18. Visceral anomalies in the developmental toxicity study of F_3C offspring				
Description	Dose			
	0 ppm	500 ppm	1000 ppm	2500 ppm
No. fetuses examined	64	74	69	80
No. fetuses with slight pelvic dilatation				
Left	1	1	1	0
Right	1	2	2	2
Bilateral	0	1	3	0

Data taken from Table 54, p. 108, MRID 92142092.

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TABLE 19. Skeletal findings in the developmental toxicity study of F_3C offspring [no. fetuses (%)]				
Description	Dose			
	0 ppm	500 ppm	1000 ppm	2500 ppm
No. animals examined	60	74	67	81
No. litters examined	11	12	11	12
Skull: wide fontanale	1 (1.7)	8 (10.8)**	3 (4.5)	7 (8.6)**
Cervical vertebrae: centra 1-5 (not ossified)	0 (0.0)	0 (0.0)	3 (4.5)**	1 (1.2)
centra 2-3 (not ossified)	0 (0.0)	1 (1.4)	5 (7.5)**	4 (4.9)*
Vertebral centrum: centra 1, 4, and 5 (not ossified)	1 (1.7)	8 (10.8)**	3 (4.5)	4 (4.9)
Caudal vertebrae: 3 ossified	2 (3.3)	6 (8.1)	4 (6.0)	9 (11.1)
4 ossified	13 (21.7)	24 (32.4)	16 (23.9)	33 (40.7)
Ribs: 14 bilateral – short	1 (1.7)	7(9.5)*	1 (1.5)	7 (8.6)*
Ribs: no. animals with extra ribs	5 (8.3)	15 (20.3)*	8 (11.9)	13 (16.0)*
Ossification of digits – grade 1 (good)	27 (45.0)	27 (36.5)	35 (52.2)	28 (34.6)
grade 2	30 (50.0)	33 (44.6)	29 (43.3)	53 (65.4)
grade 3	3 (5.0)	12 (16.2)**	3 (4.5)	0 (0.0)
grade 4 (poor)	0 (0)	2 (2.7)	0 (0.0)	0 (0.0)

Significantly different from controls, *p<0.05, **0.01 (based on fetal incidence).
Data taken from Table 55, pp.109-111, MRID 92142092.

III. DISCUSSION

A. INVESTIGATOR'S CONCLUSIONS

The investigators concluded that in animals fed PP557 over three generations, toxic effects were limited to the tremors observed in parents and pups (at weaning) at 1000 and 2500 ppm. The investigators considered the condition to be sufficient evidence that the dose range selected for the study was appropriate. With regard to persistent pupillary membrane, a congenital lesion of the eye that appeared in <3% of the F_1B animals and subsequent litters, mainly at 1000 and 2500 ppm, the investigators could not exclude an interaction between PP557 as a possible cause, but felt that it could be explained by differences in the selection of F_0 parents. There were no effects on reproductive performance of the rats bred over three generations when fed PP557 at levels up to and including 2500 ppm and no evidence of neonatal toxicity or teratogenicity. The authors did not identify a LOAEL or NOAEL for systemic or reproductive toxicity.

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B. REVIEWER'S DISCUSSION

1. Maternal Toxicity

No animals of the parental generations died during the study, although a few were killed because of conditions not related to administration of PP557. Clinical signs included whole body tremors, occurring in all parental generations at 2500 ppm (except the F_0 males) during the premating period. Tremors were also observed in pregnant and lactating females exposed to 2500 ppm PP557, the highest incidences occurring in the F_1 females during weeks 1-3 of lactation of the A and B litters. F_2A weanlings in the high-dose group were also affected. There were no tremors at 0 ppm. The tremors were intermittent and transient and the investigators noted that the tremors had been observed in other studies with PP557. Neuropathy was not observed in a special microscopic examination of selected neurological tissues.

Tremors were also observed at very low incidences in the low- and mid-dose F_0 females. However, the effect at these doses was not persistent across generations and only occurred one time during premating. While it is possible that tremors in these animals were due to test article administration, the isolated incidence is not considered to be adverse. In addition, tremors observed in one F_1 male during week 3 are considered incidental to treatment.

The statistically significant changes in premating body weights, body weight gains and food consumption were sporadic and not dose-related. Many of the increases or decreases were less than 10%, compared to control values, and although statistically significant, were of questionable biological significance. No gross lesions were observed.

Therefore, the LOAEL for systemic toxicity is 2500 ppm (125 mg/kg/day) based on whole body tremors observed in females of the F_0 generation and in males and females of the F_1 and F_2 generations. The systemic toxicity NOAEL is 1000 ppm (50 mg/kg/day).

2. Reproductive Toxicity

Mating performance, fertility, and pup growth and survival were not affected by treatment in any generation. The F_2A pups exhibited increased mortality, but this was neither dose-related nor repeated in subsequent litters or generations. Additional studies on these animals suggested that respiratory infection and/or external noise and vibration from building work may have contributed to the deaths and ruled out a treatment-related cause.

Variations in pup body weights corresonded to litter sizes to a large extent; i.e., pups in smaller litters tended to be larger and pups in larger litters tended to be smaller.

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In the F_3C offspring, there were no developmental effects associated with the administration of PP557 over three generations. The percentages of male fetuses were lower than controls at 1000 and 2500 ppm, but the effect was not associated with increased resorptions, and was not dose-related. Also, no consistent effect on sex ratios was observed in other litters or generations of the study.

Persistent pupillary membrane is a congenital effect and although an interaction between genetic factors and PP557 might be possible, the low incidence of the lesion (<3%) would preclude consideration of the effect in the determination of a NOAEL or LOAEL for the current report.

Therefore, the reproductive toxicity NOAEL is ≥2500 ppm (125 mg/kg/day) and the reproductive toxicity LOAEL is not identified.

3. Offspring Toxicity

Microscopic examination of F_3B weanlings revealed dose-related increases in centrilobular hypertrophy of the liver. The incidences of slight and moderate centrilobular hypertrophy were dose-related, ranging from 0 to 80% for the males and from 10 to 100% for the females. On April 18, 2002, the HIARC evaluated the toxicology database of permethrin and determined that the hypertrophy of the liver is an adaptive and reversible effect and is not considered as an adverse effect. This conclusion is supported by a 90-day rat feeding study (MRID 00054737) where the hepatocellular hypertrophy was observed at 185 mg/kg/day with a NOAEL of 92.9 mg/kg/day. In addition, similar findings might have been observed if histopathological examinations were conducted during the parental evaluation.

The NOAEL for offspring toxicity is ≥2500 ppm (125 mg/kg/day) and the offspring LOAEL is not established.

C. STUDY DEFICIENCIES

This study was conducted in 1977, prior to the implementation of GLP and makes no claim to GLP compliance. The reformatted document was revised in 1990. That author acknowledges that the study does not fully satisfy OPP guidelines and discusses its deficiencies. Only major deficiencies are noted here.

Homogeneity analyses were not conducted on the test diets: "analyses were carried out in duplicate and this would give some indication of homogeneity."

Stability of PP557 was not measured in this study: "as it had been verified previously."

F_0 rats were weanlings at the start of the study, rather than 8-10 weeks old: "a more stringent regime than OPP guidelines." 187

Histopathology of reproductive organs was done only on those animals considered to be infertile: "there was no functional evidence of any chemically-induced infertility nor any indication of an effect on reproductive organs in chronic or subchronic studies." The author further states that histopathological data from other studies overcomes this deficiency.

One male was mated with two females: "an acceptable regime according to OECD, rather than one to one as specified in the OPP guidelines."

The current reviewer finds these explanations reasonable and agrees with the 1990 reviewer that acceptance criteria are met for the study.

D. CORE CLASSIFICATION

This study is classified as **Acceptable/Guideline** and satisfies the requirements for a reproduction study (OPPTS 870.3800 [§83-4a]) in rats.

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DATA FOR ENTRY INTO ISIS

Reproductive Study - rats (870.3800)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
109701	92142092	reproductive	rats	3 generations	oral	diet	25-125	0, 25, 50, 125	50	125	neurological	Parental/ systemic
109701	92142092	reproductive	rats	3 generations	oral	diet	25-125	0, 25, 50, 125	≥125	Unidentified		Offspring
109701	92142092	reproductive	rats	3 generations	oral	diet	25-125	0, 25, 50, 125	≥125	Unidentified		Reproductive

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DATA EVALUATION RECORD

PERMETHRIN

STUDY TYPE: REPRODUCTION AND FERTILITY EFFECTS STUDY - RAT
OPPTS 870.3800 [§83-4]; OECD 416
MRID 00097443

Prepared for

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Office of Pesticide Programs
U.S. Environmental Protection Agency
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Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
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Work Assignment No. 02-04

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Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson NOV 26 2001

Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

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[PERMETHRIN/109701] Multigeneration Reproduction Study

EPA Reviewer: Yung Yang, Ph.D.
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, HED (7509C)

Signature: Yung G. Yang
Date 12/6/2001
Signature: Joycelyn Stewart
Date 1/22/2002

DATA EVALUATION RECORD

TXR# 0050649

This is an updated DER of HED Doc. No. 008163. The study classification has been changed to unacceptable/guideline due to major deficiencies.

STUDY TYPE: Multigenerations Reproduction Study - Rat; OPPTS 870.3800 [§83-4]

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin (94.5 % total isomers; cis-/trans-isomer ratio: 26.3/73.7)

SYNONYMS: 21Z73

CITATION: James, D. (1979) A multigeneration reproduction study of 21Z73 (Permethrin) in the rat. Wellcome Group Research and Development, Beckenham, UK. Laboratory study number 9858, January 26, 1979. MRID 00097443. Unpublished.

SPONSOR: Wellcome Veterinary Research Laboratories, Berkhamsted, U.K.

EXECUTIVE SUMMARY: In a 3-generation reproduction study (MRID 00097443) Permethrin (94.5 % total isomers; cis-/trans-isomer ratio: 26.3/73.7; Lot no.: C8165-106, Batch no.: ZJ) was administered to 20 Wistar (COBS) rats/sex/dose in the diet at dose levels of 0, 4.9, 28.1, or 174.0 mg/kg bw/day for males and 0, 5.3, 29.8, and 173.6 mg/kg bw/day for females. Two litters were produced by each generation. F_2 dams were sacrificed and necropsied on gestation day 20, and the F_{3B} offspring were then processed and examined according to the test facility's protocol for a developmental toxicity study.

There were no treatment-related effects on parental mortality, clinical signs, body weights, or food consumption. **Under the conditions of this study, the parental systemic NOAEL is greater than or equal to 173.6 mg/kg bw/day for females and 174 mg/kg/day for males, and the parental systemic LOAEL is unidentified.**

There were no treatment-related effects on mating performance, fertility, pup sex ratios, pup growth or pup survival. However, it must be noted that standardization (culling) of the litters was conducted on LD 0, thereby decreasing the likelihood of detecting any subtle effects on survival. Although not considered a treatment-related effect, swollen eyes with or without intraocular hemorrhage were observed in one or two treated litters from all pregnancies and were

not seen in control group litters; the significance of this finding is unknown. In the F_{3B} litters, there were no developmental effects associated with the administration of Permethrin over three generations.

Therefore, under the conditions of this study, the reproductive toxicity NOAEL is greater than or equal to 173.6 mg/kg body weight/day for females and 174 mg/kg/day for males, and the systemic toxicity LOAEL is not identified.

In addition, the NOAEL for offspring growth and survival is greater than or equal to 173.6 mg/kg bw/day for females and 174 mg/kg/day for males, and the LOAEL for offspring is not identified.

Due to the absence of any systemic and/or reproductive toxicity at the highest dose level tested, this study is classified **unacceptable (non-upgradable)/guideline** and does not satisfy the guideline requirement for a multi-generation reproductive study (OPPTS 870.3800); OECD 416 in the rat. Additionally, numerous major deficiencies were noted in the conduct of this study.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test Material:	Permethrin
Description:	Technical; further characterization not provided
Lot/Batch #:	Lot no.: C8165-106; Batch no.: ZJ
Purity:	94.5 % total isomers; cis-/trans-isomer ratio: 26.3/73.7
Compound Stability:	Stable for at least one year when stored at room temperature, 35°C., or 50 °C.
CAS No. of TGAI:	

2. **Vehicle and/or positive control:** Powdered rat diet (see below) was used as a vehicle. There was no positive control used in the study.

3. **Test animals:**

Species:	Rat
Strain:	Wistar (COBS)
Age at study initiation:	(P) 6 wks; (F_{1B}) 3 wks; (F_{2B}) 3 wks
Wt. at study initiation:	(P) Males: group means = 139.2-140.6 g; Females: group means = 122.0-125.8 g (F_1) Males: group means = 83.6-90.9 g; Females: group means = 75.3-83.5 g (F_2) Males: group means = 86.2-101.1 g, Females: group means = 86.5-100.0 g
Source:	Charles River, U.K. Ltd.
Housing:	In suspended cages (not otherwise described). Males: in groups of four except during mating. Females: in pairs during pre-mating and individually after mating.
Diet:	Powdered BP EM No. 3 expanded breeding diet (BP nutrition) *ad libitum*
Water:	Not reported

Environmental conditions:	**Temperature:**	22 ± 2 °C
	Humidity:	Not reported
	Air changes:	Not reported
	Photoperiod:	10 hours dark / 14 hours light
Acclimation period:	Not reported	

B. PROCEDURES AND STUDY DESIGN:

1. **Mating procedure:** Females in proestrous were each caged overnight with one male from the same test group. The following morning, mating was confirmed by the presence of sperm cells in the vagina, and the day evidence of mating was observed was designated gestation day (GD) 0. It was unclear from the study report whether the animals were paired for more than one estrous cycle. Sibling matings were avoided for F_1 and F_2 matings. The day of birth was considered Day 0 of lactation (LD 0). F_1, F_2, and F_3 generations were produced and two litters were produced in each generation.

2. **Study schedule:** The P parental animals were given test diets for 12 weeks before they were mated and were approximately 18 weeks of age at mating. Selection of F_1 and F_2 parental animals was made when the pups from the B litters were 21 to 35 days of age; these animals were not mated until 7 weeks after selection and were approximately 10-12 weeks of age at mating. With the exception of F_2 adults, parental animals were sacrificed after weaning of their B litters. F_2 dams were sacrificed and necropsied on gestation day 20 of the B litters, and the offspring were then processed and examined according to the test facility's protocol for a developmental toxicity study. F_2 sires were sacrificed shortly after the F_2 dams. Paired animals that failed to produce a pregnancy at the B matings were switched to the control diet and test-mated with untreated partners. Animals that still failed to produce a pregnancy were maintained on the control diet for a month and again test-mated with untreated partners. Animals that failed to produce a pregnancy during test-mating were sacrificed and necropsied.

3. **Animal assignment:** P animals were assigned to test groups in Table 1 by an unspecified method. Selection of F_1 and F_2 parental animals was done on the dates when the largest possible number of litters from the B pregnancies were 21 to 35 days postpartum. When possible, one male and one female pup were randomly selected from each litter of the appropriate age. Additional animals were then chosen from randomly selected litters in order to make up the required numbers of animals per group.

TABLE 1. Animal Assignment							
Test Group	Nominal Dose [a] (mg/kg/day)	Animals/group					
		P Males	P Females	F_1 Males	F_1 Females	F_2 Males	F_2 Females
Control	0	20	20	20	20	20	20
Low (LDT)	5	20	20	20	20	20	20
Mid (MDT)	30	20	20	20	20	20	20
High (HDT)	180	20	20	20	20	20	20

Data taken from text, pp. 3-4, MRID 00097443.
[a]Dietary concentrations were adjusted weekly using the previous week's food consumption and body weight data. Diets were administered from beginning of the study until sacrifice or inclusion in the test mating group.

4. **Dose selection rationale:** The study did not provide a rationale for dose selection.

5. **Dosage preparation and analysis:**

Each month, a standard pre-mix of the test substance in feed was prepared by an unspecified method and delivered to the test facility. For the first 10 weeks of the study, the pre-mix was a 5% w/w mixture, and for the remainder of the study the pre-mix was a 1% w/w mixture. The concentration of the test material in the pre-mix was changed due to difficulties in obtaining a homogeneous final mix. Formulations were prepared weekly at the test facility by mixing appropriate amounts of the pre-mix with additional food (powdered BP EM No. 3 expanded breeding diet, BP nutrition) using an electric, rotating oblique drum (Manesty). Diets for the initial 3 weeks of the study were mixed for 30 minutes, and diets for the remaining weeks of the study were mixed for an hour. In order to achieve the nominal daily doses of test material, the concentrations of the test substance in the diet were adjusted each week according to the previous week's mean body weight and food consumption of each group, and separate diet formulations were prepared for each sex. A computer program ("Drug-mix," DAJ) was used to calculate the appropriate dietary concentrations. The study report did not mention the storage conditions of the weekly diet formulations. Stability of the test substance in feed was not evaluated. Homogeneity of the mixtures was either not analyzed or not reported. During the study, samples of final mixtures of all dietary concentrations for both sexes were analyzed for concentration at weeks 3, 5, 7, 12, 16, 34, 56, 67, 68, 69, and 72. Samples of all monthly batches of pre-mix were also analyzed for concentration.

Results -
Homogeneity Analysis: The study report stated that there were difficulties in obtaining a homogeneous final mix from the 5% w/w premix used during the initial 10 weeks of the study. No homogeneity data were reported.

Stability Analysis: Stability analysis was not conducted.

Concentration Analysis: The actual concentrations of all monthly batches of pre-mix were within ± 5% of target. Actual concentrations of most of the analyzed samples of final diet mixes for males were within ± 15% of target, with the exception of week 3 mid- and high-dose diets, week 67 low- and mid-dose diets, week 68 low- and high-dose diets, and week 69 low-dose diet; actual concentrations of these diet mixes deviated from target by 16.7 to 135.9%. For females, the actual concentrations of 11/33 submitted samples deviated from target by 17.2 to 81.3%, and the actual concentrations of the remaining 22 samples were within ± 15% of target.

Homogeneity data were not available; therefore the mixing procedure could not be evaluated. There was considerable variance between target and actual dietary concentrations on numerous occasions. The investigators used the results of the concentration analyses to adjust their estimates of the actual dosages of test material received by the animals on study; however, the intervals between analyses were as long as 22 weeks. It is therefore possible that there was significant variance between nominal and actual dosages to the study animals.

C. OBSERVATIONS:

1. **Parental animals:** The animals were observed daily for mortality and clinical signs. Body weights were recorded weekly, and dams were also weighed on days 0, 4, 12, and 21 post-partum. Food consumption per cage was recorded daily. There was no mention of detailed examinations, estrous cyclicity, or sperm parameters, although it is assumed that vaginal smears were at least made and examined during mating. Animals that died or were sacrificed moribund were necropsied, as were animals that failed to produce a pregnancy during test-mating. F_{2B} dams were sacrificed and necropsied on GD 20, and numbers of corpora lutea, implantations, early and late resorptions, and live fetuses were recorded.

2. **Litter observations:** The following litter observations (X) were made (see Table 2). Anogenital distance was not measured, and sexual maturation was not evaluated.

TABLE 2. $F_1/F_2/F_{3A}$ Litter Observations [a]					
Observation	Time of observation (lactation day)				
	Day 0 [b]	Day 0 [c]	Day 4	Day 12	Day 21
Number of live pups	X	X	X	X	X
Pup weight	X	–	X	X	X
Number of dead or missing pups	X	X	X	X	X
Examination [d]	X	X	X	X	X
Sex of each pup (M/F) [e]	X	–	–	–	X
External abnormalities [f]	–	–	–	–	X

Data taken from text, p. 8, MRID 00097443.
[a] F_{3B} litters were evaluated according to the test facility's protocol for developmental toxicity studies.
[b] Before standardization (culling)
[c] After standardization (culling)
[d] No details were provided regarding what observations were made at each examination.
[e] Fetal sex ratio data were reported only in summary form for PND 0 and 21.
[f] External abnormalities were only reported for PND 21, although in one case the abnormality was first noted on PND 18.

On day 0 postpartum, litters were standardized to a maximum of 8 pups/litter (4/sex/litter, as nearly as possible). For litters with less than 4 pups of one sex, the opposite sex was still culled to a maximum of 4, if possible. Excess pups were discarded.

There was no mention of necropsies of pups born or found dead.

3. **Postmortem observations:**

a. **Parental animals:** All surviving parental animals (except the F_2 parents) were sacrificed during the same week the parental animals of the next generation were selected. F_2 dams were sacrificed on GD 20 of the B litters, and F_2 males were sacrificed shortly thereafter. There was no mention of either gross or microscopic examination of these animals, with the exception of histopathological examination of one eye from one P generation female and *in vivo* and histopathological examination of both eyes from one F_1 generation female. Animals failing to produce a pregnancy during test-mating were submitted for necropsy; however, necropsy results for these animals were not reported.

b. **Offspring:** All F_{1A}, F_{2A}, and F_{3A} offspring were sacrificed at 21 days of age. The F_{1B} and F_{2B} offspring not selected as parental animals were also sacrificed at 21 days of age. These animals were subjected to postmortem examinations consisting of macroscopic external and internal examinations. A single eye from one F_{2A} pup was submitted for histopathological examination. Tissues were not routinely collected for microscopic examination.

4. **Developmental Toxicity Study:**

a. **Maternal evaluations:** All F_2 dams were sacrificed on GD 20 of litter B by chloroform inhalation and necropsied. The numbers of corpora lutea, implantations, early dead embryos, late dead embryos, and live fetuses were recorded.

b. **Fetal evaluations:** All fetuses from the F_{3B} litters were weighed, sexed, and examined for external abnormalities. Approximately one-third of the fetuses were examined for visceral abnormalities by open dissection, one-third were examined for visceral abnormalities using the Wilson method, and one-third were examined for skeletal abnormalities using Staples and Schnell's skeletal preparation method.

D. DATA ANALYSIS:

1. **Statistical analyses:** Parental body weights and body weight gains, and pup body weights and numbers of live and dead pups were analyzed using Bartlett's test for homogeneity of variances followed by Student's 't' test. If variances were homogeneous, pairs of groups were compared using the pooled variance estimate with total degrees of freedom, and if variances were heterogeneous, pairs of groups were compared using variance estimates for each group with adjustment of the degrees of freedom to take into account the unequal variances. Independent occurrences such as pregnancy success were analyzed using the Chi-squared test. Dependent occurrences, such as implantation/corpora lutea, were analyzed using a modified Chi-squared test that took intra-group variance into account when making inter-group comparisons. All data were analyzed without transformation. The dosed sire or dam was considered to be the basic unit of comparison.

   The reviewer considers the analyses used to be appropriate. It must be noted that in most cases, the data were reported as group means and standard errors. The term "standard error" can either be a synonym of "standard deviation"or a separate statistical measure of variance that can be converted to standard deviation by multiplying by the square root of n. As it was unknown which was the case in the study report, the data were summarized as reported.

2. **Test substance intake:** Weekly test substance intake was calculated for each group as follows:

   Calculated Dose (mg/kg body weight/day) = (weekly food consumption) x (dietary concentration in g test substance/100 g food) x 100/(7 x mid-week body weight).

   The Calculated Doses were adjusted to account for the results of diet concentration analyses.

3. **Indices:**

Reproductive indices: The following reproductive index was calculated from breeding and parturition records of animals in the study.

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"Pregnancy success" = Number of females pregnant/number of females mated

Offspring viability indices: The following viability indices were calculated by the reviewer from lactation records of litters in the study:

Live birth index = (Number of pups born alive/total number of pups born) x 100;

Viability index = (Number of pups alive on day 4/Number of pups alive on day 0 post cull) x 100;

Lactation index = (Number of pups alive on day 21/Number of pups alive on day 4) x 100.

4. **Historical control data:** Historical control data were not provided.

II. RESULTS:

A. PARENTAL ANIMALS:

1. Mortality and clinical signs:

One control P generation female died on (presumed) GD 2 of the B litter. One control F_1 generation male died during week 2 of the premating interval for the A litter. One low-dose F_2 female died during week 2 of the premating interval for the A litter. No abnormal clinical signs prior to death were reported for these animals, and necropsies did not determine any cause of death.

One mid-dose F_1 female was sacrificed near the beginning of the premating interval for the A litter due to developing a swollen left eye during week 2. One control F_2 female was sacrificed after delivering 15 dead fetuses of the A litter and subsequently exhibiting vaginal discharge/hemorrhage and a sick appearance, with no underlying cause identified at necropsy. One high-dose F_2 female was sacrificed during lactation of the A litter due to a large abscess in the tissue of an abdominal mammary gland.

The following clinical signs were reported. Convulsions or twitching were exhibited between weeks 9 and 24 of the study by one to four animals from all treated and control P generation groups except mid-dose females; no dose-response pattern was evident. One low-dose P generation female exhibited progressive opacity of the left eye beginning in week 19 and continuing through termination. During lactation of litter A, 2 control, 2 low-dose, and 3 high-dose F_1 generation females developed mastitis of 2 or 3 days duration in most cases but lasting for 2 to 3 weeks for 2 of the high-dose females. One F_2 generation female from each of the mid- and high-dose groups exhibited eye irritation during the pre-mating interval for the A litter.

2. **Body weight and food consumption:**

Body weight and food consumption results for males throughout the study and for females during pre-mating are summarized in Tables 3, 4 and 5. There were no treatment-related effects on body weights or food consumption of males throughout the study or females during pre-mating. Statistically significant increases were noted in absolute body weights and/or body weight gains by some of the treated groups as compared with the controls, but the differences were not considered an adverse effect due to the direction of the change (increases vs. decreases) and/or lack of a dose-response pattern.

Group mean absolute body weight, body weight gain, and food consumption values for the treated pregnant or nursing dams were generally similar to those of controls. Decreased cumulative body weight gain by the high-dose F_1 generation dams during lactation of the A litter was noted (34.6 g vs. 47.5 g for controls; $p<0.05$). However, this finding was not considered treatment related because absolute body weights were not affected, the decrease was due to a non-significant decrease in body weight gain by the high-dose group during LD 0-4, which was attributable to a higher mean absolute body weight on LD 0, and a similar effect was not observed during lactation of the B litter. Other sporadic statistically significant increases or decreases in body weights or body weight gains were noted but were not considered biologically relevant due to the direction of the change (increases vs. decreases) and/or lack of a dose-response pattern.

TABLE 3. Mean Body Weight and Food Consumption of the P Generation - Pre-mating

Observations/study week	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
P Generation Males				
Mean body weight (g) / Week 0	140.3±1.8	140.6±2.5	140.4±3.1	139.2±2.2
Week 2	226.3±2.8	226.3±3.1	230.7±4.6	226.4±3.3
Week 4	308.9±4.7	308.3±2.9	318.5±6.0	306.0±4.7
Week 6	360.2±5.6	362.7±3.6	369.7±7.3	356.3±5.3
Week 8	412.3±7.2	412.0±4.3	421.5±9.0	407.3±6.9
Week 10	445.2±7.7	449.7±4.6	457.1±9.9	439.8±7.7
Week 12 (end of pre-mating)	473.1±8.5	477.8±4.9	486.2±10.9	469.6±8.3
Week 16	507.5±9.4	515.8±5.9	522.1±11.9	500.9±9.0
Week 20	518.2±9.7	534.9±6.5	539.3±11.8	523.4±10.9
Week 24 [a]	550.0±11.0	559.6±7.1	563.3±12.8	546.3±10.8
Mean weight gain (g) Weeks 0-12	332.8±8.4	337.2±5.0	345.9±9.2	330.4±7.4
Weeks 0-24 [a]	409.7±11.1	419.0±7.2	423.0±11.2	407.1±10.1
Mean food consumption [b] (g/animal/day) Weeks 1-12	190.6	192.6	196.7	193.2
Weeks 13-24	180.0	187.7	189.6	189.0
P Generation Females				
Mean body weight (g) / Week 0	123.6±2.4	122.0±2.2	125.8±2.4	123.6±3.0
Week 2	168.2±3.1	164.6±3.9	165.6±2.7	168.0±3.2
Week 4	201.3±4.4	201.9±4.5	203.3±3.3	202.9±4.0
Week 6	227.8±4.3	220.2±7.2	227.6±4.1	226.1±3.8
Week 8 [c]	247.6±5.5	244.4±6.0	245.2±4.2	245.6±4.3
Mean weight gain (g) / Weeks 1-8 [c]	124.1±4.1	122.5±4.6	119.4±4.2	122.0±3.4
Mean food consumption [b] (g/animal/day) Week 1-8 [c]	144.8	144.9	149.6	146.5

Data taken from Tables II.1, II.2, II.3, and II.4, pp. 49-50, 52, 56, and 57, respectively, MRID 00097443.
[a] P generation animals were terminated in week 27 of the study; however, week 24 is the last week for which body weight data were reported.
[b] Calculated by reviewer using weekly mean food consumption data.
[c] Week 12 was the end of pre-mating; however, body weight data for weeks 9-12 were not reported for P generation females.

TABLE 4. Mean Body Weight and Food Consumption of the F_1 Generation-Premating

Observations/study week	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
F_1 Generation Males				
Mean body weight (g) / Week 0 [a]	83.6±4.4	90.9±2.2	89.2±3.4	87.1±2.5
Week 2	167.6±6.5	174.4±3.4	171.7±4.6	171.7±4.3
Week 4	254.3±7.4	270.2±4.8	277.1±5.5 ** (109) [b]	270.4±5.3
Week 6	338.2±8.9	351.6±5.9	362.5±5.8 * (107)	356.8±5.9
Week 8 (end of pre-mating)	382.0±9.5	399.4±6.6	414.0±5.9 ** (108)	407.6±6.6 * (107)
Week 10	420.6±10.2	440.9±8.0	451.4±6.9 ** (107)	440.9±7.0
Week 12	452.5±11.4	468.7±8.9	483.2±7.5 * (107)	471.2±7.9
Week 16	488.1±12.6	506.1±9.8	533.5±8.0 ** (109)	515.4±8.7
Week 20	515.1±12.4	535.3±11.2	555.4±8.7 ** (108)	537.9±9.4
Week 24 [c]	549.6±13.7	571.4±13.6	587.4±8.8 * (107)	570.1±9.4
Mean weight gain (g) / Weeks 0-8	298.7±7.7	308.6±6.4	324.9±6.0 ** (109)	320.6±5.8 * (107)
Weeks 0-24	466.4±12.2	480.5±13.5	498.3±10.0 * (107)	483.0±8.7
Mean food consumption [d] (g/animal/day) Weeks 1-12	172.6	173.8	179.5	174.4
Weeks 13-24	200.8	205.2	213.1	208.2
F_1 Generation Females				
Mean body weight (g) / Week 0	75.3±3.7	83.5±2.4 * (111)	83.3±3.1	75.7±2.1
Week 2	140.3±4.1	148.7±3.4	153.3±3.3 * (109)	142.0±3.2
Week 4	186.6±3.7	193.8±4.4	199.5±3.7 * (107)	184.7±4.1
Week 6	221.3±3.8	226.7±4.9	232.9±4.2	215.2±4.9
Week 8	236.5±4.3	243.8±4.6	249.3±4.9	233.6±5.6
Mean weight gain (g) / Weeks 1-8	161.3±4.3	160.3±3.2	164.9±5.1	157.9±4.5
Mean food consumption [d] (g/animal/day) Week 1-8	139.4	140.1	149.8	137.1

Data taken from Tables III.1, III.2, III.3, and III.4, pp. 82-83, 85-86, 89, and 90, respectively, MRID 00097443.
[a] Week 0 for the F_1 generation was week 27 of the entire study.
[b] Numbers in parentheses are percent of control, calculated by reviewer.
[c] F_1 generation animals were terminated in week 26; however, week 24 is the last week for which body weight data were reported.
[d] Calculated by reviewer using weekly mean food consumption data.
Significantly different from control: * $p \leq 0.05$; ** $p \leq 0.01$.

TABLE 5. Mean Body Weight and Food Consumption of the F_2 Generation-Premating

Observations/study week	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
F_2 Generation Males				
Mean body weight (g) / Week 0 [a]	86.2±3.1	101.1±4.0 ** (117) [b]	90.9±3.6	100.8±4.7 ** (117)
Week 2	179.8±5.1	202.1±5.7 ** (112)	187.2±5.6	206.6±6.6 ** (115)
Week 4	273.7±7.3	303.4±6.1 ** (111)	287.4±8.3	306.0±7.6 ** (112)
Week 6	361.2±7.4	376.6±6.7	371.3±9.9	384.4±7.4 * (106)
Week 8 (end of pre-mating)	403.7±8.2	423.1±7.9	431.1±12.1 * (107)	429.9±8.3 * (106)
Week 10	439.1±9.3	460.1±9.2	471.8±13.1 * (107)	463.8±9.8
Week 12	467.0±10..1	487.4±40.8	504.4±15.0 * (108)	490.7±10.2
Week 16	504.3±10.7	526.9±12.5	549.9±17.0 * (109)	537.4±12.9
Week 20	533.3±11.9	558.8±14.6	583.4±18.5 * (109)	566.2±13.6
Mean weight gain (g) / Weeks 0-8	317.5±6.8	322.1±8.0	340.2±10.5	329.1±7.8
Weeks 0-20	446.6±10.9	456.6 [c]	492.5±17.2 * (110)	465.4±13.7
Mean food consumption [d] (g/animal/day) Weeks 1-12	168.8	174.2	176.8	179.8
Weeks 13-24	177.8	184.0	189.2	188.4
F_2 Generation Females				
Mean body weight (g) / Week 0	86.5±2.6	96.9±4.4	87.4±3.2	100.0±4.7 * (116)
Week 2	156.3±2.9	161.3±4.1	158.0±3.1	164.6±4.7
Week 4	196.3±4.1	203.2±5.1	205.0 [c]	204.6±4.0
Week 6	225.5±4.6	231.8±6.1	232.6±4.1	234.4±4.0
Week 8	251.1±5.0	255.7±7.3	256.0±5.1	253.6±4.6
Mean weight gain (g) Weeks 1-8	164.6±4.8	158.8±5.7	168.6±3.7	153.6±4.0
Mean food consumption [d] (g/animal/day) Week 1-8	137.7	139.0	138.9	139.9

Data taken from Tables IV.1, IV.2, IV.3, and IV.4, pp. 116-117, 119-120, 123, and 124, respectively, MRID 00097443.

[a] Week 0 for the F_2 generation was week 53 of the entire study.

[b] Numbers in parentheses are percent of control, calculated by reviewer.

[c] The standard error for this mean was illegible.

[d]Calculated by reviewer using weekly mean food consumption data.
Significantly different from control: * p≤0.05; ** p≤0.01.

3. **Test Substance Intake:** The doses expressed as mean daily mg test substance/kg body weight throughout the entire study are given in Table 6. These data are based on food consumption, body weight, and nominal weekly dietary concentrations both with and without adjustment for the results of diet analyses. The values that are adjusted for the results of diet analyses are considered to be representative of the test substance intake for the entire study.

TABLE 6. Mean calculated substance intake (mg/kg body weight/day)

Generation / Study Interval	Male			Female		
	LDT	MDT	HDT	LDT	MDT	HDT
P	5.1±0.43[a]	30.5±2.57	184.6±17.69	5.2±1.13	31.2±6.50	185.8±34.73
F_1	5.2±0.41	31.1±2.68	186.8±17.61	5.4±1.33	32.0±6.79	195.8±45.64
F_2	5.2±0.38	31.3±2.99	186.4±13.46	5.2±1.26	31.3±6.48	190.3±45.89
Total Study	5.1±0.41	31.0±2.72	186.1±16.39	5.3±1.23	31.5±6.50	190.4±41.55
Adjusted[b]	4.9±0.47	28.1±3.04	174.0±17.46	5.3±1.46	29.8±7.38	173.6±45.86

Data taken from Table I.5, p. 48, MRID 00097443.
[a] Data expressed as mean ± one standard deviation.
[b]The mean calculated test substance intake for the entire study was readjusted to account for chemical analysis results.

4. **Reproductive function:**

a. **Estrous cycle length and periodicity:** There were no data provided for estrous cycle length, and no results from evaluation of vaginal smears were included. One control female and one high-dose P female remained anestrous throughout the mating period for the A litter; however, both did become pregnant with a B litter. According to the study report, all F_1 and F_2 generation females had normal estrous cycles at both A and B matings.

b. **Sperm measures:** Sperm parameters were not evaluated in this study.

5. **Reproductive performance:**

There were no biologically relevant effects on reproductive performance. Results for the parental animals are summarized in Tables 7, 8, and 9. Mean precoital intervals were not reported. Reproductive indices were not calculated by the reviewer because no mating records, results of vaginal smears, or necropsy results from females failing to produce litters at both matings were provided. However, high pregnancy rates (85-100 %) were achieved for all groups of all generations.

Test mating results for the P generation were as follows: one mated control group pair failed to produce a pregnancy at both matings and were assumed to be infertile; two low-dose pairs

and one high-dose pair produced A litters but failed to produce a B litter, and all animals except the high-dose female proved fertile at test-mating.

Test-mating results for the F_1 generation were as follows: one control group pair failed to become pregnant at both matings, and one, one, two, and one pair from each of the control, low-, mid-, and high-dose groups, respectively, produced A litters but failed to become pregnant at the B mating; all 12 animals proved fertile at test-mating.

There were no test-matings conducted for the F_2 generation, as all mated pairs became pregnant at the B mating.

TABLE 7. Reproductive Performance of the P Generation [a]

Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
Litter A				
Number males/females paired	19/19 [b]	20/20	20/20	19/19 [b]
Number females with evidence of mating	19	20	20	19
Number females gravid (%)	18 (95)	20 (100)	18 (90)	19 (100)
Number of males siring a litter	18	20	18	19
Mean gestation length (days)	22.3±0.11	22.2±0.17	22.1±0.10	22.4±0.27
Number of litters	18	20	18	19
Litter B				
Number males/females paired	19/19 [c]	20/20	20/20	20/20
Number females with evidence of mating	19	20	20	20
Number females gravid (%)	18 (95)	18 (90)	20 (100)	19 (95)
Number of males siring a litter	18	18	20	19
Mean gestation length (days)	22.2±0.17	21.7±0.23	22.2±0.09	21.9±0.06
Number of litters	18	18	20	19

Data taken from Tables II.5, II.7, II.12, and II.14, pp. 61, 64, 71, and 73, respectively, MRID 00097443.
[a]Data expressed as mean ± standard error, where appropriate.
[b]One female failed to cycle and was not mated.
[c]One female died two days after mating, and pregnancy could not be confirmed; therefore, this female was excluded from analysis.

TABLE 8. Reproductive Performance of the F_1 Generation [a]

Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
Litter A				
Number males/females paired	19 [b] /20	20/20	19/19	20/20
Number females with evidence of mating	20	20	19	20
Number females gravid (%)	19 (95)	20 (100)	18 (95)	20 (100)
Number of males siring a litter	18 or 19	20	18	20
Mean gestation length (days)	22.1±0.07	22.2±0.08	22.1±0.06	22.0±0.09
Number of litters	19	20	18	20
Litter B				
Number males/females paired	19/20	20/20	19/19	20/20
Number females with evidence of mating	20	20	19	20
Number females gravid (%)	18 (90)	19 (95)	17 (90)	19 (95)
Number of males siring a litter	17 or 18	19	17	19
Mean gestation length (days)	22.2±0.10	21.9±0.07	22.1±0.08	22.2±0.11
Number of litters	18	19	17	19

Data taken from Tables III.5, III.7, III.12, and III.14, pp. 94, 97, 104, and 106, respectively, MRID 00097443.
[a]Data expressed as mean ± standard error, where appropriate.
[b]One control male died during week 2; therefore, an alternative mate from the same dose group was provided for one female.

TABLE 9. Reproductive Performance of the F_2 Generation[a]

Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
Litter A				
Number males/females paired	20/20	19/19	20/20	20/20
Number females with evidence of mating	20	19	20	20
Number females gravid (%)	20 (100)	19 (100)	17 (85)	19 (95)
Number of males siring a litter	20	19	17	19
Mean gestation length (days)	22.2±0.13	22.1±0.14	22.4 [b]	22.1±0.13
Number of litters	20	19	17	19
Litter B				
Number males/females paired	19/19	19/19	20/20	19/19
Number females with evidence of mating	19	19	20	19
Number females gravid (%)	19 (100)	19 (100)	20 (100)	19 (100)
Number of males siring a litter	19	19	20	19
Mean gestation length (days)	N/A	N/A	N/A	N/A
Number of litters	19	19	20	19

Data taken from Tables IV.5, IV.7, and IV.12, pp. 128, 131, and 138, respectively, MRID 00097443.
[a]Data expressed as mean ± standard error, where appropriate.
[b]Standard error for this mean is illegible.

6. **Parental postmortem results:** Parental animals were not routinely submitted for necropsy. Necropsy results for animals that died were inconclusive. There were no necropsy results reported for the infertile P generation animals. Eyes from one low-dose P female and one high-dose F_2 female were examined via indirect ophthalmoscopy and/or histopathologically. Both animals had changes characteristic of unilateral glaucoma, and one also had intra-ocular hemorrhage.

B. OFFSPRING:

1. Viability and clinical signs:

Mean litter size and viability (survival) results from F_1, F_2, and F_3 pups during lactation are summarized in Tables 10, 11, and 12, respectively. Two control P dams had whole litter losses of the F_{1A} pups by day 4. One mid-dose F_1 dam had whole litter loss of the F_{2B} pups by day 4. One control F_2 dam had a litter of 15 dead pups in the A litter, and it is unknown whether there were any live pups in the litter. One mid-dose F_2 dam and one high-dose F_2 dam had whole litter losses of the A litter between the LD 0 pre-cull and LD 0 post-cull counts. There were unusually high numbers of dead or missing pups from control, low-, and

mid-dose F_{1A} litters between LD 0-4 and from control and mid-dose F_{1A} litters between LD 4-21; however, the high-dose group was unaffected, and this finding was not considered treatment-related. All other calculated survival indices of all treated and control groups of all litters of all generations were similar. However, it must be noted that scheduling the standardization (culling) of litters on LD 0 drastically limits the usefulness of the viability index and somewhat limits the usefulness of the lactation index. Fetal sex ratios at birth and weaning of all litters of all treated and control groups of all generations were similar. Significant increases in the mean live litter sizes of treated F_{2A} litters and a significant decrease in the mean litter size of the low-dose F_{3A} litters were not considered treatment-related as the direction of the change was not biologically relevant (increased vs. decreased) and/or a dose-response pattern was not evident.

Anogenital distances were not measured and offspring clinical signs were not reported. One low-dose P dam delivered the B litter on GD 18, but all offspring appeared normal. During lactation of the F_{2A} pups, 2 control, 2 low-dose, and 3 high-dose F_1 generation females developed mastitis, which lasted 2-3 days in most cases but persisted for 2-3 weeks for 2 of the high-dose females. One control F_2 dam exhibited vaginal discharge and hemorrhage following parturition of 15 dead F_{3A} pups and was sacrificed.

TABLE 10. Litter size and viability parameters for F_1 generation litters [a]

Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
Litter A				
Number of viable litters	18	20	18	19
Number born alive	200	273	245	253
Number born dead	19	2	9	2
Sex Ratio (% male) -				
Day 0 pre-cull	48.9	46.9	50.8	48.0
Day 0 post-cull	48.8	49.7	51.4	50.3
Day 21	48.5	50.7	51.7	51.0
Mean live litter size -				
Day 0 pre-cull	11.1±0.94	13.7±0.55 *	13.6±0.32 *	13.3±0.43 *
Day 0 post-cull	7.2±0.33	7.8±0.16	8.0±0.00 *	7.9±0.06 *
Day 4	6.8±0.41	7.2±0.28	7.2±0.30	7.7±0.13 *
Day 12	7.1±0.39	7.0±0.33	6.6±0.35	7.6±0.14
Day 21	7.1±0.39	7.0±0.33	6.6±0.37	7.6±0.13
No. alive Day 0 post cull	129	155	144	151
No. dead or missing - Days 0-4	20	12	15	4
No. dead or missing - Days 4-21	10	3	11	2
Whole litter loss - Days 0-4	2	0	0	0
Live birth index (%) [b]	91.3	99.3	96.5	99.2
Viability index (%) [b]	84.5	92.3	89.6	97.4
Lactation index (%) [b]	90.8	97.9	91.5	98.6

TABLE 10. Litter size and viability parameters for F_1 generation litters [a]

Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
Litter B				
Number of viable litters	18	18	20	19
Number born live	230	250	275	261
Number born dead	3	1	6	4
Sex Ratio (% male) - Day 0 pre-cull	57.9	55.4	43.8	53.6
Day 0 post-cull	53.4	51.0	50.0	50.7
Day 21	54.0	51.8	49.7	50.3
Mean live litter size -				
Day 0 pre-cull	12.8±0.88	13.9±0.43	13.8±0.48	13.7±0.60
Day 0 post-cull	7.4±0.35	7.9±0.06	8.0±0.00	7.8±0.12
Day 4	7.0±0.38	7.7±0.13	7.8±0.12	7.7±0.13
Day 12	6.9±0.41	7.7±0.13	7.8±0.12	7.7±0.13
Day 21	6.9±0.41	7.7±0.13	7.8±0.12	7.7±0.13
No. alive Day 0 post cull	133	143	160	148
No. dead or missing - Days 0-4	7	4	4	1
No. dead or missing - Days 4-21	2	0	1	0
Whole litter loss - Days 0-4	0	0	0	0
Live birth index (%) [b]	98.7	99.6	97.9	98.5
Viability index (%) [b]	94.7	97.2	97.5	99.3
Lactation index (%) [b]	98.4	100.0	99.4	100.0

Data taken from Tables II.9, II.10, II.16, and II.17, pp. 67, 68-69, 75, and 76-77, respectively, MRID 00097443.
[a]Data expressed as mean ± standard error, where appropriate and/or available.
[b]Calculated by reviewer, as follows: live birth index = (# of pups born alive/total # of pups born) x 100; viability index = (# of pups alive on day 4/# of pups alive on day 0 post cull) x 100; and lactation index = (#number of pups alive on day 21/# of pups alive on day 4) x 100.
Significantly different from control: * $p \leq 0.05$.

TABLE 11. Litter size and viability parameters for F_2 generation litters [a]				
Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
Litter A				
Number of viable litters	29	20	18	20
Number born alive	239	258	249	254
Number born dead	3	0	3	0
Sex Ratio (% male) -				
Day 0 pre-cull	52.1	46.6	49.2	46.5
Day 0 post-cull	50.7	47.8	48.6	48.1
Day 21	50.3	48.1	48.6	48.4
Mean live litter size -				
Day 0 pre-cull	12.6±0.56	12.9±0.45	13.8±0.43	12.7±0.55
Day 0 post-cull	8.0±0.00	8.0±0.06	7.9±0.11	7.9±0.07
Day 4	7.8±0.08	7.9±0.07	7.9±0.11	7.9±0.08
Day 12	7.8±0.08	7.9±0.07	7.9±0.11	7.9±0.08
Day 21	7.8±0.08	7.9±0.07	7.9±0.11	7.9±0.08
No. alive Day 0 post cull	152	159	142	158
No. dead or missing - Days 0-4	3	1	0	1
No. dead or missing - Days 4-21	0	0	0	0
Whole litter loss - Days 0-4	0	0	0	0
Live birth index (%) [b]	98.8	100.0	100.0	100.0
Viability index (%) [b]	98.0	99.4	100.0	99.4
Lactation index (%) [b]	100.0	100.0	100.0	100.0
Litter B				
Number of viable litters	18	19	17	19
Number born alive	245	269	225	259
Number born dead	3	2	5	1
Sex Ratio (% male) - Day 0 pre-cull	48.2	52.4	50.4	48.5
Day 0 post-cull	50.7	50.3	50.0	51.3
Day 21	51.4	51.0	48.7	50.7
Mean live litter size -				
Day 0 pre-cull	13.6±0.41	14.2±0.49	13.2±0.80	13.6±0.50
Day 0 post-cull	7.9±0.08	7.9±0.06	7.6±0.30	7.9±0.07
Day 4	7.7±0.14	7.7±0.15	7.4±0.39	7.8±0.09
Day 12	7.7±0.14	7.7±0.15	7.2±0.48	7.8±0.10
Day 21	7.7±0.14	7.7±0.15	7.2±0.48	7.8±0.10

TABLE 11. Litter size and viability parameters for F_2 generation litters [a]

Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
No. alive Day 0 post cull	142	151	130	150
No. dead or missing - Days 0-4	4	4	11	1
No. dead or missing - Days 4-21	0	0	4	1
Whole litter loss - Days 0-4	0	0	1	0
Live birth index (%) [b]	98.8	99.3	97.8	99.6
Viability index (%) [b]	97.2	97.4	91.5	99.3
Lactation index (%) [b]	100.0	100.0	96.6	99.3

Data taken from Tables III.9, III.10, III.16, and III.17, pp. 100, 101-102, 108, and 109-110, respectively, MRID 00097443.

[a]Data expressed as mean ± standard error, where appropriate and/or available.

[b]Calculated by reviewer, as follows:live birth index = (# of pups born alive/total # of pups born) x 100; viability index = (# of pups alive on day 4/# of pups alive on day 0 post cull) x 100; and lactation index = (#number of pups alive on day 21/# of pups alive on day 4) x 100.

TABLE 12. Litter size and viability parameters for F_3 generation litters [a]				
Observation	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
Litter A				
Number of viable litters	19 [b]	19	17 [c]	19 [c]
Number born live	251	259	201	240
Number born dead	3	1	3	3
Sex Ratio (% male) - Day 0 pre-cull	50.8	48.5	52.5	45.7
Day 0 post-cull	50.0	50.7	53.8	50.0
Day 21	50.0	51.0	53.4	48.5
Mean live litter size - Day 0 pre-cull	13.2±0.42	13.6±0.57	12.6±0.97	13.3±0.57
Day 0 post-cull	8.0±0.0	8.0±0.00	7.3±0.40	7.9±0.11
Day 4	7.9±0.07	7.6±0.16	7.3±0.40	7.8±0.22
Day 12	7.9±0.07	7.6±0.16	7.3±0.40	7.7±0.24
Day 21	7.9±0.07	7.5±0.16 *	7.3±0.43	7.6±0.26
No. alive Day 0 post cull	152	152	117	142
No. Dead or missing - Days 0-4	2	8	0	2
No. Dead or missing - Days 4-21	0	1	1	4
Whole litter loss - Days 0-4	0	0	1 [c]	1 [c]
Live birth index (%) [d]	98.8	99.6	98.5	98.8
Viability index (%) [d]	98.7	94.7	100	98.6
Lactation index (%) [d]	100.0	99.3	99.1	97.1

Data taken from Text, p. 32, and Tables IV.9 and IV.10, pp. 134, and 135-136, respectively, MRID 00097443.

[a] Data expressed as mean ± standard error, where appropriate and/or available.

[b] An additional dam had a litter of 15 dead offspring and was subsequently sacrificed due to vaginal discharge, hemorrhage, and sick appearance. It is unknown whether there were any live offspring in the litter. Data for this litter were not included in Table IV.9.

[c] The study reported 17 and 19 mid- and high-dose litters on Day 0 pre cull, and 16 and 18 mid- and high-dose litters on Day 0 post cull. Data from the missing litters were not included in the numbers of live and dead births or mean numbers per litter on Day 0 pre cull. No explanation was provided.

[d] Calculated by reviewer, as follows: live birth index = (# of pups born alive/total # of pups born) x 100; viability index = (# of pups alive on day 4/# of pups alive on day 0 post cull) x 100; and lactation index = (#number of pups alive on day 21/# of pups alive on day 4) x 100.

Significantly different from control: * $p \leq 0.05$.

2. Body weight: Mean F_1, F_2, and F_3 pup body weight data are given in Tables 13, 14, and 15, respectively. The increased LD 21 mean pup weight for combined sexes of high-dose F_{3B} pups compared to controls was due to increased mean pup weight of high-dose males; however, this difference was not considered biologically relevant due to the direction of the

H2

change (increase vs. decrease). All other mean pup weights were similar to those of the respective control groups of each generation and litter.

TABLE 13. Mean Pup Weights (g) for F_1 Offspring [a]

Lactation Day	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
F_{1A} Pups				
0 [b]	6.0±0.23	5.6±0.11	5.9±0.14	5.9±0.13
4	10.9±0.95	10.3±0.45	10.4±0.45	10.3±0.46
12	27.4±1.48	25.5±1.16	24.2±1.01	25.4±0.97
21 - Combined sexes	51.7±2.55	50.2±1.47	49.3±1.50	49.3±1.24
21- Males	52.9±2.53	51.5±1.62	50.5±1.60	50.4±1.32
21 - Females	50.8±2.66	48.4±1.47	49.1±1.26	48.2±1.26
F_{1B} Pups				
0 [b]	6.1±0.14	6.1±0.11	6.1±0.15	6.0±0.07
4	10.9±0.43	11.4±0.41	11.0±0.37	11.5±0.20
12	28.3±0.88	28.2±0.74	28.0±1.01	29.0±0.44
21 - Combined sexes	55.4±1.17	53.8±1.20	55.1±1.10	54.9±0.75
21- Males	57.2±1.31	55.0±1.36	56.2±1.21	57.1±0.92
21 - Females	53.4±1.08	52.6±1.15	54.0±1.02	52.9±0.85

Data taken from Tables II.9, II.10, II.16, and II.17, pp. 67, 68-69, 75, and 76-77, respectively, MRID 00097443.
[a]Data expressed as mean ± standard error.
[b]Before standardization (culling)

TABLE 14. Mean Pup Weights (g) for F2 Offspring [a]

Lactation Day	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
	F_{2A} Pups			
0 [b]	6.1±0.12	5.9±0.08	5.9±0.08	6.0±0.09
4	11.6±0.32	11.9 [c]	11.7±0.27	11.2±0.35
12	29.6±0.83	29.5±0.50	30.1±0.63	28.9±0.82
21 - Combined sexes	52.5±1.29	54.4±0.56	55.0±1.09	51.8±1.34
21- Males	54.2±1.42	55.5±0.59	56.9±1.13	53.5±1.28
21 - Females	52.0±1.22	53.2±0.60	53.2±1.08	50.2±1.47
	F_{2B} Pups			
0 [b]	6.1±0.11	5.9±0.08	6.1±0.15	6.5±0.17
4	11.4±0.35	10.9±0.42	11.2±0.38	12.3±0.29
12	28.7±0.87	28.6±0.75	27.0±1.05	29.6±0.72
21 - Combined sexes	55.3±1.41	54.3±1.37	51.2±2.01	56.7±1.40
21- Males	56.4±1.53	55.7±1.32	53.5±2.27	58.2±1.41
21 - Females	54.2±1.39	52.7±1.60	49.4±1.88	55.4±1.43

Data taken from Tables III.9, III.10, III.16, and III.17, pp. 100, 101-102, 108, and 109-110, respectively, MRID 00097443.

a Data expressed as mean ± standard error.

b Before standardization (culling)

c The standard error for this mean was illegible in the study report.

TABLE 15. Mean Pup Weights (g) for F_3 Offspring [a]

Lactation Day	Nominal Dose Group (mg/kg body weight/day)			
	Control	5	30	180
	F_{3A} Pups			
0 [b]	6.0±0.10	5.7±0.14	6.0±0.17	6.1±0.15
4	11.0±0.29	10.2±0.35	11.3±0.42	11.0±0.31
12	26.4±0.65	25.2±0.70	28.1±0.90	28.2±0.81
21 - Combined sexes	51.4±1.21	49.8±1.14	54.8±1.40	55.1±1.33 * (107) [b]
21- Males	52.6±1.21	51.0±1.28	56.1±1.38	57.1±1.40 (109)
21 - Females	50.2±1.25	48.5±1.14	53.3±1.43	53.3±1.33 (106)

Data taken from Tables IV.9 and IV.10, pp. 134, and 135-136, respectively, MRID 00097443.

[a] Data expressed as mean ± standard error.

[b] Before standardization (culling)

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Significantly different from control: * $p \leq 0.05$.

Sexual maturation: Sexual maturation of the pups was not evaluated.

2. **Offspring postmortem results:**

a. **Organ weights:** Organ weights were not measured.

b. **Pathology**

1. **Macroscopic examination:** No treatment-related lesions were found at necropsy of the weanlings. Several eye abnormalities were noted sporadically, including opaque cornea, "cloudy pupil," and retinal hemorrhage. Swollen eyes with or without intraocular hemorrhage were observed in weanlings from all five matings (i.e. the F_{1A}, F_{1B}, F_{2A}, F_{2B}, and F_{3A} litters). Although swollen eyes were only noted in treated groups, in all cases only single pups from one or two litters were affected, and the low incidences of this finding make it unlikely to be treatment-related.

2. **Microscopic examination**: A swollen eye from one high-dose F_{3A} male pup had changes characteristic of chronic glaucoma. Abnormal tissues (including a grossly enlarged and mottled kidney and "small yellow/green disc(s) among liver lobes) from F_{2B} pups from several litters were examined; however, the results were not reported. No other tissues from weanlings were submitted for histopathology.

3. **Developmental toxicity study results:**

a. **Cesarean section:** Data collected at cesarean section are summarized in Table 16. There were no treatment-related effects on pregnancy rate, pre- and postimplantation losses, live fetuses per litter, fetal sex ratios, or mean fetal weight. There were no abortions or total litter resorptions.

b. **Fetal morphological data:** Selected fetal morphological data are given in Table 17.

1. **External examination:** A total of 266 (19), 273 (19), 280 (20), and 280 (19) fetuses (litters) from the control, low-, mid-, and high-dose groups were subjected to external examination. Abnormal external examination findings were noted in 4 (3), 2 (1), 1 (1), and 3 (2), fetuses (litters) from the same respective groups and included the following: exencephaly, skin across fontanelles distended with fluid, swollen left jaw, dropped wrist(s), hematoma along the spine, hyperflexion of right hind foot and ankle, and splayed toes of right hind foot. All were present at single or low incidences and none were considered treatment-related.

2. **Visceral examination:** Numbers of fetuses from each group subjected to visceral examination by open dissection or the Wilson method were not reported. Results of these examinations were reported only as numbers of litters containing fetuses with each abnormal visceral examination finding. Severe hydrocephaly was noted in one of the two high-dose litters that had a fetus with exenchephaly. Slight dilatation of the third and fourth ventricles of the brain was noted in one high dose litter. All abnormal visceral examination findings were present in single or low incidences and/or at similar incidences in all treated and control

groups without any dose-response pattern being evident; therefore, none were considered treatment-related.

3. **Skeletal examination:** All skeletal examination findings (normal and abnormal) were present at similar incidences in the treated and control groups, at single or low incidences, and/or without a dose response pattern, and therefore none were considered treatment-related.

TABLE 16. Cesarean Section Observations (F3B Litters) [a]

Observation	Treatment Group (mg/kg body weight/day)			
	Control	5 mg/kg/day	30 mg/kg/day	180 mg/kg/day
No. Animals Assigned	19	19	20	19
No. Animals Pregnant	19	19	20	19
Pregnancy Rate (%)	100	100	100	100
Delivered Early/Aborted	0	0	0	0
Total corpora lutea	313	313	354	313
Corpora Lutea/dam	16.5±0.47	16.5±0.56	17.7±0.49	16.5±0.55
Total implantations	276	291	301	296
Implantations/dam	14.5±0.55	15.3±0.70	15.1±0.53	15.6±0.51
Preimplantation losses (%) [b]	11.8	7.0	15.0	5.4
Total live fetuses	266	273	280	280
Live fetuses/litter	14.0±0.73	14.4±0.78	14.0±0.68	14.7±0.61
Postimplantation losses (%) [c]	3.2	6.2	7.0	5.4
Total "early deaths"	10	18	20	16
Total "late deaths"	0	0	1	0
Fetal weight (g)	3.74±0.077	3.69±0.071	3.48±0.055	3.61±0.045
Sex Ratio (% Male)	49.6	51.3	50.7	49.6
Dams with all resorptions	0	0	0	0

Data taken from Tables IV.12, IV.14, IV.15, IV.16, IV.17, and IV.18, pp. 138, 140, 141, 142-145, 146, and 147-150, respectively, MRID 00097443.

[a]Data expressed as mean ± standard error where appropriate and/or available.

[b]Calculated by reviewer as [(number of corpora lutea - number of implantations)/number of corpora lutea] x 100.

[c]Calculated by reviewer as [(number of implantations - number of live fetuses)/number of implantations] x 100.

TABLE 17. Fetal Morphological Observations [a]				
Observation	Nominal Dose (mg/kg body weight/day)			
	Control	5	30	180
External Examination				
Number of fetuses (litters) examined	266 (19)	273 (19)	280 (20)	280 (19)
Exencephaly	0 (0)	0 (0)	0 (0)	2 (2)
Skin across fontanelles distended with	0 (0)	0 (0)	0 (0)	1 (1)
Multiple severe abnormalities [b]	0 (0)	1 (1)	0 (0)	0 (0)
Dropped wrist(s)	2 (1)	0 (0)	0 (0)	0 (0)
Hyperflexion of hind foot and ankle	1 (1)	0 (0)	0 (0)	0 (0)
Visceral Examination - Open Dissection				
Number of (litters) examined	(19)	(19)	(20)	(19)
Atria distended with blood	(0)	(0)	(1)	(0)
Large spleen	(0)	(0)	(0)	(1)
Small pancreas	(0)	(0)	(0)	(1)
Large/dark adrenal	(1)	(0)	(0)	(0)
Enlarged kidney	(0)	(0)	(1)	(0)
Visceral Examination - Wilson Method				
Number of(litters) examined	(19)	(19)	(20)	(19)
Severe hydrocephaly	(0)	(0)	(0)	(1) [c]
Slight dilatation of 3rd & 4th ventricle(s)	(0)	(0)	(0)	(1)
Dilated kidney pelvis (pelves)	(3)	(2)	(3)	(2)
Multiple renal/ureter anomalies [d]	(0)	(1)	(0)	(0)
Skeletal Examination				
Number of (litters) examined	(19)	(19)	(20)	(19)
Poorly ossified parietals	(17)	(15)	(13)	(9)
Widened fontanelle	(1)	(2)	(2)	(2)
Poorly ossified cervical centra	(8)	(7)	(10)	(2)
Vestigial 14th rib(s)	(7)	(10)	(3)	(7)
Unossified sternebra(e)	(14)	(12)	(18)	(15)

Data taken from Tables IV.15, IV.16, IV.17, IV.18, IV.19, and IV.20, pp. 141, 142-145, 151, and 152-155, respectively, MRID 00097443.

[a]Data expressed as number of affected fetuses (litters). Numbers of affected fetuses were available only for external examination findings.

[b]Included cyclopia, underdevelopment of front of head, absent right ear, complete abdominal hernia, dropped left ankle, very long tail.

[c]Present in one of the two litters that had a fetus with exencephaly.

dIncluded dilated kidney pelves & calyxes, underdeveloped medulla, grossly dilated ureters.

III. DISCUSSION and CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** The study author concluded that at nominal doses up to 180 mg/kg/day there was no parental toxicity, no adverse effects on growth, survival, or reproductive ability through three generations, and no fetotoxic or teratogenic effects. The study author did not identify LOAELs or NOAELS.

B. **REVIEWER COMMENTS:**

In the absence of any treatment-related mortality, clinical signs, or effects on body weight or food consumption, the reviewer agrees that there was no parental systemic toxicity observed in this study.

Therefore, under the conditions of this study, the systemic toxicity NOAEL is greater than or equal to 173.6 mg/kg body weight/day for females and 174 mg/kg/day for males, and the systemic toxicity LOAEL is not identified.

Mating performance, fertility, pup sex ratios, and pup growth were not affected in the P, F_1, F_2, and F_{3A} generations. Pup survival was also not affected; however, it must be noted that scheduling standardization (culling) of the litters on LD 0 decreased the likelihood of detecting a subtle effect on survival. Although swollen eyes with or without intraocular hemorrhage were observed in one or two treated litters from all pregnancies and were not seen in control group litters, the low incidence of this finding makes it unlikely to be a treatment-related effect. In the F_{3B} litters, there were no developmental effects associated with the administration of Permethrin over three generations.

Therefore, under the conditions of this study, the reproductive toxicity NOAEL is greater than or equal to 173.6 mg/kg body weight/day for females and 174 mg/kg/day for males, and the systemic toxicity LOAEL is not identified.

In addition, the NOAEL for offspring growth and survival is greater than or equal to 173.6 mg/kg bw/day for females and 174 mg/kg/day for males, and the LOAEL for offspring is unidentified.

However, in the absence of any systemic and/or reproductive toxicity at the highest dose level tested, this study is classified **unacceptable (non-upgradable)/guideline.**

C. **STUDY DEFICIENCIES:** Major deficiencies noted in the conduct of this study included the following:

- The most serious deficiency is the lack of systemic and/or reproductive toxicity at the highest dose level tested.
- Litters were standardized (culled) on LD 0, instead of LD 4, thereby decreasing the likelihood of detecting any subtle treatment-related effect on survival.
- Selection of parental animals was confined to litters of a certain age range. F_1 parents were selected from 13/18, 14/18, 16/20, and 15/19 of the available litters in the control, low-, mid-,

and high-dose groups, respectively, and F2 parents were selected from 14/18, 18/19, 11/16, and 18/19 of the available litters in the same respective groups, i.e fully 19% of the available litters were not used.

- Adult parental animals were not necropsied and neither were pups that died.
- There was considerable variance between target and actual dietary concentrations on numerous occasions, and the intervals between diet analyses were as long as 22 weeks. Also, the study report stated that there was difficulty in attaining homogeneous diet mixes as late as 10 weeks into the study. It is therefore possible that there was significant variance between nominal and actual dosages to the study animals.
- Stability and homogeneity of the test diets were not analyzed, and physical properties and impurities of the test material were not reported.
- Individual data were not included in the study report.

Minor deficiencies not affecting the classification of this study include the following:

- Test and control groups did not contain a sufficient number of mating pairs to yield approximately 20 pregnant females.
- The mating procedure was inadequately described in the study report, which stated only that each mating pair was given "2 attempts" to mate. It is unknown whether pairs were caged together until either 3 estrous periods or 2 weeks had elapsed.
- It is unknown whether estrous cycle length and pattern were evaluated during a minimum of three weeks prior to mating and through cohabitation.
- Age of vaginal opening and preputial separation were not determined for weanlings selected as parental animals.
- Sperm parameters were not evaluated.

DATA EVALUATION RECORD

PERMETHRIN/109701
STUDY TYPE: METABOLISM AND PHARMACOKINETICS - DOG
[OPPTS: 870.7485 (§85-1)]
MRID 00054721 and 00042160

Prepared for

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Office of Pesticide Programs
U.S. Environmental Protection Agency
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Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

[PERMETHRIN/PC Code 109701]

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DATA EVALUATION RECORD
Supplementary, HED Doc # 001660

TXR#: 0050649

STUDY TYPE: Metabolism - [dog]; OPPTS 870.7485 [§85-1)]; OECD 417.

PC CODE: 109701 **DP BARCODE:**D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin, purity not reported.

SYNONYMS: PP557; [3-phenoxybenzyl (±) cis:trans -2,2-dimethyl-3'(2,2-dichlorovinyl)-cyclopropane-1-carboxylate]

CITATION: 1) Mills, I., Slade, M. 1977. PP557: Absorption, distribution and excretion in the dog. Report No. CTL/F/285, January 1977. ICI Americas. MRID 00054721. Unpublished.
2) Anonymous. 1981. PP557: Permethrin: tissue retention in the dog. MRID 00042160. Unpublished.

SPONSOR: ICI Americas, Inc.

EXECUTIVE SUMMARY: Two metabolism studies were conducted using adult Beagle dogs. In MRID 0054721, groups of four male and four female beagle dogs were given [^{14}C-alcohol]permethrin (PP557; no lot/batch nos.; 59.7 mCi/mM; purity not reported) or [^{14}C-acid]permethrin (PP557; no lot/batch nos.; 1.87 mCi/mM; purity 99%) as a single oral dose (6.5 mg/kg and 6.2 mg/kg, respectively) in a gelatin capsule. Excreta were collected over a 7-day period and tissues collected and analyzed at termination. In MRID 00042160, two beagle dogs (gender not specified) were given 10 daily doses (1.0 mg/kg via gelatin capsules) of [^{14}C-alcohol]permethrin (PP557; no lot/batch nos.; 59.7 mCi/mM; purity not reported). Excreta were collected after seven days and adipose tissues analyzed at termination.

These experiments provided preliminary information regarding the metabolism and disposition of permethrin in dogs. Data were insufficient for determination of definitive mass balance for administered radioactivity. Following oral administration of a single dose of [^{14}C-alcohol]permethrin (6.5 mg/kg) or [^{14}C-acid]permethrin (6.2 mg/kg), approximately 84-87% of administered radioactivity was eliminated via the feces and urine in 24-48 hours (MRID 000054721). Fecal excretion (~45-56% of dose) was somewhat greater than urinary excretion (~30-38% of dose) and the rate of excretion was slightly less for the [^{14}C-alcohol]permethrin. At seven days postdose, radioactivity was detected in the tissues selected for analysis (peri-renal and

subcutaneous fat, liver, kidney, lung, heart, blood, and brain). The highest tissue levels (0.5-0.7 µg eq./g) were found in the fat tissues. Although radioactivity was detected in all tissues seven days following the single oral dose, levels were minimal and there was no evidence for significant sequestration. Following a single oral dose, TLC analysis of organic solvent extracts revealed up to four metabolites in the urine and six in the feces, none of which were characterized. The excretory pattern for dogs given multiple doses of [^{14}C-alcohol]permethrin (1.0 mg/kg/day for 10 days) (MRID 00042160) was similar to that observed for the single dose study. The repeat-dose study also provided preliminary data showing a shift in the cis:trans ratio (an increase in the cis isomer) of residues in peri-renal and subcutaneous fat, and noted that this shift was indicative of a preferential metabolism of the trans isomer.

These metabolism/disposition studies in the dog are classified **Unacceptable/Non-Guideline** and do not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD 417] in dogs. The unacceptability is the result of deficiencies in level of detail provided which prevent verification/validation of findings (e.g., insufficient data regarding characterization of recovered radioactivity, no dose confirmation, no lot/batch numbers for the test article, mass balance data lacking in MRID 00042160). Furthermore, the studies were conducted prior to GLP Guidelines and lacked quality assurance statements.

COMPLIANCE: The studies were conducted prior to implementation of GLP guidelines and, therefore, there was no claim regarding GLP compliance. Certification of Access to Raw Data and Data Confidentiality statements were signed subsequent to completion of the studies. provided.

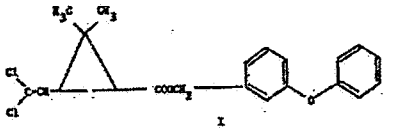
I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test compound:

Radiolabelled test material:	1) [^{14}C-alcohol]PP557 (53:47, cis:trans) 2) [^{14}C-acid]PP557 (38:62, cis:trans) 3) [^{14}C-alcohol]PP557 (42:58, cis:trans)
Radiochemical purity	1) Not provided 2) 99% 3) Not provided
Specific Activity	1) 59.7 mCi/mM 2) 1.87 mCi/mM 3) 59.7 mCi/mM
Lot/Batch #:	No lot or batch numbers were provided for any of the test articles
Non-Radiolabelled Test Material:	PP557 (40.5:59.3, cis-trans)
Description:	Not provided
Lot/Batch #:	Not provided
Purity:	93.6%
Contaminants:	None reported
CAS # of TGAI:	52645-53-1

Structure:



2. **Vehicle and/or positive control:** The test article was mixed in commercially available corn oil and administered in gelatin capsules

3. **Test animals:**

Species:	Dog (male and female)	
Strain:	Beagle, inbred	
Age/weight at study initiation:	Adults (age not specified); 10-14 kg (MRID 00045721) Adults (age not specified); 16-17 kg (MRID 00042160)	
Source:	Alderley Park, Cheshire	
Housing:	Housed individually in metabolism cages (MRID 00045721) Transferred to individual metabolism cages after 7 days of the 10-day dosing period (MRID 00042160)	
Diet:	Fed once daily, diet not specified	
Water:	*Ad libitum*	
Environmental conditions:	**Temperature:**	Not specified
	Humidity:	Not specified
	Air changes:	Not specified
	Photoperiod:	Not specified
Acclimation period:	Not specified	

4. **Preparation of dosing solutions:** In MRID 00054721, the cis and trans isomers of the stock [^{14}C-alcohol]permethrin were separated by thin-layer chromatography (TLC), extracted with methanol, recombined in a 40:60 (cis:trans) ratio and, where necessary diluted with non-labeled PP557 (40.5:59.5, cis:trans). [^{14}C-acid]permethrin, (38:62, cis:trans) was diluted with non-labeled PP557 (40.5:59.5, cis:trans). For MRID 00042160, the isomer mixture was prepared similarly resulting in the final cis:trans ratio of 42:58. The test articles and vehicle (corn oil) were administered in gelatin capsules.

B. STUDY DESIGN AND METHODS:

1. Group arrangements

The test groups for MRID 00054721 and MRID 00042160, are summarized in Table 1. For MRID 00054721, groups were established to determine the excretion following a single oral dose of either [^{14}C-alcohol]PP557 or [^{14}C-acid]PP557. No randomization procedures for establishing treatment groups were noted in the study report. For MRID 00042160, only two dogs were used.

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TABLE 1: Dosing groups for distribution/excretion studies identification studies in beagle dogs given a single oral dose of PP557 ([^{14}C-alcohol]permethrin or [^{14}C-acid]permethrin).			
Test Group	**Dose (mg/kg)**	**Number/sex**	**Remarks**
1	6.5 6.5	4♂ 4♀	Single oral dose (gelatin capsule) of [^{14}C-alcohol]permethrin (PP557); excreta collected at 24-hr intervals for 7 days; tissue analysis at termination (MRID 00054721)
2	6.2 6.2	4♂ 4♀	Single oral dose (gelatin capsule) of [^{14}C-acid]permethrin (PP557); excreta collected at 24-hr intervals for 7 days; tissue analysis at termination (MRID 00054721)
3	1	2; not specified	Repeated oral dose (gelatin capsule)of [^{14}C-alcohol]permethrin for 10 days; excreta collected after 7 days of dosing; tissue analysis at termination (24 hrs after last dose) (MRID 00042160)

Data taken from p. 4, MRID 00054721 and p. 318, MRID 00042160.

2. **Dosing and sample collection:** The test article was suspended in a corn oil vehicle and administered in a gelatin capsule for both studies.

Expired air: Expired air was not collected.

Urine: Urine was collected at 24-hour intervals for seven days (MRID 00054721). Samples were added to scintillation fluid and counted by LSC. For the multiple-exposure study in dogs (MRID 00042160), urine was collected at 48-hr intervals following Day 7 of dosing. For analysis by TLC (silica gel GF and hexane:diethyl ether solvent [10:1, v/v] system), samples were processed through a series of ether extractions and pH adjustments. Radioactive area on the gel plates were detected by Kodirex® X-ray film.

Feces: Feces were collected at 24-hour intervals for seven days (MRID 00054721). Fecal samples were air-dried and combusted to $^{14}CO_2$ which was captured in scintillation fluid and analyzed by LSC. For the multiple-exposure study in dogs (MRID 00042160), feces were collected at 48-hr intervals following Day 7 of dosing. Counting efficiencies for all LSC analyses were determined by an external standard and quench-corrected.

Blood: Blood was collected at seven days postdose (no interim collection for plasma kinetics) (MRID 00054721). Blood samples were mixed with cellulose powder, combusted to $^{14}CO_2$, and analyzed for radioactivity by LSC.

Tissues: At seven days postdose, the following tissues were collected from one male and one female dog from each group: brain, kidney, liver, fat, lung, heart, muscle, and blood (MRID 00054721). For the multiple-exposure study in dogs (MRID 00042160), peri-renal fat, subcutaneous fat, liver, kidneys, and muscles were removed at termination (24-hrs after last dose). Tissue were macerated and aliquots combusted to $^{14}CO_2$ in a sample oxidizer.

a. **Pharmacokinetic studies:** Absorption/elimination kinetics were not specific protocol elements although limited time-course excretion data were presented in MRID 00054721.

b. **Metabolite characterization studies:** Metabolite characterizations were not specific protocol elements in either report. Excreta and some tissues (peri-renal fat and subcutaneous fat in MRID 00042160) were subjected to organic solvent (ether or dimethylformamide/hexane) extractions primarily for identification of isomeric forms.

3. **Statistics:** Description of statistical analyses were limited to those applied to LSC analyses.

II. RESULTS

A. **PHARMACOKINETIC STUDIES:** Recovery of radioactivity ranged from 83.9 to 99.7% of the administered dose in MRID 00054721 (Table 2). For MRID 00042160, data were unavailable for assessing mass balance.

TABLE 2. Recovery of administered radioactivity (% of dose) in dogs following a single oral dose of [^{14}C-alcohol]PP557 or [^{14}C-acid]PP557[a]

Day	[^{14}C-alcohol]PP557 (6.5 mg/kg)		[^{14}C-acid]PP557 (6.2 mg/kg)	
	Male	Female	Male	Female
1				
urine	14.9	11.0	32.7	25.7
feces	14.5	25.5	35.8	6.7
2				
urine	9.1	9.8	5.2	13.0*
feces	35.3	19.9	8.3	15.9*
3				
urine	3.1	3.7	1.3	9.9
feces	3.6	6.5	1.9	24.2
4				
urine	1.4	2.3	0.4	0.9
feces	1.6	2.5	1.2	1.6
5				
urine	1.01	1.6	0.4	0.3
feces	1.1	0.7	0.2	0.4
6				
urine	0.6	1.2	0.3	0.3
feces	0.3	0.4	0.5	0.4
7				
urine	0.4	0.8	0.2	0.2
feces	0.3	0.3	0.3	0.2
Total[b]				
urine	**30.4**	**31.2**	**37.9**	**50.3**
feces	**55.5**	**55.7**	**45.3**	**49.4**

[a] Each value is average of two dogs except (*) which is for one dog only

[b] Totals are averages of those reported in study (MRID 00054721) except last column.

Data taken from Tables 1 and 2, pp. 10-11, MRID 00054721.

1. **Absorption:** As implied from urinary excretion data (MRID 00054721), at least 30-38% of an administered oral dose (6.2 or 6.5 mg/kg) was absorbed within 48 hours. An accurate assessment of absorption was not possible based upon the experimental protocols and resulting data. Additionally, in the absence of biliary excretion data, it is not possible to determine what portion of radioactivity recovered in the feces represents absorbed material.

2. **Tissue distribution**: A complete assessment of tissue distribution was not a protocol element in MRID 00054721 or MRID 00042160. Quantitative analysis of tissue residues in MRID 00054721 was limited to tissues noted in §B.2 of this Data Evaluation Record but the data presented in the study report were illegible. The study authors (MRID 00054721) reported that at 7 days post dose, the highest radioactivity residues (0.5-0.7 µg. eq./g) were found in fat tissue of dogs following a single oral dose of ~6 mg/kg. In the 10-day repeat dose study in dogs (MRID 00042160), radioactivity was detected in all tissues examined (subcutaneous and peri-renal fat, muscle, kidney, and liver) 24 hours after the last of 10 daily 1 mg/kg oral

doses (Table 3). The data indicate that fat tissue consistently exhibited the greatest tissue burdens. Data for the three dogs indicated similar distribution patterns with unremarkable individual variability.

TABLE 3. Distribution of radioactivity (µg eq./g tissue) in dogs at 24 hours following 10 daily oral doses of 1 mg/kg

Tissue	Dog 1	Dog 2	Dog 3
Subcutaneous fat	2.75	4.76	3.75
Peri-renal fat	4.99	6.72	5.86
Muscle	0.15	0.09	0.12
Kidney	0.94	0.84	0.89
Liver	1.31	1.06	1.19

Data taken from p. 325, MRID 00042160.

3. **Excretion:** Both feces and urine were major routes of excretion of administered radioactivity. Following a single oral dose (6.5 mg/kg) of [^{14}C-alcohol]permethrin, approximately 30-31% of the [^{14}C-alcohol]permethrin dose was excreted in the urine and 56% excreted in the feces over a seven-day period. Following a single 6.2 mg/kg oral dose of [^{14}C-acid]permethrin, urinary excretion represented 38-50% of the administered dose and fecal excretion accounted for 45-49% (see Table 2, §II. A in this Data Evaluation Record). Most excretion, regardless of route, occurred within 24-48 hours, although the rate of excretion was slightly less for the alcohol-labeled permethrin. For the 10-day repeat dose study in dogs (MRID 00042160), cumulative urinary excretion on Days 8 and 9 was 36.8% and 39.6% for one dog and 27.5% and 27.8% for the second dog. No additional data were available.

C. **METABOLITE CHARACTERIZATION STUDIES:** Metabolite characterization was limited to the determination of the isomeric form of permethrin in adipose tissue and a preliminary qualitative analysis of excreta. In MRID 00054721, TLC analysis of ether extracts of fecal samples from dogs given [^{14}C-alcohol]permethrin revealed six components according to the study authors; this was not, however, evident from the poor copy quality of the report. Both the cis and trans isomers were detected. TLC analysis of ether extracts of urine from these dogs exhibited four components, one of which according to the TLC chromatogram was PP557. In MRID 00042160, TLC analysis of adipose tissue extracts revealed both the cis and trans isomers in cis:trans ratios of 1:1.07 and 1:1.34 for peri-renal fat for the two dogs. Isomer rations for subcutaneous fat were 1:1.52 and 1:1.15 for the two dogs. Extraction efficiency was illegible.

III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** Two metabolism studies were conducted using adult Beagle dogs. In MRID 0054721, groups of four male and four female beagle dogs were given [^{14}C-alcohol]permethrin (PP557; no lot/batch nos.; 59.7 mCi/mM; purity not reported) or [^{14}C-acid]permethrin (PP557; no lot/batch nos.; 1.87 mCi/mM; purity 99%) as a single oral dose (6.5 mg/kg and 6.2 mg/kg, respectively) in a gelatin capsule. Excreta were collected over a 7-day period and tissues collected and analyzed at termination. In MRID 00042160,

two beagle dogs (gender not specified) were given 10 daily doses (1.0 mg/kg via gelatin capsules) of [^{14}C-alcohol]permethrin (PP557; no lot/batch nos.; 59.7 mCi/mM; purity not reported). Excreta were collected after seven days and adipose tissues analyzed at termination.

In MRID 00054721, approximately 43% and 46% of the administered radioactivity was excreted via the urine and feces, respectively, over the 7-day experimental period although fecal excretion was more prevalent. Most excretion occurred within 24-48 hours, although the rate of excretion was slightly less for the [^{14}C-alcohol]permethrin. At seven days postdose, radioactivity was detected in the tissues selected for analysis (peri-renal and subcutaneous fat, liver, kidney, lung, heart, blood, and brain). The highest tissue levels (0.5-0.7 µg eq./g) were found in the fat tissues. Although radioactivity was detected in all tissues seven days following the single oral dose, the study authors contended that prolonged sequestration was unlikely. TLC analysis of organic solvent extracts of urine and fecal samples revealed four components in the urine and six components in the feces. None of the fractions were characterized although it was noted that parent compound occurred in the feces but not the urine.

In MRID 00042160, dogs given 10 daily doses of [^{14}C-alcohol]permethrin excreted approximately 30-38% of the administered radioactivity in the urine within seven days. TLC analysis of peri-renal and subcutaneous fat revealed a shift in the cis:trans ratio indicative of preferential biotransformation of the trans isomer. Only parent compound was detected in the fat tissues. The authors provided a brief interspecies comparative analysis regarding metabolism/disposition noting that both the rat and dog exhibit preferential metabolism of the trans isomer (rat more so) and that dogs exhibit somewhat greater levels of permethrin in adipose tissues.

B: **REVIEWER COMMENTS:** These experiments provided semiquantitative information regarding the metabolism and disposition of permethrin in dogs. Data were insufficient for determination of definitive mass balance for administered radioactivity. Following oral administration of a single dose of [^{14}C-alcohol]permethrin (6.5 mg/kg) or [^{14}C-acid]permethrin (6.2 mg/kg), approximately 84-87% of administered radioactivity was eliminated via the feces and urine in 24-48 hours (MRID 000054721). Fecal excretion (~45-56% of dose) was somewhat greater than urinary excretion (~30-38% of dose). The single dose study indicated the presence of up to four metabolites in the urine and six in the feces although the data were not clear (poor copy quality) to fully verify this conclusion. The excretory pattern for dogs given multiple doses of [^{14}C-alcohol]permethrin (1.0 mg/kg/day for 10 days) (MRID 00042160) was similar to that observed for the single dose study. The repeat-dose study (MRID 00042160) also provided preliminary data regarding the cis:trans ratio of residues in peri-renal and subcutaneous fat, and noted that a shift in this ratio (an increase in the cis isomer in tissue residues) was indicative of a preferential metabolism of the trans isomer. Data were not presented that allowed for verification of the interspecies comparisons noted by the study authors.

These metabolism/disposition studies in the dog are classified Unacceptable/Guideline and do not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD

417] in dogs. The unacceptability is the result of deficiencies in level of detail provided which prevent verification/validation of findings (e.g., insufficient data regarding characterization of recovered radioactivity, no dose confirmation, no lot/batch numbers for the test article, mass balance data lacking in MRID 00042160). Furthermore, the studies were conducted prior to GLP Guidelines and lacked quality assurance statements.

C. **STUDY DEFICIENCIES:** The study report (MRID 00054721) text noted that no parent compound was detected in the urine but the chromatogram indicates PP557 as a component in solvent system B. Both studies were deficient in detail (e.g., no dose confirmation, no lot/batch numbers for the test article), failed to assess mass balance (MRID 00042160), and were conducted prior to GLP Guidelines and lacked quality assurance statements.

DATA EVALUATION RECORD

PERMETHRIN/109701

Study Type: Subchronic (90-day) Oral Toxicity Study (Non-rodents) OPPTS 870.3150/OECD 409

40493

MRID: 00071951 (Main study), 00071953

Prepared for
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Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
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Prepared by
Toxicology and Hazard Assessment Group
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Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed and operated by UT-Battelle, LLC., for the U.S. Dept. of Energy under contract DE-AC05-00OR22725.

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Date 5/28/2002

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Subchronic Oral Toxicity Study (Capsule) [OPPTS 870.3150 (§82-1b)] (Dog)

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): FMC 45801 [(Permethrin) Purity not stated]

SYNONYMS: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclo-propanecarboxylate; permethrin

CITATION: Gephart, L., Becci, P., Parent, R. (1980). 90-day oral subchronic dosing study with FMC 45801 in Beagle dogs. Food and Drug Research Laboratories, Inc. Laboratory Report Number 6338, June 3, 1980). MRID 00040493. Unpublished.

Osborne, B. (1977). Letter regarding potential nerve degeneration by Permethrin. Inveresk Research International. MRID 00071953. Unpublished.

SPONSOR: FMC Corporation (ICI Americas, Inc.), Princeton NJ

EXECUTIVE SUMMARY: In a 90-day oral toxicity study (MRID 00040493,), Permethrin (Purity not stated, Batch No. 79-50-2) was administered in capsules to six Beagle dogs/sex/dose at doses of 0, 5, 50 and initially 500 mg/kg that was adjusted to 364 mg/kg/day.

Severe signs of central nervous system (CNS) toxicity were observed in all high dose dogs during the first 8 days of the study. As a result, the high-dose was lowered to 364 mg/kg on day 9 through the remainder of the study. However, the CNS effects persisted, but were of shorter duration and usually disappeared between daily dosing periods. A significant decrease in body weight gain and an increase in relative liver weights were observed in high dose males. No microscopic effects attributed to treatment were found and this observation was supported by a letter from Inveresk Research International (MRID 00071953) noting that Wallerian degeneration of the sciatic nerve was not seen in a three month oral toxicity dog study with doses up to 2000 mg/kg/day.

The LOAEL for this study is 364 mg/kg/day based on decreased body weight gain of high-dose males and adverse CNS effects in high-dose male and female dogs. The NOAEL for the study is 50 mg/kg/day.

This 90-day oral toxicity study in the dog is classified **unacceptable/guideline** and does not satisfy the requirement for a 90-day oral toxicity study (OPPTS 870.3150; OECD 409) in dogs. The test article purity and stability were not provided. The pathology report text ends abruptly in midsentence on p. 193. The remainder of the text was not found, even though all pages of the MRID appear to be present in numbered sequence.

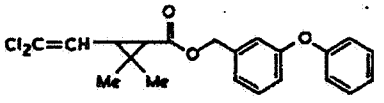
COMPLIANCE: A signed and dated GLP/Quality Assurance statement was provided. Data Confidentiality and Flagging statements and test material purity and stability were not provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test material: FMC 45801

Description:	Brown mottled solid
Lot/Batch #:	Batch No. 79-50-2
Purity:	Not reported
Compound Stability:	Not reported
CAS # if TGAI:	52645-53-1
Structure:	



2. Vehicle and/or positive control: Gelatin capsules were the delivery system and control article.

3. Test animals:

Species	Dog
Strain	Beagle
Age/weight at study initiation:	~4 to 6 months of age; males: 5.9 – 12.2 kg; females: 5.4 – 8.8 kg
Source:	Cornell University, Ithaca, NY
Housing:	Individual pens; size not specified
Diet:	Purina dog chow #5007, *ad libitum*
Water:	Tap water, *ad libitum*
Environmental conditions:	
Temperature:	Not specified
Humidity:	Not specified
Air changes:	Not specified
Photoperiod	12 hrs dark/12 hrs light
Acclimation period:	At least 2 weeks

B. STUDY DESIGN:

1. In life dates - Start: October 30, 1979 End: About January 27, 1980

2. **Animal assignment**: Animals were assigned to the test groups noted in Table 1 using a stratified randomization of body weights approach.

TABLE 1: Study design

Test Group	Dose (mg/kg/day)	# Male	# Female
Control	0	6	6
Low	5	6	6
Mid	50	6	6
High	364[a]	6	6

[a]Original dose was 500 mg/kg but was adjusted to 364 mg/kg on day 9

3. **Dose selection rationale:** The dose selection rationale was not reported. The high dose (500 mg/kg/day) was adjusted to 364 mg/kg/day on day 9 because of severe CNS effects.

4. **Dose preparation and analysis:** The doses for each dog were prepared by adding the volume of test substance for each dog to one or more gelatin capsules (Size 000) for daily administration. The low- and mid-doses were placed in one capsule and the high-dose material was placed into 5 capsules. Five capsules were used daily for each control dog. Stability analyses (identification and purity) were conducted on 1 gm samples of the test material during Weeks 1, 4, 9, and 13.

Results -

Stability analysis: Not reported
Concentration analysis: Not reported

5. **Statistics:** Body weight, body weight change, food consumption, and organ weights were evaluated using analysis of variance (ANOVA). Differences among groups were identified using the Least Significant Difference test. Pathology incidence data were done using Fisher's Exact test.

C. METHODS:

1. Observations:

1a. Cageside observations: Animals were inspected daily for signs of toxicity and twice daily for mortality.

1b. Clinical examinations: No description of detailed clinical examinations was given.

2. **Body weight:** Body weights were recorded weekly throughout the acclimation and treatment periods.

3. **Food consumption and compound intake:** Weekly food consumption values were recorded for each animal. Food efficiency was not estimated.

4. **Ophthalmology examination:** Ophthalmic examinations were done on all dogs before and at termination of treatment. A 1% tropicamide solution was instilled prior to examination.

5. **Hematology and clinical chemistry:** Blood was collected prior to study initiation and during weeks 3, 6, 10, and 13 for hematology and clinical chemistry from all surviving animals. Animals were deprived of food and water overnight (14 - 16 hours) prior to sample collection. Urine samples were collected at the same intervals. The CHECKED (X) parameters were examined.

6. a. Hematology

X	Hematocrit (HCT)*	X	Leukocyte differential count*
X	Hemoglobin (HGB)*		Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*		Mean corpusc. HGB conc.(MCHC)*
X	Erythrocyte count (RBC)*		Mean corpusc. volume (MCV)*
X	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Recommended for 90-day oral non-rodent studies based on Guideline 870.1350

b. Clinical chemistry

	ELECTROLYTES		OTHER
X	Calcium*	X	Albumin*
	Chloride*		Creatinine*
	Magnesium	X	Urea nitrogen*
	Phosphorus*	X	Total Cholesterol*
X	Potassium*	X	Globulins
	Sodium*	X	Glucose*
	ENZYMES	X	Total bilirubin*
X	Alkaline phosphatase (ALK)*	X	Total protein (TP)*
	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase		
X	Lactic acid dehydrogenase (LDH)		
X	Alanine aminotransferase (ALT also SGPT)*		
X	Aspartate aminotransferase (AST also SGOT)*		
	Sorbitol dehydrogenase*		
	Gamma glutamyl transferase (GGT)*		
	Glutamate dehydrogenase		

* Recommended for subchronic non-rodent studies based on Guideline 870.1350

7. **Urinalysis:** Urine was collected prior to study initiation and during weeks 3, 6, 10, and 13 for analysis from all surviving animals. Animals were deprived of food and water overnight (14 - 16 hours) prior to sample collection. The CHECKED (X) parameters were examined.

X	Appearance*	X	Glucose*
	Volume*	X	Ketones
X	Specific gravity / osmolality*	X	Bilirubin
X	pH*	X	Blood / blood cells*
X	Sediment (microscopic)		Nitrate
X	Protein*	X	Urobilinogen

* Recommended for subchronic non-rodent studies based on Guideline 870.1350

8. **Sacrifice and pathology:** All animals that died and those sacrificed on schedule were subjected to gross pathological and the CHECKED (X) tissues were collected and examined microscopically. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue	X	Aorta, thoracic*	XX	Brain*+
X	Salivary glands*	XX	Heart*+	X	Peripheral nerve*
X	Esophagus*	X	Bone marrow*	X	Spinal cord (3 levels)*
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*+	X	Eyes (optic nerve)*
X	Jejunum*	X	Thymus*+		GLANDULAR
X	Ileum*			XX	Adrenal gland*+
	Cecum*		UROGENITAL		Lacrimal gland
X	Colon*	XX	Kidneys*+	XX	Parathyroid*+
X	Rectum*	X	Urinary bladder*	XX	Thyroid*+
XX	Liver*+	XX	Testes*+		OTHER
	Gall bladder*+	XX	Epididymides*+	X	Bone (sternum and/or femur)
X	Pancreas*	XX	Prostate*	X	Skeletal muscle
	RESPIRATORY	XX	Ovaries*+	X	Skin*
X	Trachea*	XX	Uterus*+	X	All gross lesions and masses*
X	Lung*	X	Mammary gland*		
	Nose*	X	Cervix		
	Pharynx*				
	Larynx*				

* Recommended for 90-day oral non-rodent studies based on Guideline 870.1350
\+ Organ weight required for non-rodent studies.

II. RESULTS

A. OBSERVATIONS:

1. **Clinical signs of toxicity** - Evidence of central nervous system (CNS) toxicity was observed in all high-dose male and female dogs during the first 8 days of treatment when they received 500 mg/kg test material. These included seizures, rapid eye twitching, severe tremors, sensitivity to audio/visual stimuli, aggressive behavior, ataxia, prostration, and rigid (Straub) tails. When the daily dose was lowered to 364 mg/kg on day 9, most CNS effects persisted

through the remainder of the study, but were of shorter duration and usually disappeared between daily dosing periods. There were no signs of toxicity in the low dose group, but seizures were observed in four male dogs in the mid-dose group on Day 81 when they were inadvertently given six times the correct dose.

2. **Mortality** - All animals survived to termination.

B. **BODY WEIGHT AND WEIGHT GAIN**: Selected body weight and body weight gain data are given in Table 2. Males at the high dose level had a statistically decreased body weight gain relative to controls. No dose- or treatment-related differences in body weight gains were found in the remaining males or females during the study.

TABLE 2. Average body weights and body weight gains (kg)during 90 days of treatment[a]

Dose mg/kg/day	Body Weight (kg) ± SD				Total Weight Gain	
	Week -1	Week 1	Week 7	Terminal	kg	% of control
Male						
0	9.5 ±1.6	10.2 ± 1.7	12.9 ± 2.0	13.4 ± 2.2	3.9	100
5	8.8 ± 1.2	9.2 ± 1.0	11.8 ± 1.9	12.7 ± 2.1	3.9	100
50	9.7 ± 1.6	10.2 ± 1.8	12.7 ± 2.3	14.0 ± 2.9	4.3	110
364	8.7 ± 2.0	9.0 ± 1.9	10.9 ± 2.1	11.7 ± 2.5	3.0*	77
Female						
0	6.6 ± 0.6	6.9 ± 0.6	9.0 ± 0.4	9.8 ± 0.5	3.2	100
5	7.4 ± 1.1	8.0 ± 1.2	9.8 ± 1.3	10.6 ± 1.6	3.2	100
50	7.0 ± 0.8	7.3 ± 0.8	9.6 ± 1.2	10.0 ± 1.2	3	94
364	7.1 ± 1.2	7.5 ± 1.2	9.3 ± 1.7	10.1 ± 2.1	3	94

[a] Data obtained from pages 18-21; 160-162 of MRID 40493.
* Statistically different ($p < 0.05$) from the control at 12 weeks

C. **FOOD CONSUMPTION AND COMPOUND INTAKE**:

1. **Food consumption** - Food consumption was recorded weekly. Intermittent significant differences in food consumption were observed in both male and female dogs, however no consistent pattern was observed.

2. **Compound consumption** - Not applicable

3. **Food efficiency** - Not applicable

D. **OPHTHALMOLOGY EXAMINATION** - Ophthalmology examinations showed no treatment-related changes occurred during the study.

E. **BLOOD ANALYSES**:

1. **Hematology** - There were no consistent, notable, or dose-related variations in hematologic parameters between the control and treated groups. 2 SS

2. **Clinical Chemistry** - In the high dose male dogs, globulin was significantly increased at 10 weeks and albumin was significantly increased at study termination. These findings were considered unrelated to treatment based on historical ranges for these analytes for the laboratory.

   Alkaline phosphatase (ALK) activity was statistically increased at 3 and 6 weeks in high dose male dogs, as well as at study termination (Table 3). ALK was slightly increased in male dogs of the mid-dose level, but the activities were not statistically different. None of the increases in ALK activity were biologically significant and no correlation with histopathological data were found. No treatment-related findings for either sex were found in any of the other clinical chemistry measurements.

TABLE 3. ALK activities (IU/L) ± SD of male dogs given FMC 45801 for 90 days[a]

Week	0 mg/kg/day	5 mg/kg/day	50 mg/kg/day	364 mg/kg/day
0	129.5 ± 24.7	127.7 ± 39.0	150.8 ± 39.6	134.7 ± 44.6
3	110.8 ± 17.0	123.3 ± 28.9	135.5 ± 30.1	170.3* ± 50.7
6	86.5 ± 15.8	94.3 ± 21.0	102.2 ± 17.3	136.7* ± 42.6
10	80.3 ± 21.6	86.8 ± 18.3	98.8 ± 22.6	118.8 ± 49.6
Terminal	60.2 ± 12.9	66.7 ± 12.5	81.7 ± 17.2	125.7* ± 61.0

[a]Data taken from Table 5, pp. 34 - 48 and summary table, pp. 15, MRID 40493.
*Significantly different from control: $p \leq 0.05$.

F. **URINALYSIS** - No dose- or treatment-related changes were found for either sex.

G. **SACRIFICE AND PATHOLOGY:**

1. **Organ weight** - Liver weight data are given in Table 4. Relative liver weights were statistically increased in high-dose males, and slightly increased in high-dose females although not statistically. The elevated relative liver weights of high dose males correlated well with increased alkaline phosphatase levels in these same animals, but no correlation was evident with the histopathology findings. No other treatment-related effects were found in other organs or tissues.

TABLE 4. Absolute and relative liver (to body) weights of dogs given FMC 45801 for 90 days

Organ	0 mg/kg/day	5 mg/kg/day	50 mg/kg/day	364 mg/kg/day
Males				
Terminal body wt. (kg)	13.4 ± 2.2	12.7 ± 2.1	14.0 ± 2.9	11.7 ± 2.5
Liver				
Absolute (g)	403.9 ± 87.2	377.6 ± 51.6	415.8 ± 56.3	456.6 ± 83.2
Relative (%)	3.00 ± 0.24	2.99 ± 0.17	3.03 ± 0.40	3.94* ± 0.35
Females				
Terminal body wt. (kg)	9.8 ± 0.5	10.6 ± 1.6	10.0 ± 1.2	10.1 ± 2.1
Liver				
Absolute (g)	300.1 ± 35.8	324.1 ± 50.5	315.2 ± 41.6	367.9 ± 92.3
Relative (%)	3.08 ± 0.41	3.10 ± 0.61	3.15 ± 0.23	3.63 ± 0.30

Data taken from Table 7, pp. 59 - 61, MRID 40493.
*Significantly different from control: $p \leq 0.05$.

2. **Gross pathology** - No effects related to treatment were found.

3. **Microscopic pathology** - No microscopic effects attributed to treatment were found and this observation was supported by a letter from Inveresk Research International (MRID 00071953) noting that Wallerian degeneration of the sciatic nerve was not seen in a three month oral toxicity dog study with doses up to 2000 mg/kg/day.

III. DISCUSSION and CONCLUSIONS

A. **INVESTIGATORS' CONCLUSIONS:** The authors concluded that FMC 45801 caused acute toxicity with severe CNS effects at 500 mg/kg. These effects were reduced when the high dose was lowered to 364 mg/kg. Treatment with the test material induced a statistically significant decrease in body weight gain for high-dose males, as well as a significant increase in relative liver weights. Alkaline phosphatase activity was significantly increased in high dose male dogs at 3 and 6 weeks, as well as at study termination. Neither a LOAEL nor a NOAEL were specified by the study authors.

B. **REVIEWER COMMENTS:** Administration of FMC 45801 to male and female dogs for 90 days had no effect on food consumption, urinalysis results, or ophthalmology examinations. The high dose caused a decrease in body weight gain in males as compared to controls, as well as a significant increase in relative liver weights. In addition, the high-dose induced mild CNS effects in male and female dogs.

A slight treatment-related increase in ALK activity was found in high-dose male dogs at 3 and 6 weeks and at study termination. The increase correlated with the increased relative liver weights of this treatment group, however was of questionable biological significance since no histopathology correlation was found. A LOAEL of 364 mg/kg was demonstrated in this study based on CNS effects in high-dose male and female dogs and decreased body weight gain of male dogs. A NOAEL of 50 mg/kg was established.

C. **STUDY DEFICIENCIES:** Test article purity and stability were referred to in the report but the results were not provided. The pathology report text ends abruptly in midsentence on p. 193. The remainder of the text was not found, even though all pages of the MRID appear to be present in numbered sequence.

DATA FOR ENTRY INTO ISIS

ıbchronic Oral Study - non-rodents (870.3150)

PC code	MRID	Study	Species	Duration	Route	Admin	Dose range mg/kg/day	Doses mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ	Comments
109701	40493	subchronic	dog	90 days	oral	capsule	5 - 364	0, 5, 50, 364	50	364	weight gain decr., CNS effects	Toxicity

DATA EVALUATION RECORD

PERMETHRIN/109701
STUDY TYPE: METABOLISM AND PHARMACOKINETICS - RAT
[OPPTS: 870.7485 (§85-1)]
MRID 00065903

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-87

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Date: APR 10 2002

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

[PERMETHRIN/PC Code 109701]

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Work Assignment Manager, Health Effects Division (7509C)

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Date 5/28/2007
Signature: Joycelyn Stewart
Date 6/3/2007

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Metabolism - [rat]; OPPTS 870.7485 [§85-1)]; OECD 417.

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin

SYNONYMS: PP557; [3-phenoxybenzyl (±) cis:trans -2,2-dimethyl-3'(2,2-dichlorovinyl)-cyclopropane-1-carboxylate]

CITATION: Gaughan, L. Unai, T., Casida, J. 1976. Permethrin metabolism in rats. Division of Entomology and Parasitology, Univ. Calif., Berkeley, California, 94720. Manuscript prepared for submission to J. Agr. Food Chem. May 11, 1976.

SPONSOR: ICI Americas, Inc.

EXECUTIVE SUMMARY: In a metabolism study (MRID 00065903), groups of rats were given oral doses (1.6-4.8 mg/kg) of radiolabeled isomers ([^{14}C-acid] or [^{14}C-alcohol] labeled) of permethrin (radiochemical purity >99%; no lot/batch nos.) in dimethylsulfoxide vehicle. Metabolism and disposition was assessed over a 4 to12-day period

Recovery of administered radioactivity was 97-100% at 12 days after administration of the test article. The test material appeared to be rapidly absorbed and excreted in the urine and feces. Quantitative differences in excretion profile were characterized by greater amounts of *trans*-permethrin in the urine suggesting greater metabolism of the *trans* isomer than the *cis* isomer. Most of the urinary metabolites and some fecal metabolites appeared to be hydroxylation products, and glucuronide and sulfate conjugates of these products. Qualitative differences in metabolite profiles were also noted for the two isomers. Excretion of radioactivity via expired air was negligible. Fat tissue, liver, and kidney contained the highest levels of radioactivity, although there did not appear to be potential for sequestration at the dose regimens studied. The study authors concluded that the metabolism in rats of the *cis* and *trans* isomers of permethrin was characterized by ester cleavage, oxidation at the *cis* or *trans* methyl group of the dimethyl moiety, and oxidation at the 2' or 4' position of the phenoxy group.

This review is conducted on a best available copy of the report. However, most data tables and some text were not legible and, therefore, verification of the study authors' interpretations and conclusions was not possible. This metabolism study in the rat (MRID 00065903), apparently a

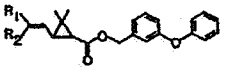
draft manuscript for submission to the J. of Agricultural and Food Chemistry, is classified **Unacceptable/Guideline** and does not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD 417] in rats. *Although the study appeared to be an in-depth examination of the metabolism of the *cis* and *trans* isomers of permethrin in the rat and could potentially achieve guideline requirements, the resulting study report was generally unreadable and exhibited notable deficiencies.

COMPLIANCE: No GLP, Quality Assurance or Data Confidentiality statements accompanied the study report.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test Compound:

Radiolabeled test material:	[^{14}C-alcohol]permethrin *cis* and *trans* isomers [^{14}C-acid]permethrin *cis* and *trans* isomers
Radiochemical purity	>99% for each test article
Specific Activity	1.7-58.2 mCi/mM
Lot/Batch #:	Not provided
Non-Radiolabelled test material:	None noted
Description:	Not provided
Lot/Batch #:	Not provided
Purity:	Not applicable
Contaminants:	None reported
CAS # of TGAI:	52645-53-1
Structure:	

2. Vehicle and/or positive control: The test article was suspended (1:10) in dimethylsulfoxide (DMSO) and administered by stomach tube. The administration was followed by a DMSO (150 µl) rinse of the tube to assure complete dose delivery.

3. Test animals:

Species:	Rat, male	
Strain:	Sprague-Dawley	
Age/weight at study initiation:	Adults (160-200g)	
Source:	Horton Laboratories, Oakland, CA	
Housing:	Housed individually in all-glass metabolism cages	
Diet:	Ground chow *ad libitum* (no additional details)	
Water:	*Ad libitum*	
Environmental conditions:	**Temperature:**	22±2°C
	Humidity:	50±5% r.h.
	Air changes:	Not specified
	Photoperiod:	12 hrs/12 hrs
Acclimation period:	3 Days	

4. **Preparation of dosing solutions:** Details regarding preparation of dosing solutions beyond description of the vehicle and actual dosing technique were unavailable or unreadable.

B. STUDY DESIGN AND METHODS:

1. **Group arrangements**: The test groups for MRID 00065903 were established that received varying doses and isomeric forms of [^{14}C-alcohol]permethrin and [^{14}C-acid]permethrin. Doses ranged from 1.6 to 4.8 mg/kg. It is unclear which groups received single or repeated doses. Additional details were obscure due to the poor copy quality of the report.

2. **Dosing and sample collection**: The test article was administered by gavage as previously described.

Expired air: Expired air and $^{14}CO_2$ was collected in monethanolamine-methyl cellosolve (2:1) from four treatment groups over a 12-day period. No additional details were available.

Urine: Urine was collected over a 12-day period. No additional details were available.

Feces: Feces were collected over a 12-day period. No additional details were available.

Tissues: Tissues (subcutaneous fat, peri-renal fat, muscle, liver, and kidney) were collected at sacrifice after 4 or 12 days of treatment. No further details were provided.

a. **Pharmacokinetic studies:** Pharmacokinetic studies were not performed.

b. **Metabolite characterization studies:** Chromatographic analysis with known standards was used to characterize metabolites isolated form excreta.

3. **Statistics:** There was no detailed descriptions of statistical methods provided. Measures of central tendency (means) were provided.

II. RESULTS:

A. PHARMACOKINETIC STUDIES: Recovery of radioactivity after 12 days was reported as 97-100%. Data tables were unreadable and verification was not possible.

1. **Absorption**: Based upon the study authors' report that most administered radioactivity was recovered in the urine after four days, absorption was extensive. Due to poor copy quality of the report, the contention could not be verified.

2. **Tissue distribution**: Tissue distribution was reported for rats given daily doses of [^{14}C-alcohol]permethrin (1 mg/kg/day) for 11 weeks. During both the treatment period and up to seven weeks post treatment, the highest concentrations (up to 2.05 µg eq./g) were found in the subcutaneous and peri-renal fat. Levels in liver, kidney and muscle were substantially lower. Autoradiographic findings affirmed the tissue distribution findings.

4. **Excretion:** Based on the study authors' conclusions, excretion via expired air was inconsequential. According to the study authors, hydrolysis products of both [^{14}C-alcohol]permethrin and [^{14}C-acid]permethrin are eliminated in the urine but not the feces. At 12 days post treatment, 97-100% of the administered radioactivity of either [^{14}C-alcohol]permethrin or [^{14}C-acid]permethrin was recovered in excreta with very little tissue residue being detected. At four days post treatment, 76-87% of the administered dose was recovered in excreta. The authors noted that only 45-54% of excreted radioactivity from the *cis* isomer occurs in the urine while 81-90% of radioactivity from the *trans* isomer is found in the urine. Data to confirm this could not be deciphered.

B. METABOLITE CHARACTERIZATION STUDIES: Metabolite characterization was performed using TLC analysis and known standards. The study authors contended that hydrolysis followed by various conjugation processes were primary reactions characterizing the biotransformation of permethrin in the rat. Both quantitative and qualitative differences were noted between the *cis* and *trans* isomers; the *cis* isomer appears to be more resistant to metabolism. A proposed metabolic pathway was provided. Verification of the study authors' conclusions was, however, not possible due to the poor copy quality of the report.

III. DISCUSSION and CONCLUSIONS:

A. INVESTIGATORS' CONCLUSIONS: In a metabolism study (MRID 00065903), groups of rats were given oral doses (1.6-4.8 mg/kg) of radiolabeled isomers ([^{14}C-acid] or [^{14}C-alcohol] labeled) of permethrin (radiochemical purity >99%; sp. act. 1.7-58.2 mCi/mmol no lot/batch nos.) in dimethylsulfoxide vehicle. Metabolism and disposition was assessed over a 4 to12-day period

Recovery of administered radioactivity was 97-100% at 12 days after administration of the test article. The test material appeared to be rapidly absorbed and excreted in the urine and feces. Quantitative differences in excretion profile were characterized by greater amounts of *trans*-permethrin in the urine suggesting greater metabolism of the *trans* isomer than the *cis* isomer. Most of the urinary metabolites and some fecal metabolites appeared to be hydroxylation products, and glucuronide and sulfate conjugates of these products. Qualitative differences in metabolite profiles were also noted for the two isomers. Excretion of radioactivity via expired air was negligible. Fat tissue, liver, and kidney contained the highest levels of radioactivity, although there did not appear to be potential for sequestration at the dose regimens studied. The study authors concluded that the metabolism in rats of the *cis* and *trans* isomers of permethrin was characterized by ester cleavage, oxidation at the *cis* or *trans* methyl group of the dimethyl moiety, and oxidation at the 2' or 4' position of the phenoxy group.

B: REVIEWER COMMENTS: The copy quality of the study report MRID 00065903 was such that review of most data was not possible and, therefore, verification of the authors' conclusions was not possible. Only portions of the text were readable and study protocol details appeared to be deficient for 85-1 Guidelines.

This metabolism study in the rat (MRID 00065903) is classified **Unacceptable/Guideline** and does not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD 417] in rats. Although the study appeared to be an in-depth examination of the metabolism of the *cis* and *trans* isomers of permethrin in the rat and could potentially achieve guideline requirements, the resulting study report was generally unreadable and exhibited notable deficiencies. Most data tables and some text were not legible and, therefore, verification of the study authors' interpretations and conclusions was not possible. Furthermore, no GLP, quality assurance or data confidentiality statements were available.

C. **STUDY DEFICIENCIES:** The report was generally unreadable, especially the data tables. Text descriptions of the experimental protocol (e.g., precise delineations of treatment groups, test article data), dose confirmation, and environmental conditions were lacking.

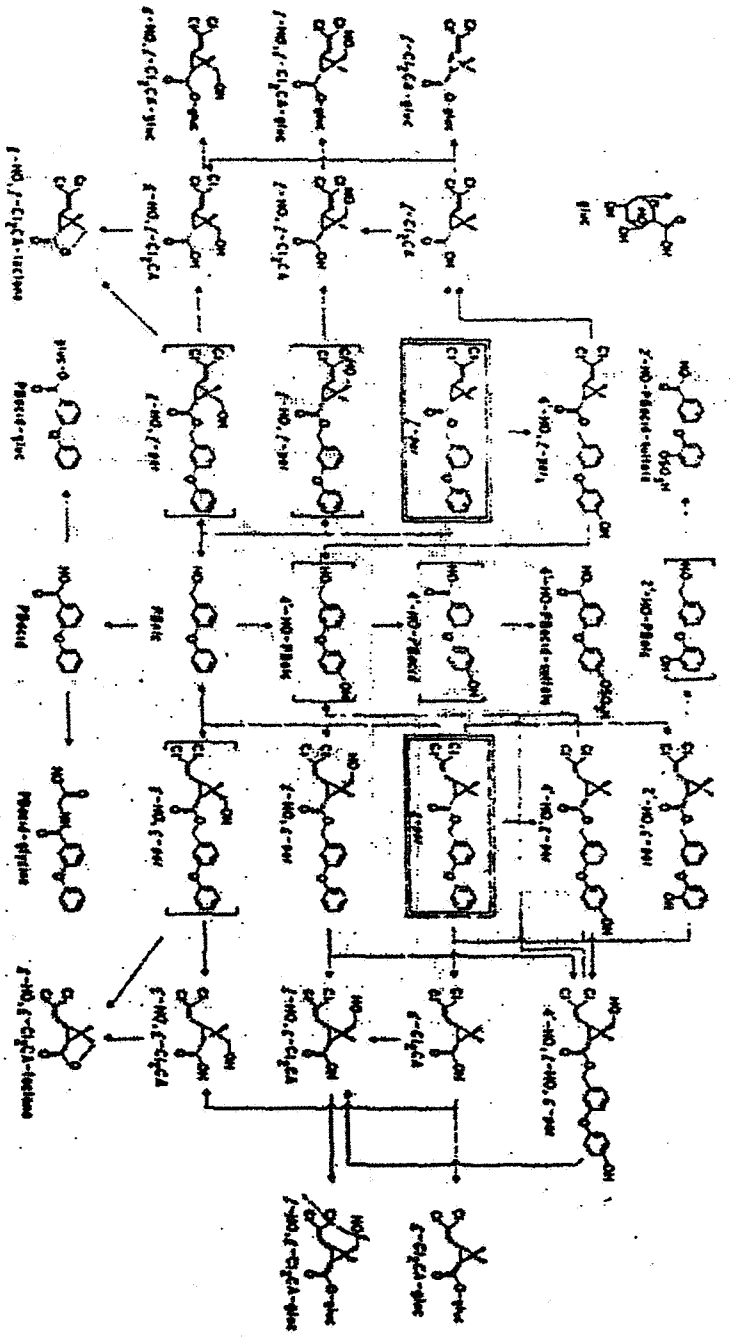


Figure 1. Proposed metabolic pathway of permethrin in the rat.
Taken from p. 48, MRID 00065903.

DATA EVALUATION RECORD

21Z73 (PERMETHRIN)/109701

STUDY TYPE: SUBCHRONIC (10-DAY) ORAL TOXICITY STUDY -Rat
MRID: 00050198

Prepared for
Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37830
Task No.02-05

≡7

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Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory managed and operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

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Date 5/22/2002

DATA EVALUATION RECORD
Supplementary, HED Doc # 007392

TXR#: 0050649

STUDY TYPE: 10-Day Oral Toxicity *[gavage]-[rat]*; Non-guideline

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 21Z73 (permethrin) (Batch WF), purity not stated
SYNONYMS: permethrin, cis/trans not reported

CITATION: Wallwork, L., R. Clampitt, and J. Malone (1974) 10-Day cumulative oral toxicity study with 21Z73 in rats. Wellcome Research Laboratories, Berkhamsted. Doc. Code HEFG 74-10, June 13, 1974. MRID 50198. Unpublished.

SPONSOR: The Wellcome Foundation, Ltd., Research and Development (V&A)

EXECUTIVE SUMMARY:

In a 10-day oral toxicity study (MRID 50198) 21Z73 (permethrin, Batch# WF) was administered to 5-6 female Wistar rats/dose in a corn oil gavage at dose levels of 0, 200, 400, and 800 mg/kg/day. Animals dosed with 800 mg/kg/day experienced marked muscle spasms and convulsions after each dose with 3/6 animals dying after the second dose and another rat dying after the fifth dose. The spasms began "shortly" after the first dose and persisted for 6 hours after dosing. The 400 mg/kg/day group showed muscle fasciculation and hypersensitivity about 6 hours after the first two doses and appeared normal thereafter. No overt toxicity signs were seen in the 200 mg/kg/day and vehicle control groups. There was a significant decrease in body weight in the two remaining 800 mg/kg/day rats over the 10-day study. Significant increases in GOT and GPT were noted in the 200 mg/kg/day treatment group and are supported by a dose-related increase in relative liver weight at all treatment levels and increased absolute liver weight in the high dose group. These liver effects were considered adaptive effects and not adverse effects.

A LOAEL is 400 mg/kg/day based on clinical signs of neurotoxicity (muscle fasciculation and hypersensitivity). A NOAEL is 200 mg/kg/day.

This 10-day oral toxicity study in the rat is **unacceptable/non-guideline** as a range finding study. Refer to the deficiency section for additional deficiencies.

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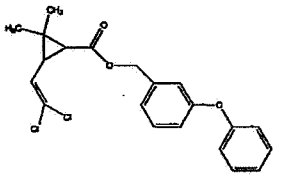
COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. **Test material:** 21Z73

Description:	Not provided
Lot/Batch #:	Batch WF
Purity:	Not provided
Compound Stability:	Not provided
CAS # of TGAI:	CAS NO. 52645-53-1



2. **Vehicle and/or positive control:** 40% (w/v) in corn oil

3. **Test animals:**

Species:	Rat (female)		
Strain:	Wistar		
Age/weight at study initiation:	230-245 g		
Source:	Charles River, England		
Housing:	5-6 animals per metal cage		
Diet:	Rat and mouse breeding diet ex Heygates *ad libitum*		
Water:	Not described		
Environmental conditions:	**Temperature:**	Not described	
	Humidity:	Not described	Not described
	Air changes:	Not described	
	Photoperiod:		
Acclimation period:	Not described		

B. STUDY DESIGN:

1. **In life dates** - Start: 2/1974 End: 3/1974 (no other specific dates)

2. **Animal assignment**: Animals were assigned to the test groups noted in Table 1. The assignment method was not described. The study design is taken from the study but the number of animals in each group is not in agreement with the bodyweight tables which show vehicle control and mid-level groups to have 5 animals.

TABLE 1: Study design for 10-day maximum tolerated dose of 21Z73 in female rats.				
Test Group	**Dose to Animal (mg/kg)**	**No. of Doses**	**# Male**	**# Female**
Absolute Control	0	0	0	6
Vehicle Control	0	10	0	6
Low	200	10	0	6
Mid	400	10	0	6
High	800	10	0	6

3. **Dose selection rationale:**

The rationale for dose selection was not provided and not needed since this was a range finding study.

4. **Dose solution preparation and analysis:** The dose was prepared in corn oil (40%, w/v). All doses were given in a constant volume of 10 ml/kg. Additional details or confirmatory analyses were not provided.

5. **Statistics:**The Students "t" test was the only statistical method employed. The limited data available to the reviewer prevents a full assessment of the suitability of the analyses but trend and linear analysis for dose dependence of response is necessary.

C. **METHODS:**

1. **Observations:**

1a. **Cageside observations:**Animals were inspected frequently (not specifically stated, probably more than once daily) for signs of toxicity and mortality after dosing.

1b. **Clinical examinations:** Clinical examinations were not described.

1c. **Neurological Evaluations**: No neurological studies were performed although neurological effects were noted.

2. **Body weight:** Animals were weighed daily.

3. **Food consumption and compound intake:** Not reported.

4. **Ophthalmoscopic examination:** Eyes were not examined.

5. **Hematology and clinical chemistry:** Blood was collected at termination for hematology and clinical chemistry from all surviving animals. The time of collection and feeding state of the animals were not provided. The CHECKED (X) parameters were examined.

a. Hematology:

X	Hematocrit (HCT)*		Leukocyte differential count*
	Hemoglobin (HGB)*		Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*		Mean corpusc. HGB conc.(MCHC)*
X	Erythrocyte count (RBC)*		Mean corpusc. volume (MCV)*
	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Recommended for 90-day oral rodent studies based on Guideline 870.3100

b. Clinical chemistry:

	ELECTROLYTES		OTHER
	Calcium		Albumin*
	Chloride		Creatinine*
	Magnesium	X	Urea nitrogen*
	Phosphorus	X	Total Cholesterol*
	Potassium*		Globulins
	Sodium*	X	Glucose*
	ENZYMES		Total bilirubin
	Alkaline phosphatase (ALK)*		Total protein (TP)*
	Cholinesterase (ChE)	X	Triglycerides
	Creatine phosphokinase		Serum protein electrophoresis
X	Lactic acid dehydrogenase (LDH)		
X	Alanine aminotransferase (ALT/also SGPT)*		
X	Aspartate aminotransferase (AST/also SGOT)*		
	Sorbitol dehydrogenase*		
	Gamma glutamyl transferase (GGT)*		
	Glutamate dehydrogenase		

* Recommended for 90-day oral rodent studies based on Guideline 870.3100

6. **Urinalysis:** Urine was not collected.

7. **Sacrifice and pathology:** All animals that died and those sacrificed on schedule by carbon dioxide and exsanguination on schedule were subjected to gross pathological examination and the CHECKED (X) tissues were collected for histological examination. The (XX) organs, in addition, were weighed. There was no report of histopathology findings.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue		Aorta*	XX	Brain*+
	Salivary glands*	XX	Heart*+		Peripheral nerve*
	Esophagus*		Bone marrow*		Spinal cord (3 levels)*
	Stomach*		Lymph nodes*		Pituitary*
	Duodenum*	XX	Spleen*+		Eyes (optic nerve)*
	Jejunum*		Thymus*+		GLANDULAR
	Ileum*			XX	Adrenal gland*+
	Cecum*		UROGENITAL		Lacrimal gland
	Colon*	XX	Kidneys*+		Parathyroid*
	Rectum*		Urinary bladder*	XX	Thyroid*
XX	Liver*+		Testes*+		OTHER
	Gall bladder (not rat)*		Epididymides*+		Bone (sternum and/or femur)
	Bile duct (rat)		Prostate*		Skeletal muscle
	Pancreas*		Seminal vesicles*		Skin*
	RESPIRATORY		Ovaries*+		All gross lesions and masses*
	Trachea*		Uterus*+		
XX	Lung*		Mammary gland*		
	Nose*				
	Pharynx*				
	Larynx*				

* Recommended for 90-day oral rodent studies based on Guideline 870.3100
+ Organ weights required for rodent studies.

II. RESULTS

A. OBSERVATIONS:

1. **Clinical signs of toxicity:** Clinical signs of toxicity included marked muscle spasms in the 800 mg/kg dose group. The spasms began shortly after the first dose (exact onset not defined) and persisted for 6 hours after dosing. After the second 800 mg/kg dose all rats showed muscle spasms and convulsions but twenty-four hours after dosing surviving rats appeared normal. The 400 mg/kg group animals showed muscle fasciculation and hypersensitivity 6 hours after the first and second doses.

2. **Mortality:** Dose related mortality was seen in the 800 mg/kg group in which 3/6 rats died within 6 hours of receiving the second dose and a fourth rat died after the fifth dose. One 400 mg/kg and one vehicle control rat died as the result of a dosing error after the seventh dose.

B. BODY WEIGHT AND WEIGHT GAIN: The table of body weights was not readable. The authors state no dose-associated effects were observed in control, 200 and 400 mg/kg groups but there was a slight but significant decrease in body weights of the surviving (2) rats in the 800 mg/kg group.

C. FOOD CONSUMPTION AND COMPOUND INTAKE: No food consumption data were reported.

D. OPHTHALMOSCOPIC EXAMINATION: No ophthalmoscopic examination was performed.

E. BLOOD ANALYSES:

1. **Hematology:** Tables of hematology parameters were unreadable but the authors state that there were no important or significant changes in red or white blood cell parameters.

2. **Clinical chemistry:** The raw clinical chemistry data table was also unreadable. A summary table was provided and it is reproduced here (Table 2). The authors provided the following summary statements for each parameter; the reviewer is unable to provide additional comments. The AP and TRIG were increased in all corn oil treated rats. No significant changes were noted in cholesterol. Treated groups showed slight to moderate increases in GOT in all treated groups. The GPT response was irregular but was increased in all corn oil treatment groups with significance in the 200 mg/kg group. The LDH shows non-significant increases in all treated groups. There were no important changes in glucose or urea N in any group. The increases in GOT, GPT, and LDH suggest slight drug induced liver damage.

TABLE 2: Summary of clinical chemistry results for 10-day maximum tolerated dose of 21Z73 in female rats. Comparisons made to absolute control.

Dose	GOT	GPT	LDH	UN	GLUC	CHOL	TRIG	AP
Corn Oil	slight decrease NS	slight increase NS	slight decrease NS	slight decrease p<0.05	NVR	slight increase NS	marked increase NS	marked increase p<0.05
200 mg/kg	slight increase p<0.05	moderate increase p<0.05	slight increase NS	slight decrease p<0.05	NVR	slight increase NS	slight increase NS	marked increase p<0.05
400 mg/kg	moderate increase p<0.05	marked increase NS	slight increase NS	NVR	NVR	slight decrease NS	marked increase p<0.05	marked increase p<0.05
800 mg/kg	moderate increase p<0.05	sight increase NS	slight increase NS	NVR	slight decrease NS	NVR	NVR	marked increase p<0.05

Data from MRID 50198, pg. 5.
NS= not significant, NVR= no value reported similar to control.

F. URINALYSIS: Not performed.

G. SACRIFICE AND PATHOLOGY: The data tables for organ weights are unreadable. Other data on sacrifice and pathology were summarized in the textual presentation.

1. **Organ weight:** The raw data tables for organ weights were unreadable. The following summary table was presented by the study authors (Table 3). The study authors note there was a dose related increase in the absolute and relative liver weights with the increase in relative weights being statistically significant at all doses. The authors do not consider any other organ weight changes to be toxicologically important. There was a statistically significant moderate increase in relative heart weight for the 800 mg/kg group. Except for the brain, most other relative and absolute organ weights in the 800 mg/kg group were

increased in spite of the noted statistically significant decrease in body weight of the two surviving animals.

TABLE 3: Summary of organ weights for 10-day maximum tolerated dose of 21Z73 in female rats. Comparisons made to absolute control.

Dose	Value	Brain	Kidney	Liver	Lung	Spleen	Heart	Adrenal	Thyroid
Corn Oil	Absolute	NVR	NVR	NVR	Slight increase NS	NVR	NVR	NVR	NVR
	Relative	NVR	NVR	NVR	Slight increase NS	NVR	NVR	NVR	NVR
200 mg/kg	Absolute	Slight decrease NS	NVR	Slight increase NS	Slight increase NS	Slight increase NS	NVR	NVR	NVR
	Relative	Slight decrease NS	NVR	Slight increase p<0.05	Slight increase NS	NVR	NVR	Slight increase NS	Slight increase NS
400 mg/kg	Absolute	NVR	NVR	Slight increase NS	NVR	NVR	NVR	Slight increase NS	Slight increase NS
	Relative	NVR	NVR	Slight increase p<0.05	NVR	NVR	NVR	Slight increase NS	slight increase NS
800 mg/kg	Absolute	Slight decrease NS	Slight increase NS	Marked increase p<0.05	NVR	Slight increase NS	Slight increase NS	Slight increase NS	NVR
	Relative	Slight decrease NS	Moderate increase NS	Marked increase p<0.05	Slight increase NS	Moderate increase NS	Moderate increase p<0.05	Moderate increase NS	NVR

Data from MRID 50198, pg. 7.
NS= not significant, NVR= no value reported- similar to control.

2. **Gross pathology:** Limited gross pathology results for the terminal observations were presented without statistical comparisons. No comments were provided about animals that died earlier in the study. No absolute control animals showed abnormal observations. In the vehicle control 1/4 animals had a pale fatty liver. In the 200 mg/kg groups 1/6 animals had a pale fatty liver and 1/6 had a slight hydropericardium. In the 400 mg/kg group 1/4 had a pale fatty liver and 1/4 had congested kidneys. In the 800 mg/kg group ½ had congested kidneys. The study authors attributed the pale fatty liver to the corn oil vehicle gavage. Congested kidneys were only noted in the 400 and 800 mg/kg groups.

3. **Microscopic pathology:** No microscopic pathology findings were reported.

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III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** The investigators note that in the 10-day oral toxicity test of 21Z73 (permethrin) animals dosed with 800 mg/kg/day experienced marked muscle spasms and convulsions after each dose with 3/6 animals dying after the second dose and another rat dying after the fifth dose. The 400 mg/kg/day group showed muscle fasciculation and hypersensitivity after the first two doses and appeared normal thereafter. No toxicity signs were seen in the 200 mg/kg/day and vehicle control groups. No important changes in body-weight or hematological parameters wee noted. Changes in GOT, GPT and LDH indicate slight liver dysfunction and there was a corresponding significant and dose related increase in the relative liver weights at all dose levels and a significant increase in absolute liver weight in the high dose group. The preliminary test indicates that 21Z73 is of low oral toxicity to female rats.

B. **REVIEWER COMMENTS:** In the 10-day oral toxicity test of 21Z73 (permethrin), animals dosed with 800 mg/kg/day experienced marked muscle spasms and convulsions after each dose with 3/6 animals dying after the second dose and another rat dying after the fifth dose. The spasms began shortly after the first dose and persisted for 6 hours. The 400 mg/kg/day group showed muscle fasciculation and hypersensitivity about 6 hours after the first two doses and appeared normal thereafter. No overt toxicity signs were seen the 200 mg/kg/day and vehicle control groups. There was a significant decrease in bodyweight in the two remaining 800 mg/kg/day rats over the 10-day study but the reviewer is not able to correlate it with feed consumption or toxicological effects and the reviewer disagrees with the study authors that this is not noteworthy.

It is not possible to comment on hematological parameters due to illegible tables. Significant increases in GOT and GPT were noted in all treatment groups and are supported by a dose related increase in relative liver weight at all treatment levels. (The reviewer was unable to recalculate dose response trends). This indicates a dose-related liver dysfunction even at the low dose level. The corn oil control allowed for the interpretation of effects of the vehicle such as the consistent increase in AP, elevated triglycerides, and appearance of fatty liver in controls. These liver effects were judged to be adaptive effect and not to be considered as adverse effects. A NOAEL is estimated as 200 mg/kg/day and a LOAEL is 400 mg/kg/day based on clinical signs of neurotoxicity (muscle fasciculation and hypersensitivity).

C. **STUDY DEFICIENCIES:** There were no information on test material (i.e., purity, stability) reported. No quality assurance statements were provided. The study used unequal numbers in groups (initially 5 or 6 mature female rats) but by the end of the 10-day trial only 2 rats were remaining in the 800 mg/kg test group. This provided for limited statistical comparisons. There were very sparse descriptions of housing conditions. No necropsy reports were presented for animals that died early in the study. No food consumption results were reported which would be necessary to further evaluate the weight loss in the 800 mg/kg treatment group. Only a limited number of organs were collected for observation and weighing and limited clinical chemistry and hematological parameters were assessed. No microscopic pathology or urinanalysis results were reported. The exact time of onset of overt toxicity symptoms was not noted. This study is classified as unacceptable/non-guideline as a range finding study.

DATA FOR ENTRY INTO ISIS

ıbchronic (10 day) Oral Study - rodents (non-guideline)

PC code	MRID	Study	Species	Duration	Route	Admin	Dose range mg/kg/day	Doses mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ	Comments
09701	50198	subchronic	rat	10 days	oral	gavage	200-800	0, 200, 400, 800	-	200	Liver, nervous system, body weight dec.	Toxicity

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DATA EVALUATION RECORD

PERMETHRIN/109701
(21Z73)
STUDY TYPE: PRENATAL DEVELOPMENTAL TOXICITY STUDY - MOUSE
OPPTS 870.3700a [§83-3a]; OECD 414
MRID 00057100

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
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Work Assignment No. 02-05

Primary Reviewer: Donna L. Fefee, D.V.M.	Signature: Date:	Robert H. Ross for D.L. Fefee APR 0 5 2002
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Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross APR 0 5 2002
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson APR 0 5 2002

Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

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EPA Reviewer: Y. Yang, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: J. Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yang G. Yang
Date 5/8/2002
Signature: Jaycelyn Stewart
Date 5/21/2002

DATA EVALUATION RECORD

TXR#: 0050649

STUDY TYPE: Prenatal Developmental Toxicity Study - Mouse; OPPTS 870.3700a [§83-3a]; OECD 414.

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 21Z73 (Permethrin; purity and cis-/trans- ratio not provided)

SYNONYMS: NRDC 143

CITATION: James, D. (1974) Preliminary foetal toxicity study in the mouse given 21Z73 (NRDC 143) orally. Wellcome Research Laboratories, Beckenham. Laboratory report number Path 169, June 20, 1974. MRID 00057100; Assession No. 00110645. Unpublished.

SPONSOR: The Wellcome Foundation, Ltd.

EXECUTIVE SUMMARY: In a developmental toxicity study (MRID 00057100) 21Z73 (Permethrin; purity, cis-/trans- ratio, and batch/lot # not reported) was administered to 22-23 mated female CDI mice/dose in corn oil by gavage at dose levels of 0 or 400 mg/kg/day from days 6 through 15 of gestation. An additional group of 20 animals served as "environmental controls." Dams were sacrificed on GD 18, and all fetuses were weighed, sexed, and subjected to external examination. Approximately one-third of the fetuses from each litter were examined for skeletal alterations, and the remaining two-thirds were examined for visceral alterations.

There were no treatment-related effects on maternal body weight. Food consumption was not measured and clinical signs were not reported. It is unknown whether a single intercurrent death in the treated group (on GD 17) was treatment-related. **The maternal toxicity NOAEL for 21Z73 in CDI mice is ≥400 mg/kg/day, and the maternal toxicity LOAEL is not identified.**

Increased postimplantation loss of the treated group occurred (9.9% vs. 6.9% for controls) due to an increased number of dams exhibiting at least one resorption (76.2% vs. 39% of control dams; $p<0.05$). There were no treatment related increases in fetal deaths or the incidences of fetal structural alterations, and there was no evidence of altered growth. **The developmental toxicity LOAEL for 21Z73 in CDI mice is 400 mg/kg bw/day, based on increased resorptions. The developmental toxicity NOAEL is not identified.**

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This developmental toxicity study in the mouse is classified **unacceptable/guideline).** Due to numerous major deficiencies noted in the conduct of this study (see deficiency section), the study alone still would not satisfy the guideline requirements for a developmental toxicity study in the mouse (OPPTS 870.3700); however, the study may provide supplemental information when combined with additional, more recent, studies. It must be noted that the study report did not describe the method of dosing formulation preparation and that concentration and homogeneity analyses were not done; it is therefore not known whether the mixing procedure was adequate or whether the variance between nominal and actual dosages to the animals was acceptable

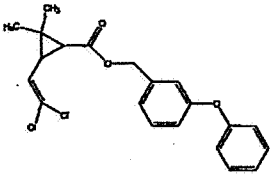
COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided; however, the study was conducted in 1974.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. **Test material:** 21Z73

Description:	not reported
Lot/Batch #:	not reported
Purity:	not reported
Compound Stability:	not reported
CAS #of TGAI:	52645-53-1



2. **Vehicle and/or positive control:** The vehicle was corn oil (Lot/Batch # and purity not reported). There was no positive control used.

3. **Test animals:**

Species:	Mouse	
Strain:	Females: CDI; Males: XP	
Age/weight at study initiation:	Females: approximately 8 weeks; 26-37 g. at mating; Males: approximately 10 weeks; weights not provided	
Source:	Charles River (UK) Ltd	
Housing:	Not reported	
Diet:	Rat cake (supplied by Lillico, manufactured by Heygates) *ad libitum ad libitum*	
Water:	*ad libitum*, not otherwise described	
Environmental conditions:	**Temperature:**	Not reported
	Humidity:	Not reported
	Air changes:	Not reported
	Photoperiod:	Not reported
Acclimation period:	Not reported	

B. PROCEDURES AND STUDY DESIGN:

1. **In life dates** - Not reported. The entire study took place March-June 1974.

2. **Mating:** Females were mated with XP strain males. The day on which a vaginal plug was observed was designated as gestation day (GD) 0.

3. **Animal assignment:** Animals were assigned by an unspecified method to dose groups as indicated in Table 1. On GD 0, the group mean body weights and their variances were homogeneous (Bartlett's test and Dunnett's comparison performed by reviewer). In addition, an environmental control group consisting of 20 animals was monitored throughout the study.

TABLE 1. Animal assignment

Dose (mg/kg bw/day) [a]	0 (Vehicle controls)	400
# Females	23	22

Data taken from text, p. 3, MRID 00057100.
a Animals were treated on GD 6-15, inclusive. Dose is expressed in terms of the active ingredient.

4. **Dose selection rationale:** There was no dose selection rationale provided. The title of the study report [Preliminary foetal toxicity study in the mouse given 21Z73 (NRDC 143) orally] implies that this study may have been intended as a range-finding study; however, this is not stated explicitly in the report.

5. **Dosage preparation and analysis** :The preparation method of the test material-vehicle mixture was not described. The study report stated that the treated group received 400 mg active ingredient/kg body weight; it is therefore assumed that the concentration of the test material in the vehicle was adjusted to account for purity.

   There was no mention of the frequency of preparation or storage conditions of mixtures of test substance with the vehicle. There was also no mention of evaluation of the stability of the test substance in the vehicle or concentration and homogeneity of the test mixtures.

   There were no analytical data provided to indicate whether the mixing procedure was adequate or that the variance between nominal and actual dosage to the study animals was acceptable.

6. **Dosage administration:** All doses were administered once daily by gavage, on gestation days 6 through 15, inclusive. The vehicle control group received a volume of 10 mL/kg of body weight/day; however, the volume given to the treated group was not reported. The study report did not state whether dosing was based on the body weight from the most recent body weight determination; however, this is assumed to be the case as the animals were weighed daily during gestation.

C. OBSERVATIONS

1. **Maternal observations and evaluations:** The animals were checked once daily for mortality or clinical signs. Body weights were recorded daily throughout gestation but were

reported only for GDs 0, 6, 16, and 18. Food consumption was not measured. Dams were sacrificed on GD 18. Examinations at sacrifice consisted of recording numbers of implantations, live fetuses, dead fetuses, early or late resorptions, and externally abnormal fetuses. There was no mention of examination of ovaries for corpora lutea or further gross necropsies of the dams.

2. **Fetal evaluations**: All fetuses were weighed and subjected to external examination. Approximately one-third of the fetuses from each litter were subjected to soft tissue examination of the organs of the neck, thorax, and abdomen by gross dissection; one-third were examined by the Wilson method of serial free-hand sectioning; and one-third were subjected to skeletal examination after staining by the Staples and Schnells Alizarin red S method. External, visceral, and skeletal anomalies were not further characterized as variations and malformations. Litter incidences of fetal morphological data were not reported and fetal individual morphological data were not provided.

D. DATA ANALYSIS:

1. **Statistical analyses:** The following data were analyzed using ANOVA: maternal body weight gains during dosing and gestation, numbers of corpora lutea, implantations, live fetuses, normal fetuses, litter weights, and weights of normal fetuses. Litter incidences of external, visceral, and skeletal anomalies were not reported, and fetal incidences of these endpoints were not compared statistically.

   "Standard errors" were reported for some of the data. The term "standard error" can either be a synonym of "standard deviation."or a separate statistical measure of variance that can be converted to standard deviation by multiplying by the square root of n. As it was unknown which was the case in the study report, the data were summarized as reported.

2. **Indices:** The reviewer calculated the postimplantation loss index as follows:

   Postimplantation loss (%) = (total number of implantations minus total number of live fetuses/number of implantations) x 100.

3. **Historical control data:** Historical control data (cumulative control data) were provided for fetal visceral and skeletal anomalies to allow comparison with concurrent controls. However, these data were provided only in aggregate form rather than by individual study with appropriate descriptive statistics, and there was no information provided regarding the number of studies included, dates of the studies, source of the animals, or the vehicle(s) and route(s) of administration.

II. RESULTS:

A. MATERNAL TOXICITY:

1. **Mortality and clinical observations:** Clinical observations were not reported. One dam in the treated group died on GD 17. In the absence of clinical signs or necropsy data, it is not possible to rule out the possibility that this death may have been treatment-related.

2. **Body weight:** Body weight data are summarized in Table 2. Mean absolute body weights throughout gestation and the estimated corrected mean terminal body weight and corrected body weight gains of the treated and vehicle control groups were similar. Increased body weight gains by the treated dams during the treatment interval followed by decreased body weight gain by this group post treatment were not considered treatment-related.

TABLE 2. Mean maternal body weights and body weight gains (g)

GD	Dose in mg/kg bw/day (# of Dams)	
	Vehicle control (23)	400 (21)
Absolute body weights [a]		
0	30.35	30.67
6	33.91	34
16	49.74	51.48
18	57.17	58.29
Corrected body weight [b]	42.85	42.32
Body weight changes		
0-6 (Pre-treatment) [c]	3.57	3.33
6-16 (Treatment) [c]	15.83	17.48 (110) [d]
16-18 (Post-treatment) [c]	7.43	6.81 (75.2)
0-18 [e]	26.83±1.22	27.62±0.83
Corrected body weight gain [f]	12.5	11.63

Data taken from Tables 1, 5, and 7, pp. 7, 12-13, and 16-17, respectively, MRID 00057100.
a Data reported as means, standard deviations or standard errors were not provided.
b Estimated by reviewer as Corrected body weight = Mean GD 20 weight minus mean litter weight. Mean litter weights for vehicle controls and treated groups were 14.32 and 15.97 g, respectively.
c Calculated by reviewer using mean absolute body weight data. Not subjected to statistical analysis.
d Number in parentheses is percent of vehicle control; calculated by reviewer.
e Data given as Mean ± standard error; no statistically significant intergroup differences.
f Estimated by reviewer as Corrected body weight gain = Corrected body weight minus GD 0 body weight.

3. **Food Consumption** - Food consumption was not measured.

4. **Gross Pathology** - There were no gross pathology data reported.

5. **Cesarean Section Data** - Data collected at cesarean section are summarized in Table 3. There were no remarkable differences between pregnancy rates, implantations per dam, mean live litter size, mean fetal weights, or fetal sex ratios of the treated and vehicle control groups. There were no abortions, early deliveries, or total litter resorptions in either group. Postimplantation loss of

the treated group was increased compared to that of the vehicle control group.. Although the increases in early and late resorptions were very slight, this was considered a treatment-related effect, as there was a corresponding statistically significant increase noted in the number of dams exhibiting at least one resorption.

TABLE 3. Cesarean section observations [a]

Observation	Dose (mg/kg bw/day)	
	0 (Vehicle control)	400
# Animals Assigned (Mated)	23	22
# Animals Pregnant	23	21 [b]
Pregnancy Rate (%)	100	100 [b]
# Nonpregnant	0	0 [b]
Maternal Wastage		
# Died	0	1
# Aborted	0	0
# Premature Delivery	0	0
Total # Implantations	262	272
Implantations/Dam	11.39±0.68	12.95±0.50
Total # Litters	23	21
Total # Live Fetuses	244	245
Live Fetuses/Dam	10.61±0.64	11.67±0.46
Total # Dead Fetuses	2	3
Dead Fetuses/Dam	0.09	0.14
Total # Resorptions	16	24
Early	11	15
Late	9	5
Resorptions/Dam [c]	0.7	1.14
Early	0.48	0.71
Late	0.22	0.43
Litters with Total Resorptions	0	0
# [percent] of litters with at least one resorption [d]	9 (39.1%)	16 (76.2%) *
# [percent] of litters with at least one postimplantation loss [d]	11 (47.8%)	16 (76.2%)
Mean Fetal Weight (g)	1.35	1.37
Sex Ratio (% Male)	58.61	60
Postimplantation Loss (%) [e]	6.9	9.9 (143)[f]

Data taken from Tables 1, 6, and 7, pp. 7, 14-15, and 16-17, respectively, MRID 00057100.

a Data expressed as mean ± standard error where appropriate; however, standard errors were not provided and/or legible for all means.

b One dam that died during the study is excluded because her pregnancy status was not reported.

c Calculated by reviewer and not subjected to statistical analysis.

d Calculated by reviewer from individual data and subjected to statistical analysis using the Fisher Exact test.

e Calculated by reviewer as Postimplantation Loss = (Total implantations minus total live fetuses/Total implantations) x 100.

f Number in parentheses equals percent of controls, calculated by reviewer.

* Significantly different from control ($p<0.05$); Fisher Exact test performed by reviewer.

B. **DEVELOPMENTAL TOXICITY**: The total numbers of live fetuses (litters) in the vehicle control and treated groups were 244 (23) and 245 (21), respectively. The study report did not include litter incidences of fetal structural alterations, and it was unclear whether all findings, both major and minor were reported. Selected fetal morphological observations are given in Table 4.

1. **External examination:** There were no external abnormalities observed at caesarian section.

2. **Visceral examination:** Visceral abnormalities included a very pale spleen, dilated renal pelves, and a thickened and dilated urinary bladder in treated fetuses, all of which were present at single or low incidences. None were considered treatment-related. A dilated renal pelvis was also observed in one control fetus.

3. **Skeletal examination:** All observations from fetal skeletal examinations were found at similar incidences in the treated and vehicle control groups. However, the reported observations concerned only the axial skeleton; it is therefore unknown whether the appendicular skeleton was routinely examined. Sternebral ossification was reported as the number and percent of fetuses with either one or more unossified sternebra(e) or one or more poorly ossified sternebra(e).

TABLE 4. Fetal morphological data [a]		
Observations [b]	Dose in mg/kg bw/day (number of litters)	
	Vehicle controls (23)	400 (21)
Visceral Examination - Open Dissection		
Number of fetuses examined	82	82
Very pale spleen	0	1
Visceral Examination - Wilson Method		
Number of fetuses examined	81	82
Pelvis of left kidney dilated	1	1
Pelvis of right kidney dilated	0	2
Thickened and dilated urinary bladder	0	1
Skeletal Examination		
Number of fetuses examined	81	81
Hyoid poorly ossified	2	8
Occipitals poorly or irregularly ossified	17	19
Sternebrae - 1 or more bifid	1	0
Sternebrae - 1 or more not ossified	0	2
13th ribs very small bilaterally	1	0

Data taken from Tables 3 and 4a-4c, pp. 9 and 10-11, respectively, MRID 00057100.

a Data are given as the number of fetuses with the specified abnormality. Litter incidences were not reported, and individual data for fetal morphological endpoints were not included in the study report. Abnormalities were not further categorized as malformations/variations or any other classifications of anomalies.

b Some observations may be grouped together.

III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** The study author concluded that there were no effects on maternal weight or body weight gain during gestation, or numbers of implantations, live fetuses, malformations, or fetal deaths. The study author did not identify LOAELs or NOAELS.

B. **REVIEWER COMMENTS:**

1. **Maternal toxicity:** It is unknown whether there were any treatment-related clinical signs or whether the death in the treated group was treatment-related. There were no treatment-related effects on maternal body weight, and food consumption was not measured. Increased postimplantation loss by the treated group was due to an increased number of dams exhibiting at least one resorption. [This conclusion differs from that of the study author.] **Therefore, the maternal toxicity NOAEL for 21Z73 in CDI mice is ≥400 mg/kg bw/day, and the maternal toxicity LOAEL is not identified.**

2. **Developmental toxicity:**

a. **Deaths/resorptions:** Maternal treatment resulted in an increase in fetal resorptions.

b. **Altered growth:** Maternal treatment did not result in decreased fetal weights. It was not possible to adequately evaluate fetal ossification rates due to insufficient data.

c. **Developmental abnormalities:** Treatment with the test article did not result in an increased incidence of fetal structural alterations.

Therefore, the developmental toxicity LOAEL for 21Z73 in CDI mice is 400 mg/kg bw/day based on increased resorptions, and the developmental toxicity NOAEL is not identified.

C. **STUDY DEFICIENCIES:**

Major deficiencies include the following:

- The study had a single dose level and a concurrent control group.
- The study report did not include full identification of the test material (chemical identification, percentage active, batch or lot number, physical properties, purity/impurities, expiration date). It is also unknown whether the technical form of the active ingredient was used.
- There were no analyses for test material stability, homogeneity and concentration in the dosing medium.
- Individual daily clinical observations were not included in the study report; nor were clinical observations reported in summary form.
- Gross necropsies of the dams were not conducted.

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- Individual and group mean body weights were only reported for GDs 0, 6, 16, and 18 instead of at 3-day intervals during the dosing period as specified in the guideline.
- Gravid uterine weights and body weight changes adjusted for gravid uterine weights were not reported.
- Food consumption was not measured.
- Litter incidences were not included for fetal morphological observations, and the litter was not considered the basic unit of analysis in the evaluation of fetal structural alterations. Individual data for these endpoints were also not provided; therefore the reviewer was unable to calculate numbers and percent of litters with structural alterations.

The following deficiencies were considered minor and did not affect the classification of the study:

- The test substance was administered during GD 6-15 (inclusive) rather than through the day prior to cesarean section as specified in the guideline. However, the dosing interval included the period of major organogenesis in the mouse.
- Females were mated with males of a different strain.
- Mean fetal body weights were only reported for combined sexes.
- Approximately one-third of the fetuses were examined for skeletal alterations rather than one-half as specified by the guideline.

DATA FOR ENTRY INTO ISIS

evelopmental Study - mice (870.3700a)

PC code	MRID	Study	Species	Duration	Route	Admin	Dose range mg/kg/day	Doses mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ	Comments
09701	57100	developmental	mice	GD 6-15	oral	gavage	400	0, 400	≥400 mg/kg/day	not determined	not identified	Maternal
09701	57100	developmental	mice	GD 6-15	oral	gavage	400	0, 400	not determined	400	increased resorptions	Developmental

8/2

EXECUTIVE SUMMARY

PERMETHRIN

STUDY TYPE: DEVELOPMENTAL TOXICITY - RAT
[OPPTS 870.3700 (§83-3A)]
MRID NO. 40943603

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order 01-85

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Date: JAN 11 2001

Secondary Reviewers:
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Signature: Cheryl B Bast
Date: JAN 11 2001

Robert H. Ross, M.S., Group Leader

Signature: Robert H. Ross
Date: JAN 11 2001

Disclaimer

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PERMETHRIN

Developmental Toxicity Study [OPPTS 870.3700 (83-3a)]

EPA Reviewer: Yung G. Yang, Ph.D. Yung G. Yang Date: 10/10/2001
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Joycelyn Stewart Date: 1/28/2002
Toxicology Branch, HED (7509C)

TXR# 0050649

DATA EVALUATION RECORD

This is an updated executive summary of MRID 40943603, HED Doc. No. 008344. The final conclusion has not been changed.

STUDY TYPE: Developmental Toxicity - Rat; OPPTS 870.3700 (§83-3a)

DP BARCODE: D269531
PC CODE: 109701

SUBMISSION CODE: S504352
TOX CHEM NO: 652BB

TEST MATERIAL: Permethrin (93.9% a.i.; 38 cis:62 trans isomers)

CHEMICAL NAME: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; permethrin; NRDC 143; PP557

CITATION: Hodge, M.C.E. (1988). Permethrin: Teratogenicity study in the rat. ICI Central Toxicology Laboratory, Alderley Park Macclesfield, Cheshire, UK. Study No. CTL/P/2269. September 20, 1988. MRID 40943603. Unpublished.

SPONSOR: ICI Americas, Inc.

EXECUTIVE SUMMARY: In a developmental toxicity study (MRID 40943603), 24 presumed pregnant Wistar rats per group were administered 0, 15, 50, or 150 mg/kg/day of permethrin (93.9% a.i.; 38 cis:62 trans isomers; Reference No. RS 78/E) by gavage on gestation days (GD) 7-16, inclusive. The vehicle was corn oil. On GD 22, all surviving dams were sacrificed and all fetuses were weighed, sexed, and examined for external malformations/variations. All fetuses were examined for visceral anomalies and the heads cut along the fronto-parietal suture line. All carcasses were processed for skeletal examination.

All animals survived to scheduled termination and no treatment-related abnormalities were noted at gross necropsy. No maternal effects on clinical signs of toxicity, body weight gains, or food consumption were observed in the low- or mid-dose groups. In the high-dose group, clinical signs of toxicity seen between GD 8-19 included tremors in 21/24 rats and head flicking in 6/24 rats. Body weight gains by the high-dose dams were significantly ($p \leq 0.05$ or 0.01) less than that of the controls throughout the dosing interval. For GD 7-10, 10-13, and 13-16, body weight gains were decreased by 88%, 32%, and 18%, respectively, as compared with the controls. Food consumption by the high-dose group was significantly ($p \leq 0.05$ or 0.01) less than that of the controls during the dosing interval.

Therefore, the maternal toxicity LOAEL is 150 mg/kg/day based on clinical signs of toxicity and decreased body weight gain and food consumption. The maternal toxicity NOAEL is 50 mg/kg/day.

No dose- or treatment-related effects were observed on gravid uterine weights, fetal sex ratios, pre- or post-implantation losses, or numbers of corpora lutea/dam or live fetuses/dam. Mean fetal body weight of the high-dose group was 3.2% ($p \leq 0.05$) less than that of the controls. However, mean litter weight of the high-dose group was 3% (n.s.) greater than that of the controls. Therefore, the reduced fetal body weights were considered a questionable toxic response.

No treatment-related external or visceral fetal malformations/variations were noted. The fetal and litter incidence rates of short length extra ribs were significantly ($p \leq 0.05$ or 0.01) increased in the high-dose group as compared with the controls. Short length extra ribs were observed in 31% of the high-dose fetuses vs. 11% of the control fetuses and in 87% of high-dose litters vs. 57% of control litters.

Therefore, the developmental toxicity LOAEL is 150 mg/kg/day based on decrease in fetal body weights and an increase in the incidence rate of short length extra ribs. The developmental toxicity NOAEL is 50 mg/kg/day.

This study is classified as **Acceptable/Guideline** and does satisfy the requirements for a developmental toxicity study [OPPTS 870.3700 (83-3a)] in rats.

DATA EVALUATION RECORD

PERMETHRIN
(FMC 33297)

STUDY TYPE: SUBCHRONIC ORAL TOXICITY [CAPSULE] - DOG [870.3150 (82-1b)]
MRID 00110647

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

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Life Sciences Division
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Task No. 02-05

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Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

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[PERMETHRIN/109701] Subchronic (90-Day) Dog Oral Toxicity Study

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Date 1/29/2002

TXR#: 0050649

DATA EVALUATION RECORD

STUDY TYPE: Subchronic Oral Toxicity *[capsule]-[dog]*;OPPTS 870.3150 [§82-1b]

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): FMC 33297 (Permethrin; purity not given)

SYNONYMS: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclo-propanecarboxylate

CITATION: Killeen, J.C. and Rapp, W.R. (1976) A three month oral toxicity study of FMC 33297 in beagle dogs. BioDynamics Inc., East Millstone, NJ. Laboratory report number 75-1188B, January 28, 1976. MRID 00110647. Unpublished.

SPONSOR: FMC Corporation, Middleport, NY 14105

EXECUTIVE SUMMARY: In a 90-day oral toxicity study (MRID 00110647) technical Permethrin (purity not given, batch/lot # C-6699-65) was administered to 4 beagle dogs/sex/dose by capsule at dose levels of 0, 5, 50, or 500 mg/kg/day.

All animals survived to scheduled sacrifice. Tremors were observed on several occasions between weeks 6 and 14 in three males and two females in the high-dose groups. One high-dose male also exhibited transient narcosis with nystagmus on one occasion during week 13. No other clinical signs of toxicity were described for any animal.

No statistical differences in weekly body weights were observed during the study between the treated and control groups of either sex. In general, weight gains by the treated groups were less than those of the control groups throughout the study although a clear dose-related decrease was not evident. Food consumption was not measured.

No treatment-related effects were observed in hematology, clinical chemistry, or urinalysis parameters, ophthalmoscopic examinations, gross necropsy, or microscopic evaluations. For the high-dose groups at necropsy, increases in organ weights were not of a magnitude as to be considered biologically significant and increases in relative organ weights could be attributed to slightly lower final body weights.

Therefore, the systemic toxicity LOAEL for permethrin in male and female dogs is 500 mg/kg/day based on clinical signs of toxicity. The systemic toxicity NOAEL is 50 mg/kg/day.

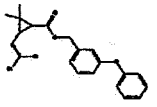
This 90-day oral toxicity study in the dog is **Unacceptable/Guideline** (upgradeable) and does not satisfy the guideline requirement for a 90-day oral toxicity study (OPPTS 870.3150; OECD 409) in dogs. Upon submission of data on purity and stability of the compound the study may be upgraded to acceptable.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test material: FMC 33297

Description:	Yellow liquid
Lot/Batch #:	C-6699-65
Purity:	Not given
Compound Stability:	Not given
CAS # if TGAI:	52645-53-1
Structure:	

2. Vehicle and/or positive control: The test article was administered in capsules. No vehicle or positive control was used in this study.

3. Test animals:

Species:	Dog	
Strain:	Beagle	
Age/weight at study initiation:	7-8 Months; males: 9.7-11.7 kg; females: 7.2-10.3 kg	
Source:	Marshall Research Animals, Inc., North Rose, NY	
Housing:	Animals were housed individually in elevated metal grid cages.	
Diet:	Dogs were given standard laboratory diet daily; additional details were not given.	
Water:	Water (source not stated) was available *ad libitum*.	
Environmental conditions:	**Temperature:**	Not given
	Humidity:	Not given
	Air changes:	Not given
	Photoperiod:	Not given
Acclimation period:	28 days	

B. STUDY DESIGN:

1. **In life dates** - Start: July 10, 1975; End: October 13-21, 1975

2. **Animal assignment**: Animals were assigned by body weight to the test groups noted in Table 1 in an attempt to equalize mean group weights.

TABLE 1: Study design

Test Group	Dose to Animal (mg/kg/day)	No. Male	No. Female
Control	0	4	4
Low	5	4	4
Mid	50	4	4
High	500	4	4

3. **Dose selection rationale**: A dose selection rationale was not given in the report.

4. **Capsule preparation:** Capsules were prepared daily. The amount of test article placed into each capsule was based on the most recently recorded body weight.

5. **Statistics:** The treated groups were compared to the control group at each time interval. Body weight and hematology and clinical chemistry parameters were analyzed by the F-test and Student's t-test. When variances differed significantly, Student's t-test was appropriately modified and Cochran's approximation was utilized. Organ weights and organ-to-body weight ratios were analyzed by Dunnett's test.

C. METHODS:

1. **Observations:**

a. **Cageside observations:** Animals were inspected daily for signs of toxicity and for mortality.

b. **Clinical examinations:** Clinical examinations were conducted weekly.

2. **Body weight:** Animals were weighed prior to study initiation, weekly during treatment, and at termination after fasting.

3. **Food consumption:** Food consumption for each animal was determined by visual estimation prior to study initiation and four times weekly thereafter.

4. **Ophthalmoscopic examination:** Eyes of all animals were examined by indirect ophthalmoscopy prior to study initiation and at termination. Tropicamide was used to induce mydriasis.

5. **Hematology & Clinical Chemistry:** Blood was collected prior to study initiation, after 1 month of treatment, and at termination for hematology and clinical chemistry from all surviving animals. The CHECKED (X) parameters were examined. The report did not state whether the animals were fasted prior to collection, however glucose was listed as "fasting glucose."

a. Hematology:

X	Hematocrit (HCT)*	X	Leukocyte differential count*
X	Hemoglobin (HGB)*		Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*		Mean corpusc. HGB conc.(MCHC)*
X	Erythrocyte count (RBC)*		Mean corpusc. volume (MCV)*
	Platelet count*	X	Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)	X	RBC morphology
X	(Clotting time)		
X	(Prothrombin time)		

b. Clinical Chemistry:

	ELECTROLYTES		OTHER
	Calcium*	X	Albumin*
	Chloride*		Creatinine*
	Magnesium	X	Urea nitrogen*
	Phosphorus*		Total Cholesterol*
	Potassium*	X	Globulins
	Sodium*	X	Glucose*
	ENZYMES		Total bilirubin*
X	Alkaline phosphatase (ALK)*	X	Total protein (TP)*
	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase		Serum protein electrophores
	Lactic acid dehydrogenase (LDH)		
X	Alanine amino-transferase (also SGPT)*		
X	Aspartate amino-transferase (also SGOT)*		
	Sorbitol dehydrogenase*		
	Gamma glutamyl transferase (GGT)*		
	Glutamate dehydrogenase		

c. Urinalysis

Urine was collected from all animals prior to study initiation, after 1 month of treatment, and at termination. The CHECKED (X) parameters were examined.

X	Appearance*	X	Glucose*
	Volume*	X	Ketones
X	Specific gravity / osmolality*	X	Bilirubin
X	pH*	X	Blood / blood cells*
	Sediment (microscopic)		Nitrate

X	Protein*		Urobilinogen

* Recommended for subchronic non-rodent studies based on Guideline 870.1350

7. **Sacrifice and pathology**: All animals that died and those sacrificed on schedule were subjected to gross pathological examination. Animals were sacrificed by exsanguination under sodium pentobarbital anesthesia. The CHECKED (X) tissues were collected for histological examination. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue	X	Aorta, thoracic*	XX	Brain*+
X	Salivary glands*	X	Heart*+		Peripheral nerve*
	Esophagus*	X	Bone marrow*	X	Spinal cord (cervical)*
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	X	Spleen*+	X	Eyes (optic nerve)*
	Jejunum*		Thymus*+		GLANDULAR
X	Ileum*			XX	Adrenal gland*+
	Cecum*		UROGENITAL		Lacrimal gland
X	Colon*	XX	Kidneys*+		Parathyroid*+
	Rectum*	X	Urinary bladder*	XX	Thyroid*+
XX	Liver*+	XX	Testes*+		OTHER
X	Gall bladder*+		Epididymides*+		Bone (sternum and/or femur)
X	Pancreas*	X	Prostate*	X	Skeletal muscle
	RESPIRATORY	XX	Ovaries*+	X	Skin*
	Trachea*	X	Uterus*+	X	All gross lesions and masses*
X		X	Mammary gland*		
	Lung*				
	Nose*				
	Pharynx*				
	Larynx*				

* Recommended for 90-day oral non-rodent studies based on Guideline 870.1350
+ Organ weight required for non-rodent studies.

II. RESULTS:

A. OBSERVATIONS:

1. **Clinical signs of toxicity**: The study authors stated that tremors were observed between weeks 6 and 14 in three males and two females in the high-dose groups. One high-dose male also exhibited transient narcosis with nystagmus on one occasion during week 13. No other clinical signs of toxicity were described for any animal. Neither a summary table nor individual animal observations were included in the report.

2. **Mortality**: All animals survived to scheduled termination.

B. BODY WEIGHT AND WEIGHT GAIN:

No statistical differences in weekly body weights were observed during the study between the treated and control groups of either sex. In general, weight gains (calculated by the reviewer from group means) by the treated groups were lower than those of the control groups throughout the study although a clear dose-

related decrease was not evident. However, overall body weight gains by the low-, mid-, and high-dose groups were 109%, 51%, and 51%, respectively, of the control levels for males and 60%, 69%, and 35%, respectively, of the controls for females.

TABLE 2. Average body weights (kg) and body weight gains[a] (kg) of beagle dogs during 90 days of treatment with Permethrin.

Week of study	0 mg/kg/day	5 mg/kg/day	50 mg/kg/day	500 mg/kg/day
Males				
Week 0	10.77 ± 0.74	10.65 ± 0.59	10.60 ± 0.82	11.10 ± 0.34
Week 1	11.02 ± 0.66	11.27 ± 0.68	10.82 ± 1.15	11.20 ± 0.29
Week 4	11.20 ± 0.40	11.62 ± 0.78	10.90 ± 1.32	11.20 ± 0.59
Week 8	11.98 ± 0.47	12.08 ± 0.88	11.18 ± 1.46	11.65 ± 0.72
Week 13	12.33 ± 0.87	12.35 ± 0.85	11.40 ± 1.62	11.90 ± 0.75
Wt. gain 0-4	0.43	0.97	0.3	0.1
Wt. gain 4-8	0.78	0.46	0.28	0.45
Wt. gain 8-13	0.35	0.27	0.22	0.25
Wt. gain 0-13	1.56	1.7	0.8	0.8
Females				
Week 0	8.50 ± 0.74	8.57 ± 1.13	8.80 ± 1.15	8.30 ± 0.82
Week 1	8.15 ± 0.80	8.40 ± 1.16	8.70 ± 1.39	8.37 ± 0.62
Week 4	8.77 ± 1.05	8.65 ± 1.29	8.82 ± 1.29	8.37 ± 0.71
Week 8	9.10 ± 1.16	8.88 ± 1.25	8.95 ± 1.40	8.38 ± 0.66
Week 13	9.30 ± 1.08	9.05 ± 1.32	9.35 ± 1.30	8.58 ± 0.61
Wt. gain 0-4	0.27	0.08	0.02	0.07
Wt. gain 4-8	0.33	0.23	0.13	0.01
Wt. gain 8-13	0.2	0.17	0.4	0.2
Wt. gain 0-13	0.8	0.48	0.55	0.28

Data taken from Table 1, p. 12, MRID 00110647.
[a]Calculated by reviewer from group means.

C. FOOD CONSUMPTION AND COMPOUND INTAKE:

1. **Food consumption**: Food consumption was not measured in this study, but was estimated by visual inspection. The study authors stated that "evaluation of ... food consumption data ... did not reveal any effects" of treatment.

2. **Compound consumption**: The test article was administered by capsule.

3. **Food efficiency**: Food efficiency was not calculated by the study authors and cannot be calculated since food consumption was not measured.

D. **OPHTHALMOSCOPIC EXAMINATION:** No treatment-related ophthalmoscopic lesions were observed in any dog.

E. **BLOOD ANALYSES:**

1. **Hematology**: Statistically significant differences in several hematological parameters were found between the treated and control groups. However, these differences were sporadic, not dose-related, not sustained over time, and not consistent between sexes. In addition, the study authors noted that all values were within "normal limits" for the testing laboratory.

2. **Clinical Chemistry**: Statistically significant differences in several clinical chemistry parameters were found between the treated and control groups. However, these differences were sporadic, not dose-related, not sustained over time, and not consistent between sexes. In addition, the study authors noted that all individual values were within the historical control ranges of the testing facility.

F. **URINALYSIS**: No dose- or treatment-related differences in any urinalysis parameter were found between the treated and control groups at any time during the study.

G. **SACRIFICE AND PATHOLOGY:**

1. **Organ weight**: At study termination, the mid- and high-dose males and the high-dose females had slightly lower final body weights (89-91% of controls, n.s.), slightly increased absolute liver weights (108-118% of controls, n.s.), and significantly increased relative liver weights ($p \leq 0.05$; 122-128% of controls). High-dose females also had increased absolute thyroid weights (118% of the control group, n.s.) which resulted in significantly ($p \leq 0.05$) increased relative thyroid weights (130% of controls). No other differences in absolute or relative organ weights were noted between the treated and control groups of either sex.

2. **Gross pathology**: Results of gross necropsy were not given in the study report or discussed by the study authors. The reviewer assumes that gross findings were unremarkable.

3. **Microscopic pathology**: No treatment-related microscopic lesions were observed in any dog. Lesions in the lungs of several treated and control animals were directly attributable to a parasitic infection.

III. **DISCUSSION and CONCLUSIONS**:

A. **INVESTIGATORS' CONCLUSIONS:** The study authors concluded that treatment of dogs with the test article for 90 days resulted in clinical signs of toxicity in males and females at 500 mg/kg/day and in increases in absolute and relative liver weights in males at 50 and 500

mg/kg/day and in females at 50 mg/kg/day. No effects were noted on body weights, food consumption, laboratory studies, and histopathologic examination. Neither a NOAEL nor a LOAEL was specified.

B. REVIEWER COMMENTS: Clinical signs of toxicity were evident in high-dose males and females beginning in week 6 and persisting until the end of the study. The main clinical finding was tremors which indicates that the chemical may be neurotoxic.

Body weight gains by most treated groups were generally less than those of the controls throughout the study, but no clear dose-related pattern was evident. Food consumption was determined by subjective observation and the study authors stated that no effects were seen. Due to the variability in weight gains and the lack of food consumption data, the reviewer does not think that the lower weight gains by the treated groups can be definitively attributed to treatment with the test article.

No treatment-related effects were observed in hematology, clinical chemistry, or urinalysis parameters, ophthalmoscopic examinations, gross necropsy, or microscopic evaluations. The study authors noted increased liver weights for the mid- and high-dose males and the mid-dose females. The reviewer thinks that the inclusion of mid-dose females is a typographical error and this should have been high-dose females. Regardless, none of the increases in absolute or relative organ weights were considered to be toxicologically significant and no histopathological correlates were found. Increased liver weight is a common adaptive response to exposure to a xenobiotic and is not considered to be adverse.

Therefore, the systemic toxicity LOAEL for permethrin in male and female dogs is 500 mg/kg/day based on clinical signs of toxicity. The systemic toxicity NOAEL is 50 mg/kg/day.

This 90-day oral toxicity study in the dog is **Unacceptable/Guideline** (upgradeable) and does not satisfy the guideline requirement for a 90-day oral toxicity study (OPPTS 870.3150; OECD 409) in dogs. Upon submission of data on purity and stability of the compound the study may be upgraded to acceptable.

C. STUDY DEFICIENCIES: Several deficiencies were noted in the conduct of this study: purity of the compound was not submitted; individual clinical observations were not reported; food consumption was not measured; several hematology, clinical chemistry, and urinalysis endpoints were not measured; not all recommended tissues were weighed or collected for microscopic evaluation. This study was conducted prior to implementation of current guidelines.

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DATA FOR ENTRY INTO ISIS

bchronic Oral Study - non-rodents (870.3150)

C code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
09701	110647	subchronic	dog	90 days	oral	capsule	5-500	0, 5, 50, 500	50	500	neurological (tremors)	

DATA EVALUATION RECORD

PERMETHRIN/109701

STUDY TYPE: 28-Day Oral Toxicity (Feeding) Range-Finding in Rats
MRID 00120267

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
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Work Assignment No. 02-05

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	Date:	APR 10 2002
Quality Assurance: Lee Ann Wilson, M.A.	Signature:	L. A. Wilson
	Date:	APR 10 2002

Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract no. DE-AC05-00OR22725.

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Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/13/2002
Signature: Joycelyn E. Stewart
Date 5/22/2002

TXR# 0050649

DATA EVALUATION RECORD

STUDY TYPE: 28-Day Oral Toxicity (Feeding) Range-Finding in Rats

PC CODE: 109701 **DP BARCODE:**D269531
SUBMISSION NO.:S504352

TEST MATERIAL (PURITY): PP557, permethrin (90.5 %), cis:trans (38:52.5)

SYNONYMS: Permethrin; 3-phenoxybenzyl (±) cis : trans-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-1-carboxylate

CITATION: Clapp. M.J.L., P.B. Banham, I.S. Chart, et al. (1977) PP557: 28 Day feeding study in rats. Imperial Chemical Industries, Limited, Central Toxicology Laboratory (location not given). Laboratory Report No. CTL/P/355, December 20, 1977. MRID 00120267. Unpublished.

SPONSOR: Wellcome Co.

EXECUTIVE SUMMARY: In a 28-day oral toxicity range-finding study (MRID 120267), PP557 (permethrin, 90.5% a.i., batch no. 17) was administered to Wistar rats (8/sex/dose) in the diet at dose levels of 0, 200, 500, 1000, 2500, 5000, or 10,000 ppm (equivalent to 0, 20, 51, 106, 267, 319, and [highest dose] mg/kg/day in males and 0, 20, 51, 76, 207, 428, and [highest dose] mg/kg/day in females.) The mg/kg equivalent of the 10,000 ppm dose could not be calculated, since all rats receiving that dose died within three days.

All rats receiving the 10,000 ppm dose died within three days. At 5000 ppm, one female and four males died by Day 18. No other deaths were observed. Surviving rats receiving 2500 ppm and above became hypersensitive during the first week of treatment and remained so throughout the study. Body weight of the 5000 ppm males was 16-22% below that of controls throughout the treatment period, and overall body weight gain of that group was decreased by 31%. Liver weight was increased 28-33% in the 2500- and 5000 ppm females. The liver/body weight ratio was increased in the 2500- and 5000 ppm males, and in all treated females except those receiving 500 ppm. The liver changes were judged to be an adaptive response. There were no treatment-related changes in food consumption, hematology, clinical chemistry, or urinalysis of any group of rats.

Under the conditions of this study, the LOAEL for PP557 in rats is 2500 ppm (267 mg/kg/day for males, 207 mg/kg/day for females) based on hypersensitivity. The NOAEL is 1000 ppm (106 mg/kg/day for males, 76 mg/kg/day for females.

This 28-day oral toxicity study in the rat is **Acceptable/Non-guideline** and satisfies its intended purpose as a range-finding study.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided. This study was completed prior to current regulatory requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1. **Test material:** PP557, permethrin (38.0% cis, 52.5% trans)

Description:	None provided
Lot/Batch #:	17
Purity:	90.5 % a.i.
Compound Stability:	Not provided
CAS # of TGAI:	52645-53-1

2. **Vehicle and/or positive control:** The test material was incorporated into the diet. No positive control was used.

3. **Test animals:**

Species:	Rats	
Strain:	Wistar-derived	
Age/weight at study initiation:	5-6 weeks	
Source:	Alderley Park	
Housing:	4/wire mesh cage	
Diet:	Stock ration (Oakes Limited, Congleton, Cheshire, England), *ad libitum*.	
Water:	Not specified, *ad libitum*	
Environmental conditions:	**Temperature:**	(Illegible)°C
	Humidity:	Not provided
	Air changes:	Not provided
	Photoperiod:	12 hrs dark/12 hrs light
Acclimation period:	One week	

B. STUDY DESIGN:

1. **In life dates** - Start: June 16, 1975 End: July 14, 1975

2. **Animal assignment**: Animals were randomly assigned by body weight to the test groups noted in Table 1.

TABLE 1: Study design					
Test Group	Conc. in Diet (ppm)	Estimated Dose to Animal (mg/kg/day)[a]		# Male	# Female
		Male	Female		
1	0	0	0	8	8
2	200	20	20	8	8
3	500	51	51	8	8
4	1000	106	76	8	8
5	2500	267	207	8	8
6	5000	319	428	8	8
7	10000	–[b]	–[b]	8	8

[a]Calculated by reviewer
[b]All animals died within 3 days

3. **Dose selection rationale:** The purpose of this study was to set dose levels of PP557 for a two-year feeding study in rats. The high dose was chosen to provide the equivalent of half the oral LD_{50} when the test material was administered in aqueous suspension. The other doses were to provide a progression below the high dose.

4. **Diet preparation and analysis:** Diet was prepared weekly by mixing appropriate amounts of test substance with the control diet. The control diet consisted of 77 parts stock ration (Oakes Limited, Congleton, Cheshire, England), 18 parts malt extract, and 5 parts corn oil. A small amount of water was added to aid mixing. The diet storage temperature was not provided. Homogeneity of the test material in the diet was not determined. Treated diet was tested for stability 37 days after preparation. Samples of treated food were not analyzed for stability and concentration during the study.

Results -
Homogeneity analysis: Homogeneity was not determined.

Stability analysis: After 37 days, the concentration of the test material in the 500-, 2500-, and 10,000-ppm diets was 90%, 96%, and 81% of nominal, respectively. The study authors could not explain the low value for the 10,000-ppm diet, but stated it was unlikely to be due to instability, as the stability over 4 weeks was subsequently verified (no data were given, but a reference was provided).

Concentration analysis: Concentration was not determined during the study.

5. **Statistics:** Body weight, food consumption, organ weight, organ/bodyweight ratio, hematological, and clinical chemistry values were analyzed using Student's t-test to compare group means with those of controls.

C. METHODS:

1. **Observations:**

1a. **Cageside observations:** Animals were inspected daily for signs of toxicity and mortality.

1b. **Clinical examinations:** Clinical examinations were not conducted.

1c. **Neurological evaluations**: Neurological evaluations were not performed.

2. **Body weight**:Animals were weighed at study start and weekly thereafter.

3. **Food consumption and compound intake:** Food consumption was determined weekly for each cage. Compound intake (mg/kg bw/day) values were calculated by the reviewer from the consumption and body weight gain data.

4. **Ophthalmoscopic examination:** Ophthalmoscopic examinations were not conducted.

5. **Hematology and clinical chemistry:** Blood was collected from four males and four females per group prior to the study start and at 28 days for hematology and clinical chemistry. It was not stated whether the animals were fasted prior to blood collection. The CHECKED (X) parameters were examined.

a. **Hematology**

x	Hematocrit (HCT)*	x	Leukocyte differential count*
x	Hemoglobin (HGB)*		Mean corpuscular HGB (MCH)*
x	Leukocyte count (WBC)*		Mean corpusc. HGB conc.(MCHC)*
	Erythrocyte count (RBC)*	x	Mean corpusc. volume (MCV)*
x	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Clotting time)		
x	(Prothrombin time)		

* Recommended for 90-day oral rodent studies based on Guideline 870.3100

b. Clinical chemistry

	ELECTROLYTES		OTHER
	Calcium		Albumin*
	Chloride		Creatinine*
	Magnesium	x	Urea nitrogen*
	Phosphorus		Total Cholesterol*
	Potassium*		Globulins
	Sodium*	x	Glucose*
	ENZYMES		Total bilirubin
	Alkaline phosphatase (ALK)*		Total protein (TP)*
	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase		Serum protein electrophores
	Lactic acid dehydrogenase (LDH)		
x	Alanine aminotransferase (ALT/also SGPT)*		
x	Aspartate aminotransferase (AST/also SGOT)*		
	Sorbitol dehydrogenase*		
	Gamma glutamyl transferase (GGT)*		
	Glutamate dehydrogenase		

* Recommended for 90-day oral rodent studies based on Guideline 870.3100

6. **Urinalysis**[1]: Urine was collected over 18 hours from one cage of males and one cage of females per group prior to the study start and at 28 days. It was not stated if the animals were fasted. The CHECKED (X) parameters were examined.

	Appearance*	x	Glucose
x	Volume*		Ketones
x	Specific gravity/osmolality*	x	Bilirubin
x	pH*		Blood/blood cells*
	Sediment (microscopic)		Nitrate
x	Protein*		Urobilinogen

* Recommended for 90-day oral rodent studies

7. **Sacrifice and pathology**: All animals that died during the study and 4 males and 4 females from treatment groups 1, 3, 5, and 6 at scheduled sacrifice were subjected to gross pathological examination and the CHECKED (X) tissues underwent histological examination. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue	x	Aorta*	xx	Brain*+
x	Salivary glands*	xx	Heart*+	x	Peripheral nerve*
	Esophagus*	x	Bone marrow*	x	Spinal cord (3 levels)*
x	Stomach*	x	Lymph nodes*	xx	Pituitary*
x	Duodenum*	xx	Spleen*+		Eyes (optic nerve)*
x	Jejunum*	xx	Thymus*+		GLANDULAR
x	Ileum*			xx	Adrenal gland*+
x	Cecum*		UROGENITAL		Lacrimal gland
x	Colon*	xx	Kidneys*+		Parathyroid*
	Rectum*	x	Urinary bladder*	x	Thyroid*
xx	Liver*+	xx	Testes*+		OTHER
	Gall bladder (not rat)*	x	Epididymides*+		Bone (sternum and/or femur)
	Bile duct (rat)	x	Prostate*	x	Skeletal muscle
x	Pancreas*	x	Seminal vesicles*		Skin*
	RESPIRATORY	xx	Ovaries*+		All gross lesions and masses*
	Trachea*	x	Uterus*+		
xx	Lung*	x	Mammary gland*		
	Nose*				
	Pharynx*				
	Larynx*				

* Recommended for 90-day oral rodent studies based on Guideline 870.3100
+ Organ weights required for rodent studies.

II. RESULTS:

A. OBSERVATIONS:

1. **Clinical signs of toxicity:** The premature decedents had persistent whole body tremors, hyperactivity, and piloerection. The surviving animals of the 5000-ppm group had piloerection, moderate body tremors, urinary incontinence, and hypersensitivity beginning during the first week and continuing throughout the study. Rats of the 2500-ppm group had piloerection and slight body tremors during the first week only, but were hypersensitive (not defined in the study) throughout treatment. Rats in the 1000-ppm group had slight tremors for the first day only. The tremors at all doses were more severe in the morning and decreased considerably by late afternoon. No abnormalities were seen in the 500- or 200-ppm groups.

2. **Mortality:** All rats receiving the 10,000 ppm dose died within three days. Of those, six males and three females died within 24 hours. In the 5000-ppm group, one female died on Day 3, three males were found dead on Day 4, and one male died on Day 18. All other animals survived to scheduled sacrifice.

3. **Neurological evaluations:** Neurological examinations were not conducted.

B. BODY WEIGHT AND WEIGHT GAIN: Body weight and body weight gain are shown in Table 2. Mean body weight of the 5000-ppm males was significantly lower than that of controls throughout the treatment period. Mean body weight of the 5000-ppm females was reduced 12% at Week 1 only. All other group body weights were comparable to controls. The

5000-ppm males also had significantly reduced body weight gain; female body weight gain was comparable to that of controls.

TABLE 2. Average body weights and body weight gain during 28 days of treatment[a]

Dose rate (ppm)	Body Weights (g±SD)					Total Weight Gain	
	Week 0	Week 1	Week 2	Week 3	Week 4	g	% of control
Male							
0	104.5±5.0	135.3±6.3	182.1±10.5	223.1±11.33	247.8±15.7	143.3±15.7	–
200	105.8±4.8	138.3±6.9	179.3±8.5	224.8±9.2	254.8±14.8	149.0±15.5	104
500	105.8±6.6	141.3±5.9	185.3±12.7	229.5±13.7	255.9±10.4	150.1±6.1	105
1000	109.1±5.0	137.1±10.4	181.4±9.8	229.5±11.6	250.3±15.9	141.1±17.6	98
2500	106.5±5.6	136.1±7.0	176.9±10.4	223.1±11.0	248.3±6.9	141.8±8.4	99
5000	108.0±7.3	113.8±6.6** (84)[b]	141.6±15.2** (78)	184.8±17.7** (83)	205.0±18.8** (83)	98.8±18.8**	69
10000	104.0±6.8	–[c]	–	–	–	–	–
Female							
0	90.5±5.8	124.5±4.9	149.9±6.9	170.8±8.1	176.4±15.9	85.9±12.1	–
200	91.8±5.8	127.8±8.1	154.4±7.3	177.3±8.3	182.4±24.2	90.6±10.1	105
500	90.4±3.6	124.6±8.1	153.9±10.3	181.3±11.0	182.4±18.0	92.0±16.9	107
1000	93.5±4.2	124.9±6.6	152.8±6.6	181.6±18.9	177.9±12.5	84.4±12.9	98
2500	98.3±4.1	125.9±8.7	158.8±7.6* (106)	180.5±12.5	190.3±33.0	97.0±32.2	113
5000	93.3±7.3	109.9±11.5** (88)	136.7±16.2	156.9±22.3	173.0±18.8	77.7±18.3	90
10000	92.3±7.2	–[c]	–	–	–	–	–

[a]Data obtained from pages 16-17 in the study report.
[b]Numbers in parentheses are percent of control value, calculated by the reviewer.
[c] All animals died.
* Significantly different ($p < 0.05$) from the control.
** Significantly different ($p < 0.01$) from the control.

C. FOOD CONSUMPTION AND COMPOUND INTAKE:

1. **Food consumption**: Overall food consumption for the 5000-ppm group was reduced by 49% in males and 19% in females compared to controls, but the study authors did not find these values to be statistically significant. The reviewer found the 49% reduction to be significantly ($p \leq 0.5$) different from the control value using a t-test for unequal variances. The large decrease in food consumption by the 5000-ppm males was likely due to inappetence and food spillage, based on the standard deviation for the value given in MRID 120267 Table 5. Females of the 2500-ppm group had a slight but statistically significant increase (5%, $p \leq 0.01$) in overall food consumption. Food consumption for all other groups was comparable to controls.

2. **Compound consumption**: Estimated compound consumption is given in Table 1. Calculations were based on the average of the week 0 and week 4 body weights.

3. **Food efficiency**: Food efficiency was not calculated. The study authors determined food utilization as mean food intake/g bw gained. There were no significant differences between the treated and control rats.

D. OPHTHALMOSCOPIC EXAMINATION: Ophthalmoscopic examinations were not conducted.

E. BLOOD ANALYSES:

1. **Hematology:** There were no treatment-related hematology findings.

2. **Clinical chemistry:** There were no treatment-related clinical chemistry findings. Blood urea levels were slightly elevated in females receiving 500 ppm and above, but were within normal limits. Plasma glucose and AST and ALT activities were normal in both sexes, although the study authors noted that one 200-ppm female showed an isolated increase in AST (individual animal values were not included in MRID 120267).

F. Urinalysis: Urinalysis was unremarkable. Urinary protein was decreased in 5000-ppm males compared to controls (0 and 3.5 mg/rat, respectively), but this difference was not biologically significant.

G. SACRIFICE AND PATHOLOGY:

1. **Organ weight:** Organ weights are given in Table 3. Liver weight was increased 28-33% in 2500- and 5000-ppm females. Relative (to body weight) liver weight was increased 17-44% in 2500 and 5000-ppm males and 8-48% in all treated groups of females except those receiving 500 ppm. Testis weight was decreased 10-13% in 2500- and 5000-ppm males, and thymus weight was decreased 31% in the 5000-ppm males. Relative kidney weight and relative brain weight were increased (24% and 20%, respectively) in 5000-ppm males. Relative weight changes in 5000-ppm females occurred in the kidneys (29% decrease), adrenals (29 % increase), and lungs (10% increase). The relative spleen weight of 2500-ppm females was decreased by 15%. None of these organ weight changes showed a dose-response relationship.

TABLE 3. Average organ weights and organ/body weight ratios after 28 days of treatment[a]

	Dose rate (ppm)						
	0	200	500	1000	2500	5000	10000
Male							
Testis[b] (g)	3.034±0.187	2.991±0.104	2.742±0.167** (90)[c]	2.803±0.132** (92)	2.743±0.064** (90)	2.650±0.074** (87)	–[d]
Thymus (g)	0.701±0.120	0.580±0.056	0.658±0.218	0.566±0.076	0.625±0.135	0.487±0.095* (69)	–
Liver/ body weight (%)	3.962±0.275	4.089±0.065	4.105±0.106	3.884+0.340	4.648±0.311* (117)	5.691±0.495** (144)	–
Kidney/ body weight (%)	0.651±0.032	0.731±0.059	0.679±0.025	0.682±0.071	0.653±0.056	0.809±0.058* (124)	–
Brain/ body weight (%)	0.756±0.037	0.762±0.056	0.759±0.042	0.742±0.037	0.740±0.025	0.905±0.054** (120)	–
Female							
Liver (g)	6.856±0.414	7.422±0.426	6.528±0.349	7.287±0.564	8.8??±0.056** (128)	9.120±0.562** (133)	–
Liver/ body weight (%)	3.584±0.090	3.874±0.211 * (108)	3.342±0.068** (93)	3.889±0.197* (109)	4.277±0.472* (119)	5.290±0.297** (148)	–
Kidney/ body weight (%)	0.731±0.038	0.782±0.091	0.743±0.080	0.729±0.038	0.740±0.109	0.522±0.014** (71)	–
Adrenal/ body weight (%)	0.028±0.005	0.033±0.004	0.025±0.009	0.032±0.007	0.029±0.010	0.036±0.002* (129)	–
Spleen/ body weight (%)	0.239±0.015	0.231±0.040	0.229±0.041	0.237±0.025	0.204±0.020* (85)	0.281±0.035	–
Lung/ body weight (%)	0.458±0.015	0.528±0.066	0.453±0.063	0.456±0.055	0.418±0.043	0.505±0.032* (110)	–

[a] Data obtained from pages 27-30 in the study report.
[b] Significance for this row calculated by reviewer using ANOVA.
[c] Numbers in parentheses are percent of control value, calculated by the reviewer.
[d] All animals died
* Significantly different ($p < 0.05$) from the control

** Significantly different ($p < 0.01$) from the control.

2. **Gross pathology** - Gross pathology revealed no treatment-related abnormalities.

3. **Microscopic pathology** - Histological findings included degenerating tubules in the kidney, peribronchial lymphoid hyperplasia, pituitary cysts, and lymphocytic foci in the liver. These findings are generally associated with the strain of rats used, and their incidence was not noticeably different from that in the controls.

III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** The study authors concluded that effects of PP557 at 5000 ppm and above were severe and sometimes resulted in death. While animals in the 2500-ppm group had body tremors in the early stages of the study, they evidently became tolerant to PP557. The increased liver weight at 2500 ppm and above was believed to be an adaptive change, and changes in the liver/bodyweight ratio in females at lower doses were due to biological variation. The study authors did not determine LOAEL or NOAEL values. The dietary levels recommended for a two-year rat study were 500, 1000, and 2500 ppm PP557, with the first two doses believed to be possible no-effect levels.

B. **REVIEWER COMMENTS:** PP557 was obviously toxic at 5000 ppm and above. Surviving rats receiving 2500 ppm and above became hypersensitive during the first week of treatment, and remained so throughout the study. While body weights of 5000-ppm males were significantly reduced throughout the treatment, this effect was not seen in the females, and males at lower doses were not affected. The large decrease in food consumption by the 5000-ppm males was likely due to inappetence and food spillage. The liver changes in rats receiving 2500 or 5000 ppm PP557 had no histological or plasma enzyme correlates, and the reviewer agrees that these were likely an adaptive change. The decreased testis weights in males receiving 500 ppm PP557 and above were likely related to their decreased body weights, as there were no histopathological correlates. Hematology, clinical chemistry, and urinalysis parameters were within normal limits. Gross and microscopic examinations revealed no treatment-related findings.

Under the conditions of this study, the LOAEL for PP557 in rats is 2500 ppm (267 mg/kg/day for males, 207 mg/kg/day for females) based on hypersensitivity. The NOAEL is 1000 ppm (106 mg/kg/day for males, 76 mg/kg/day for females.

This 28-day oral toxicity study in rats is classified as **acceptable/nonguideline** and satisfies its intended purpose as a range-finding study.

C. **STUDY DEFICIENCIES:** While this is a non-guideline study, several deficiencies were noted. Only eight animals/sex/group were used. The concentration and homogeneity of the test material in the diet were not reported. Not all the recommended hematology, clinical chemistry, or pathology parameters were analyzed. However, this study is adequate for its purpose as a range-finding study.

DATA FOR ENTRY INTO ISIS

3-Day Oral Range-Finding Study - rodents

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
109701	120267	range-finding	rat	28 days	oral	dietary	20-428	Males: 0, 20, 51, 106, 267, 319 Females: 0, 20, 51, 76, 207, 428	males: 106 females: 76	males: 267 females: 207	Hypersensitivity	

DATA EVALUATION REPORT

PERMETHRIN
(NRDC 143)

STUDY TYPE: SUBCHRONIC ORAL NEUROTOXICITY – RAT [Nonguideline]
MRID 00059066, 00070627

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-88

Primary Reviewer: Carol S. Forsyth, Ph.D., D.A.B.T.	Signature: Date:	Carol S Forsyth JAN 30 2001
Secondary Reviewers: Cheryl B. Bast, Ph.D., D.A.B.T.	Signature: Date:	Cheryl B Bast JAN 30 2001
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross JAN 30 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson JAN 30 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract DE-AC05-00OR22725

Permethrin, tech./PC Code 109701 Subchronic Oral Neurotoxicity - rodent (Nonguideline) / 1

EPA Reviewer: Linnea Hansen, Ph.D.
Toxicology Branch (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch (7509C)

Signature: Linnea Hansen
Date: February 1, 2002
Signature: Joycelyn Stewart
Date: 11 Feb 14, 2002

TXR#: 0050649

DATA EVALUATION RECORD

STUDY TYPE: Subchronic Oral Neurotoxicity, Dietary - Rat (Nonguideline)

PC CODE: (1) 109701 (NRDC 143); (2) 109704 (NRDC 149) **DP BARCODE:** D269531
(3) 127901 (S3206; (4))109301 (S5602) **SUBMISSION CODE:** S504352

TEST MATERIAL: (1) NRDC 143 (isomer ratio 45 *cis*:55 *trans*; purity 93.3%) [also (2) NRDC 149 (*cis*:*trans* 47:53, 96.5% a.i.);(3) S3206 (97.3% a.i.) and (4) S5602 (97.7% a.i.)]

SYNONYMS: (1) **NRDC 143** - 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; permethrin; PP 557; (2) **NRDC 149** - cypermethrin; (3) **S3206** - fenpropathrin; (RS)-α-cyano-3-phenoxybenzyl 2,2,3,3-tetramethylcyclopropane carboxylate and (4) **S5602** - fenvalerate; (RS)-α-cyano-3-phenoxybenzyl (RS)-2-(4-chlorophenyl)-3-methylbutyrate.

CITATIONS:

(1) Okuno, Y. and Miyamoto, J. (1976) Neurotoxic effects of some synthetic pyrethroids by short-term feeding in rats. Testing facility not given. Report No. (none), November 1976. MRID 00059066, Unpublished.

Miyamoto J

(2) Okuno, Y. and Kadota, T. (1977) No-effect level of neurotoxicity in rats by short-term feeding of NRDC 149 and NRDC 143. Testing facility not given. Report No. (none), March 1977. MRID 00070627, Unpublished.

SPONSOR: not specified in report; Shell Chemical listed on work assignment sheet

NOTE: These two MRIDs summarized findings of 8-day feeding studies on several synthetic pyrethroids on Sprague-Dawley rats. MRID 00070627 describes effects of NRDC 143 (permethrin) and NRDC 149 (cypermethrin), each at 3 dose levels. MRID 00059066 provides comparative evaluation of the same high dose animals presented in MRID 00070627, plus rats fed single dose levels of S3206 (fenpropathrin) and S5602 (fenvalerate). The evaluation of NRDC 143 in rats is presented in this DER and the evaluations of other synthetic pyrethroids, NRDC 149, S3206 and S5602, are summarized in the Appendix to this DER.

EXECUTIVE SUMMARY: In a nonguideline repeated dose oral neurotoxicity study (MRIDs 00059066 and 00070627), groups of 10-16 Sprague-Dawley rats/sex/dose were administered 700, 2000 or 6000 ppm of NRDC 143 (Lot No.: 60307, 93.3% a.i.; 45 *cis*:55 *trans*) in the diet for 8 days. Additional groups of 8-10 animals/sex served as controls. Doses for the treated groups were 57, 160 or 454 mg/kg/day, respectively, males and 58, 198 or 453 mg/kg/day, respectively,

females. Toxicity assessments were limited to clinical observations, body weights, food consumption and microscopic evaluation of the brain, spinal cord and sciatic nerve. In addition, groups of 16 Sprague-Dawley rats/sex/dose group were treated with three other synthetic pyrethroids: NRDC 149 at 500, 1500 or 3000 ppm (average daily dose levels 42, 72 or 126 mg/kg/day, males and 37, 80 or 115 mg/kg/day, females); S3206 at 1000 ppm (77 mg/kg/day, males or 58 mg/kg/day, females) and S5602 at 3000 ppm (146 mg/kg/day, males or 142 mg/kg/day, females) and were similarly evaluated.

At 6000 ppm permethrin, a total of 3 males and 2 females died during the study; one each on day 5 and the remainder on day 6. In addition, 4 moribund high-dose rats of each sex were sacrificed on day 7 and again on day 8. Clinical signs of toxicity, including severe tremor and muscle twitch, were reported in high-dose males and females beginning on day 1, but the frequency of these signs was not given. Body weight gains by the high-dose males and females (taken on day 7) were -74% and -58% lower than their respective control group levels (mean body weights were about -8.4% below controls, both sexes). Food consumption was not affected at any dietary concentration. No clinical signs of toxicity or mortalities and no effects on body weight gains occurred in the low- and mid-dose groups. Very slight or slight swelling of the sciatic nerve fibers was seen in 5/5 high-dose males and females, but only very slight swelling was observed in 6/15 control males, 5/13 control females, 1/8 low-dose males and 1/9 mid-dose females. No abnormalities were noted in the brains or spinal cords from any high-dose or control animal. Findings in the brains and spinal cords from the low- and mid-dose groups were not reported. **The LOAEL is 6000 ppm (453 mg/kg/day, females; 454 mg/kg/day, males) based on mortality, clinical signs of toxicity, decreased body weight gain and microscopic lesions in the sciatic nerve. The NOAEL is 2000 ppm (160 mg/kg/day, males; 198 mg/kg/day, females).**

Similar clinical findings (mortality, clinical signs in addition to tremor including hindlimb ataxia, erratic jumping and hypersensitivity) and neuropathology (sciatic nerve swelling, fiber disintegration and/or occasional nodal demyelination) were observed at variable incidence with NRDC 149 (3000 ppm), S3206 (1000 ppm)and S5602 (3000 ppm). Body weight/weight gain decreases were observed in all groups. Effects at 1500 ppm NRDC 149 included slight hypersensitivity, decreased body weight/weight gain and in females, very slight sciatic nerve fiber swelling and disintegration. No findings were reported at 500 ppm NRDC 149. NOAELs were not established for S3206 or S5602 in these studies.

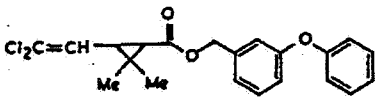
This study is classified **Unacceptable/nonguideline (upgradable)** and does not satisfy the requirements for a subchronic oral neurotoxicity study [OPPTS 870.6200 (§82-7)] in rats. These studies were performed as a comparative evaluation of neurobehavioral observations and neuropathology. The study was not conducted to fulfill a guideline requirement and a new study is not required. However, this study may be upgraded to acceptable if the deficiencies listed in the Discussion section of this review can be satisfactorily addressed.

COMPLIANCE: Signed and dated Quality Assurance, Data Confidentiality, Good Laboratory Practice Compliance and Flagging statements were not provided. This study was conducted prior to promulgation of the US EPA GLP guidelines.

I. MATERIALS AND METHODS

A. MATERIALS

1. **Test compound:** NRDC 143
   Description: not given
   CAS No.: 52645-53-1
   Lot No.: 60307
   Purity: 93.3% a.i.; isomeric ratio 45 cis:55 trans
   Contaminants: none given
   Structure:



2. **Vehicle:** The test article was suspended in corn oil, then mixed into feed and administered in the diet. The brand of feed used was not specified. No positive control was used in this study.

3. **Test animals**
   Species: rat
   Strain: Sprague-Dawley
   Age and mean weight at study initiation: about 5 weeks: males 152-202 g; females 119-143 g
   Source: Shizuoka Agricultural Cooperative Association for Experimental Animals
   Housing: aluminum cages of 35x24x20 cm
   Food: not described
   Water: not described
   Environmental conditions:
   Temperature: 24 ± 2°C
   Humidity: 60 ± 10%
   Air changes: not stated
   Photoperiod: not stated
   Acclimation period: not stated

B. STUDY DESIGN

1. **In life dates:** The study report did not indicate the in-life dates, but the following study dates were provided: Start: October 15, 1976; End: March [illegible], 1977.

2. **Animal assignment:** Animal assignment and dose selection for rats treated with NRDC 143 are shown below in Table 1 (see the Appendix for animal assignment for animals treated with NRDC 149, S3206 and S5602). The method of randomization was not described. The evaluation of NRDC 143 was conducted in two phases: first, control group 1 and the high dose animals (6000 ppm) were tested. Additional animals were subsequently tested at the lower dose levels (Control group 2 and the 700 and 2000 ppm groups) to establish a neuropathology NOAEL for NRDC 143. The duration of treatment was 8 days for all groups.

TABLE 1: Study design

Test Group	Conc. in Diet, ppm	Dose to Animal, mg/kg/day	# Male	# Female
Control 1	0	0	10	10
Control 2	0	0	8	8
Low	700	57♂, 58♀	10	10
Mid	2000	160♂, 198♀	10	10
High	6000	454♂, 453♀	16	16

Data taken from Tables 2-3, pp. 7- 8,and Table 5, p. 10, MRID 00070627.

3. **Validation of test methods:** Positive control data were not included for evaluation of the ability of the testing laboratory to conduct FOB or neuropathological studies because these evaluations were not performed.

4. **Rationale for dose selection:** A rationale for selection of the initial high dose used in this study was not provided. The high-dose of 6000 ppm caused mortality, neurobehavioral effects and microscopic lesions in rats following short-term administration in rats. Therefore, the low- and mid-dose levels were chosen in an attempt to identify a no-effect level for histopathological lesions.

5. **Preparation and analysis of test diets:** The test article was dissolved in corn oil to facilitate incorporation into the diet, but further details of the mixing procedures and frequency of preparation were not given. Analyses of the test diet for concentration, homogeneity and stability were not discussed or reported.

6. **Statistical analysis:** The data were not analyzed statistically.

C. METHODS

1. **Observations:** All animals were examined daily for clinical signs of toxicity and mortality.

2. **Body weight:** Body weights were recorded at the beginning and end of the study (day 7, as indicated in Table 4 of MRID 00070627).

3. **Food consumption and food efficiency:** Food consumption was "recorded twice" (on unspecified days) during the study and was expressed as g consumed/animal/day.

4. **Functional observational battery (FOB):** A full FOB was not conducted.

5. **Motor activity:** Motor activity was not assessed.

6. **Ophthalmology:** Ophthalmoscopic examinations were not conducted.

7. **Hematology, clinical chemistry and urinalyses:** Clinical laboratory evaluations were not performed.

8. **Sacrifice/necropsy/neurohistopathology:** Animals were sacrificed by ether anesthesia or CO_2 inhalation and perfused through the thoracic aorta with normal saline followed by 10% formalin. The brain, spinal cord and sciatic nerve were dissected out and fixed in 10% formalin. Fixed tissues were embedded in paraffin, sectioned and stained with Glee's. Histopathological examination of the nervous tissues was conducted on all control 2, low- and mid-dose animals and on 5 randomly selected animals/sex in the control 1 and high-dose group (6000 ppm).

II. RESULTS

A. CLINICAL OBSERVATIONS AND MORTALITY

In the high-dose group, a total of 3 males and 2 females died during the study; one each on day 5 and the remainder on day 6. In addition, 4 moribund rats of each sex were sacrificed on day 7 and again on day 8. Clinical signs of toxicity, including tremor and muscle twitch, were observed in high-dose males and females beginning on day 1. The number of animals affected was not reported and individual animal data were not included in the report. No clinical signs of toxicity or mortalities were observed in the low- or mid-dose groups.

B. BODY WEIGHTS AND BODY WEIGHT GAINS

Body weight and body weight gain data are given in Table 2. These data were not analyzed statistically. Body weight gains of the low- and mid-dose groups were similar to their respective control group levels. However, body weight gains by the high-dose males and females were 74% and 58% less than their respective control group levels. The final mean body weights of high dose males were reduced by 8.4% relative to controls. In high dose females, mean body weight after 8 days reduced by 8.3% relative to controls.

TABLE 2: Body weights and body weight gains (g) of male and female rats administered NRDC 143 in the diet for 8 days (final measurements taken on day 7)

Group (ppm)	Males			Females		
	Initial	Final	Wt. gain	Initial	Final	Wt. gain
Control 1	152±6	201±9	50±11	123±4	154±6	31±7
700	152±7	214±11	63±15	128±4	157±9	30±12
2000	154±6	206±7	52±11	119±7	149±6	30±7
Control 2	177±7	238±11	62±4	143±10	168±13	25±4
6000	202±10	218±20	16±13 (-74)[a]	143±6	154±5	11±6 (-58)

Data taken from Table 4, p. 9, MRID 00070627. For initial weight measurements, N = 8, control group 2; N = 10, control group 1, 700 and 2000 ppm groups and N = 16, 6000 ppm groups. For final weights, N = 8, control group 2; N = 10, control group 1, 700 and 2000 ppm groups. For the 6000 ppm group, N appears to be 9 for males and 10 for females based on mortality data, but this is unclear. It is assumed, but cannot be verified from the data provided in this report, that the four animals/sex sacrificed moribund on day 7 were not included in the terminal weighing.
[a]Number in parentheses is percent decrease relative to appropriate control group; calculated by reviewer and not analyzed statistically.

C. FOOD CONSUMPTION AND COMPOUND INTAKE

Food consumption was similar between the treated and control groups of both sexes. Compound intake for the low-, mid- and high-dose groups was 57, 160 and 454 mg/kg/day, respectively, for males and 58, 198 and 453 mg/kg/day, respectively, for females.

D. NECROPSY

Findings at gross necropsy were not reported.

E. NEUROPATHOLOGY

No abnormalities were noted in the brains or spinal cords from any high-dose or control animal. Findings in the brains and spinal cords from the low- and mid-dose groups were not reported. Very slight or slight swelling of the sciatic nerve fibers was seen in 5/5 high-dose males and females, but only very slight swelling was observed in 6/15 control males, 5/13 control females, 1/8 low-dose males, and 1/9 mid-dose females. In addition, slight nodal demyelination and very slight disintegration were each observed in 1/5 high-dose males and 1/5 high-dose females compared with none of the controls or other treated animals.

III. DISCUSSION

A. INVESTIGATORS' CONCLUSIONS: Treatment-related effects were observed in male and female rats administered 6000 ppm NRDC 143 in their diet for 8 days. Three males and two females died during treatment, beginning on day 5. An additional 4

animals/sex were sacrificed moribund on both day 7 and day 8. Clinical signs were characterized as severe and included tremors and muscle twitch. Body weight gain was markedly depressed relative to controls, although food consumption was not affected. Neuropathological evaluation of the sciatic nerve showed very slight to slight nerve fiber swelling and very slight axonal disintegration in one male and one female. There were no significant signs of toxicity at the lower doses of 700 and 2000 ppm. The study authors noted that the observed effects were seen at a lethal dose level.

B. REVIEWER'S COMMENTS: The reviewer agreed with the conclusions of the investigators. The objective of this study was to evaluate neuropathology (and neurobehavioral signs of toxicity) in rats administered NRDC 143 in the diet. Findings for NRDC 143 were consistent with the other synthetic pyrethroids examined in this study (see Appendix), suggesting a common or similar mechanism of action. Clinical signs of toxicity were observed at the highest dose beginning on the first day of treatment. Several intercurrent deaths of both males and females were considered due to test article administration.

Body weight gains were markedly depressed in high-dose males and females as compared with that of the controls. However, food consumption was not affected by treatment, indicating a direct treatment-related toxicity.

Lesions in the nervous tissues were slight and limited to the sciatic nerve of the high-dose animals. Swelling of the nerve fibers was observed in all high-dose animals, in several of the controls but in only one animal from each of the low- and mid-dose groups. This lesion was graded as very slight in some of the controls and lower dose groups, but very slight or slight in the high-dose groups. The slightly increased severity and similarity of findings in animals treated with other pyrethroids, along with the finding of very slight sciatic nerve disintegration only in the high dose group, suggest that these findings were related to treatment.

Therefore, the LOAEL for male and female rats is 6000 ppm (453-454 mg/kg/day) based on decreased body weight gain and microscopic lesions in the sciatic nerve. The NOAEL for male and female rats is 2000 ppm (160-198 mg/kg/day).

This study is classified **Unacceptable/Nonguideline (upgradable)** and was not conducted to satisfy the requirements for a subchronic oral neurotoxicity study (OPPTS 870.6200 [§82-7]) in rats. Because this is a nonguideline study, a new study is not required. However, the study may be upgraded upon submission of the information listed below in the study deficiencies section.

C. STUDY DEFICIENCIES: The studies described in these two MRIDs are nonguideline studies, therefore, a new study is not required. However, information missing from the reports included the following: (1) concentration, homogeneity and stability analyses of the test diets; (2) individual animal clinical observation data to evaluate incidence, frequency and severity of clinical signs and for chemical S3206, determine the first day of observation; (3) individual body weight data to verify "N" for the final weight determinations. In

addition, food consumption data for individual animals were not provided. This study was not conducted as a guideline neurotoxicity study and endpoints not evaluated included an FOB, motor activity and clinical chemistries.

Because this study is considered nonguideline, a new study is not required. However, this study may be upgraded to acceptable if the issues listed above in items 1 through 3 can be satisfactorily addressed.

APPENDIX
Summary of studies on synthetic pyrethroids NRDC 149, S5602 and S3206. MRIDs 00059066 and 00070627

In addition to NRDC 143 (permethrin), several other chemicals were examined in these studies: (1) NRDC 149 (cypermethrin) - 96.5% a.i., lot no. T-1; (2) S5602 (fenvalerate) - 97.7% a.i., lot no. 1-1A and (3) S3206 (fenpropathrin) - 97.3% a.i., lot no. T-1. The test animals, methods and evaluations for these chemicals are the same as those described for NRDC 143. The dates of the study reported in MRID 00059066 are cited as October 15, 1976 through November 8, 1976 (for NRDC 149 low and mid dose groups, see dates in main study report). The groups tested with NRDC 149 at all dose levels are described in MRID 00070627 (along with NRDC 143); groups tested with S5602, S3206 and the high dose of NRDC 149 are described in MRID 00059066 (along with the high dose group of NRDC 143).

Animal assignment to test groups for each chemical are shown below in Table 1a:

TABLE 1a: Study design

Test Group	Conc. in Diet, ppm	Dose to Animal, mg/kg/day	# Male	# Female
Control 1 (for NRDC Low and Mid dose only)	0	0	10	10
Control 2 (all other groups)	0	0	8	8
NRDC 149 Low	500	42♂, 37♀	10	10
NRDC 149 Mid	1500	72♂, 80♀	10	10
NRDC 149 High	3000	126♂, 115♀	16	16
S3206	1000	77♂, 58♀	16	16
S5602	3000	146♂, 142♀	16	16

Data taken from Tables 2 and 3, pp. 503-504, MRID 00070627.

NRDC 149 (Cypermethrin): There were no treatment-related effects reported at 500 ppm. At 1500 ppm, slight hypersensitivity was reported for both males and females during daily clinical observations, but the frequency of occurrence and total number of animals affected could not be determined. No individual animal data for clinical signs of toxicity were provided in the study report. Neither males nor females gained weight during treatment, vs. 44 g gain for males and 23 g gain for females in the control group. Food consumption during the treatment period was reduced by approximately -50% for both sexes. Very slight sciatic nerve fiber swelling was observed in 6/10 females, along with 3/10 females showing axonal disintegration. Because only 3/10 controls showed swelling and none showed disintegration, this is considered a possible threshold effect (the study authors did not consider this a treatment-related finding due to the low severity of the lesion). Sciatic nerve findings in males were similar to controls. There were no lesions observed in the brain or spinal cord.

At 3000 ppm, a total of 3 males and 3 females died during treatment, beginning on day 6. An additional 4/sex animals were sacrificed in moribund condition on both the 7th and 8th day.

Clinical signs, characterized as severe and reported in all animals, were first observed on day 2 for both males and females, and included hind limb ataxia, tremor, erratic jumping and hypersensitivity. It could not be determined whether these findings were observed daily in all animals. Males lost weight during treatment (weight loss of -21 g vs. gain of 62 g, controls) and females did not gain weight (-1 g loss vs 25 g gain in controls). Food consumption was also sharply decreased compared to controls (approximately -50% less than controls, both sexes). Sciatic nerve fiber swelling was observed in all animals (slight to moderate, males and very slight to moderate, females) along with nerve fiber disintegration (very slight to slight, 5/5 males and slight, 4/5 females). Slight nodal demyelination was also reported in 1/5 each male and female in MRID 00059066, but was not reported for the same animals in MRID 00070627. No findings were reported in brains or spinal cords.

S3206 (Fenpropathrin): At 1000 ppm, the only dose tested, a total of 3/16 males and 8/16 females died during treatment, beginning on day 3. An additional 4 animals/sex were sacrificed in moribund condition on day 7 and again on day 8. Clinical signs of toxicity were reported to be the same as for NRDC 149, with hindlimb ataxia, tremors, erratic jumping and hypersensitivity observed in males and females. No individual animal data were provided for clinical signs and although the report stated that all animals showed severe signs of toxicity, it could not be determined whether they were observed daily throughout treatment and whether all animals showed all signs. The summary table was also unclear as to when clinical signs were first observed in these animals, although all treatment groups reportedly showed signs by 24-48 hrs, according to the study report. Mean body weight/weight gain was reduced in males (-16.8%/-76% less than controls), but there were only slight decreases in females (body weight -4.8%/gain -12%). Food consumption was comparable to the control group. Histopathological examination showed lesions in the sciatic nerve (very slight to slight swelling in 3/5 males, slight to moderate swelling in 5/5 females) and nerve fiber disintegration in 2/5 each males and females (all slight). The only findings in control animals were very slight swelling in 3/5 males and 2/5 females. No treatment-related findings were observed in the brain or spinal cord.

S5602 (Fenvalerate): At 1000 ppm, the only dose level tested, a total of 8/16 males and 6/16 females died while on the study, with the first death observed on study day 4. An additional 4 animals/sex were sacrificed in moribund condition on day 7 and again on day 8. Clinical signs were observed beginning on day 1 and included hind limb ataxia, tremor, erratic jumping and hypersensitivity. No individual animal data were provided for clinical signs and although the report stated that all animals showed signs of toxicity, it could not be determined whether they were observed daily or whether all animals showed all signs. Mean body weight/weight gain (through day 7) were decreased in both males (-14%/-44%) and females (-4.8%/-60%). Food consumption was reduced by -50% in males and -36% in females. Microscopic findings included sciatic nerve fiber swelling in all animals (5/sex) examined (slight to moderate in females, very slight to moderate in males). In addition, very slight sciatic nerve fiber disintegration was observed in 3/5 males and 1/5 females and slight nodal demyelination in 1/5 female. Control animals showed only very slight sciatic nerve fiber swelling in 3/5 females and 2/5 males. No treatment-related findings were observed in the brain or spinal cord.

TABLE 2a: Body weights and body weight gains (g) of male and female rats administered NRDC 149, S3206 and S5602 in the diet for 8 days (final measurements taken on day 7)

Group (ppm)		Males			Females		
		Initial	Final	Wt. gain	Initial	Final	Wt. gain
Control 1	0	152±6	201±9	50±11	123±4	154±6	31±7
NRDC 149:	700	152±7	199±9	44±10	125±6	147±13	23±13
NRDC 149:	1500	154±4	155±23	1±25	122±7	119±13	-3±13
Control 2	0	177±7	238±11	62±4	143±10	168±13	25±4
NRDC 149:	3000	162±8	140±9	-21±9	149±4	148±5	-1±3
S3206:	1000	183±10	198±21	15±16	138±7	160±9	22±10
S5602:	3000	169±10	204±10	35±12	150±6	160±8	10±6

Data taken from Table 2, p. 9, MRID 00059066 and Table 4, p. 9, MRID 00070627. For initial weight measurements, N = 8, control group 2; N = 10, control group 1, 700 and 2000 ppm groups for NRDC 149; and N = 16, NRDC 3000 ppm, S3206 1000 ppm and S5602 3000 ppm groups. For final weights, N = 8, control group 2; N = 10, control group 1, 500 and 1500 ppm groups for NRDC 149. For the remaining treated groups, N appears to be 10 males and 11 females for NRDC 3000 ppm; 9 males and 4 females for S3206 and 6 males and 6 females for S5602, but this is unclear. It is assumed, but cannot be verified from the data provided in this report, that the four animals/sex sacrificed moribund on day 7 were not included in the terminal weighing for these groups.

[1]Number in parentheses is percent decrease relative to appropriate control group; calculated by reviewer and not analyzed statistically.

DISCUSSION: The reviewer agreed with most of the conclusions of the investigators, with the exception that the reviewer considered the increased incidence of sciatic nerve swelling and disintegration in females at 1500 ppm NRDC 149 to be a possible threshold effect level. The synthetic pyrethroids tested in these studies all show similar neurobehavioral and neuropathological effects. The dose levels tested (except for the low and mid dose NRDC 149 groups) resulted in significant mortality and moribundity for all chemicals. Mortality was comparable between males and females for S5602, whereas mortality in females was higher from S3206 and higher in males from NRDC 149. Clinical signs of toxicity were the same for all chemicals, but the frequency of these findings throughout the study in each animal could not be determined from the data presented in this report. The most pronounced effect on body weight was observed in the 3000 ppm NRDC 149 groups, although decreased weight gain was observed in all other treated groups. Only S3206 showed no significant effect on food consumption. Disintegration and swelling of the sciatic nerve fibers and occasional nodal demyelination, were observed for all chemicals, with NRDC 149 and S5206 showing the most pronounced effects. S3206 had more pronounced neuropathological effects in females, while other compounds tested did not appear to show sex-related differences in terms of severity or incidence. The neuropathology correlates with the types of clinical signs that were observed.

The study deficiencies for these reports are listed in the Discussion section of the main study report.

DATA FOR ENTRY INTO ISIS

ıbchronic (8 day) Oral Neurotoxicity Study - rodents (nonguideline)

PC code	MRID #s	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
09701	00059066, 00070627	subchronic neurotoxicity	rat	8 days	oral	diet	57-454	57, 160, 454 ♂; 58, 198, 453 ♀	160	453	body wt decr, peripheral nerves	Toxicity

DATA EVALUATION REPORT

PERMETHRIN
(PP 557)

STUDY TYPE: ACUTE DELAYED NEUROTOXICITY – HEN [OPPTS: 870.6100 (81-7)]
MRID 00112933

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-87

Primary Reviewer: Carol S. Forsyth, Ph.D., D.A.B.T.	Signature: Date:	Carol S Forsyth JAN 3 0 2001
Secondary Reviewers: Cheryl B. Bast, Ph.D., D.A.B.T.	Signature: Date:	Cheryl B Bast JAN 3 0 2001
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross JAN 3 0 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson JAN 3 0 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract

Permethrin

Delayed Neurotoxicity [870.6100(§81-7)]

EPA Reviewer: Yung G. Yang, Ph.D. Yung G. Yang, Date 10/31/2001
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Joycelyn Stewart, Date 1/24/2002
Toxicology Branch, HED (7509C)

TXR# 0050649

DATA EVALUATION RECORD

STUDY TYPE: Acute Delayed Neurotoxicity – Hen; [OPPTS 870.6100 (§81-7)]

DP BARCODE: D269531
PC CODE: 109701

SUBMISSION CODE: S504352
TOX CHEM NO: 652BB

TEST MATERIAL: Permethrin (isomer ratio 36 cis:58.9 trans; purity 94.9%)

SYNONYMS: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; NRDC 143; PP 557

CITATION: Ross, D.B., Roberts, N.L., Cameron, M.M., Prentice, D.E., and Cooke, L. (1977) Examination of permethrin (PP 557) for neurotoxicity in the domestic hen. Huntingdon Research Centre, Huntingdon, Cambridgeshire. Report No. (none), 30 September 1977. MRID 00112933, Unpublished.

SPONSOR: Imperial Chemical Industries Ltd., Central Toxicology Laboratory, Alderley Park, Nr. Macclesfield, Cheshire

EXECUTIVE SUMMARY: In a delayed neurotoxicity study (MRID 00112933), a group of 15 domestic hens were administered 15 mL of permethrin (Lot No.: not given; isomer ratio 36 cis:58.9 trans, 94.9% a.i.) by oral gavage. Based on a specific gravity of 1.2, mean body weight on study day 0, and not correcting for purity of the test article, the dose to the hens was approximately 9000 mg/kg. Additional groups were given water as the negative control (n = 10) or 500 mg TOCP/kg as the positive control. All birds were given a single oral dose on study day 0 and observed for 21 days. Birds in the permethrin and negative control groups were redosed on study day 21 and observed for an additional 21 days. Prior to redose, birds in the permethrin group were protected with 10 mg atropine/kg and 50 mg 2-PAM/kg given by intramuscular injection.

Toxicity assessments were limited to clinical observations, assessment of ataxia, measurements of body weights and food consumption, and microscopic evaluation of the brain, spinal cord, and sciatic nerve. Acetylcholinesterase and neurotoxic esterase activities were not measured.

No treatment-related clinical signs of toxicity and no effects on body weights or food consumption were observed in birds administered permethrin. Ataxia was not seen in birds treated with the test article and no treatment-related lesions were observed on microscopic examination of the nervous tissues.

Following treatment with TOCP, clinical signs and neurohistopathological lesions indicative of delayed neuropathy were observed in these birds.

Therefore, under the conditions of this study, oral administration of permethrin does not produce delayed neuropathy in the hen.

This study is classified **Acceptable/Guideline** and does satisfy the requirements for a delayed neurotoxicity study [OPPTS 870.6100 (§81-7)] in hens. Although a major deficiency was that AChE and NTE activities were not measured, the study is considered sufficient for determining the potential of permethrin to produce delayed neurotoxicity in the hen. This study was conducted prior to implementation of current guidelines.

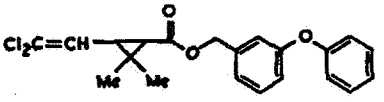
COMPLIANCE: Signed and dated Quality Assurance, Data Confidentiality, Good Laboratory Practice Compliance, and Flagging statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound: permethrin

Description: brown solid, becoming liquid at warm room temperature
CAS No.: 52645-53-1
Lot No.: not given
Purity: 94.9% a.i.; isomeric ratio 36 cis:58.9 trans
Contaminants: none given
Structure:



2. Vehicle and/or positive control

The test article was administered as supplied; no vehicle was used. Distilled water was the negative control. The positive control used in this study was TOCP in corn oil.

3. Test animals

Species: domestic hen
Strain: not specified
Age and mean weight at study initiation: 1-1.5 years: 1650-2260 g
Source: Stanley Brown Ltd., Bucks
Housing: Birds were housed in floor pens measuring 1.5 m x 1.5 m in a controlled environment poultry building.

Food: A commercial laying ration was available *ad libitum.*
Water: Water was available *ad libitum.*
Environmental conditions:
Temperature: 17°C
Humidity: not stated
Air changes: not stated
Photoperiod: 17 hours light/ 7 hours dark
Acclimation period: not stated

B. STUDY DESIGN

1. In life dates

Start: April 1977; End: June 1977

2. Animal assignment and dosing protocol

Animal assignment and dose selection are listed in Table 1. The method of randomization was not described. The amount of the test article administered was stated as the maximum practicable dose volume. An actual dose was not calculated. However, based on a specific gravity of 1.2, mean body weight on study day 0 of 1940 g, and not correcting for purity of the test article, the dose to the hens was approximately 9000 mg/kg.

All birds were given a single oral dose on study day 0 and observed for 21 days. Birds in the permethrin and negative control groups were redosed on study day 21 and observed for an additional 21 days. Prior to redose, birds in the permethrin group were protected with 10 mg atropine/kg and 50 mg 2-PAM/kg given by intramuscular injection.

TABLE 1. Study design

Group	Dose Volume	No. of Animals
Negative Control (water)	15 mL	10
Permethrin	15 mL	15
Positive Control (TOCP)	500 mg/kg	5

Data taken from text, p. 7, MRID 00112933.

3. Validation of test methods

A positive control group was run concurrently with the test article group to show the ability of the testing facility to detect delayed neuropathy in the hen.

4. Rationale for dose selection

The dose of the test article was stated as the maximum practicable dose volume. An actual dose to the animals based on body weight was not calculated or stated.

5. Dose solution preparation and analyses

The reviewer assumes that the test article was administered as supplied. No preparation was described.

6. Statistical analysis

The data were not analyzed statistically.

C. METHODS

1. Observations

All animals were examined daily for clinical signs of toxicity and mortality.

2. Body weight

Body weights were measured on study days 0, 7, 14, 21, 28, 35, and 42.

3. Food consumption and food efficiency

Food consumption was measured over 7-day periods. Food efficiency was not calculated.

4. Neurotoxicity assessment

Each bird was moved several times daily to assess muscle coordination. The type of forced activity was not described. Ataxia was graded according to the following scale:

1. doubtful; slight incoordination, not always present.
2. slight but definite incoordination.
3. frequent incoordination or stumbling.
4. more severe incoordination or "drunken gait".
5. shuffling on hocks and difficulty in maintaining upright stance.
6. bird stands for short period only, normally moves by shuffling on hocks.
7. very weak limb movements, reflexes markedly affected.
8. total inability to rise and walk.

5. Biochemical measurements

Acetylcholinesterase (AChE) and neurotoxic esterase (NTE) activities were not measured in this study.

6. Sacrifice/necropsy/neurohistopathology

Birds in the test article and negative control groups were sacrificed after the second 21-day observation period; birds in the positive control group were sacrificed after the first observation period. Ten hens in the permethrin group and all positive and negative control hens were killed, examined grossly, and tissues processed for histological examination. The brain, spinal cord, sciatic nerve, and leg muscle were removed and fixed in 10% buffered formalin. Fixed tissues were embedded in paraffin and sectioned. Brain and spinal cord sections were stained with either H&E or by the Glees Marsland technique. Muscle sections were stained with H&E.

II. RESULTS

A. CLINICAL OBSERVATIONS AND MORTALITY

All hens survived to scheduled sacrifice. Following the initial dosing, no clinical signs of toxicity were observed in the permethrin or negative control groups. Shortly after redosing, birds in the permethrin group were quiet, lethargic, and showed signs of slight muscular incoordination. Hens in the positive control group appeared lethargic after dosing and showed some evidence of incoordination. Incidence rates of these clinical signs were not given and individual animal observation data were not included in the report.

C. BODY WEIGHTS AND BODY WEIGHT GAINS

Body weight and body weight gain data are given in Table 2. These data were not analyzed statistically. Absolute body weights and body weight changes for the permethrin and negative control groups were considered within normal limits. In the negative control group, bullying was associated with a loss of body weight during the first 7 days of the study and three birds were moved to a separate pen for the remainder of the study. Marked weight loss was observed in the positive control animals.

TABLE 2: Body weights and body weight gains (g) of hens administered permethrin or a positive control

Day of Study	Negative Control (water)	Permethrin	Positive Control (TOCP)
0	1948	1940	1925
7	1846	2000	1908
14	1902	1996	1853
21	1932	1990	1801
28	1938	1996	–
35	1915	1999	–
42	1920	2027	–
Wt. change 0-21	-16	50	-124
Wt. change 21-42	-12	37	–

Data taken from Table 3, p. 12, MRID 00112933.

D. FOOD CONSUMPTION

No treatment-related effects on food consumption were observed during the study. The data were somewhat variable due to an appreciable amount of spillage.

E. NEUROTOXICITY ASSESSMENT

No signs of ataxia were observed in birds treated with the test article or in the negative control birds. Hens in the positive control group showed signs of ataxia ranging from slight muscular incoordination to difficulty in standing. Signs in these animals were first observed on study day 11 and increased in severity throughout the remainder of the study.

F. BIOCHEMICAL MEASUREMENTS

Acetylcholinesterase (AChE) and neurotoxic esterase (NTE) activities were not measured.

G. NECROPSY

No treatment-related abnormalities were seen in any animal at gross necropsy.

H. NEUROPATHOLOGY

Occasional myelinophages were observed mainly in the lumbar spinal cord from 5/10 birds in each of the permethrin and negative control groups. This was considered a normal finding in birds of this age. One bird treated with permethrin had swollen and

vacuolated neurons in the thalamus and brain stem that were considered artifacts of preparation. Perivascular edema was observed in the brains from one negative control animal and one animal in the permethrin group. No abnormalities were seen in the sciatic nerves from hens treated with the test article or negative control.

Degenerative changes were observed in the spinal cord and sciatic nerve of birds in the positive control group.

III. DISCUSSION

A. DISCUSSION

The objective of this study was to determine whether permethrin causes delayed neurotoxicity in the domestic hen. An actual dose to the animals was not given, but the reviewer calculated the dose based on the available information to be approximately 9000 mg/kg. This greatly exceeds the limit dose required in Subdivision F guidelines and was repeated for a second 21-day observation period.

Body weights and body weight gains were not affected by treatment with the test article. Food consumption data were highly variable due to spillage by the animals, but no treatment-related trends were apparent. Clinical signs of toxicity were observed following the second treatment, and may have been due to the atropine and 2-PAM rather than a direct effect of the test article. These signs were observed shortly after treatment and are not considered to be indicative of delayed neuropathy.

Ataxia was not seen in birds treated with the test article and no treatment-related lesions were observed on microscopic examination of the nervous tissues.

TOCP was used as a positive control in this study. Clinical signs and neurohistopathological lesions indicative of delayed neuropathy were observed in these birds. Therefore, the testing facility should have been able to detect neuropathy induced by the test article.

Therefore, under the conditions of this study, oral administration of permethrin does not produce delayed neuropathy in the hen.

This study is classified **Acceptable/Guideline** and does satisfy the requirements for a delayed neurotoxicity study [OPPTS 870.6100 (§81-7)] in hens.

B. STUDY DEFICIENCIES

AChE and NTE activities were not measured and nervous tissues were not fixed *in situ* by perfusion. However, the dose administered greatly exceeded the limit dose and was administered on two separate occasions without resulting in clinical signs or ataxia. Therefore, the study is considered sufficient for determining the delayed neurotoxicity potential of permethrin.

EPA Reviewer: Yung Yang, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/21/2002
Signature: Joycelyn Stewart
Date 6/5/2002

DATA EVALUATION RECORD

TXR#: 0050649

STUDY TYPE: ***In vitro* Bacterial Gene Mutation** *Salmonella typhimurium* / mammalian activation gene mutation assay; OPPTS 870.5100[2] [§84-2]

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Compound 33297 (Permethrin; 95.7% a.i.)

SYNONYMS: FMC 33297

CITATION: Simmon, V.F. (1976) *In vitro* microbiological mutagenicity study of an FMC corporation compound. Stanford Research Institute, Menlo Park, California 94025. SRI Project LSC-4768, January 1976. MRID 110646 (Other MRID# 43729, 57102, 70582 for the same study). Unpublished.

SPONSOR: FMC Corporation, Agricultural Chemical Division, 100 Niagara Street, Middleport, New York 14105

EXECUTIVE SUMMARY: In a reverse gene mutation assay in bacteria (MRID 110646, 43729, 57102 or 70582), strains TA98, TA100, TA1535, TA1537 and TA1538 of *S. typhimurium* and strain WP2(her') of *E. coli* were exposed to Compound 33297 (permethrin, 95.7% a.i., batch/lot # not provided) in an unspecified solvent at concentrations of 1, 50, 100, 250, 500 and 1000 µg/plate in the presence and absence of mammalian metabolic activation (S9-mix). The S9-fraction was obtained from Aroclor 1254 induced male mouse liver.

Compound 33297 was tested up to cytotoxic concentrations with a reduction in the number of spontaneous revertants per plate seen at 1000 µg/plate in several strains. No increases in the average number of revertants per plate over the solvent control values were seen in any tester strain at any test material concentration with or without S9-mix. Any given tester strain was treated with a positive control that was either active with or without S9-mix but no strain was treated with positive controls for both activation conditions. No historical control values were provided. The components of the S9-mix were given but the percent S9-fraction in the mix and

[2] 870.5100 Reverse Mutation Assay [WP2 and WP2 uvrA; S. typhimurium TA 98, TA100, TA1535, TA1537

the concentrations of cofactors were not given. No confirmatory assay was conducted. Where available, the positive control values were appropriate in the corresponding strains. **Under the condition of this study, compound 33297 was not mutagenic either with or without S9-mix.**

This study is classified as **Unacceptable/Guideline.** It does not satisfy the guideline requirements for Test Guideline OPPTS 870.5100[1]; OECD 471 for *in vitro* mutagenicity (bacterial reverse gene mutation) data. Individual plate counts with means and standard deviations were not provided. In addition, assay acceptance and evaluation criteria were not provided, the solvent was not specified, no signed and dated GLP statement was provided and other than the purity, the test material was not further characterized. Because of the number of omissions, the age of the study and likely unavailability of original data and the short time required to conduct an Ames Test, the study should be repeated.

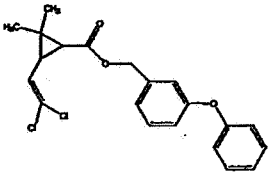
COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test material: Compound 33297, Permethrin

Description:	Not provided
Lot/Batch #:	Not provided
Purity:	95.7% a.i.
CAS # of TGAI:	52645-53-1
Solvent Used:	Not specified



2. Control materials:

Negative:	None
Solvent (final conc'n):	Not specified / 100 µL/plate
Positive:	Nonactivation:
	Sodium azide ____ µg/plate
	2-Nitrofluorene ____ µg/plate
	9-Aminoacridine 100 µg/plate TA1537
	Other:
	MNNG 2 µg/plate TA1535, WP2(her')
	Activation:
	2-Aminoanthracene (2-anthramine) ____ µg/plate usually all strains
	Other:
	4-*o*-Tolylazo-*o*-toluidine 25 µg/plate TA98, TA1538
	Benzo(*a*)pyrene 20 µg/plate TA100

3. **Activation: S9 derived from**

x	induced	x	Aroclor 1254		Rat	x	Liver
	non-induced		Phenobarbitol	x	Mouse		Lung
			None		Hamster		Other
			Other		Other		

Describe S9 mix composition: Unspecified concentrations of the following cofactors were added–$MgCl_2$, KCl, glucose-6-phosphate, TPN and sodium phosphate (pH 7.4).

4. **Test organisms:** ***S. typhimurium*** **strains**

	TA97	x	TA98	x	TA100		TA102		TA104
x	TA1535	x	TA1537	x	TA1538	x	list any others: *E. coli* WP2(her)		

Properly maintained?	x	Yes		No
Checked for appropriate genetic markers (*rfa* mutation, R factor)?	x	Yes		No

5. **Test compound concentrations used:**

Mutagenicity assay:
Nonactivated and activated conditions: 1, 50, 100, 250, 500, 1000 µg/plate in all strains.
The number of replicates was not provided.

B. TEST PERFORMANCE:

1. **Type of *Salmonella* assay:**
x standard plate test
__ pre-incubation (__ minutes)
__ "Prival" modification *(i.e. azo-reduction method)*
__ spot test
__ other

2. **Protocol:** A standard plate test was conducted by adding, in order, 2 mL of molten 0.6% agar containing 0.05 mM histidine or tryptophan and 0.05 mM biotin, 0.1 mL of an overnight culture of tester bacteria, 0.15 mL of S9-mix for tests with activation, and up to 100 µL of test substance solution or 100 µL of solvent to a test tube. After mixing, the contents of each tube were poured onto minimal agar plates, the top agar allowed to solidify, the plates incubated at 37°C for two days and the number of revertant colonies then counted.

3. **Statistical analysis:** No statistical analysis was performed.

4. **Evaluation criteria:** The testing laboratory's evaluation criteria were not described.

II. REPORTED RESULTS: 320

A. **PRELIMINARY CYTOTOXICITY ASSAY:** No preliminary cytotoxicity assay was reported.

B. **MUTAGENICITY ASSAY:** Six concentrations of Compound 33297 ranging from 1 to 1000 µg/plate were tested in the mutagenicity assay. The number of replicate plates was not given and only the average number of revertants per plate was reported. There was no evidence of a mutagenic effect in any tester strain at any test substance concentration with or without S9-mix. Based on the summary table provided with the study (and included here as Appendix Table 1, MRID 110646, p.7), positive controls in the absence of S9-mix were used with strains TA1535, TA1537 and WP2(her') only and positive controls in the presence of S9-mix were used with strains TA98, TA100 and TA1538 only. The control values presented appear appropriate for the respective strains.

III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** The investigators concluded that Compound 33297 was not mutagenic in any of the tester strains at any concentration up to 1000 µg/plate, with or without S9-mix.

B. **REVIEWER COMMENTS:** The reviewer agrees with the investigators' conclusion of a negative response based on the information provided; however, this study was completed in January 1976 and does not meet many of the current acceptance criteria. Individual plate counts with means and standard deviations were not provided, assay acceptance and evaluation criteria were not provided, other than the purity, the test material was not further characterized, the solvent was not specified, no signed and dated GLP statement was provided and any given tester strain was treated with a positive control either active with or without S9-mix but no strain was treated with positive controls for both activation conditions. No historical control values were provided but the study was done near the start of Ames Test usage and few historical data may have been available. The components of the S9-mix were given but the percent S9-fraction in the mix and the concentrations of cofactors were not given. No confirmatory assay was conducted.

The study is unacceptable as presented but would be acceptable if the testing laboratory could provide the missing information. Due to the age of the study, the missing data may no longer be available and a repeat assay is more practical.

C. **STUDY DEFICIENCIES:** Study deficiencies were described in the Reviewer Comments section. Most of the deficiencies are likely data presentation deficiencies rather than experimental deficiencies; however, because of the number of omissions, the age of the study and the short time required to conduct an Ames Test, the study should be repeated.

APPENDIX

(MRID 110646)

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Tox Review # 50649

Page 185 is not included in this copy.

Pages _____ through _____ are not included in this copy.

The material not included contains the following type of information:

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_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

DETAILED EXECUTIVE SUMMARY

PERMETHRIN/109701

SALMONELLA/ESCHERICHIA/MAMMALIAN ACTIVATION GENE MUTATION ASSAY [OPPTS 870.5100[1] (§84-2)]
MRID 00110646

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Health Sciences Research Division
Oak Ridge National Laboratory*
Oak Ridge, TN 37831
Task Order No. 02-05

Primary Reviewer: B.L. Whitfield, Ph.D.	Signature: Date:	B L Whitfield MAR 2 6 2002
Secondary Reviewers: Cheryl B. Bast, Ph.D., D.A.B.T.	Signature: Date:	Cheryl B Bast MAR 2 6 2002
Robert H. Ross, Group Leader	Signature: Date:	Robert H. Ross MAR 2 6 2002
Quality Assurance: LeeAnn Wilson, M.A.	Signature: Date:	L. A. Wilson MAR 2 6 2002

Disclaimer

This review may have been altered subsequent to the contractor's signature above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

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[1]870.5100 - Reverse mutation *E. coli* WP2 and WP2uvrA
870.5140 - Gene mutation *Aspergillus nidulans*

EXECUTIVE SUMMARY

PERMETHRIN

STUDY TYPE: DEVELOPMENTAL TOXICITY - RABBIT
[OPPTS 870.3700 (§83-3B)]
MRID NO. 92142091

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order 01-85

Primary Reviewer:
Carol S. Forsyth, Ph.D., D.A.B.T.

Signature: Carol S Forsyth
Date: JAN 10 2001

Secondary Reviewers:
Cheryl B. Bast, Ph.D., D.A.B.T.

Signature: Cheryl B Bast
Date: JAN 10 2001

Robert H. Ross, M.S., Group Leader

Signature: Robert H. Ross
Date: JAN 10 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle. LLC., for the U.S. Department of Energy under contract number DE-AC05-96OR22464.

PERMETHRIN

Developmental Toxicity Study [OPPTS 870.3700 (83-3b)]

EPA Reviewer: Yung G. Yang, Ph.D. Date: 12/10/2001
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Date: 1/28/2002
Toxicology Branch, HED (7509C)

TXR# 0050649

DATA EVALUATION RECORD

This is an updated executive summary of MRID 92142091, HED Doc. No. 008344. The final conclusion of the study has not been changed.

STUDY TYPE: Developmental Toxicity - Rabbit; OPPTS 870.3700 (§83-3b)

DP BARCODE: D269531
PC CODE: 109701

SUBMISSION CODE: S504352
TOX CHEM NO: 652BB

TEST MATERIAL: Permethrin (92.5% a.i.; 32.3 cis:60.2 trans isomers)

CHEMICAL NAME: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; permethrin; NRDC 143; PP557

CITATION: Richards, D., Banham, P.B., and Kilmartin, M. (1980). Permethrin: Teratogenicity study in the rabbit. Central Toxicology Laboratory, Alderley Park Macclesfield, Cheshire, UK. Study No. RB0138, Report No. CTL/P/523. August, 1980. MRID 92142091. Unpublished.

SPONSOR: ICI Corporation

EXECUTIVE SUMMARY: In a developmental toxicity study (MRID 92142091), presumed pregnant Dutch rabbits were administered 0, 600, 1200, or 1800 mg/kg/day of permethrin (92.5% a.i.; 32.3 cis:60.2 trans isomers; Batch No. D108136E) by gavage on gestation days (GD) 6-18, inclusive. The number of does mated for each group was 19, 21, 20, and 23, respectively. The vehicle was 0.5% aqueous Tween 80. On GD 29, all surviving does were sacrificed and all fetuses were weighed and examined for external malformations/variations. Approximately one-half of the fetuses was processed for skeletal examination and the remaining one-half was fixed and examined for visceral anomalies. Maternal food consumption was not measured.

A total of 0, 5, 5, or 4 does died or were sacrificed moribund in the control, low-, mid-, or high-dose groups, respectively. Due to the lack of a dose-response, the deaths could not be definitively attributed to test article administration. Clinical signs of toxicity included body tremors observed in 5 of the high-dose animals only. Little or no feces or urine was noted on at least one occasion for 2/19 (11%), 4/21 (19%), 6/20 (30%), and 8/23 (35%) animals in the control, low-, mid-, and high-dose groups, respectively.

Absolute body weights were similar between the treated and control groups throughout the study. However, after examining the replotted body weight data, there was a sharp drop in weight for

the low, mid, and high dose groups after day 6 and only a slight drop for the control that was noticeable after day 12. Body weight gain by the low-, mid-, and high-dose groups was 21%, 50%, and 9%, respectively, of the control level during GD 0-18 with statistical significance ($p \leq 0.05$) attained for the low- and high-dose groups. During the post-dosing interval, recovery of body weights was noted for the low- and mid-dose groups, but not for the high-dose group.

The maternal toxicity LOAEL is estimated to be <600 mg/kg/day based on decreased body weight gain. The maternal toxicity NOAEL is not identified.

The number of live fetuses and mean litter size was decreased for all dose groups compared to the control group (110(15), 80(13), 69(14), and 72(13) for control, low-, mid-, and high-dose groups, respectively). However, no dose-response was evident or statistical significance noted.

Post-implantation loss was significantly ($p \leq 0.05$) increased in the mid- and high-dose groups to 155% and 248% of the control level. Correspondingly, the number of early and late resorptions were higher in these groups as compared to the control group values (statistical significance was not reported). Mean fetal body weights in the high-dose group were slightly (-9%; n.s.) less than that of the controls and attributed to maternal body weight decreases. No dose-related or statistical differences were observed between the treated and control groups for number of fetuses/litter or mean gravid uterine weights.

No treatment-related external or visceral fetal malformations/variations were noted. In the mid- and high-dose groups, reduced ossification of the fore- and hind-limbs was indicated by slightly (n.s.) greater ossification scores as compared with the controls. Mean scores for the control, low-, mid-, and high-dose groups were 1.92, 1.99, 2.00, and 2.25, respectively, for the forelimb and 1.65, 1.56, 1.89, and 1.90, respectively, for the hindlimb.

Therefore, the developmental toxicity LOAEL is 1200 mg/kg/day based on increased post-implantation loss, greater numbers of early and late resorptions and an equivocal decrease in ossification of the fore- and hind-limbs. The developmental toxicity NOAEL is 600 mg/kg/day.

This study is classified as **Acceptable/Guideline** and does satisfy the guidelines for a developmental toxicity study [OPPTS 870.3700 (83-3b)] in rabbits. It should be noted that this study was conducted prior to implementation of the current guidelines. Because the mid- and high-doses exceeded the limit dose of 1000 mg/kg/day, the study is considered sufficient for determining the developmental toxicity potential of permethrin in the rabbit even though a maternal toxicity NOAEL was not identified.

DETAILED EXECUTIVE SUMMARY

PERMETHRIN/109701

SALMONELLA/ESCHERICHIA/MAMMALIAN ACTIVATION GENE MUTATION ASSAY [OPPTS 870.5100[1] (§84-2)]
MRID 00054738

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Health Sciences Research Division
Oak Ridge National Laboratory*
Oak Ridge, TN 37831
Task Order No. 02-05

Primary Reviewer: B.L. Whitfield, Ph.D.	Signature: Date:	B L Whitfield MAR 2 6 2002
Secondary Reviewers: Cheryl B. Bast, Ph.D., D.A.B.T.	Signature: Date:	Cheryl B Bast MAR 2 6 2002
Robert H. Ross, Group Leader	Signature: Date:	Robert H. Ross MAR 2 6 2002
Quality Assurance: LeeAnn Wilson, M.A.	Signature: Date:	L. A. Wilson MAR 2 6 2002

Disclaimer

This review may have been altered subsequent to the contractor's signature above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

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[1]870.5100 - Reverse mutation *E. coli* WP2 and WP2uvrA
870.5140 - Gene mutation *Aspergillus nidulans*
870.5250 - Gene mutation *Neurospora crassa*

EPA Reviewer: Yung Yang, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/22/2002
Signature: Joycelyn B Stewart
Date 6/3/2002

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: ***In vitro* Bacterial Gene Mutation** *Salmonella typhimurium* / mammalian activation gene mutation assay [OPPTS 870.5100[2] (§84-2)]

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin (95.1% a.i., 42.1% cis and 57.9% trans)

SYNONYMS: FMC 33297

CITATION: Longstaff, E. (1976) Permethrin: Short-term predictive tests for carcinogenicity: Results from the Ames test. Report No. CTL/P/301, MRID 00054738. Unpublished

SPONSOR: ICI Plant Protection Division

EXECUTIVE SUMMARY: In a reverse gene mutation assay in bacteria (MRID 54738), strains TA98, TA100, TA1535 and TA1538 of *S. typhimurium* were exposed to Permethrin (95.1% a.i., batch/lot # not provided) in DMSO at concentrations of 4, 20, 100, 500 and 2500 µg/plate in the presence and absence of mammalian metabolic activation (S9-mix). The S9-fraction was obtained from Aroclor 1254 induced rat liver.

Permethrin was tested up to 2500 µg/plate but the justification for this upper dose was not given. No significant increases (defined as a two-fold or greater increase) in the average number of revertants per plate over the corresponding solvent control values were seen in any tester strain at any test material concentration with or without S9-mix; however, a number of the current acceptance criteria were not met. **Under the condition of this study, compound 33297 was not mutagenic either with or without S9-mix.**

This study is classified as **Unacceptable/Guideline.** It does not satisfy the guideline requirements for Test Guideline OPPTS 870.5100[1]; OECD 471 for *in vitro* mutagenicity

[2] 870.5100 Reverse mutation assay E. coli WP2 and WP2 uvrA S. typhimurium TA 97, TA98, TA100, TA1535, TA1537
870.5140 Gene mutation in Aspergillus nidulans
870.5250 Gene mutation in Saccharomyces cerevisiae

(bacterial reverse gene mutation) data. No signed and dated GLP statement was provided, individual plate counts were not reported, the composition of the S9-mix was not detailed, the batch/lot number of the test material was not given and no historical control values were provided. Four, rather the recommended five strains were used and one strain which was used, TA1538, is not recommended. In addition, the information normally included in the Citation section such as the testing laboratory, laboratory study number and study completion date were not provided. The solvent and positive control values were appropriate in the corresponding strains. Because of the number of omissions, the age of the study and likely unavailability of original data and the short time required to conduct an Ames Test, the study should be repeated.

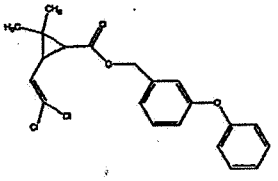
COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test material: Permethrin

Description:	Colored liquid
Lot/Batch #:	Not provided
Purity:	95.1% a.i.
CAS # of TGAI:	Not provided (52645-53-1 from ChemIDplus)
Solvent Used:	DMSO



2. Control materials:

Negative: None

Solvent (final conc'n): DMSO / 100 µL/plate

Positive: Nonactivation:

Sodium azide ____ µg/plate

2-Nitrofluorene 4, 20, 100, 500 µg/plate TA98 (not 4 µg/plate), TA1538

9-Aminoacridine ____ µg/plate

Other (list):

1,3-Propane sultone 20, 100, 500µg/plateTA100, TA1535

Activation:

2-Aminoanthracene (2-anthramine) ____ µg/plate

Other (list):

Acetylaminofluorene 4, 20, 100, 500 µg/plate TA98 (not 4 µg/plate), TA1538

3. Activation: S9 derived from

x	Induced	x	Aroclor 1254	x	Rat	x	Liver
	Non-induced		Phenobarbitol		Mouse		Lung
			None		Hamster		Other
			Other		Other		

Describe S9 mix composition: Composition of the S9-mix was not detailed but it "was prepared according to the method of Ames et al (1975)."

4. **Test organisms: *S. typhimurium* strains**

	TA97	x	TA98	x	TA100		TA102		TA104
x	TA1535		TA1537	x	TA1538		list any others:		

Properly maintained? [x] Yes [ ] No
Checked for appropriate genetic markers (*rfa* mutation, R factor)? [x] Yes [ ] No

5. **Test compound concentrations used:**

Mutagenicity assay:
Nonactivated and activated conditions: 4, 20, 100, 500, 2500 µg/plate in all strains. Five plates/dose/activation condition.

B. TEST PERFORMANCE:

1. **Type of *Salmonella* assay:**

x standard plate test
_ pre-incubation (_ minutes)
_ "Prival" modification *(i.e. azo-reduction method)*
_ spot test
_ other

2. **Protocol**: A standard plate test was conducted by adding 0.1 mL of test material solution, 0.2 mL of S9-mix for tests with activation and 2 mL of molten top-agar (0.6% agar containing 0.5% NaCl, 0.5 mM histidine and 0.5 mM biotin) at 45°C to 0.1 mL of broth culture of the desired bacterial tester strain (approximately 10^9 cells/mL). After mixing, the contents of each tube were poured onto minimal glucose agar plates, the top agar allowed to solidify, the plates incubated at 37°C for two to three days and the number of revertant colonies then counted using an automatic colony counter.

3. **Statistical analysis:** No statistical analysis was performed.

4. **Evaluation criteria:** The number of revertants per plate at each experimental point was determined and the means and standard deviations calculated. If the mean number of revertants per plate was at least twice that of the corresponding solvent control value the results were considered positive.

II. REPORTED RESULTS:

A. **PRELIMINARY CYTOTOXICITY ASSAY:** No preliminary cytotoxicity assay was reported.

B. MUTAGENICITY ASSAY: Five concentrations of Permethrin ranging from 4 to 2500 µg/plate were tested in the mutagenicity assay using, presumably, five replicate plates for each strain/dose/activation combination. The report stated that the average number of revertants was determined from five independent assays. There was no evidence of a mutagenic effect in any tester strain at any test material concentration with or without S9-mix. The positive control values are presented as fold-increases over the corresponding solvent control values and appear appropriate for the respective strains. Results of the mutagenicity assay are summarized in Appendix Table 1 and 2 (MRID 54738, pp. 6 and 7) for the mean number of revertants per plate and the fold-increase over the solvent controls, respectively.

III. DISCUSSION AND CONCLUSIONS

A. INVESTIGATORS' CONCLUSIONS: The investigators concluded that Permethrin was not mutagenic in any of the tester strains at any concentration up to 2500 µg/plate, with or without S9-mix.

B. REVIEWER COMMENTS: The reviewer agrees with the investigators' conclusion of a negative response based on the information provided; however, this study is quite old (completion date not given) and does not meet some of the current acceptance criteria. Individual plate counts were not provided, no signed and dated GLP statement was provided, no historical control values were provided, four rather than five tester strains were used and one was TA1538, not recommended now, and the components of the cofactor solution and the percent S9-fraction in the mix were not given. No justification was given for limiting the upper dose to 2500 µg/plate rather than the limit value of 5000 µg/plate. The information normally included in the Citation section such as the testing laboratory, laboratory study number and study completion date were not provided.

The study is **Unacceptable/guideline** as presented but would be acceptable if the testing laboratory, if identified, could provide the missing information. Due to the age of the study, the missing data may no longer be available and a repeat assay is more practical.

C. STUDY DEFICIENCIES: Study deficiencies were described in the Reviewer Comments section. Most of the deficiencies are likely data presentation deficiencies rather than experimental deficiencies; however, because of the number of omissions, the age of the study and the short time required to conduct an Ames Test, the study should be repeated.

APPENDIX

(MRID 54738)

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Page ___ is not included in this copy.

Pages 176 through 197 are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product inert impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

EXECUTIVE SUMMARY

PERMETHRIN

STUDY TYPE: REPEATED DOSE DERMAL- RAT
[OPPTS 870.3200 (§82-2)]
MRID NOs. 41143801; 42653301

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order 01-85

Primary Reviewer:
C. B. Bast, Ph.D., D.A.B.T.

Signature: Cheryl B Bast
Date: JAN 05 2001

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Date: JAN 05 2001

Robert H. Ross, M.S., Group Leader

Signature: Robert H. Ross
Date: JAN 05 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle. LLC., for the U.S. Department of Energy

PERMETHRIN

Repeated Dose Dermal Study [OPPTS 870.3200 (82-2)]

EPA Reviewer: Yung G. Yang, Ph.D. Date: 10/31/2001
Reregistration Branch 2 (7509C)
EPA Work Assignment Manager: Joycelyn. Stewart, Ph.D. Date: 1/28/2002
Toxicology Branch (7509C)

TXR# 0050649

DATA EVALUATION RECORD

This is an updated executive summary of MRIDs 41143801 & 42653301 (HED # 010385). The NOAEL/LOAEL has been changed.

STUDY TYPE: Repeated Dose Dermal-Rat; OPPTS 870.3200

OPP Number: 82-2
DP BARCODE: D269531
PC CODE: 109701

OPPTS Number: 870.3200
SUBMISSION CODE: S504352
TOX CHEM NO: 652BB

TEST MATERIAL: Permethrin (95.6%)

CHEMICAL NAME: Not provided

CITATION: Milburn, G. M. (1989). Permethrin: 21-day dermal study in rats. ICI Central Toxicology Laboratory, Alderly Park, Macclesfield, Cheshire SK10 4TJ, UK, Study No. CTL/P/2445 and LR0533. May 11, 1989. MRIDs 41143801 & 42653301. Unpublished.

SPONSOR: ICI Americas, Inc.

EXECUTIVE SUMMARY: In a 21-day repeated dose dermal toxicity study (MRIDs 41143801 & 42653301), groups of 5 male and 5 female Wistar Alpk:Apfsd SPF rats were treated with undiluted Permethrin (95.6%, Batch No. Y00040/85, RS/38F). Animals were treated by dermal occlusion for 6 hours/day for 21 days at doses of 0, 50, 150, or 500 mg/kg/day.

There were no treatment-related deaths and no effects on body weight, food consumption, hematology, clinical chemistry, or gross or microscopic lesions. Increases in absolute ($p<0.05$; 10.3% increase) and relative ($p<0.05$; 10.6% increase) liver weight were noted in high-dose females only. No histopathological evidence of adaptive liver change was seen in any treatment group. Therefore, the increase of liver weight in females was not considered biologically significant. Skin irritation was observed at the application site of all treatment groups.

The systemic NOAEL was 500 mg/kg/day (the highest dose tested), the systemic LOAEL was not established. The dermal LOAEL was 50 mg/kg/day based on skin irritation. A dermal NOAEL was not identified.

This study is classified as **Acceptable/Guideline** and does satisfy the guideline requirements for a repeated-dose dermal study [OPPTS 870.3200 (§82-2)] in rats.

DATA EVALUATION REPORT

PERMETHRIN
(PP 557)

STUDY TYPE: SUBCHRONIC ORAL NEUROTOXICITY – RAT [Nonguideline]
MRID 00071952

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-87

Primary Reviewer:
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Robert H. Ross, M.S., Group Leader

Signature: Robert H. Ross
Date:

Quality Assurance:
Lee Ann Wilson, M.A.

Signature: L. A. Wilson
Date:

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract

Permethrin

Subchronic Oral Neurotoxicity (Nonguideline)

EPA Reviewer: Yung G. Yang, Ph.D. Yung G. Yang Date 11/16/2001
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Joycelyn Stewart Date 1/24/2002
Toxicology Branch, HED (7509C)

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Subchronic Oral Neurotoxicity – Rat (Nonguideline)

DP BARCODE: D269531
PC CODE: 109701

SUBMISSION CODE: S504352
TOX CHEM NO: 652BB

TEST MATERIAL: PP 557 (permethrin, purity 90.4%, 39.9% cis, 60.1% trans isomers)

SYNONYMS: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; permethrin; NRDC 143

CITATION: Glaister, J.R., Pratt, I., and Richards, D. (1977) Effects of high dietary levels of PP557 on clinical behaviour and structure of sciatic nerves in the rat. Testing facility not given. Report No. CTL/P/317, March 1977. MRID 00071952, Unpublished.

SPONSOR: ICI Ltd., Plant Protection Division, Bracknell, Berks.

EXECUTIVE SUMMARY: In a preliminary subchronic oral neurotoxicity study (MRID 00071952), groups of 10 male Wistar rats were administered 2500, 3000, 3750, 4500, 5000, or 7500 ppm of PP 557 in the diet for 14 days. The isomeric ratio of the test article (Batch No. P48; 90.4% a.i.) was 39.9% cis and 60.1% trans. Based on a food factor of 0.05 for the rat, doses for the treated groups were 125, 150, 187.5, 225, 250, and 375 mg/kg, respectively. Each treated group had a paired control group consisting of litter mates with similar body weights. Toxicity assessments were limited to clinical observations, measurements of body weights and food consumption, and light and electron microscopic evaluation of the sciatic nerve.

At 7500 ppm six rats were found dead on day 1 and the remainder were sacrificed *in extremis* on day 1 or 2. Prior to sacrifice the animals were observed with convulsive tremors and excessive salivation and those animals for which data were available showed marked weight loss and decreased food consumption. In the 5000-ppm group, two rats were found dead on day 1 and six were sacrificed on day 2; convulsive tremors were observed in one animal prior to death.

Slight to moderate whole body tremors were observed initially in all animals in the 2500 and 3000 ppm groups but almost complete remission occurred by day 5. Moderate tremors were seen in most animals of the 3750 and 4500 ppm groups which lessened during the study but were still evident on day 14. Also at 3750 and 4500 ppm hyperactivity and hypersensitivity to noise were

observed mainly during the first 7 days. In the two surviving 5000-ppm animals, slight to moderate tremors were observed until day 10.

Mean absolute body weights of the 3000-, 3750-, and 4500-ppm groups were significantly ($p \leq 0.05$ or 0.01) less than their paired control group weights beginning on day 1 and continuing until termination. Body weights of the surviving 5000-ppm animals were also clearly less than the control. Body weight gains by the 2500-, 3000-, 3750-, 4500-, and 5000-ppm groups were 81%, 60%, 61%, 28%, and 22%, respectively, of their control group level during the first week. However, during the second week body weight gains by all treated groups were 98-104% of the control levels with the exception of the 5000-ppm group which was 83% of the controls.

Food consumption for the first week was significantly ($p \leq 0.01$) reduced in all treated groups to 67-84% of their paired control group levels. Consequently, food utilization was increased in a dose-related manner for all treated groups as compared with the control groups.

The number of rats with degenerating nerve fragments in the treated and paired control groups was 5/10 each at 2500 ppm, 8/10 and 2/9, respectively, at 4500 ppm, and 6/10 and 2/10, respectively, at 5000 ppm. The number of fragments per nerve ranged from 1-5 for animals in the control, 2500-, and 4500-ppm groups and for animals in the 5000 ppm group that died or were killed intercurrently. In contrast, the two surviving rats in the 5000 ppm group had 19 and 44 fragments respectively.

Nerves from rats in the 2500- and 5000-ppm groups were also examined by electron microscopy. No treatment-related abnormalities were observed in the 2500-ppm group. At 5000 ppm, the ultrastructural changes observed were similar in animals that died and in the two rats that survived to scheduled termination. In the unmyelinated nerves, 7/7 rats given 5000 ppm had degenerative changes including axonal swelling, disorganization of the neurofilaments, an increase in multivesicular-type and vesicular structures, and vacuolation. Only a minimal increase in vesicular structures was observed in 3/7 paired controls. Mild to marked vacuolation of the Schwann cell cytoplasm was seen in 5/7 rats treated with 5000 ppm and mild vacuolation was seen in 2/7 controls. Also in the Schwann cells, dense bodies occurred in the cytoplasm of 6/7 treated rats vs. 0/7 controls and hypertrophy and increased nuclear chromatin with multiple nucleoli were seen in 5/7 treated and 1/7 control rats. Intercellular vacuolation was observed in 4/7 treated and 1/7 control rats.

Therefore, the systemic and neurotoxicity LOAEL is 2500 ppm (125 mg/kg) based on clinical signs of toxicity and decreases in body weight gain and food consumption. The systemic and neurotoxicity NOAEL was not identified for this preliminary study.

This study is classified **Acceptable/Nonguideline** and does not satisfy the requirements for a subchronic oral neurotoxicity study [OPPTS 870.6200 (§82-7)] in rats. The study is sufficient for the purposes for which it was intended, as an evaluation of the effects of feeding high concentrations of PP 557 to male rats on body weights, food consumption, clinical signs, and microscopic lesions in the sciatic nerve.

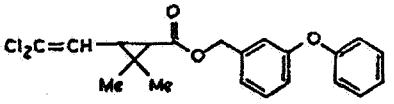
COMPLIANCE: Signed and dated Quality Assurance, Data Confidentiality, Good Laboratory Practice Compliance, and Flagging statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound: PP 557 (permethrin)

Description: not given
CAS No.: 52645-53-1
Batch No.: P48
Purity: 90.4% a.i., isomers cis : trans = 39.9:60.1
Contaminants: none given
Structure:



2. Vehicle

Commercial diet supplied by Oakes of Congleton, Cheshire was used as the vehicle and untreated control. No positive control was used in this study.

3. Test animals

Species: rat
Strain: Wistar
Age and weight at study initiation: "weanling": males 85-129 g
Source: Alderley Park
Housing: not described
Food: Commercial diet was available *ad libitum.*
Water: Water was available *ad libitum.*
Environmental conditions:
Temperature: not stated
Humidity: not stated
Air changes: not stated
Photoperiod: not stated
Acclimation period: 2 weeks

B. STUDY DESIGN

1. In life dates

Start: not given End: not given

2. Animal assignment

Animal assignment and dose selection are listed in Table 1. Rats were randomly assigned to treated groups and each treated animal had a paired litter mate control of similar body weight. The method of randomization was not described. Only male rats were used in this study. The duration of treatment was 14 days.

TABLE 1. Study design

Treated Animals		Paired Controls	
Diet Conc. (ppm)	No. of Males	Diet Conc. (ppm)	No. of Males
2500	10	-	10
3000	10	-	10
3750	10	-	10
4500	10	-	10
5000	10	-	10
7500	10	-	10

Data taken from text table p. 6, MRID 00071952.

3. Validation of test methods

Positive control data were not included for evaluation of the ability of the testing laboratory to conduct FOB or neuropathological studies.

4. Rationale for dose selection

A dose selection rationale was not included in the current report. The study authors noted that tremors were observed in a preliminary dietary study in rats with "high levels" of the test article and that males appeared to be more sensitive than females. No details of the preliminary study were included in the current report.

5. Preparation and analysis of test diets

An appropriate amount of the test article was incorporated into the diets. Further details of the mixing procedures and frequency of preparation were not given. Concentration was determined once in all test diets. Analyses of the test diets for homogeneity and stability were not discussed or reported.

Results -
Concentration analysis: Mean concentrations of the test article in the diets ranged from 104% to 114% of nominal.

Homogeneity analysis: not determined

Stability analysis: not determined

6. Statistical analysis

Body weight and food consumption data were analyzed by Student's t test comparing each treated group with its paired control group.

C. METHODS

1. Observations

All animals were examined for behavioral changes and clinical signs of toxicity, but the frequency of observation was not stated.

2. Body weight

Individual body weights were recorded daily for the first 7 days and at sacrifice.

3. Food consumption and food efficiency

Food consumption was measured daily for the first 7 days and in total for the second week of the study. Food utilization data were reported but the method of calculation was not given.

4. Functional observational battery (FOB)

A full FOB was not conducted.

5. Motor activity

Motor activity was not assessed.

6. Ophthalmology

Ophthalmoscopic examinations were not conducted.

7. Clinical chemistry

Clinical chemistry evaluations were not done.

8. Sacrifice/necropsy/neurohistopathology

Animals found dead, sacrificed moribund, and sacrificed at scheduled termination were subjected to gross necropsy. Each treated rat and its littermate control were necropsied at approximately the same time and the tissues processed together. Brain, spinal cord, sciatic nerve, skeletal muscle, and vagus nerve were preserved in buffered formol saline. The sciatic nerve and skeletal muscle from the other leg were stored in 3% buffered glutaraldehyde and liquid nitrogen, respectively. Only the sciatic nerves from the control, 2500-ppm, 4500-ppm, and 5000-ppm groups were processed for light and/or electron microscopic examination.

II. RESULTS

A. CLINICAL OBSERVATIONS AND MORTALITY

At 7500 ppm six rats were found dead on day 1 and the remainder were sacrificed *in extremis* on day 1 or 2. Prior to sacrifice the animals were observed with convulsive tremors and excessive salivation. In the 5000-ppm group, two rats were found dead on day 1 and six were sacrificed on day 2; convulsive tremors were observed in one animal prior to death. One rat in the 4500 ppm group developed hematuria and was killed on day 7; necropsy revealed ulceration of the bladder. All other animals survived to scheduled termination.

Among survivors, the main clinical sign of toxicity was slight to moderate whole body tremors. Incidence rates were not summarized and the individual animal data were difficult to read due to poor copy quality, however, it appeared that 80-100% of the animals were affected during the study. Slight to moderate tremors were observed initially in all animals in the 2500 and 3000 ppm groups but almost complete remission occurred by day 5. Moderate tremors were seen in most animals of the 3750 and 4500 ppm groups which lessened during the study but were still evident on day 14. Also at 3750 and 4500 ppm hyperactivity and hypersensitivity to noise were observed mainly during the first 7 days. In the two surviving 5000-ppm animals, slight to moderate tremors were observed until day 10.

B. BODY WEIGHTS AND BODY WEIGHT GAINS

Selected data for body weights and body weight gains is given in Table 2. Data for the 7500-ppm group have been omitted from the table because all animals were dead by day

2 of the study. However, those animals for which data were available showed marked weight loss during the first two days of treatment.

Dose-related decreases in body weights and body weight gains were observed throughout the study with the most pronounced effects during the first week. Mean absolute body weights of the 3000-, 3750-, and 4500-ppm groups were significantly ($p \leq 0.05$ or 0.01) less than their paired control group weights beginning on day 1 and continuing until termination. Body weights of the surviving 5000-ppm animals were also clearly less than the control, however, because only two animals were available for calculation statistical significance was not attained at all timepoints. During the first week of the study, body weights of the 3000-, 3750-, 4500-, and 5000-ppm groups were 89-91%, 87-89%, 82-88%, and 72-79%, respectively, of their controls.

Weight loss occurred in all treated groups for the first day of the study. Body weight gains by the 2500-, 3000-, 3750-, 4500-, and 5000-ppm groups were 81%, 60%, 61%, 28%, and 22%, respectively, of their control group level during the first week. However, during the second week body weight gains by all treated groups were 98-104% of the control levels with the exception of the 5000-ppm group which was 83% of the controls.

TABLE 2: Body weights and body weight gains (g) of rats given PP 557 for 14 days

Day of Study	2500 ppm		3000 ppm		3750 ppm		4500 ppm		5000 ppm	
	Treat.[a]	PC	Treat.	PC	Treat.	PC	Treat.	PC	Treat.	PC
Mean body weights										
0	106.0	104.2	116.6	112.4	112.3	112.3	110.8	109.7	104.7	102.7
1	103.8	111.3	106.9**	119.5	105.0**	119.4	103.1**	116.6	96.4	111.3
3	115.3	122.6	119.1**	131.7	114.7**	130.6	108.1**	127.8	101.5	133.5
5	129.5	136.3	131.4**	146.0	127.4**	144.3	116.5**	140.6	108.5	149.0
7	143.8	150.6	143.9**	158.1	139.4**	156.6	123.1**	154.1	118.0*	163.0
14[b]	191.5	196.5	191.3*	205.5	186.5**	202.1	168.9**	200.8	157.0*	210.0
Body weight changes[c]										
0-1	-2.2	7.1	-9.7	7.1	-7.3	7.1	-7.7	6.9	-8.3	8.6
0-7	37.8	46.4	27.3	45.7	27.1	44.3	12.3	44.7	13.3	60.3
7-14	47.7	45.9	47.4	47.4	47.1	45.5	45.8	46.7	39.0	47.0
0-14	85.5	92.3	74.7	93.1	74.2	89.8	58.1	91.1	52.3	107.3

Data taken from Tables 3 and 4, pp. 19 and 20, respectively, MRID 00071952.
[a]Treat. = treated group; PC = paired control group.
[b]Due to poor copy quality some day 14 means may be inaccurate.
[c]Calculated by reviewer from group means.
Significantly different from paired control: *$p \leq 0.05$; **$p \leq 0.01$.

C. FOOD CONSUMPTION AND COMPOUND INTAKE

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Selected food consumption and food utilization data are given in Table 3. Data for the 7500-ppm group have been omitted from the table because all animals were dead by day 2 of the study. However, those animals for which data were available showed marked reduction in food consumption the first day of treatment.

Dose-related decreases in food consumption were observed with the most pronounced effects during the first week of the study. Food consumption for the first week was significantly ($p \leq 0.01$) reduced in all treated groups to 67-84% of their paired control group levels. Slight recovery was observed during the second week of the study when food consumption by the groups administered ≤4500 ppm was 83-94% that of the controls. Food consumption by the 5000-ppm group during the second week was 123% of the control level, but only two animals were available and the data were variable.

Food utilization was increased in a dose-related manner for all treated groups as compared with the control groups during the first week of the study. Thereafter, food utilization by the treated groups was similar to the controls with the exception of the 5000-ppm group which remained slightly greater than the control values during the second week.

Compound consumption was not calculated by the study authors. Based on a food factor of 0.05 for the rat, doses for the 2500, 3000, 3750, 4500, 5000, and 7500 ppm groups were 125, 150, 187.5, 225, 250, and 375 mg/kg, respectively.

TABLE 3: Selected food consumption data (g/animal) for rats given PP 557 for 14 days

Group	Days 0-7	Days 7-14	Days 0-14
Food Consumption			
2500 ppm Treated Paired Control	124.3** (84)[a] 148.1	157.3 (94) 167.1	281.6** (89) 315.2
3000 ppm Treated Paired Control	122.1** (78) 156.6	151.9** (91) 167.5	274.0* (85) 324.1
3750 ppm Treated Paired Control	127.1** (85) 150.2	149.0 (94) 158.9	276.1* (89) 309.1
4500 ppm Treated Paired Control	110.1** (72) 152.0	135.7** (83) 162.9	252.0** (79) 318.1
5000 ppm Treated Paired Control	107.0** (67) 159.0	211.0* (123) 171.5	318.0 (96) 330.5
Food Utilization[b]			
2500 ppm Treated Paired Control	3.3 3.2	3.2 3.6	3.3 3.4
3000 ppm Treated Paired Control	3.8 3.4	3.2 3.5	3.4 3.5
3750 ppm Treated Paired Control	4.7 3.4	3.2 3.5	3.7 3.4
4500 ppm Treated Paired Control	9.0 3.4	3.1 3.4	4.4 3.5
5000 ppm Treated Paired Control	53.5 3.2	5.4 3.6	7.8 3.4

Data taken from Tables 5-8, pp. 21-24, respectively, MRID 00071952.
[a]Number in parentheses is percent of control; calculated by reviewer.
[b]Food utilization data were not analyzed statistically.
Significantly different from paired control: *p ≤ 0.05; **p ≤ 0.01.

D. FUNCTIONAL OBSERVATIONAL BATTERY (FOB)

Clinical signs of toxicity are described above. A detailed FOB was not conducted.

E. MOTOR ACTIVITY

Motor activity was not measured.

F. OPHTHALMOLOGY

Ophthalmologic examinations were not performed.

G. CLINICAL CHEMISTRY

Clinical chemistry parameters were not evaluated in this study.

H. NECROPSY

Findings at gross necropsy were not reported.

I. NEUROPATHOLOGY

Sciatic nerves from rats administered 2500, 4500, and 5000 ppm and their control groups were examined histologically. Sections from the 7500-ppm group were not examined because morphological changes were not expected to develop in less than 24 hours and 6/10 of these animals died by day 1. Degenerating nerve fragments were observed in rats from the treated and control groups. The number of rats with degenerating nerve fragments in the treated and paired control groups was 5/10 each at 2500 ppm, 8/10 and 2/9, respectively, at 4500 ppm, and 6/10 and 2/10, respectively, at 5000 ppm. The number of fragments per nerve ranged from 1-5 for animals in the control, 2500-, and 4500-ppm groups and for animals in the 5000 ppm group that died or were killed intercurrently. In contrast, the two surviving rats in the 5000 ppm group had 19 and 44 fragments respectively.

Nerves from rats in the 2500- and 5000-ppm groups were also examined by electron microscopy. At 5000 ppm, the ultrastructural changes observed were similar in animals that died and in the two rats that survived to scheduled termination. In the unmyelinated nerves, 7/7 rats given 5000 ppm had degenerative changes including axonal swelling, disorganization of the neurofilaments, an increase in multivesicular-type and vesicular structures, and vacuolation. Only a minimal increase in vesicular structures was observed in 3/7 paired controls. Fragmentation of the myelin sheath was seen in 6/7 treated and 5/7 control animals, but was particularly marked in one of the surviving rats. Mild to marked vacuolation of the Schwann cell cytoplasm was seen in 5/7 rats treated with 5000 ppm and mild vacuolation was seen in 2/7 controls. Also in the Schwann cells, dense bodies occurred in the cytoplasm of 6/7 treated rats vs. 0/7 controls and hypertrophy and increased nuclear chromatin with multiple nucleoli

DLA

were seen in 5/7 treated and 1/7 control rats. Intercellular vacuolation was observed in 4/7 treated and 1/7 control rats.

At 2500 ppm, fragmentation of the myelin axons was seen in 6/10 treated animals and in 3/10 control animals. Minor lesions in unmyelinated nerves, including axonal swelling, vacuolation, and increased vesicular structures, occurred in 8/10 animals administered 2500 ppm compared with 3/10 controls, but the severity was greater in the control animals than in the treated animals. Multiple nuclei, mild vacuolation of Schwann cell cytoplasm, and dense bodies in the cytoplasm were seen at similar rates between the treated 2500-ppm rats and their controls. Intercellular vacuolation was observed in 9/10 rats fed 2500 ppm and 7/10 control rats and was less marked than that seen in the 5000-ppm animals. These changes in rats treated with 2500 ppm are of questionable biological significance and may not be treatment-related.

III. DISCUSSION

A. DISCUSSION

The objective of this study was to evaluate the effects of PP557 (permethrin) on body weight, food consumption, clinical signs of toxicity, and the structure of sciatic nerves when administered to rats in the diet. A dietary concentration of 7500 ppm was lethal to all animals on the first day of treatment and only 2 rats in the 5000-ppm group survived to terminal sacrifice. Whole body tremors were observed in animals of all dose groups and increased in severity with increasing concentration. Complete regression of slight to moderate tremors occurred in the lower dose groups, whereas, convulsive tremors preceded death at the two highest concentrations.

Dose-related decreases in body weights, body weight gains, and food consumption were observed in all treated groups with the most pronounced effects during the first week of the study. Although decreased food consumption paralleled the reduction in body weight gains, the increases in food utilization indicate a direct toxicity as well.

Evaluation of the structural changes in the nervous tissue was limited to the sciatic nerve. An increase in the number of rats with degenerating nerve fragments was found in the 4500- and 5000-ppm groups. The number of fragments per nerve was also greatly increased in the surviving rats of the 5000 ppm group. Ultrastructural examination of nerves from the rats in the 2500 and 5000 ppm groups revealed numerous changes at 5000 ppm. The relationship between the microscopic lesions and the clinical signs of toxicity is unknown, as is the mechanism by which the test article could affect the lesions.

Therefore, the systemic and neurotoxicity LOAEL is 2500 ppm (125 mg/kg) based on clinical signs of toxicity and decreases in body weight gain and food consumption. The systemic and neurotoxicity NOAEL was not established for this nonguideline study.

This study is classified **Acceptable/Nonguideline** and does not satisfy the requirements for a subchronic oral neurotoxicity study [OPPTS 870.6200 (§82-7)] in rats. The study is sufficient for the purposes for which it was intended, as an evaluation of the effects of feeding high concentrations of PP 557 to male rats on body weights, food consumption, clinical signs, and microscopic lesions in the sciatic nerve.

B. STUDY DEFICIENCIES

This is a nonguideline study; however, information missing from the report included homogeneity and stability of the test diets and validation of testing methods. Endpoints not evaluated included an FOB, motor activity, and clinical chemistry. The study was conducted in males only.

DATA EVALUATION RECORD

PERMETHRIN/109701
(21Z73)

STUDY TYPE: PRENATAL DEVELOPMENTAL TOXICITY STUDY - RABBIT
OPPTS 870.3700b [§83-3b]; OECD 414.
MRID 00057101

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
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Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

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Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/13/2002
Signature: Joycelyn C. Stewart
Date 5/17/2002

Template version 11/01

TXR#: 0050649

DATA EVALUATION RECORD
Supplementary, HED Doc # 008163

STUDY TYPE: Prenatal Developmental Toxicity Study - Rabbit; OPPTS 870.3700b

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 21Z73 (Permethrin; purity, cis-/trans- ratio, and lot/batch number not reported)

SYNONYMS: NRDC 143

CITATION: James, D. (1974) Preliminary foetal toxicity study in the rabbit given 21Z73 (NRDC 143) orally. The Wellcome Research Laboratories, Beckenham. Laboratory report number Path 176, October 22, 1974. MRID 00057101. Unpublished.

SPONSOR: The Wellcome Foundation, Ltd.

EXECUTIVE SUMMARY: In a developmental toxicity study (MRID 00057101) 21Z73 (Permethrin; % a.i., cis-/trans- ratio, and batch/lot # not reported) was administered to an unknown number of inseminated female Dutch strain rabbits/dose in corn oil by gavage at dose levels of 0 or 400 mg/kg bw/day from days 6 through 18 (inclusive) of gestation, and an additional group of animals served as "environmental controls." Dams were sacrificed on day 28 of gestation and all fetuses were weighed, sexed, and examined externally. One-third of the fetuses were examined for skeletal alterations, and two-thirds were examined for visceral alterations.

There were no treatment-related effects on survival, body weight, or cesarean section parameters. Food consumption was not measured and clinical signs were not reported. Three intercurrent deaths occurred and were attributed to respiratory insufficiency due to pulmonary infections. **The maternal LOAEL for 21Z73 is not identified, and the maternal NOAEL is greater than or equal to 400 mg/kg bw/day.**

There were no treatment-related effects on developmental parameters. However, the number of fetuses (litters) examined in the vehicle control and treated groups was only 29 (5) and 28 (7), respectively. The small numbers of fetuses and litters that resulted from low pregnancy rates, intercurrent deaths, and/or high preimplantation losses severely limited the sensitivity of this

study with respect to developmental toxicity. **The developmental LOAEL for 21Z73 is not identified, and the developmental NOAEL is greater than or equal to 400 mg/kg bw/day.**

This developmental toxicity study in the rabbit is classified **unacceptable/guideline (non-upgradable)** and does not satisfy the guideline requirements for a developmental toxicity study (OPPTS 870.3700; OECD 414) in the rabbit. In addition to compromised maternal health during the study, there were insufficient numbers of fetuses and litters available to adequately evaluate developmental toxicity. Additional deficiencies are listed in the deficiency section.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided; however, the study was conducted in 1974.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. **Test material:** 21Z73 (Permethrin)

Description:	Not reported
Lot/Batch #:	Not reported
Purity:	Not reported
Compound Stability:	Not reported
CAS # of TGAI:	52645-53-1

2. **Vehicle and/or positive control:** The vehicle control group was given corn oil at 2.0 mL/kg (Lot/Batch # and purity not reported). There was no positive control used.

3. **Test animals:**

Species:	Rabbit	
Strain:	**Dutch belted**	
Age/weight at study initiation:	Females: assumed to be young adult; approximately 1.8-3.6 kg.	
Source:	Ranch Rabbits	
Housing:	Individually. Cages were not described.	
Diet:	Pelleted diet 18 (supplied by Lillico and Co. Ltd., manufactured by Oxoid) *ad libitum*	
Water:	not otherwise described *ad libitum*	
Environmental conditions:	**Temperature:**	Not reported
	Humidity:	Not reported
	Air changes:	Not reported
	Photoperiod:	Not reported
Acclimation period:	Not reported	

B. PROCEDURES AND STUDY DESIGN:

1. **In life dates:** Not reported. The entire study took place August-October 1974.

2. **Mating:** The does were artificially inseminated, and each doe received an intravenous injection of 25 I.U. of Chorulon in 0.25 mL of distilled water. Mating records and the source of the semen used were not provided; it is therefore unknown whether males were the same

strain and from the same source as the females and whether sibling matings were avoided. The day of artificial insemination was designated as gestation day (GD) 0.

3. **Animal assignment:** Animals were assigned by an unspecified method to the dose groups indicated in Table 1.

TABLE 1: Animal assignment

Dose	Vehicle controls	Environmental controls	400 mg/kg/day
# Females	6	9	7

Data taken from text p. 1, and Table 1, p. 7, MRID 00057101.
a Animals were treated on GD 6-18 inclusive. Dose is expressed in terms of the active ingredient.

4. **Dose selection rationale:** There was no dose selection rationale provided.

5. **Dosage preparation and analysis** The preparation method of the test material-vehicle mixture was not described in the study report. The study report stated that the treated group received 400 mg active ingredient/kg body weight; it is therefore assumed that the concentration of the test material in the vehicle was adjusted to account for purity.

   There was no mention of the frequency of preparation or storage conditions of mixtures of test substance with the vehicle. There was also no mention of evaluation of the stability of the test substance in the vehicle or concentration and homogeneity of the test mixtures.

   There were no analytical data provided to indicate whether the mixing procedure was adequate or that the variance between nominal and actual dosage to the study animals was acceptable.

6. **Dosage administration:** All doses were administered once daily via gavage, on gestation days 6 through 18, in a volume of 2.0 mL/kg of body weight/day. The study report did not state whether dosing was based on the body weight from the most recent body weight determination; however, this is assumed to be the case.

C. OBSERVATIONS:

1. **Maternal observations and evaluations:** The animals were checked for mortality or clinical signs daily. Body weights were recorded at artificial insemination, then either on alternate days during dosing and approximately every fourth day thereafter (p. 3) or daily throughout pregnancy (p. 5); however, body weight data were reported only for GD 0, 6, 20, and 28 and only for does that were pregnant at cesarean section. Food consumption was not measured. Does were sacrificed on day 28 of gestation. Examinations at sacrifice consisted of recording numbers of corpora lutea, implantations, live fetuses, dead fetuses, early or late resorptions, and externally abnormal fetuses. There was no mention of further gross necropsies of the dams.

2. **Fetal evaluations:** All fetuses were weighed and subjected to external examination. Approximately one-third of the fetuses from each litter were subjected to soft tissue examination of the organs of the neck, thorax, and abdomen by gross dissection; one-third were examined by the Wilson method of serial free-hand sectioning; and one-third were subjected to skeletal examination after staining by the Staples and Schnells Alizarin red S method. External, visceral, and skeletal anomalies were not further characterized as variations and malformations. Litter incidences of fetal morphological data were not reported and fetal individual morphological data were not provided.

D. DATA ANALYSIS:

1. **Statistical analyses:** The following data were analyzed using ANOVA: maternal body weight gains during dosing and gestation, numbers of corpora lutea, implantations, live fetuses, and normal fetuses, litter weights, and weights of normal fetuses. Litter incidences of external, visceral, and skeletal anomalies were not reported, and fetal incidences of these endpoints were not compared statistically.

   "Standard errors" were reported for some of the data. The term "standard error" can either be a synonym for "standard deviation."or a separate statistical measure of variance that can be converted to standard deviation by multiplying by the square root of n. As it was unknown which was the case in the study report, the data were summarized as reported.

2. **Indices:** The following index was calculated from cesarean section records of animals in the study:

   Implant/corpora lutea ratio = The number of implantations/the number of corpora lutea

   The reviewer calculated pre- and postimplantation loss indices as follows:

   Preimplantation loss (%) = (total number of corpora lutea minus total number of implantations/total number of corpora lutea) x 100.

   Postimplantation loss (%) = (total number of implantations minus total number of live fetuses/number of implantations) x 100.

3. **Historical control data:** Historical control data were not provided to allow comparison with concurrent controls.

II. RESULTS:

A. MATERNAL TOXICITY:

1. **Mortality and clinical observations:** Clinical signs were not reported. One doe from each of the environmental control, vehicle control, and treated groups died prior to scheduled sacrifice, with the deaths occurring on GDs 26, 20, and 16, respectively. All three deaths were attributed to pulmonary infection and respiratory insufficiency. No abortions or early deliveries were reported.

2. **Body weight** - Body weight data are given in Table 2. There were no treatment-related effects on mean absolute body weights and body weight changes. The lower mean absolute body weights of the treated group on GDs 6, 20, and 28 are most likely due to a mean weight loss by this group during the pre-treatment interval.

TABLE 2. Mean maternal body weight data (kg) [a]

Gestation Day	Dose in mg/kg bw/day (# of Dams)	
	Vehicle Control (6)	400 (7)
Absolute Body Weights		
0	2.53±0.49	2.31±0.34
6	2.55±0.46	2.27±0.31 (89) [b]
20	2.65±0.50	2.34±0.31 (88)
28	2.68±0.40	2.44±0.24 (91)
Corrected body weight [c]	2.48	2.29
Body Weight Changes		
0-6 (Pretreatment)	0.02±0.08	-0.04±0.05
6-20 (~Treatment) [d]	0.10±0.11	0.07±0.10
20-28 (~Posttreatment) [d]	0.03±0.14	0.10±0.08
0-28 (Gestation)	0.15±0.19	0.13±0.15
Corrected body weight change [e]	-0.05	-0.02

Data taken from Tables 1, 5, and 8, pages 7, 12, and 15, respectively, MRID 00057101.
a Calculated by reviewer using individual body weight data and expressed as Mean±SD
b Number in parentheses equals percent of control; calculated by reviewer.
c Estimated by reviewer as Corrected body weight = Mean GD 28 weight minus mean litter weight. Mean litter weights for vehicle control and treated groups were 199.00 and 144.29 g, respectively.
d Absolute body weight data were reported only for GD 0, 6, 20, and 28; therefore, the reviewer used GD 6-20 and GD 20-28 to approximate the treatment and post-treatment intervals, respectively.

3. **Food consumption**: Food consumption was not measured.

4. **Gross pathology**: Does were not routinely subjected to gross necropsy.

5. **Cesarean section data**: Data collected at cesarean section are summarized in Table 3. Pregnancy rates could not be determined by the reviewer due to poor copy quality; however, the study author stated that there was a low conception rate in all groups. The preimplantation losses of both groups were high, possibly as a consequence of either poor artificial insemination technique or compromised maternal health status (pulmonary infection). There were no remarkable differences between postimplantation losses, early and late resorptions per dam, mean live litter size, mean fetal weights, or fetal sex ratios of the treated and control groups. There were no abortions or early deliveries in either group. One doe from the vehicle control group had a total litter resorption.

TABLE 3. Cesarean section observations [a] [Mean±SD, as appropriate]		
Observation	Dose (mg/kg bw/day)	
	0 (Vehicle controls)	400
# Animals Assigned (Mated) [b]	Unknown	Unknown
# Animals pregnant at cesarean section	6	7
Pregnancy Rate (%) [b]	Unknown	Unknown
# Nonpregnant [b]	Unknown	Unknown
Maternal Wastage:		
# Died	1	1
# Died Pregnant	1	1
# Died Nonpregnant	0	0
# Aborted or delivered early	0	0
Total # Corpora Lutea	55	63
Corpora Lutea/Dam	9.17	9
Total # Implantations	34	29
Implantations/Dam	5.67	4.14
Total # Litters	5	7
Total # Live Fetuses	29	28
Live Fetuses/Dam	4.83	4
Total # Dead Fetuses	0	0
Dead Fetuses/Dam	0	0
Total # Resorptions	5	1
Early (Complete)	4	1
Late (Partial)	1	0
Resorptions/Dam	0.83	0.14
Early (Complete)	0.67	0.14
Late (Partial)	0.17	0
Litters with Total Resorptions	1	0
Mean Fetal Weight (g) [c]	35.18	37.3
Sex Ratio (% Male)	44.83	50
Preimplantation Loss (%) [d]	38.2	54
Postimplantation Loss (%) [e]	14.7	3.4

Data taken from Tables 1, 6, 7, and 8, pp. 7, 13, 14, and 15, respectively, MRID 00057101.

a Data expressed as mean ± standard error where appropriate; however, standard errors were not provided and/or legible for all means.

b The number of animals assigned to each group was illegible in the study report.

c Mean fetal weight was reported only for combined sexes.

d Calculated by reviewer as Preimplantation Loss = (Total corpora lutea minus total implantations/total corpora lutea) x 100.

e Calculated by reviewer as Postimplantation Loss = (Total implantations minus total live fetuses/Total implantations) x 100.

B. DEVELOPMENTAL TOXICITY: The total number of fetuses (litters) in the vehicle control and treated groups were 29 (5) and 28 (7), respectively. The study report did not include litter incidences of fetal structural alterations, and it was unclear whether all findings,

both major and minor were reported. Selected fetal morphological observations are given in Table 4.

1. **External examination**: There were no external anomalies observed at caesarian section.

2. **Visceral examination:** Ten fetuses from each of the vehicle control and treated groups were examined for visceral alterations by open dissection, and no visceral alterations were reported. Ten and 9 fetuses from the vehicle control and treated groups, respectively, were examined for visceral alterations by Wilson's technique; the multiple visceral abnormalities noted in a single fetus from the treated group were not considered treatment-related. It could not be determined from the study report whether fetuses from all litters were examined for visceral alterations.

3. **Skeletal examination**: Nine fetuses from each of the vehicle control and treated groups were examined for skeletal alterations. It could not be determined from the study report whether fetuses from all litters were examined for skeletal alterations. All observations from the skeletal examinations occurred at similar incidences in the treated and vehicle control groups; however, the data were not reported in sufficient detail to permit evaluation of ossification rates.

TABLE 4. Fetal morphological observations [a]

Observations [b]	Dose (mg/kg bw/day)	
	0 (Vehicle controls)	400
Visceral examination by Wilson's technique		
# Fetuses examined	10	9
Multiple visceral abnormalities [c]	0	1
Hyperplasia of small intestines	1	0
Skeletal examination		
# Fetuses examined	9	9
Poor ossification of interparietals	0	1
13th vestigial rib on one side	0	2
One or more poorly ossified sternebrae	7	8
Forelimb phalanges poorly ossified	1	0

Data taken from Tables 4b and 4c, pages 10 and 11, respectively, MRID 00057101.

a Data are given as the number of fetuses with the specified abnormality. Litter incidences were not reported, and individual data for fetal morphological endpoints were not included in the study report; it therefore could not be determined how many litters were included in each type of visceral or skeletal examination.

b Some observations may be grouped together.

c Included the following: stomach displaced to right hand side; lobe of liver lying dorsal to stomach and enfolding the right kidney; and spleen in normal position on ectopic stomach, abnormally positioned in body.

III. DISCUSSION AND CONCLUSIONS:

A. INVESTIGATORS' CONCLUSIONS:

The study author concluded that there were no significant treatment-related effects on maternal weights, numbers of live fetuses, fetal malformations, or fetal deaths. The study author did not identify LOAELs or NOAELS.

B. REVIEWER COMMENTS:

1. **Maternal toxicity:** The three intercurrent deaths that were attributed to "pulmonary infection and respiratory insufficiency" indicate that maternal health status during the study may have been compromised. There was no maternal toxicity reported at the highest (and only) dose level.

   Therefore, the maternal toxicity LOAEL for 21Z73 in Dutch belted rabbits is not identified, and the maternal toxicity NOAEL is greater than or equal to 400 mg/kg bw/day.

2. **Developmental toxicity:** It must be noted that the small numbers of fetuses and litters that resulted from low pregnancy rates, intercurrent deaths, and/or high preimplantation losses as well as the method of partitioning the fetuses into separate groups for either visceral or skeletal examination severely limit the sensitivity of this study with respect to developmental toxicity.

a. **Deaths/resorptions:** Maternal treatment did not result in an increase in fetal deaths or resorptions.

b. **Altered growth:** Maternal treatment did not result in decreased fetal weights. It was not possible to adequately evaluate fetal ossification rates due to insufficient data.

c. **Developmental abnormalities:** Treatment with the test article did not result in an increased incidence of fetal structural alterations.

   Therefore, the developmental toxicity LOAEL for 21Z73 in Dutch belted rabbits is not identified, and the developmental toxicity NOAEL is greater than or equal to 400 mg/kg bw/day.

C. STUDY DEFICIENCIES: No data can be provided to satisfy the following major deficiencies, which resulted in the classification of the study as unacceptable and non-upgradable:

- The study did not have three dose levels and a concurrent control (or a limit dose of 1000 mg/kg/day) but instead used a single dose level.
- At the highest dose level, there was no maternal toxicity reported and developmental toxicity was not observed.

- The groups did not contain a sufficient number of animals to yield approximately 12 animals per group with implantation sites at cesarean section. There were 6 and 7 animals with implantation sites in the vehicle control and treated groups, respectively.
- Only one-third of the fetuses were examined for skeletal alterations, and the guideline specifies that for rabbits all fetuses should be examined for both soft tissue and skeletal alterations. Due to low pregnancy rates, intercurrent deaths, and/or high preimplantation losses and the small group size mentioned above, only 19-20 fetuses from the vehicle control and treated groups were examined for visceral alterations, and only 9 fetuses from each of these groups were examined for skeletal alterations.
- Maternal health during the study was compromised (pulmonary infections).

Additional major deficiencies included the following:

- Full identification of the test material (chemical identification, percentage active,batch or lot number, physical properties, purity/impurities, expiration date, vehicle used, if any).
- It is also unknown whether the technical form of the active ingredient was used.
- Analyses for test material stability, homogeneity and concentration in dosing medium were not conducted.
- Litter incidences were not included for fetal morphological observations, and the litter was not considered the basic unit of analysis in the evaluation of fetal structural alterations. Individual data for these endpoints were also not provided; therefore the reviewer was unable to calculate numbers and percent of litters with structural alterations.
- The females used in the study were uniparous, rather than nulliparous as specified in the guideline.
- Clinical signs were not reported in either summary or individual form.
- On p. 173, the report stated that does were weighed on alternate days during dosing, then approximately every fourth day thereafter; however, on p. 175, the report stated that does were weighed at artificial insemination. and daily throughout pregnancy. Body weights were actually reported only for GD 0, 6, 20, and 28 and only for does that were pregnant at cesarean rather than at least at 3-day intervals during the dosing period, as specified in the guidelines.
- Uteri that were grossly non-gravid were not examined by a technique (such as ammonium sulfide staining) to confirm nonpregnant status.
- Gross necropsies of the dams were not conducted.

The following minor deficiencies were also noted:

- The test substance was administered on GD 6-18 (inclusive), rather than daily from implantation to the day before cesarean section, as specified in the guidelines; however, the period of major organogenesis was included in the dosing interval.
- The source of the sperm used for artificial insemination was not reported, so it is unknown whether the males were of the same strain as the females or whether sibling matings were avoided, as specified in the guidelines.
- Gravid uterine weights, as well as body weight changes adjusted for gravid uterine weights were not reported.
- Food consumption was not recorded.

- Fetal body weight data were reported only for the combined sexes.

DATA FOR ENTRY INTO ISIS

evelopmental Study - rabbits (870.3700b)

PC code	MRID	Study	Species	Duration	Route	Admin	Dose range mg/kg/day	Doses mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ	Comments
09701	57101	developmental	rabbits	GD 6-18	oral	gavage	400	0, 400	≥400	not identified	none	Maternal
09701	57101	developmental	rabbits	GD 6-19	oral	gavage	400	0, 400	≥400	not identified	none	Developmental

DATA EVALUATION RECORD

PERMETHRIN (PP557)

STUDY TYPE: CHRONIC ORAL TOXICITY/ONCOGENICITY-RAT
[OPPTS 870.4300 (§83-5a)
MRID 92142123

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-88

Robert H. Ross

Primary Reviewer: Nancy B. Munro, Ph.D.	Signature:	
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	Date:	JUN 0 8 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

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PERMETHRIN **Chronic Toxicity/Oncogenicity Study [OPPTS 870.4300 (§83-5a)]**

EPA Reviewer: Yung G. Yang, Ph.D. Yung G. Yang, Date 11/14/2001
Reregistration Action Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Joycelyn Stewart, Date 1/22/2002
Toxicology Branch, HED (7509C)

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Chronic Oral Toxicity/Oncogenicity – Rat; [OPPTS 870.4300 (§83-5a)]

DP BARCODE: D269531 SUBMISSION CODE: S504352
P.C. CODE: 109701 TOX. CHEM. NO.: 652BB

TEST MATERIAL (PURITY): Permethrin (purity 93.1-98.9%); cis:trans= 40:60

SYNONYMS: 3-phenoxybenzyl (±) cis:trans-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-1-carboxylate; PP557

CITATION: Richards, D. et al. (1977) Phase 3 reformat of MRIDs 69701 and 120268. Permethrin (PP557): 2 year feeding study in rats (Volume I of II). ICI Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire SK10 4TJ, UK. CTL Report Number: CTL/P/357, December 19, 1977; reformatted April 30, 1990. MRID 92142123.

SPONSOR: ICI Americas Inc., Agricultural Products, Wilmington, Delaware 19897.

EXECUTIVE SUMMARY: **In a chronic oral toxicity/oncogenicity study, Permethrin was administered to 60 rats/sex/group in the feed at doses of 0, 500, 1000, or 2500 ppm. The mean estimated compound intake for males was 0, 19.4, 36.9, or 91.5 mg/kg/day, respectively, and for females was 0, 19.1, 40.2, or 104 mg/kg/day. Of these animals, 12/sex/group were sacrificed at** 52 weeks and the surviving rats were sacrificed at 104 weeks' exposure.

No treatment-related effect on mortality was observed during the the study. No treatment-related effects were seen on tumor induction. During the first two weeks of the study, treatment-related tremors and hypersensitivity were observed in both the high-dose male and female groups. No . other treatment-related clinical effects were observed. There were no toxicologically significant effects on body weight, body weight gain, food consumption, or food efficiency. There were no treatment-related effects on ophthalmologic endpoints, hematologic endpoints, clinical chemistry or urinalysis parameters.

Liver changes suggestive of adaptive hypertrophy included increased aminopyrine-N-demethylase activity in all male treatment groups, in the mid- and high-dose female at 52 weeks, and in the high-dose male and female groups at 104 weeks. This was coupled with modestly increased absolute and relative liver weights in the high-dose males and high and low-dose

Batch	Purity %	*cis:trans* %	Periods of Use (Weeks of Study
P21	93.6	37.9:55.7	0-8
P29	93.1	39.3:53.8	8-10
P32	95.7	37.8:57.9	10-13
P34	98.9	43.9:55.0	13-25
P35	97.2	39.5:57.7	25-26 and 42-49
P36	98.5	39.4:58.1	26-32
P52	95.3	36.3:59.0	32-42
P44	94.0	37.9:56.1	49-81
BX4	94.1	37.0:57.1	81-91
BX6	97.2	36.2:61.0	91-104

Data from Table 1, p. 15, MRID 92142123.

Stability of compound: 4 weeks in diet (Bradbrook et al., 1977)
CAS No.: 52645-53-1
Structure: See attached page

2. Vehicle and/or positive control: corn oil

3. Test animals

Species: rat
Strain: Wistar derived, specific pathogen-free
Age/weight at study initiation: Five to six weeks; males: 92-166 g; females: 86-170 g
Source: Alderley Park colony
Housing: Barrier-maintained area; females 4/cage and males 2/cage; cages of galvanized wire mesh on 3 sides and floor; solid wood back
Diet: Stock ration from Oakes Ltd., Congleton, Cheshire, U.K. plus malt extract, *ad libitum*
Water: *ad libitum*
Environmental conditions:
Temperature: 21-25 °C
Humidity: 45-60%
Air changes: Not specified
Photoperiod: 12-hour light/dark cycle
Acclimation period: 1 week

B. STUDY DESIGN

1. In life dates

Start: August 1975; end: August 1977; animals were started on study at four one-week intervals.

2. Animal assignment

The animals were randomized by weight to give 60 males and 60 females per group in which group mean weights were similar.

TABLE 1: Study design

Dose Group	Concentration in Diet (ppm)	Dose (mg/kg/day)[a]		Number of Rats 52-Week Exposure		Number of Rats 104-Week Exposure	
		Male	Female	Male	Female	Male	Female
1	0	0	0	12	12	48	48
2	500	19.4	19.1	12	12	48	48
3	1000	36.9	40.2	12	12	48	48
4	2500	91.5	104	12	12	48	48

Data taken from Tables 2, 12-13, and 16-17, pp. 17, 46-55, and 58-61, MRID 92142123.
[a]Dose estimated by reviewer from nominal concentration in diet, food consumption in Week 104, and group mean body weights as of Week 104.

3. Dose selection rationale

Doses were based on the results of a 28-day range-finding study in which the test substance was fed at levels up to 10,000 ppm (Clapp et al., 1977). Total mortality occurred in both sexes at 10,000 ppm within 3 days; 50% of males fed 5000 ppm died during the study. The dietary LD_{50} of approximately 5000 ppm in males was confirmed in a later study (Glaister et al., 1977). The maximum dose in the present study was chosen to be 50% of the dietary LD_{50} for males.

4. Diet preparation and analysis

The diet batches were prepared weekly except during the last 13 weeks of the study when they were occasionally prepared every 2 weeks. Stock ration was mixed (77 parts by weight) with 18 parts malt extract and 5 parts corn oil. A small amount of tap water was added to aid mixing. The ingredients were mixed for 10 minutes and then pelleted. The test substance was mixed with a small portion of the corn oil (allowing for purity of the compound) and mixed with the malt extract and water to achieve the desired nominal concentration. The dietary concentrations and the

cis:trans isomer content were determined by chemical analysis on ten sampling dates; the 500 ppm diet was analyzed from two additional diet preparations. The method of chemical extraction and analysis changed four times during the two-year course of the study; no data were presented on the comparable accuracy of the methods. Homogeneity analyses were apparently not performed. Stability analysis was apparently not performed in this study but stability in the diet over a four-week period had been confirmed in an earlier study at the same laboratory (Bradbrook et al., 1977).

Results –
Homogeneity Analysis: No results are presented.
Stability Analysis: No results are presented.

Concentration Analysis: The values for the twelve 500 ppm diet batches ranged from 69 to 125% of the nominal level with a mean value of 95% (3 values >10% different from nominal). For the 1000 ppm batches, the range was 77 to 117% of nominal with a mean of 98% but 7/10 values were >10% different from nominal. During the first 6 months of the study, 3/4 values from the 1000 ppm batches were <90% of nominal but the values tended to be higher during the remainder of the study. For the 2500 ppm batches, the range was 90 to 116% of nominal with a mean of 100%; only 2 values were >10% above nominal.

Dietary analyses gave no indication regarding stability or homogeneity, but generally indicated that the doses to the animals were within acceptable limits. A previous study indicated adequate stability in the diet.

5. Statistics

Most endpoints were evaluated by analysis of variance; organ weights were compared by analysis of variance and analysis of covariance with body weight. Where appropriate, Student's t-test was used. Mortality data were compared using the logrank test. Incidence data in the histopathology evaluations were analyzed using Fisher's exact test. Where severity scores were obtained, they were compared using Student's t-test.

C. METHODS

1. Observations

All animals were inspected daily. Abnormalities of clinical condition or behavior were recorded at first observation and at each weighing.

2 Body weight

Individual body weights were recorded on all animals initially, weekly for the first twelve weeks and at two-week intervals thereafter for the duration of the study.

3. Food consumption and compound intake

Food consumption was recorded for each cage of 4 females and 2 pooled cages of 2 males each weekly for the first twelve weeks and thereafter for each of the following weeks: 16, 20, 23-26, 28, 32, 37-40, 49-52, 63-66, 75-78, 89-92, and 101-104.

4. Water consumption

Water consumption was not recorded.

5. Ophthalmoscopic examination

The eyes of at least 3 males and 5 females per group were examined using an ophthalmoscope prior to and 15 minutes after the instillation of 0.5% tropicamide during weeks 78-80, 96-97, and 103-104.

6. Blood was collected from the tail vein from 8 males and 8 females per group pre-experimentally and at weeks 4, 13, 26, 39, 65, 78, and 91. Four females per group were bled at week 93 rather than week 91. At the 52 and 104-week kills, blood samples were taken by cardiac puncture from 8 males and 8 females per group immediately before necropsy. The CHECKED (X) parameters were examined.

a. Hematology

x	Hematocrit (HCT)*	x	Leukocyte differential count*
x	Hemoglobin (HGB)*	x	Mean corpuscular HGB (MCH)*
x	Leukocyte count (WBC)*	x	Mean corpusc. HGB conc. (MCHC)*
x	Erythrocyte count (RBC)*	x	Mean corpusc. volume (MCV)*
x	Platelet count*	x	Reticulocyte count
x	Blood clotting measurements*		Bone marrow smears
	(Thromboplastin time)	x	
	(Clotting time)		
	(Prothrombin time)		
x	(Kaolin-Cephalin index)		
x			

*Recommended for chronic toxicity/oncogenicity based on Subdivision F Guidelines.

b. Clinical chemistry

Tail vein blood samples were taken from subsets of 8 males and 8 females per group pre-experimentally and at weeks 4, 13, 26, 39, 52, 65, 78, and 91. At 104 weeks, cardiac puncture samples were taken from the survivors of the designated animals. The CHECKED (X) parameters were examined. Samples of liver from 4 males and 4 females per group were taken at autopsy at the 52 and 104 week kills. The samples were placed on ice prior to assay for hepatic aminopyrine-N-demethylase (APDM) activity.

x	Calcium	x	Albumin*
	Chloride		Albumin:globulin ratio
	Magnesium		Blood creatinine*
	Phosphorus	x	Blood urea nitrogen*
	Potassium*		Total cholesterol*
	Sodium*		Globulins
	ENZYMES	x	Glucose*
			Total bilirubin
	Alkaline phosphatase (ALK)*		Total serum protein*
	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase		
	Lactic acid dehydrogenase (LDH)		
x	Alanine aminotransferase (SGPT)*		
x	Aspartate amino-transferase (SGOT)*		
	Gamma glutamyl transferase (GGT)*		
	Glutamate dehydrogenase		

* Recommended for chronic toxicity/oncogenicity studies based on Subdivision F Guidelines; more than two hepatic enzymes should be measured.

7. Urinalysis

Eighteen-hour urine samples, pooled for each group and sex, were collected in containers on ice from 4 males and 4 females per group pre-experimentally and at weeks 13, 26, 39, 52, 65, 78, 91, and 103. Fresh urine was collected over a period of 1.5 hours from water-loaded rats (4 males and 4 females/group) at weeks 61-62 (pooled for each group and sex) and individually at weeks 79-80, 92-93, and 103-104 for examination of the sediment. The CHECKED (X) parameters were examined.

X		x	
x	Appearance*	x	Glucose
x	Volume*		Ketones
x	Specific gravity*	x	Bilirubin
x	ph*		Blood*
x	Sediment (microscopic)		Nitrate
x	Protein		Urobilinogen

*Recommendred for chronic toxicity/oncogenicity studies based on Subdivision F Guidelines.

8. Sacrifice and pathology

Moribund rats and surviving rats other than those used at 52 and 104 week kills for blood samples were anesthetized with halothane vapor and necropsied immediately after death. Rats designated for organ weight analyses and specified blood assays were exsanguinated by cardiac puncture before autopsy. The CHECKED (X) tissues in the table below were collected, preserved, and examined microscopically. The (XX) organs, in addition, were weighed. In a few rats that died, brain, spinal cord and gastrointestinal tract were not submitted because of autolysis. Hematoxylin and eosin staining was used routinely; special stains such as those for reticulin or collagen fibers were used as an aid to diagnosis where necessary. The left sciatic nerve of all rats killed at week 52 and of 5 rats/sex/group of those killed at week 104 was fixed in 3% v/v glutaraldehyde and embedded for electron microscopic examination. Extra

sections of the right sciatic nerve from the rats killed at 52 weeks were stained with Luxol fast blue-cresyl violet and in some cases by Palmgren's silver technique. Livers from 4 male and 4 female rats per group were taken for electron microscopic examination at week 52; at 104 weeks the number was increased to 6 rats/sex/group (including those used for hepatic APDM activity).

X	DIGESTIVE SYSTEM	X	CARDIOVASC/HEMAT.	X	NEUROLOGIC
	Tongue	X	Aorta*	XX	Brain*+
X	Salivary glands*	XX	Heart*+	X	Periph. nerve*
	Esophagus*		Bone marrow*	X	Spinal cord (3 levels)*
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*+	X	Eyes (retina, optic n.)*
X	Jejunum*	XX	Thymus*		
X	Ileum*				GLANDULAR
X	Cecum*		UROGENITAL		
X	Colon*			XX	Adrenal gland*+
	Rectum*	XX	Kidneys*+		Lacrimal gland
XX	Liver*+	X	Urinary bladder*	X	Mammary gland*
X	Pancreas*	XX	Testes*+		Parathyroids*
	Gallbladder*	X	Epididymides* +	X	Thyroids*
		X	Prostate*		
	RESPIRATORY	X	Seminal vesicle*		OTHER
		XX	Ovaries*+		
	Trachea*	X	Uterus*+		Bone
XX	Lung*		Oviduct	X	Skeletal muscle
	Nose*		Vagina		Skin*
	Pharynx*	X	Cervix	X	Ears
	Larynx*				All gross lesions and masses*

*Required for chronic toxicity/oncogenicity studies based on Subdivision F Guidelines.
+Organ weight required in chronic toxicity/oncogenicity studies.
*Thymus was weighed only at 52 weeks.

II. RESULTS

A. OBSERVATIONS

1. Toxicity

Slight whole-body tremors associated with hypersensitivity to localized noise and disturbance, and piloerection were noted for all 2500 ppm males during the first two weeks of the study. For the 2500 ppm females, tremors (43/60) and piloerection (29/60) were observed during the first three weeks and hypersensitivity was observed in 27/60 animals during the first two weeks of the study. One male showed tremors at week 44 and one female at week 8. These were considered to be treatment-related effects. No control animals showed these signs nor did any animal of other treatment groups with the exception of one low-dose female that had piloerection in week 2. Later in the study animals of all groups showed hypersensitivity and piloerection in the absence of tremors. These signs were not considered treatment-related. Yellow

staining of the fur in the genital area and brown staining of the tail occurred in all groups but affected more animals (males) and was more pronounced (males and females) in the treated groups. Two types of infection were seen in the animals during the course of the study but were not considered treatment-related; firstly, during weeks 2-5, conjunctivitis was noted for approximately 20% of males of all groups. Secondly, moderate respiratory distress was noted at about weeks 70 to 80 in up to 20% of the animals. Other clinical findings seen were common to this strain of rat, were of a minor nature, and were not treatment-related.

2. Mortality

No treatment-related effect on mortality was observed.

B. BODY WEIGHT

No evidence of a treatment-related effect on body weight was observed for either sex (Table 2). A small decrement in body weight gain was reported in treated groups during the first six weeks; this was statistically significant in some cases but not dose-related, less than 5% different from control values and thus within the range of biological variability. After this initial period, all treated groups of both sexes grew as well or better than controls.

TABLE 2. Mean body weights for male and female rats treated orally with Permethrin for 104 weeks, g, ± S.D.

Week	Male treatment groups, ppm				Female treatment groups, ppm			
	0	500	1000	2500	0	500	1000	2500
0	123.5 ±15.1	123.4 ±14.4	122.6 ±13.6	122.7 ±17.3	108.2 ±10.4	109.0 ±10.5	109.4 ±13.1	108.0 ±11.2
6	354.9 ±30.7	353.6 ±30.9	343.0 ±31.6	348.4 ±29.4	233.1 ±19.2	229.0 ±17.0	228.4 ±19.5	229.2 ±13.8
52	608.6 ±73.2	621.0 ±69.7	605.8 ±82.3	603.0 ±67.7	341.0 ±46.8	337.0 ±41.4	344.2 ±58.8	345.3 ±45.9
104	644.6 ±62.5	642.9 ±100.9	613.0 ±79.0	626.2 ±62.9	413.7 ±70.4	429.4 ±69.4	445.3 ±87.5	421.5 ±66.0

C. FOOD CONSUMPTION AND COMPOUND INTAKE

1. Food consumption

There were scattered differences from controls early in the study with a tendency to less food consumption in some of the treated groups, with the differences reaching statistical significance, but no biologically significant differences were observed.

2. Compound consumption

Compound consumption (Table 1) was estimated by the reviewer from the nominal concentration of test substance in the diets, Week 104 group mean food consumption data, and body weights.

3. Food efficiency

No treatment-related differences were observed in food efficiency over the first 12 weeks in either sex.

D. WATER CONSUMPTION

Water consumption was not monitored.

E. OPHTHALMOSCOPIC EXAMINATION

No treatment-related observations were reported.

F. BLOOD WORK

1. Hematology

There were no toxicologically significant treatment-related effects in either sex.

2. Clinical chemistry

There were no toxicologically significant treatment-related effects in either sex. Statistically significant reductions in plasma ALT activity in treated males and females at 39 weeks and in AST activity in female groups at 39, 65, and 78 weeks were attributed to high control mean values at these time periods due to a few abnormally high individual values.

A statistically significant increase in liver aminopyrine demethylase (APDM) activity was seen in treated animals at both 52 and 104 weeks (Table 3). This was evident in male rats of all treatment groups at 52 weeks and in 2500 ppm male group at 104 weeks; in females, the increase at 52 weeks was in the middle and high dose groups, while at 104 weeks, it was limited to the high-dose animals.

TABLE 3. Liver aminopyrine demethylase (APDM) activity, µmoles/ /g/hr, in rats treated orally with Permethrin for 104 weeks

Week	Male treatment groups, ppm				Female treatment groups, ppm			
	0	500	1000	2500	0	500.00	1000	2500
52.00	0.210	0.382** (182%)	0.408** (194%)	0.730** (348%)	0.042	0.058	0.112** (267%)	0.174** (414%)
104.00	0.057	0.102	0.134	0.316** (554%)	0.050	0.065	0.064	0.157** (314%)

Data taken from Table 31, p. 93, MRID 92142123.
**Significantly different from the control at the 1% level.
Numbers in parentheses are the percent of untreated controls, calculated by the reviewer.

G. URINALYSIS

No treatment-related changes were observed in urinalysis endpoints.

H. SACRIFICE AND PATHOLOGY

1. Organ weight

At the 52-week sacrifice, liver weights were significantly increased in the 2500 ppm male group and in the high and low-dose female groups when adjusted for body weight (Table 4). At the 104-week terminal sacrifice, absolute liver weights were significantly increased to about the same extent in all male treatment groups and in the 1000 ppm female group. The female relative liver weights were clearly elevated only in the 1000 ppm group at 104 weeks. Kidney weights were elevated in the high-dose males at 52 weeks and in the low and middle dose groups at 104 weeks. In females, however, the kidney weights of the 1000 ppm group were modestly but significantly decreased at 52 weeks; no differences from control values were evident at 104 weeks. Group mean terminal body weights were not presented and individual terminal body weights were not legible, so organ weights relative to terminal body weight could not be calculated by the reviewer.

TABLE 4. Liver and kidney weights of male and female rats fed Permethrin for 52 or 104 weeks					
Organ Weight	**Weeks**	**Male Treatment groups, ppm**			
		0	**500**	**1000**	**2500**
Liver (g)	52	19.112	20.212	19.511	22.0** (115%)
Liver (g)	104	18.725	20.9* (112%)	20.7* (111%)	21.2** (113%)
Kidney (g)	52	3.2612	3.3812	3.3911	3.56* (109%)
Kidney (g)	104	4.25	4.97* (117%)	5.03* (118%)	4.712
		Female treatment groups, ppm			
		0	**500**	**1000**	**2500**
Liver (g)	52	10.411	11.512	10.312	11.812
Liver (g)	104	12.83	14.034	14.8** (116%)	14.037
Kidney (g)	52	2.0811	2.1012	1.88*	1.9412
Kidney (g)	104	2.7131	2.6134	2.8036	2.6337

Data taken from Tables 46, 47, and 52, pp. 110-111, 116, MRID 92142123.
**Significantly different from control value at the 1% level.
Numbers in parentheses are the percent of untreated controls, calculated by the reviewer.
*Significantly different from control value at the 5% level.

2. Gross pathology

No treatment-related gross pathological changes were reported.

3. Microscopic pathology

a. Non-neoplastic

At 52 weeks, abnormalities were generally infrequent, minor, and characteristic for rats of this strain and age. However, liver changes were observed that included an increased incidence of vacuolated hepatocytes in the midzonal and centrilobular regions of the 2500 ppm male livers (Table 5). The vacuolation was of two types, one in which there were several vacuoles giving a foamy appearance, the other consisting of a single large vacuole with an indistinct or ragged margin. At 104 weeks, most abnormalities were again of a nature commonly seen in rats of this strain and age, or were scattered and not dose-related. Hypertrophy of centrilobular hepatocytes with increased cytoplasmic eosinophilia was elevated in the 1000 and 2500 ppm groups of both sexes, especially in the males. Vacuolization was also observed as at 52 weeks, with a significant increase over controls in the high-dose male and female groups. Using light microscopy, some of the vacuoles appeared to be fatty but could not be

reliably distinguished from anoxic vacuoles of the type produced during halothane euthanasia.

Electron microscopic examination of liver sections was performed and the results are presented in Table 6. At 52 weeks, only the 1000 and possibly the 2500 ppm males showed increased fatty vacuoles; none were seen in any female group. At 104 weeks, fatty vacuoles were again seen in both the mid- and high-dose males, although only 2/6 of the 1000 ppm males showed this effect. At this time point, the high-dose females' hepatocytes also had increased fatty vacuoles. Smooth endoplasmic reticulum proliferation increased in all treatment groups at week 52, but at week 104 an increase was seen only in the mid- and high-dose groups and was statistically significant only in the high-dose animals when evaluated quantitatively. Light and electron microscopic examination of sciatic nerve sections revealed no observable neurotoxicity.

TABLE 5. Incidences of treatment-related microscopic changes in male and female rats fed Permethrin for 52 or 104 weeks

Endpoint/N	Week	Male treatment groups, ppm			
		0	500	1000	2500
Liver: vacuolated hepatocytes	52	11/12	12/12	11/11	11/11
Occasional		10	11	8	4
Few		0	1	2	0
Several		1	0	1	6
Many		0	0	0	1
Liver: Number of animals with vacuolated hepatocytes (5 high-power fields)	52	8/12	8/12	9/11	10/11
Mean no. vacuolated hepatocytes per animal		2.0	1.9	2.3	22.6
Liver: centrilobular hypertrophy	104	0/25	2/28	8/28**	12/20**
Liver: vacuolated hepatocytes	104				
Occasional		6	7	5	2
Few		1	7	9	4
Several		4	4	3	4
Many		3	5	6	8
Female treatment groups, ppm					
Liver: vacuolated hepatocytes	52	6/11	11/12	12/12	8/12
Occasional		6	8	9	7
Few		0	2	3	1
Several		0	1	0	0
Many		0	0	0	0
Liver: Number of animals with vacuolated hepatocytes (5 high-power fields)	52	3/11	10/12	5/12	9/12
Mean no. vacuolated hepatocytes per animal		0.5	2.7	1.0	1.8
Liver: centrilobular hypertrophy	104	0/31	1/34	2/36	17/38**
Liver: vacuolated hepatocytes	104				
Occasional		12	11	9	11
Few		6	10	9	7
Several		4	4	5	3
Many		1	2	5	13

Data taken from Tables 60-62, 65, 66, pp. 124-128, 131-139, MRID 92142123.

**Significantly different from control value at the 1% level, calculated by reviewer using Fishers's exact test.

TABLE 6. Selected ultrastructural findings in livers of rats fed Permethrin for 52 or 104 weeks

Endpoint/N	Week	Male treatment groups, ppm			
		0	500	1000	2500
Proliferation of SER[a]	52	1/4	3/4	4/4*[b]	4/4*
	104	2/6	3/6	5/6	6/6*
Increased fat vacuoles	52	1/4	0/4	3/4	2/4
	104	0/6	0/6	2/6	4/6*
	Female treatment groups, ppm				
Proliferation of SER	52	1/4	4/4*	4/4*	4/4*
	104	2/6	3/6	5/6	6/6*
Increased fat vacuoles	52	0/4	0/4	0/4	0/4
	104	2/6	0/6	0/6	4/6

Data taken from Table 73 (p. 152), MRID 92142123.
[a] SER = smooth endoplasmic reticulum.
*Significantly different from control value at the 5% level.
[b] Incidence evaluated by Fisher's exact test or 2x2 Chi Square by reviewer.

b. Neoplastic

No treatment-related changes in the incidence of tumors of any type in male or female rats were reported.

III. DISCUSSION

A. INVESTIGATOR'S CONCLUSIONS

In a chronic toxicity/oncogenicity study, Permethrin was administered to 60 rats/sex/group in the feed at doses of 0, 500, 1000, or 2500 ppm for males or females. Of these animals, 12/sex/group were sacrificed at 52 weeks and those which survived were sacrificed at 104 weeks' exposure. The mean estimated (by the reviewer) compound intake in males was 0, 19.4, 36.9, or 91.5 mg/kg/day and in females was 0, 19.1, 40.2, or 104 mg/kg/day.

No treatment-related effects on mortality were observed during the study.

The only changes the study authors considered to be worthy of comment were the slight clinical effect consisting of tremors and hypersensitivity at 2500 ppm for male and female animals during the first two weeks of the study and liver changes.

The study authors considered the hepatocellular hypertrophy, increased liver weights, increased microsomal enzyme (APDM) activity, and endoplasmic reticulum proliferation to be indicators of an adaptive response to Permethrin exposure since other studies had shown these effects to be reversible. They considered anoxic vacuolation to be a non-specific phenomenon occurring around the time of death, particularly when using halothane to euthanize the animals. They noted fatty vacuoles, particularly in the high-dose male and female groups, but thought the significance to be uncertain, possibly part of the adaptive response or possibly a slight toxic effect associated with an alteration in the lipid metabolism of some cells. They apparently did not consider fat vacuoles in the 1000 ppm males at both 52 weeks and 104 weeks to be of any toxicological significance.

Increased staining of the fur in the genital area and tail was also associated with Permethrin exposure but was not considered an adverse effect by the authors but rather ascribed to excretion of the parent compound or a metabolite. Increased kidney weights in males were attributed to the nephropathy which occurs spontaneously in this age and strain of rats rather than to compound exposure.

Two year's exposure to Permethrin did not affect the incidence of tumor-bearing animals or the incidence of any specific tumor type in either sex.

The authors concluded that Permethrin was not carcinogenic to the rat, that it had low chronic toxicity even at 2500 ppm (half the oral LD_{50} in males), causing only slight tremors and hypersensitivity during the first two weeks' exposure and possibly slight toxicity associated with hepatocyte fat vacuolation at the highest dose. The adaptive liver responses were not considered adverse. A NOAEL of 1000 ppm for both sexes was identified.

B. REVIEWER'S DISCUSSION/CONCLUSIONS

The reviewer concurs with the study authors' analysis. On April 18, 2002, the HIARC evaluated the toxicology database of permethrin and determined that the hypertrophy of the liver is an adaptive and reversible effect and is not considered as an adverse effect. This conclusion is supported by a 90-day rat feeding study (MRID 00054737) where the hepatocellular hypertrophy was observed at 185 mg/kg/day with a NOAEL of 92.9 mg/kg/day.

Under the conditions of this study, the LOAEL is 2500 ppm (91.5 and 104 mg/kg/day for males and females, respectively) based on tremors and hypersensitivity. The NOAEL is 1000 ppm (36.9 and 40.2 mg/kg/day for males and females, respectively).

C. STUDY DEFICIENCIES

A number of deficiencies were noted. No flagging statement was provided. Although 60 rats/sex/group were studied, 12/sex/group were sacrificed at 52 weeks, leaving 48/sex/group for the full 104 week study, rather than the required 50/sex/group. However, survival was good and more than the minimum number of animals survived to 104 weeks for statistical purposes.

Analysis of the diet for homogeneity was not carried out, but the analyses for concentration were generally carried out in duplicate, giving some indication of homogeneity. Stability was not confirmed in the course of this study, having been demonstrated in a previous study in the same laboratory. Concentration analyses showed that at the 1000 ppm level, the actual concentrations tended to be >10% lower than nominal over the first 6 months of the study but higher thereafter.

No animals received ophthalmological examination prior to the onset of the study and few animals per group (3-5) were examined prior to termination rather than all high-dose and control animals.

Blood clinical chemistry and hematology determinations were performed on 8 animals/sex/group rather than at least 10, but the tests were performed every 3 months rather than the minimum 6-month intervals required by the guidelines. The required clinical chemistry endpoints of sodium, potassium, albumin, creatinine, cholesterol, and total serum protein were not performed and only two liver enzymes were determined rather than the required three or more. Urinalysis endpoints did not include the required appearance or blood determinations. Histopathology endpoints lacked the required esophagus, rectum, trachea, nose, pharynx, larynx, and skin. Only abnormal eyes were examined from animals that died or had to be sacrificed before Week 104, but at terminal sacrifice, eyes from all groups were preserved and examined. However, the interpretation of results does not appear to be affected by these deficiencies.

females at 52 weeks and in all male treatment groups and mid-dose females at 104 weeks. Further evidence for adaptive changes included hypertrophy of centrilobular hepatocytes with increased cytoplasmic eosinophilia in the mid- and high-dose male and females at 104 weeks' exposure and increased smooth endoplasmic reticulum proliferation in all treatment groups except low-dose males at 52 weeks and high-dose groups at 104 weeks. Liver changes also included fatty vacuoles that were confirmed by electron microscopy in the mid- and high-dose males at both 52 and 104 weeks and in the high-dose females at 104 weeks. The HIARC evaluated the toxicology database of permethrin and determined that the increased liver weight and hypertrophy observed in the liver are adaptive and reversible effects and are not considered adverse effects.

Under the conditions of this study, the LOAEL is 2500 ppm (91.5 and 104 mg/kg/day for males and females, respectively) based on tremors and hypersensitivity. The NOAEL is 1000 ppm (36.9 and 40.2 mg/kg/day for males and females, respectively).

At the doses tested, Permethrin did not affect the incidence of tumor-bearing animals or the incidence of any specific tumor type in either sex. Permethrin was not carcinogenic to the rat. Dosing was considered adequate based on liver effects and on tremors and hypersensitivity in male and female rats.

This chronic toxicity/oncogenicity study in the rat is **Acceptable/Guideline** and satisfies the guideline requirements for a chronic toxicity/oncogenicity oral study [OPPTS 870.4300 (§83-5a)] in the rat. No deficiencies were noted that would affect the interpretation of results.

COMPLIANCE: Signed and dated GLP, Data Confidentiality and QA statements were provided. The study was completed prior to the implementation of GLP and thus no claim was made as to its GLP compliance.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test material: Permethrin

   Description: isomeric cis:trans ratio nominally 40:60
   Lot/Batch No. and purity:

DATA EVALUATION RECORD
PERMETHRIN
(NRDC 143)

STUDY TYPE: SUBCHRONIC ORAL TOXICITY-RAT
Nonguideline
MRID 00054737

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
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Work Assignment No. 01-89

Primary Reviewer: Nancy B. Munro, Ph.D.	Signature: Date:	Robert H. Ross for Nancy B. Munro APR 04 2001
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Quality Assurance: Gary A. Sega, Ph.D.	Signature: Date:	Gary Sega APR 04 2001

Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract DE-AC05-00OR22725

PERMETHRIN, TECH./PC CODE 109701

Subchronic (6-Month) Oral Toxicity Study (rodents) / 1
OPPTS 870.3100/OECD 408

EPA Reviewer: Linnea Hansen, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
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Toxicology Branch, Health Effects Division (7509C)

Signature: Linnea T. Hansen
Date: January 25, 2002
Signature: Joycelyn E. Stewart
Date: 3/1/2002

TXR#: 0050649

DATA EVALUATION RECORD

STUDY TYPE: Six-Month Oral Toxicity (Dietary - rat); OPPTS 870.3100 [82-1a](rodent); OECD 408

P.C. CODE: 109701

DP BARCODE: D269531
SUBMISSION CODE: S504352

TEST MATERIAL: Permethrin, technical grade (93.3% a.i.)

SYNONYMS: 3-phenoxybenzyl dl, cis, trans 2,2-dimethyl-3-(2,2-dichlorovinyl) cyclopropane-1-carboxylate; NRDC 143

CITATION: Kadota, T. (1975) Six-month subacute oral toxicity of NRDC 143 in Sprague Dawley rats. Research Department, Pesticides Division of Sumitomo Chemical Co., Ltd., June 25, 1975. No study report number provided. MRID 00054737. Unpublished.

SPONSOR: Not reported but ICI Americas, Inc. listed on assignment sheet.

EXECUTIVE SUMMARY: In a subchronic oral toxicity study (MRID 00054737), Permethrin (tech., Lot # M-1, 93.3% purity) was administered in the diet to 16 Sprague Dawley rats/sex/dose group at dose levels of 0, 375, 750, 1500 or 3000 ppm (0, 22.5, 46.0, 92.9 or 185 mg/kg/day for males, and 0, 27.5, 52.3, 110 or 221 mg/kg/day for females) for 6 months.

At 3000 ppm, clinical observations revealed tremors and hypersensitivity which were observed in both males (16/16) and females (at least 6/16) beginning on day 1 of treatment with both the number of animals and the severity of symptoms lessening subsequently. No individual animal data were presented; thus, it is not possible to determine the total number of females that were symptomatic in week 1. All indications of this effect disappeared by week 6 in males and week 7 in females. Absolute liver weights were increased significantly (23 and 20% greater than controls, respectively) in both males and females. The liver-to-body-weight ratio was likewise increased significantly in both sexes (24% higher than controls in both males and females). The group mean liver-to-brain-weight ratios were compared and these ratios were similarly increased in the high-dose male and female groups (23 and 17%) over control values. These changes correlated with histopathological evidence of slight hypertrophy (6 males, 7 females) and slight fatty changes in the livers of 2 high-dose males and 3 high-dose females. Statistically significant small increases in cholesterol levels in the high-dose males and 1500 and 3000 ppm female groups were within

normal range and not considered toxicologically significant, although the increase may also reflect a borderline disturbance in liver metabolism. The HIARC determined that the hypertrophy of the liver is an adaptive and reversible effect and is not considered as an adverse effect. There were no treatment-related effects on survival, body weight, food consumption, hematology, ophthalmological parameters or gross observations. **The LOAEL is 3000 ppm (185 mg/kg/day, females and 221 mg/kg/day, males), based on transient tremor and hypersensitivity in both sexes (the clinical signs beginning on day 1). The NOAEL is 1500 ppm (92.9 mg/kg/day, females and 110 mg/kg/day, males).**

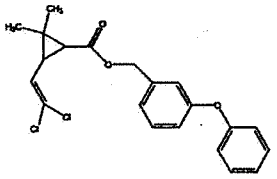
This subchronic oral toxicity study in rats is classified as **Unacceptable/guideline (upgradable)** as a subchronic feeding study in the rodent. As reported, this study does not fulfill FIFRA guideline requirements for a subchronic oral toxicity study [870.3100 (§82-1a)] in the rat; however, it may be upgraded to Acceptable/Guideline upon submission of the following information: (1) Homogeneity, concentration and stability data on diet preparations and (2) confirmation of animal fate and that each animal was examined microscopically. The permethrin chronic toxicity/carcinogenicity study in the rat may be used to satisfy this guideline requirement.

COMPLIANCE: Signed and dated GLP, Quality Assurance, Flagging and Data Confidentiality statements were not provided. This study was conducted prior to promulgation of the EPA GLP Guidelines.

I. MATERIALS AND METHODS

A. MATERIALS

1. **Test compound:** Permethrin
   Description: cis/trans ratio ~40/60
   Lot No.: M-1
   Purity: 93.3%
   Stability of compound: Unspecified
   CAS No: 52645-53-1
   Structure:



2. **Vehicle and/or positive control:** Powdered basal diet (CE-2, Nohon Crea Co.) was used as the vehicle. The chemical was dissolved in corn oil; the final concentration of corn oil in the diet was 2%; the control group diet (0 ppm) contained 2% corn oil.

3. **Test animals**
   Species: rat
   Strain: Sprague Dawley
   Age/weight at study initiation: five weeks; males: 107-161 g; females: 97-141 g
   Source: Shizuoka Jikkendobutsu Kyodokumiai (Animal Breeding Division of Shizuoka Agricultural Cooperative Association)
   Housing: In groups of four per aluminum cage, 35 x 42 x 20 cm.
   Diet: Powdered basal diet (CE-2, Nohon Crea Co.) containing 2% corn oil, *ad libitum*
   Water: Source unspecified, *ad libitum*
   Environmental conditions:
   Temperature: 24 ± 1 °C.
   Humidity: 60 ± 10%
   Air changes: Not provided
   Photoperiod: Not provided
   Acclimation period: One week

B. STUDY DESIGN

1. **In life dates:** Start: not specified; end: not specified.

2. **Animal assignment**: Animals were assigned to the tet groups noted in Table 1, below:

TABLE 1: Study Design

TABLE 1. Study design					
Dose Group	Concentration in Diet (ppm)	Dose (mg/kg/day)		Number of Rats	
		Male	Female	Male	Female
Control	0	0	0	16	16
	375	22.5	27.5	16	16
	750	46.0	52.3	16	16
	1500	92.9	110	16	16
	3000	185	221	16	16

Data taken from p. 5 and Table 5, p. 18, MRID 00054737.

3. **Dose selection rationale**

A dose selection rationale was not provided.

4. **Diet preparation and analysis**

The test substance was dissolved in corn oil at the above concentrations such that the final concentration of corn oil in the diet was 2%. It was mixed into the diet with a ribbon mixer. The diet was freshly prepared weekly and refrigerated prior to use. No mention is made of sampling and analysis for homogeneity or stability.

5. **Statistical analysis**

Mean value and standard errors were calculated; the results were analyzed by t-test.

C. METHODS

1. **Observations**

The animals were observed daily for behavior and mortality.

2. **Body weight**

Body weights were recorded weekly.

3. **Food consumption and compound intake**

Food and water consumption were recorded weekly.

4. **Ophthalmoscopic examination**

Ophthalmic examinations were not performed.

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5. **Blood was collected** via the abdominal aorta after a 24-hour fast under ether anesthesia at termination of feeding (end of 6 months' exposure) from all rats in the control, 1500 and 3000 ppm groups. The CHECKED (X) parameters were examined.

a. **Hematology**

X		X	
X	Hematocrit (HCT)*		Leukocyte differential count*
X	Hemoglobin (HGB)*		Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*		Mean corpusc. HGB conc.(MCHC)*
X	Erythrocyte count (RBC)*		Mean corpusc. volume (MCV)*
X	Platelet count*		Reticulocyte count
	Blood clotting measurements*		Blood cell morphology
	(Activated thromboplastin time)	X	Sedimentation rate
	(Clotting time)		
	(Prothrombin time)		

*Required for subchronic studies based on Subdivision F Guidelines.

b. Clinical chemistry

X	ELECTROLYTES	X	OTHER
X	Calcium*	X	Albumin*
	Chloride		Albumin/globulin ratio
	Magnesium		Blood creatinine*
	Phosphorus*	X	Blood urea nitrogen*
X	Potassium*	X	Total cholesterol*
X	Sodium*		Globulins
		X	Glucose*
	ENZYMES	X	Total bilirubin
X	Alkaline phosphatase (ALK)*	X	Total serum protein*
X	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase		Serum protein electrophoresis
X	Lactic acid dehydrogenase (LDH)*		
X	Alanine aminotransferase (also SGPT)*		
X	Aspartate aminotransferase (also SGOT)*		
	Gamma glutamyl transferase (GGT)		
	Glutamate dehydrogenase		
X	Leucine aminopeptidase		

* Required for subchronic toxicity studies based on Subdivision F Guidelines.

6. Urinalysis

Urinalysis was carried out after 3 and 6 months' exposure on 8 males and 8 females of the 0 and 3000 ppm groups. The study report did not indicate whether animals were water-deprived prior to collection.

X		X	
	Appearance	X	Glucose
	Volume	X	Ketones
	Specific gravity	X	Bilirubin
	pH	X	Blood
	Sediment (microscopic)		Nitrites
X	Protein	X	Urobilinogen

7. Sacrifice and pathology

At the end of 6 months' exposure, all animals were necropsied; the major organs were grossly examined. The CHECKED (X) tissues in the table below were microscopically examined in animals from the 0, 1500 and 3000 ppm groups. The (XX) organs, in addition, were weighed.

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X	DIGESTIVE SYSTEM	X	CARDIOVASC./HEMAT.	X	NEUROLOGIC
	Tongue		Aorta*	XX	Brain*+
X	Salivary glands*	XX	Heart*+		Periph. nerve*
X	Esophagus*	X	Bone marrow*	X	Spinal cord (3 levels)*
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*+	X	Eyes (optic n.)*
X	Jejunum*	XX	Thymus*+		
X	Ileum*				GLANDULAR
X	Cecum*		UROGENITAL	XX	Adrenal gland*+
X	Colon*	XX	Kidneys*+		Lacrimal gland
	Rectum*	X	Urinary bladder*		Mammary gland*
XX	Liver*+	XX	Testes*+		Parathyroids*
X	Pancreas*		Epididymides*+	XX	Thyroids*
	Gallbladder*	X	Prostate*		Coagulation glands
			Seminal vesicle*		
	RESPIRATORY	XX	Ovaries*+		OTHER
X	Trachea*	X	Uterus*+		Bone
XX	Lung*		Vagina		Skeletal muscle
	Nose (nasal turbinates)*				Skin*
	Pharynx*				All gross lesions and
	Larynx*				masses*

*Required for subchronic studies based on Subdivision F Guidelines.

II. RESULTS

A. OBSERVATIONS

No deaths occurred during the study. Hypersensitivity (not described further) and tremor were observed in up to 16/16 males and up to 6/16 females in the 3000 ppm dose group on any given day in the first week of exposure. The number of animals affected and the severity of symptoms decreased subsequently and disappeared by week 6 in males and week 7 in females. Individual animal data were not presented; it therefore cannot be determined how many female animals in total exhibited clinical signs in the first week.

B. BODY WEIGHT AND BODY WEIGHT GAIN

No statistically or biologically significant differences from controls were observed in body weight or body weight gain in either males or females at any dose level.

C. FOOD CONSUMPTION AND COMPOUND INTAKE

1. Food Consumption

No statistically or biologically significant differences from controls were observed in food consumption in either sex at any dose level.

2. **Test Compound Intake**

Test compound intake was estimated weekly based on nominal diet concentration, food consumption, and body weight, and the 6-month averages are given in Table 1.

3. **Food Efficiency**

Food efficiency data were not included in the study report.

D. OPHTHALMOSCOPIC EXAMINATION

Ophthalmoscopic examination was not performed.

E. BLOOD WORK

1. **Hematology**

At 6 months, in males of the 1500 ppm group, the hematocrit and hemoglobin levels were decreased 5.5% and 5.3% respectively below control values but without a corresponding decrease in erythrocyte count. The leukocyte count was decreased 26% in the same group. These changes, although statistically significant are not considered biologically significant or treatment-related due to a lack of dose-response and the small magnitude of the changes. In the 1500 ppm female group, the hematocrit was increased by 4.6% without corresponding changes in hemoglobin or erythrocyte count. In the high-dose female group, the leukocyte count was increased 19%. These changes also were not statistically significant and not considered treatment-related or biologically significant.

2. **Clinical chemistry**

Cholesterol levels were reported only for the 0, 1500 and 3000 ppm groups. In the male 1500 and 3000 groups, levels were elevated by 15% and 21%, respectively. In the corresponding female groups, cholesterol was elevated significantly at 1500 ppm (24%) and 3000 ppm (17%). These changes are within the range of biological variability. There were other statistically significant changes in various parameters which were not considered biologically significant due to the small magnitude of the change, the parameters remaining within the normal reference range, not showing a dose response relationship and/or being in a direction lacking biological significance.

F. Urinalysis

No differences in the 3000 ppm group from control rats were reported for any parameter.

III. SACRIFICE AND PATHOLOGY

A. Organ Weight

Liver weight data are given in Table 2. Absolute liver weights were increased significantly in both the high-dose male and female groups, by 23 and 20% over the controls, respectively, as calculated by the reviewer. The liver-to-body-weight ratio was likewise increased significantly in both sexes at 3000 ppm, being 24% higher than the control value in both males and females. The group mean liver-to-brain-weight ratios were compared by the reviewer and these ratios were similarly increased in the high-dose male and female groups (23 and 17%, respectively) over control values.

TABLE 2. Mean liver weight data from rats administered Permethrin in the diet[a]

Parameter	Dietary Exposure Level (ppm)				
	0	375	750	1500	3000
Males					
Terminal body weight (g)	604±10.3	598±17.6	607±13.3	615±16.7	605±11.5
Liver: absolute wt. (g)	14.7±0.46	14.3±0.66	14.4±0.40	15.6±0.53	18.1±0.51** (23)[b]
Liver-to-body -wt. ratio (%)	2.42±0.04	2.38±0.04	2.37±0.04	2.37±0.16	2.99±0.05** (24)
Liver-to-brain-wt. ratio[c]	7.1	7.2	7.0	7.8	8.7 (23)
Females					
Terminal body wt.	314±8.69	309±9.12	296±7.86	310±7.93	307±7.82
Liver: absolute wt. (g)	6.93±0.23	6.50±0.25	6.75±0.25	7.20±0.29	8.30±0.20** (20)
Liver-to-body-wt. ratio (%)	2.19±0.04	2.20±0.05	2.29±0.05	2.32±0.07	2.71±0.03** (24)
Liver-to-brain-wt. ratio	3.67	3.65	3.61	3.93	4.30 (17)

Data taken from Tables 9 and 10, pp. 23-25, MRID 00054737.
[a]Data are expressed as mean ± standard error with n=16.
[b]Numbers in parentheses equal percent change from control value, calculated by reviewer.
**Significantly different from controls, $p<0.01$.
[c]Comparison of group means, calculated by reviewer.

2. Gross pathology

No abnormal gross findings were reported at necropsy except for slight enlargement of the livers of the 3000 ppm groups.

3. Histopathology

Selected histopathologic findings are given in Table 3. Although it is presumed that all animals in the 0, 1500 and 3000 ppm groups of this study were examined for microscopic findings, individual animal fate data or pathology reports from animals with no microscopic findings were not included in this report to verify that all the animals within those dose groups were examined. At the 3000 ppm dose level, slight hypertrophy of liver parenchymal cells were noted. In the high-dose males, 6 animals showed this effect; of those, 2 also showed slight fatty changes. In the high-dose female group, 7 animals had slight hypertrophy and an additional 3 animals showed fatty changes in the liver. No scoring of severity of these changes was provided. No fatty change was observed in controls or at lower dose levels. No histopathologic changes were reported in other tissues other than pneumonia and lung abscesses and cell infiltration in kidney interstitial and pancreatic tissue. These gave no evidence of being treatment-related.

TABLE 3: Treatment-related microscopic findings in the liver[a]

Organ/Description of finding	Dietary Exposure Level (ppm)									
	Males					Females				
	0.00	375	750	1500	3000	0.00	375	750	1500	3000
Liver: megalocytic changes of parenchymal cells	0	–	–	0	6	0	–	–	0	7
Liver: fatty changes of parenchymal cells	0	–	–	0	2	0	–	–	0	3
Total animals with changes	0	–	–	0	6	0	–	–	0	10

Data taken from Appendix VIII, pp. 100-102, MRID 00054737. N is presumed to be 16/sex/dose group except for the 375 and 750 ppm animals, which were not examined (-).

III. DISCUSSION

A. INVESTIGATORS' CONCLUSIONS: The study authors determined that male and female rats administered permethrin at 3000 ppm in the diet showed treatment-related effects. Clinical signs included tremors and hypersensitivity which began on the first day of dosing, but did not persist beyond the 7th week. Liver hypertrophy, resulting in mildly increased liver weights, was accompanied by a slight fatty change in some animals of each sex, but there were no significant alterations in clinical chemistry or urinalysis parameters. The slight increases in serum cholesterol at 1500 and 3000 ppm were not considered treatment-related. No treatment-related findings were observed at 1500 ppm or below.

B. REVIEWER'S COMMENTS: The reviewer agreed with the conclusions of the study authors. All animals survived to study termination and gross necropsy was unremarkable. Clinical observations included tremors and hypersensitivity which were observed only at the highest dose level (3000 ppm) in both males (16/16) and females (at least 6/16) during the first week of exposure with both the number of animals affected and the severity of

symptoms lessening subsequently. No individual animal data were presented; thus, it is not possible to determine the total number of females that were symptomatic in week 1. All indications of this effect had disappeared by week 6 in males and week 7 in females. No neurotoxicity testing was performed and peripheral nerves were not examined microscopically.

Absolute and relative (to body and to brain) liver weights were increased in both the highest-dose male and female groups, by 17 to 24% greater than controls. These changes were correlated with histopathological evidence of slight hepatocellular hypertrophy which together suggest an adaptive response to xenobiotic chemical exposure. On April 18, 2002, the HIARC evaluated the toxicology database of permethrin and determined that the hypertrophy of the liver is an adaptive and reversible effect and is not considered as an adverse effect. No major disturbances of serum chemistries were observed. Slight increases in cholesterol levels (15-24% above controls; both sexes) were within the range of biological variability but may also reflect altered fat metabolism.

Under the conditions of this study, the subchronic toxicity LOAEL is 3000 ppm (185 and 221 mg/kg/day for males and females, respectively), based on transient clinical signs including tremor and hypersensitivity in both sexes. The NOAEL for both sexes is 1500 ppm (92.9 and 110 mg/kg/day for males and females, respectively).

This subchronic oral toxicity study in rats is classified as Unacceptable/guideline (upgradable) for the reasons outlined below. As reported, this study does not fulfill FIFRA guideline requirements for a subchronic oral toxicity study [OPPTS 870.3100 (§82-1a)] in the rat; however, it may be upgraded to Acceptable/Guideline upon submission of the following information: (1) Homogeneity, concentration and stability data on diet preparations and (2) confirmation of animal fate and that each animal was examined microscopically. The permethrin chronic toxicity/carcinogenicity study in the rat may be used to satisfy this guideline requirement.

C. STUDY DEFICIENCIES:

This study was conducted prior to promulgation of the US EPA GLP guidelines and has several deficiencies, due in part to its age. No analyses for stability, homogeneity and concentration were reported. The test diets were prepared weekly, so stability may not have been a problem. Results were clear cut between the 3000 and 1500 ppm dose groups, so diet homogeneity was likely reasonable. The lack of concentration analyses leaves the calculated compound intakes uncertain. No ophthalmoscopic examinations were performed on the animals. Although the pathology data table shows findings for individual animals, there is no confirmation of animal fate and gross or microscopic examination for each individual rat, because those rats with no findings were not listed in the table. The dates of in-life conduct of the study are also not provided. Signed and dated GLP, Quality Assurance, Flagging and Data Confidentiality statements were not provided.

Minor deficiencies in the hematology and clinical chemistry panel included the absence of a differential leukocyte count, a blood clotting measurement, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration and mean corpuscular volume. The following required tissues for histopathological examination were not included: rectum, nose, pharynx, larynx, aorta, epididymides, seminal vesicles, peripheral nerve, mammary gland, skin, and gross lesions and masses. However, the study provides useful information on the effects of permethrin in rats over a six-month exposure period.

DATA FOR ENTRY INTO ISIS

ıbchronic (6-month) Oral Study - rodents (870.3100)

?C code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
09701	00054737	subchronic	rat	6-months	oral	dietary	22.5-221	0, 22.5, 46.0, 92.9, 185 ♂; 0, 27.5, 52.3, 110, 221 ♀	92.9	185	neuromuscular system, liver	Toxicity

DATA EVALUATION RECORD

PERMETHRIN/109701

STUDY TYPE: PRENATAL DEVELOPMENTAL TOXICITY STUDY - RAT
OPPTS 870.3700a [§83-3a]; OECD 414.
MRID 00043724 (Main Study) and 00057099

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Work Assignment No. 02-05

Primary Reviewer: Donna L. Fefee, D.V.M.	Signature: Date:	RHRoss for D.L. Fefee APR 0 5 2002
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Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross APR 0 5 2002
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson APR 0 5 2002

Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

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EPA Reviewer: Yung G. Yang, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/10/2002
Signature: Joycelyn Stewart
Date 5/27/2002

DATA EVALUATION RECORD

TXR#: 0050649

STUDY TYPE: Prenatal Developmental Toxicity Study - Rat; OPPTS 870.3700a [§83-3a]

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): ICI-PP 557 (Permethrin, 95.3% a.i.)

SYNONYMS: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; permethrin

CITATION: McGregor, D. and G. Wickramaratne (1976) Teratogenicity study in rats of ICI-PP 557. Inveresk Research International, Edinburgh, EH21 7UB, Scotland. Report No. 439, February 1976. MRID 00043724. Unpublished.

James, D. (1974) Preliminary foetal toxicity study in the rat given 21Z73 (NRDC 143) orally. Wellcome Research Laboratories, Beckenham, UK. Laboratory report number Path 167, May 28, 1974. MRID 00057099. Unpublished.

SPONSOR: Imperial Chemical Industries Ltd., Alderley Park, Macclesfield, Cheshire SK10 5TJ.

EXECUTIVE SUMMARY: In a developmental toxicity study (MRID 00043724) ICI-PP 557 (Permethrin; 95.3% a.i., cis-/trans-ratio 37.5/57.8, batch # 25) was administered to timed-mated female CD rats (20/dose) in corn oil by gavage at dose levels of 0, 22.5, 71.0, or 225.0 mg/kg bw/day from days 6 through 16 (inclusive) of gestation. On GD 20, all surviving dams were sacrificed and necropsied, and all fetuses were weighed, sexed, and examined externally. Approximately one-half of the fetuses from each litter were examined for visceral alterations, and the remaining fetuses were subjected to skeletal examination.

There were no treatment-related effects on survival, maternal body weights and food consumption, or cesarean section parameters, and clinical signs were not reported. Although there was no reported evidence of maternal toxicity at the highest dose level tested, dosing was still considered adequate because dose selection was appropriate based the results of a range-finding study, and a separate developmental toxicity study (MRID 00057099) in Wistar rats. **The maternal toxicity LOAEL was not identified and the maternal toxicity NOAEL of ICI-PP 557 in CD rats was ≥225 mg/kg/day.**

There were no treatment-related increases in fetal deaths/resorptions or incidences of

growth. **The developmental toxicity LOAEL of ICI-PP 557 in CD rats is not identified, and the developmental toxicity NOAEL is greater than or equal to 225 mg/kg bw/day.**

The developmental toxicity study in the rat is classified **unacceptable/guideline (upgradeable).** The study report did not describe the method of dosing formulation preparation and that concentration and homogeneity analyses were not done; it is therefore not known whether the mixing procedure was adequate or whether the variance between nominal and actual dosages to the animals was acceptable.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided; however, the study was conducted in 1976.

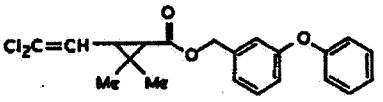
I. MATERIALS AND METHODS

A. MATERIALS::

1. Test Material: ICI-PP 557

Description:	straw-colored solid, with an ammoniacal odor
Lot/Batch #:	25
Purity:	95.3% (cis/trans ratio: 37.5/57.8)
Compound Stability:	not provided
CAS #of TGAI:	52645-53-1

Structure:



2. Vehicle and/or positive control: The vehicle control article was corn oil (*Mazola*, Brown and Polson, Esher, Surrey, England). No positive control was used in this study.

3. Test animals:

Species:	Rat	
Strain:	CD	
Age/weight at study initiation:	assumed to be young adult; 155-255 g. at the start of dosing	
Source:	Charles River (UK) Ltd., Manston, Kent, England	
Housing:	individually in polypropylene and stainless steel cages	
Diet:	Spiller's autoclaved laboratory small animal diet No. 1 *ad libitum*	
Water:	local tap water *ad libitum*	
Environmental conditions:	**Temperature:**	20±2°C
	Humidity:	50%
	Air changes:	not reported
	Photoperiod:	not reported
Acclimation period:	3 days prior to initiation of dosing	

B. PROCEDURES AND STUDY DESIGN:

1. **In life dates** - not reported

2. **Mating:** Timed mated females were supplied on gestation day (GD) 2.

3. **Animal Assignment:** Animals were randomized by an unspecified method and assigned to dose groups as indicated in Table 1. At the initiation of dosing, the group mean body weights and their variances were homogeneous (Bartlett's test and Dunnett's comparison performed by reviewer).

TABLE 1. Animal assignment

Dose (mg/kg bw/day)	0	22.5	71	225
# Females	20	20	20	20

Data taken from text, p. 198, MRID 00043724.

4. **Dose selection rationale:** The dose levels were selected based on the results of a preliminary range-finding study in the rat (MRID 00057099). A single group of 6 female rats was dosed orally for successive 5-day periods with increasing doses of the test material. Dose levels of 100, 150, 225, and 338 mg/kg/day were used. On day 12 (the third day at the 225 mg/kg/day dose level), one rat (#2) exhibited persistent trembling and had blood around the eyes and nose. On day 15 (the first day at the 338 mg/kg/day dose level) after dosing, four additional animals (Nos. 1, 4, 5, and 6) were trembling. On day 16 rat #2 continued to have blood around its eyes and exhibited a general loss of muscle tone and a highly exaggerated startle response, and animals 1, 4, 5 and 6 exhibited persistent trembling and exaggerated startle responses. The study was terminated on day 16. Animal #3 appeared normal throughout the test, and there were no consistent weight losses or reductions in food consumption observed. Based on these results, the dose levels chosen for the main study were 0, 22.5, 71.0, and 225.0 mg/kg/day.

5. **Dosage preparation and analysis:** Test material-vehicle mixtures were prepared daily; however, the preparation method was not described in the study report. There was also no mention of evaluation of the stability of the test substance in the vehicle or concentration and homogeneity of the test mixtures.

   There were no analytical data provided to indicate whether the mixing procedure was adequate or that the variance between nominal and actual dosage to the study animals was acceptable.

6. **Dosage administration:** All doses were administered once daily by gavage, on gestation days 6 through 16 (inclusive), in a volume of 10 mL/kg of body weight/day. The study report did not state whether dosing was based on the body weight from the most recent body weight determination; however, this is assumed to be the case as the animals were weighed daily during dosing.

C. OBSERVATIONS:

1. **Maternal observations and evaluations**: The animals were observed for clinical signs and mortality once a day. Body weights were recorded upon arrival from the supplier on GD 2, daily during GD 6-16, and at termination on GD 20. Daily food consumption was recorded for GD 7-16. Dams were sacrificed on GD 20 by intracardiac injection of pentobarbitone. The reproductive tract was excised, and the number of corpora lutea and the numbers and locations of all live fetuses and early and late deaths were recorded. There was no mention of gross necropsy of the dams. Gravid uterine weights were not recorded.

2. **Fetal evaluations**: The fetuses were examined in the following manner. Individual fetal and placental weights were recorded, then fetuses were subjected to external examination. Approximately one-third of the fetuses from each litter were fixed in Bouin's solution, then processed for visceral examination (including heads) by the Wilson method. The remaining fetuses were fixed in alcohol, eviscerated, stained with Alizarin Red S, then subjected to skeletal examination. The study author classified fetal structural alterations as either "abnormalities" or "variants" without providing further definitions of these categories or the criteria used for categorizing fetal structural alterations. The reviewer is assuming that "abnormalities" and "variants" roughly correspond to malformations and variations, respectively.

D. DATA ANALYSIS:

1. **Statistical analyses:** With the following two exceptions, it was not clear whether or not any of the results were subjected to statistical analysis. The study authors stated that "it was considered inappropriate to apply tests for statistical significance to the comparison" of the mean fetal weights of the control and high-dose groups since the sum of their standard deviations was greater than the difference between the two means. The data for the numbers of fetuses exhibiting hydronephrosis or hydroureter in the control and high-dose groups were subjected to a statistical test for bilateral characters [C.A.B. Smith, Appendix to Gruneberg, H. (1955) J Genetics, *53*:515-535].

2. **Indices:** The following indices were calculated by the reviewer from cesarean section records of animals in the study:

   Preimplantation loss (%) = (total number of corpora lutea minus total number of implantations/total number of corpora lutea) x 100.

   Postimplantation loss (%) = (total number of dead implantations/total number of implantations) x 100.

3. **Historical control data:** Historical control data were not provided to allow comparison with concurrent controls.

II. RESULTS:

A. MATERNAL TOXICITY:

1. **Mortality and clinical observations:** There were no maternal deaths or abortions during the study. The clinical signs of the dams throughout the study were not reported.

2. **Body weight:** Body weight data are summarized in Table 2. Mean absolute body weights throughout gestation and the estimated adjusted mean terminal body weight of the treated and control groups were similar. Increased body weight gains during treatment and increased estimated net body weight gains by the mid- and high-dose dams were not considered treatment-related, as a dose-response pattern was not evident.

   Data taken from Tables 2(a), 3, and 4, pp. 205, 207, and 208, respectively, MRID 00043724.

TABLE 2. Mean (±SD) maternal body weights and body weight gains (g)

Gestation day	Dose in mg/kg bw/day (# of Dams)			
	0 (16)	22.5 (16)	71.0 (15)	225.0 (16)
Absolute Body Weights				
6	207±14	203±16	200±26	208±21
12	243±18	240±20	246±25	248±22
16	261±23	261±21	264±25	272±24
20	303±24	297±28	319±28	314±25
Adjusted body weight[a]	273	273	287	284
Body Weight Changes[b]				
6-16	54	58	64 (119)[c]	64 (119)
16-20	42	36 (86)	55 (131)	42
6-20	96	94	119 (124)	106 (110)
Net body weight gain[d]	66	70	87 (132)	76 (115)

Data taken from Tables 2(a), 3, and 4, pp. 205, 207, and 208, respectively, MRID 00043724.
[a] Estimated by reviewer as Adjusted body weight = terminal body weight minus [mean live implantations per dam x (mean fetal weight + mean placental weight)].
[b] Calculated by reviewer using mean absolute body weight data. Not subjected to statistical analysis.
[c] Number in parentheses is percent of control; calculated by reviewer.
[d] Estimated by reviewer as Net body weight gain = Adjusted body weight minus GD 6 body weight.

3. **Food consumption:** There were no treatment-related effects on maternal food consumption. Increased mean food consumption by the mid- and high-dose groups correlated with the previously mentioned increased body weight gains by these groups.

4. **Gross pathology:** There were no gross pathology data reported.

5. Cesarean section data: Data collected at the scheduled cesarean section are summarized in Table 3. There were no abortions, early deliveries, or total litter resorptions. There were no significant differences between the treated and control groups in mean numbers of corpora lutea, implantation sites, and live fetuses per litter, pre- and postimplantation losses, early or late deaths fetal deaths, or fetal weights.

TABLE 3. Cesarean section observations [mean values±SD, as appropriate]

Observation	Dose (mg/kg bw/day)			
	0	22.5	71	225
# Animals Assigned (Mated)	20	20	20	20
# Animals Pregnant	16	16	15	16
Pregnancy Rate (%)	80	80	75	80
# Nonpregnant	4	4	5	4
Maternal Wastage				
# Died	0	0	0	0
# Aborted	0	0	0	0
# Premature Delivery	0	0	0	0
Total # Corpora Lutea [a]	194	174	177	191
Corpora Lutea/Dam	12.1±2.1	10.9±3.1	11.8±2.5	11.9±2.4
Total # Implantations	172	147	156	161
Implantations/Dam	10.6±1.8	9.1±2.5	10.4±2.7	10.1±2.7
Total # Litters	16	16	15	16
Total # Live Fetuses	164	131	153	153
Live Fetuses/Dam	10.3±1.7	8.1±3.0	10.2±2.7	9.6±3.0
Total # Dead Implantations	8	16	3	8
Early deaths	8	14	3	7
Late deaths	0	2	0	1
Dead Implantations/Dam [b]	0.5±0.5	1.0±1.8	0.2±0.4	0.5±0.7
Early deaths	0.5±0.5	0.9±1.4	0.2±0.4	0.4±0.6
Late deaths	0	0.1±0.5	0	0.1±0.3
Litters with Total Resorptions	0	0	0	0
Mean Fetal Weight (g) [c]	2.40±0.27	2.49±0.27	2.57±0.24	2.61±0.30
Sex Ratio (Male/Female)	0.98	0.75	0.96	1.19
Preimplantation Loss [d] (%)	11.3	15.5	11.9	15.7
Postimplantation Loss [e] (%)	4.7	10.9	1.9	5.0

Data taken from Tables 3 and 4, and Appendix Tables 5-8, pp 207, 208, and 215-222, respectively, MRID 00043724.

[a] One to three dams per group had one fewer corpora lutea recorded than the number of implantation sites. These data were still included in calculating pre- and postimplantation losses.

[b] Calculated by reviewer using individual data.

[c] Reported only for the combined sexes.

[d] Calculated by reviewer as Preimplantation Loss = [(Total corpora lutea-Total implantations)/T

[c] Calculated by reviewer as Postimplantation Loss = (Total dead implantations/Total implantations) x 100.

B. DEVELOPMENTAL TOXICITY: The total numbers of fetuses (litters) in the 0, 22.5, 71.0, and 225.0 mg/kg/day groups were 164 (16), 131 (16), 153 (15), and 153 (16), respectively. The study report did not include litter incidences of fetal structural alteration, and fetal individual data were provided only for malformations. Selected fetal morphological observations are given in Tables 4a-4c.

1. **External examination:** There was no mention of any external malformations or variations; however, one fetus in the control group had a filamentous tail, which was reported as either a skeletal malformation (text, p. 201) or a visceral malformation (Table 5, p. 208 and Appendix Table 6, p. 216). It is assumed that this malformation was first observed at external examination, and the reviewer reported it as an external malformation.

2. **Visceral examination**: Visceral malformations included hydronephrosis in 5/16, 7/16, 5/15, and 7/16 litters and hydroureter in 3/16, 4/16, 5/15, and 5/16 litters of the 0, 22.5, 71.0, and 225.0 mg/kg/day groups, respectively. There were no other visceral malformations observed. Visceral variations included mild ventricular enlargement, thyroid hypertrophy, thyroid hypotrophy, and slight reduction in heart size. These variations were observed at single or low fetal incidences or at similar fetal incidences in all treated and control groups. Litter incidences of variations were not reported, and since the individual data only included malformations, it was not possible for the reviewer to calculate litter incidences of the reported visceral variations.

3. **Skeletal examination**: The study report included a table of the numbers and percentages of fetuses that had retarded ossification of various bones, unossified metacarpals and/or metatarsals, ossified vertebral bodies and/or sternebrae, and/or 14th (vestigial) ribs and stated that the general pattern of ossification fell within normal limits. The increased incidence of 14th (vestigial) ribs in the high-dose group was not considered treatment-related, as a dose-response pattern was not evident. All remaining observations from the skeletal examinations occurred at similar incidences in the treated and vehicle control groups; however, the data were not reported in sufficient detail to permit evaluation of ossification rates.

TABLE 4a. External examinations [fetal (litter) incidence]

Observations[a]	Dose (mg/kg bw/day)			
	0	22.5	71	225
#Fetuses(litters) examined	164 (16)	131(16)	153 (15)	153 (16)
Malformation				
Filamentous tail[b]	1 (1)	0 (0)	0 (0)	0 (0)

Data taken from Tables 6 and Appendix Tables 5-8, pp 209-210 and 215-222, respectively, MRID 00043724.

[a] Some observations may be grouped together.

[b] The study author referred to this finding as an "abnormality." Litter incidences were calculated by the reviewer, using the individual data.

TABLE 4b. Visceral examinations [fetal (litter) incidence]				
Observations	Dose (mg/kg bw/day)			
	0	22.5	71	225
#Fetuses(litters) examined	51 (16)	43 (16)	48 (15)	52 (16)
Malformations [a]				
Hydronephrosis (uni- or bilateral)	10 (5)	9 (7)	6 (5)	13 (7)
Hydroureter (uni- or bilateral)	3 (3)	4 (4)	5 (5)	9 (5)
Variations [b]				
Mild ventricular enlargement	14	15	8	19
Thyroid hypertrophy	1	0	0	0
Thyroid hypotrophy	0	0	2	1
Slight reduction in heart size	0	1	0	1

Data taken from Table 6 and Appendix Tables 5-8, pp 209-210 and 215-222, respectively, MRID 00043724.

[a] The study author referred to these findings as "abnormalities." Litter incidences were calculated by the reviewer, using the individual data.

[b] The study author referred to these findings as "variants." Litter incidence data were not provided.

None significantly different from control; Fisher's exact test performed by reviewer.

TABLE 4c. Skeletal variations (number of fetuses)				
Observations [a]	Dose (mg/kg bw/day)			
	0	22.5	71	225
#Fetuses(litters) examined	111 (16)	88 (16)	104 (15)	101 (16)
Retarded ossification of the skull	6	9	1	4
Retarded ossification of the parietals	15	10	12	6
Pelvis retarded	11	8	2	5
14th (vestigial) rib present [b]	19	20	8#	30*#

Data taken from Table 6, pp 209-210, MRID 00043724.

a Some observations may be grouped together.

b It was unclear whether the incidences of this finding were reported as numbers of fetuses with 14th vestigial ribs or the actual number of 14th vestigial ribs. [See deficiency section.]

* Significantly different from control ($p < 0.05$); 2x2 Chi square test performed by reviewer, assuming fetal incidences.

Significantly different from control ($p < 0.05$); 2x2 Chi square test performed by reviewer, assuming rib incidences.

III. DISCUSSION and CONCLUSIONS

A. INVESTIGATORS' CONCLUSIONS:

The study author concluded that there were no treatment-related toxic or teratogenic effects on the fetuses at any treatment level. The study author did not identify LOAELs or NOAELS.

B. REVIEWER COMMENTS:

1. **Maternal toxicity:** There were no treatment-related effects on maternal body weights or food consumption during gestation, and clinical signs were not reported. Although there was no reported evidence of maternal toxicity at the highest dose level tested (225 mg/kg bw/day), dosing was still considered adequate because dose selection was appropriately based on a properly conducted preliminary study, and decreased maternal body weight gains during dosing were observed at a dose level of 200 mg/kg bw/day in a separate developmental toxicity study (MRID 00057099) in Wistar rats conducted by a different testing facility.

   The maternal toxicity LOAEL was not identified and the maternal toxicity NOAEL of ICI-PP 557 in CD rats was ≥225 mg/kg/day.

2. **Developmental toxicity:**

a. **Deaths/resorptions:** Maternal treatment did not result in an increase in fetal deaths or resorptions.

b. **Altered growth:** Maternal treatment did not result in decreased fetal weights. It was not possible to adequately evaluate fetal ossification rates due to insufficient data.

c. **Developmental variations:** Treatment with the test article did not result in an increased incidence of fetal developmental variations.

d. **Malformations:** Treatment with the test article did not result in an increased incidence of fetal malformations.

Therefore, the developmental toxicity LOAEL for ICI-PP 557 in CD rats is not identified, and the developmental toxicity NOAEL is ≥ 225 mg/kg/day.

C. STUDY DEFICIENCIES:

- There was no reported evidence of significant maternal toxicity at the highest dose level, although dose selection was appropriate based on the results of the preliminary study.
- The stability and storage conditions of the test material were not provided.
- The preparation method of the dosing formulations was not described.
- Homogeneity and concentration analyses of the test material in the vehicle were not conducted.
- Individual daily clinical observations were not included in the study report; nor were clinical

- Litter incidences were not included for fetal morphological observations, and the litter was not considered the basic unit of analysis in the evaluation of fetal structural alterations. Individual data for these endpoints were not provided; therefore the reviewer was unable to calculate numbers and percent of litters with structural alterations..

The following deficiencies did not affect study classification:

- The groups did not contain sufficient numbers of animals to yield approximately 20 animals with implantation sites at necropsy.
- The test material was administered from GD 6-16, inclusive, rather than daily from implantation to the day before cesarean section as specified in Sections (d) and (e)(2)(iv) of the guideline; however, this is considered acceptable because the treatment interval did include the period of major organogenesis.
- Randomization procedures were not provided; however, at the initiation of dosing, the group mean body weights and their variances were homogeneous.
- Sacrifice order was not provided.
- The study report said that the animals were weighed upon arrival from the supplier, but these initial body weights were not included in the report.
- The study report did not include a copy of the study protocol and any amendments.
- Food consumption was reported only as group means.
- Gravid uterine weights as well as body weight changes adjusted for gravid uterine weights were not reported.
- Uteri that were grossly non-gravid were not further examined to confirm nonpregnant status.
- Fetal body weight data were reported for the combined sexes only.
- Only one-third of the fetuses of each litter were examined for visceral alterations, rather than the one-half specified by the guideline.
- There was no mention of any observations of external malformations or variations; however, one fetus in the control group had a filamentous tail, which was reported as either a skeletal malformation (text, p. 201) or a visceral malformation (Table 5, p. 208 and Appendix Table 6, p. 216).
- The study report included a table of the numbers and percentages of fetuses that had retarded ossification of various bones, unossified metacarpals and/or metatarsals, ossified vertebral bodies and/or sternebrae, and/or 14th (vestigial) ribs; however, it was not possible to determine which fetuses may have had multiple findings.

DATA FOR ENTRY INTO ISIS

Developmental Study - rats (870.3700a)

PC code	MRID	Study	Species	Duration	Route	Admin	Dose range mg/kg/day	Doses mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ	Comments
109701	00043724, 00057099	developmental	rats	GD 6-16	oral	gavage	22.5-225	0, 22.5, 71, 225	≥225	not identified		Maternal
109701	00043724, 00057099	developmental	rats	GD 6-16	oral	gavage	22.5-225	0, 22.5, 71, 225	≥225	not identified		Developmental

DATA EVALUATION REPORT

PERMETHRIN
(NRDC 143)

STUDY TYPE: SUBCHRONIC ORAL NEUROTOXICITY – RAT [Nonguideline]
MRID 00070945

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-87

Primary Reviewer: Carol S. Forsyth, Ph.D., D.A.B.T.	Signature: Date:	Carol S Forsyth JAN 30 2001
Secondary Reviewers: Cheryl B. Bast, Ph.D., D.A.B.T.	Signature: Date:	Cheryl B Bast JAN 30 2001
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross JAN 30 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson JAN 30 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract DE-AC05-00OR22725

Permethrin **Subchronic Oral Neurotoxicity (Nonguideline)**

EPA Reviewer: Yung G. Yang, Ph.D. Date 11/7/2001
Reregistration Branch 2 (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Date 1/28/2002
Toxicology Branch (7509C)

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Subchronic Neurotoxicity – Rat (Nonguideline)

DP BARCODE: D269531 SUBMISSION CODE: S504352
PC CODE: 109701 TOX CHEM NO: 652BB

TEST MATERIAL: NRDC 143 (cis:trans isomers 90:10, 40:60, and 25:75)

SYNONYMS: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; permethrin; PP 557

CITATION: James, J.A., Creasy, D.M., and Dayan, A.D. (1976) Preliminary investigation of the neurological effects in rats offered diets containing NRDC 143. Research and Development, The Wellcome Research Laboratories, Berkhamsted. Study No. (not given), August 1976. MRID 00070945, Unpublished.

SPONSOR: The Wellcome Foundation, Ltd., Berkhamsted

EXECUTIVE SUMMARY: In a preliminary subchronic oral neurotoxicity study (MRID 00070945), groups of 10 female Wistar rats were administered 6000 ppm of NRDC 143 in the diet for up to 16 days. Three dose groups were based on the amount of cis isomer: group 1, 90:10 cis:trans, group 2, 25:75 cis:trans, group 3, 40:60 cis:trans; (Batch Nos. B1, D1, C1, respectively). Group 4 was an undosed control.

Toxicity assessments were limited to clinical observations and neurohistopathological evaluations. Body weights and food consumption were not measured. In group 1 rats treated with permethrin of 90% cis:10% trans showed mortality of all animals occurred by day 3. Prior to death, all animals in this group showed marked hypersensitivity to noise and other stimuli, piloerection, and whole body tremors and most had the gait abnormality of flat front and/or back legs. One rat in group 3 was found dead on day 5. All other animals survived to scheduled termination. In group 3, whole body tremors were observed in 1 animal on day 1 and the incidence rates of tremors and gait abnormalities increased to 100% by day 4. Beginning on day 3, 100% of the rats in group 3 also showed hypersensitivity. The authors noted that the clinical signs were more pronounced in the mornings and decreased in severity throughout the day. In group 2, the first clinical signs were observed on day 4 with the daily incidence rates throughout the study of slight body tremors and gait abnormalities ranging from 10% to 86% of animals affected.

No microscopic lesions were found in either the central or peripheral nervous tissue that could account for the clinical signs of toxicity. Vacuolation of the myelinated fibers was seen in all treated groups and in controls, but assessment of controls was limited to three animals. Therefore, further characterization of this lesion may be necessary before the relationship to treatment can be discerned.

A NOAEL was not identified for this preliminary study. The study indicated that increased content of *cis* isomer showed dose-related increases in incidence and severity of clinical signs and toxicity.

This study is classified **Acceptable/Nonguideline** and does not satisfy the requirements for a subchronic oral neurotoxicity study (OPPTS 870.6200 [82-7]) in rats. The study is sufficient for the purposes for which it was intended as a preliminary assessment of neurotoxicity in female rats.

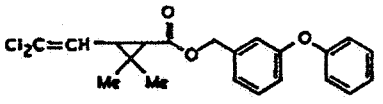
COMPLIANCE: Signed and dated Quality Assurance, Data Confidentiality, Good Laboratory Practice Compliance, and Flagging statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound: NRDC 143, cis:trans ratios, 90:10, 40:60, 25:75

   Description: not given
   CAS No.: 52645-53-1
   Batch No.: B1, C1, D1
   Purity: Not given
   Contaminants: none given
   Structure:



2. Vehicle

   Diet 41B meal was used as the vehicle and untreated control. No positive control was used in this study.

3. Test animals

Species: rat
Strain: Wistar
Age and weight at study initiation: age not stated: females 145-197 g.
Source: Charles River
Housing: "stock cages"
Food: Diet 41B meal ex Heygates was available *ad libitum.*
Water: Water was available *ad libitum.*
Environmental conditions:
Temperature: not stated
Humidity: not stated
Air changes: not stated
Photoperiod: not stated
Acclimation period: not stated

B. STUDY DESIGN

1. In life dates

Start: May 5, 1976; End: May 21, 1976

2. Animal assignment

Animal assignment and dose selection are listed in Table 1. The method of randomization was not described. Only female rats were used in this preliminary assessment of clinical neurotoxicity and neurohistopathology. The duration of treatment was stated as "until rats were moribund or there were well established clinical symptoms". Observational data were reported for days 0-16. Rats were not individually marked and no individual animal records were kept.

TABLE 1. Study design

Test group	Diet Conc. (ppm)	isomer		No. of females
		Cis (%)	Trans (%)	
Group 1	6000	90	10	10
Group 2	6000	25	75	10
Group 3	6000	40	60	10
Group 4	0	-	-	10

Data taken from text table p. 1, MRID 00070945.
All doses are expressed in term of total isomer content.

3. Validation of test methods

Positive control data were not included for evaluation of the ability of the testing laboratory to conduct FOB or neuropathological studies.

4. Rationale for dose selection

A dose selection rationale was not included in the current report.

5. Preparation and analysis of dose solutions

A 10X concentrate was prepared every 7 days and diluted to the appropriate concentrations. Further details of the mixing procedures were not given. Analyses of the test diets for homogeneity, stability, and concentration were not discussed or reported.

6. Statistical analysis

Data were not analyzed statistically.

C. METHODS

1. Observations

Treated animals were observed daily for clinical signs of toxicity; although the time of observation was not stated, it was noted in the results that the severity of signs decreased throughout the day. The frequency of examination of the control group was "limited".

2. Body weight

Body weights were presented for day 0 and day 16 for the treated animals. No body weight data were given for the control group.

3. Food consumption and food efficiency

Food consumption and food efficiency were not measured by the study authors.

4. Functional observational battery (FOB)

A full FOB was not conducted. Treated animals were observed daily with special attention to effects on gait. Observations of gait were made by placing the rats on a flat metal surface.

5. Motor activity

Motor activity was not assessed.

6. Ophthalmology

Ophthalmoscopic examinations were not conducted.

7. Clinical chemistry

Clinical chemistry evaluations were not done.

8. Sacrifice/necropsy/neurohistopathology

Animals found dead or sacrificed moribund were subjected to gross necropsy and the brain, spinal cord, nerves, diaphragm, skeletal muscle, small intestine, and adrenals were fixed in neutral buffered formalin, and the eye and optic nerve were fixed in Davidson's fixative. On day 8, animals showing the most severe clinical signs were sacrificed with the brain, spinal cord, and nerves fixed in formal acetic methanol, the skeletal muscle, small intestine, and adrenals fixed in neutral buffered formalin, and the eye and optic nerve fixed in Davidson's fixative.

The remaining surviving treated rats, plus three untreated controls, were sacrificed on day 16. These animals were perfused via the heart with 5% glutaraldehyde in phosphate buffer. The brain, spinal cord, sciatic nerve, skeletal muscle, and liver were preserved in neutral buffered formalin. Sciatic and brachial nerves from perfused animals were post fixed in osmium tetroxide and processed for electron microscopy.

Coronal slices were taken through the brain to demonstrate the cerebrum, midbrain, cerebellum, pons, and medulla. Cervical, thoracic, and lumbar regions of the spinal cord were sectioned. Sections from brain, spinal cord, and nerves were stained with Glees/LFB; sections from skeletal muscle and diaphragm were stained with Sudan black; and sections from small intestine, adrenal, eye and optic nerve, and liver were stained with H & E.

II. RESULTS

A. CLINICAL OBSERVATIONS AND MORTALITY

All rats in the group 1 were either found dead or were sacrificed *in extremis* by day 3. Prior to death, all animals in this group showed marked hypersensitivity to noise and other stimuli, piloerection, and whole body tremors and most had the gait abnormality of flat front and/or back legs. One rat in group 3 was found dead on day 5. All other animals survived to scheduled termination.

In group 3, whole body tremors were observed in 1 animal on day 1 and the incidence rates of tremors and gait abnormalities increased to 100% by day 4. Beginning on day 3, 100% of the rats in group 3 also showed hypersensitivity. The authors noted that the clinical signs were more pronounced in the mornings and decreased in severity throughout the day. In group 2, the first clinical signs were observed on day 4 with the daily incidence rates throughout the study of slight body tremors and gait abnormalities ranging from 10% to 86% of animals affected.

C. BODY WEIGHTS AND BODY WEIGHT GAINS

Body weight data for the treated animals was given for day 0 and day 16 only. No data for the control group were included. It appeared that the animals in groups 2 and 3 gained weight during the study, however, no meaningful comparisons could be made. The report also noted that the rats were not individually marked and no individual record was kept of body weight, food consumption or clinical symptoms.

D. FOOD CONSUMPTION

Food consumption was not measured. The study authors noted that the rats ate the treated diets.

E. FUNCTIONAL OBSERVATIONAL BATTERY (FOB)

A detailed FOB was not conducted.

F. MOTOR ACTIVITY

Motor activity was not measured.

G. OPHTHALMOLOGY

Ophthalmologic examinations were not performed.

H. CLINICAL CHEMISTRY

Clinical chemistry parameters were not evaluated in this study.

I. NECROPSY

Immediately prior to sacrifice, clinical signs noted in some animals were similar to the clinical signs observed during the in-life phase of the study. No abnormalities were seen at gross necropsy.

J. NEUROPATHOLOGY

No treatment-related microscopic lesions were noted in any animal. Vacuolation within myelinated fibers of the peripheral and central nervous system were observed in animals from all treated groups as well as the three control animals that were examined.

III. DISCUSSION

A. DISCUSSION

The objective of this study was to evaluate the potential neurotoxicity of NRDC 143 when administered to rats in the diet. In this preliminary study, clinical and neuro-histopathological endpoints were evaluated. In group 1, rats treated with permethrin of 90% cis:10% trans showed mortality of all animals occurred by day 3. Increased content of *cis* isomer showed a dose-related increases in incidence and severity of clinical signs. In addition, the onset of the clinical signs of toxicity was delayed in the lower cis-isomer groups. The decrease in severity of signs during the day was probably due to the night-time feeding behavior of the animals. Doses to the animals would be highest in the mornings after feeding overnight followed by elimination of the test article throughout the day.

No microscopic lesions were found in either the central or peripheral nervous tissue that could account for the clinical signs of toxicity. Vacuolation of the myelinated fibers was seen in all treated groups and in controls, but assessment of controls was limited to three animals. Therefore, further characterization of this lesion may be necessary before the relationship to treatment can be discerned.

A NOAEL was not identified for this preliminary study. The study indicated that increased content of *cis* isomer showed a dose-related increases in incidence and severity of clinical signs and toxicity.

This study is classified **Acceptable/Nonguideline** and does not satisfy the requirements for a subchronic oral neurotoxicity study [OPPTS 870.6200 (82-7)] in rats. The study is sufficient for the purposes for which it was intended as a preliminary assessment of neurotoxicity in female rats.

B. STUDY DEFICIENCIES

This is a nonguideline study, therefore, no deficiencies were identified. However, information missing from the report included concentration, homogeneity, and stability of the test diets and validation of testing methods. Endpoints not evaluated included an FOB, motor activity, food consumption, and clinical chemistry. Body weights were not measured daily and data for controls were not given.

DATA EVALUATION RECORD

PERMETHRIN/109701
(21Z73)
STUDY TYPE: PRENATAL DEVELOPMENTAL TOXICITY STUDY - RAT
OPPTS 870.3700a [§83-3a]; OECD 414.
MRID 00057099

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
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Work Assignment No. 02-05

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Date: APR 0 5 2002

Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

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Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 8/7/2007
Signature: Joycelyn Stewart
Date 8/7/2007

TXR#: 0050649

DATA EVALUATION RECORD
Supplementary, HED Doc# 007392

STUDY TYPE: Prenatal Developmental Toxicity Study - Rat; OPPTS 870.3700a [§83-3a]

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 21Z73 (Permethrin; purity and lot/batch no. not provided)

SYNONYMS: NRDC 143; Permethrin; 3-phenoxybenzyl (±) cis : trans-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-1-carboxylate

CITATION: James, D. (1974) Preliminary foetal toxicity study in the rat given 21Z73 (NRDC 143) orally. Wellcome Research Laboratories, Beckenham, UK. Laboratory report number Path 167, May 28, 1974. MRID 00057099. Unpublished.

SPONSOR: The Wellcome Foundation Ltd.

EXECUTIVE SUMMARY: In a preliminary developmental toxicity study (MRID 00057099), 21Z73 (permethrin, purity, cis-/trans- ratio, and batch/lot # not provided) was administered to 23 mated female Wistar rats/dose in corn oil by gavage at dose levels of 0 or 200 mg/kg bw/day from gestation days (GD) 6 through 16. An additional group of 22 animals served as "environmental controls." On GD 20, all surviving dams were sacrificed, and all fetuses were weighed, sexed, and examined for external abnormalities. Approximately two-thirds of the fetuses from each litter were examined for visceral alterations, and one-third were examined for skeletal observations.

Maternal toxicity was evident as decreased body weight gain during dosing (88% of controls), and there were also two potentially treatment-related deaths in the treated group (on GDs 16 and 17). There were no treatment-related effects on intrauterine parameters of the treated group compared to vehicle controls. Clinical signs were not reported; dams were not necropsied; and maternal food consumption was not measured. **The maternal toxicity LOAEL for 21Z73 in Wistar rats is 200 mg/kg bw/day based on decreased body weight gain during dosing; NOAEL is unidentified.**

There were no treatment-related increases in fetal deaths/resorptions or incidences of fetal structural alterations, and there was no evidence of altered growth. **Under the conditions of this study, the developmental toxicity NOAEL for 21Z73 in Wistar rats is greater than or equal to 200 mg/kg bw/day, and the developmental toxicity LOAEL is unidentified.**

This range-finding study is classified **acceptable/nonguideline**. Due to numerous major deficiencies noted in the conduct of this study (see deficiency section), the study alone would not satisfy the guideline requirements for a developmental toxicity study in the rat (OPPTS 870.3700); however, the study may provide supplemental information when combined with additional studies.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided; however, the study was conducted in 1974.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. **Test material:** 21Z73, permethrin

Description:	Not reported
Lot/Batch #:	Not reported
Purity:	Not reported
Compound Stability:	Not reported
CAS #of TGAI:	52645-53-1

2. **Vehicle and/or positive control:** The vehicle was corn oil (Lot/Batch # and purity not reported). There was no positive control used.

3. **Test animals:**

Species:	Rat	
Strain:	Wistar	
Age/weight at study initiation:	Females: approximately 3 months / 155-240 g.; Males: data not reported	
Source:	The animal holding unit of Wellcome Research Laboratories	
Housing:	Not reported	
Diet:	Rat cake (supplied by Lillico, manufactured by Heygates) *ad libitum*	
Water:	*ad libitum*, not otherwise described	
Environmental conditions:	**Temperature:**	Not reported
	Humidity:	Not reported
	Air changes:	Not reported
	Photoperiod:	Not reported
Acclimation period:	Not reported	

B. PROCEDURES AND STUDY DESIGN:

1. **In life dates** - Not reported. The entire study took place April-May 1974

2. **Mating:** Females were paired with males of the same strain and source. Mating was confirmed by the presence of sperm in a vaginal smear, and this day was designated as day 0 of gestation (GD 0).

3. **Animal assignment:** Animals were assigned by an unspecified method to the dose groups given in Table 1. In addition, an environmental control group consisting of 22 mated females was monitored throughout the study.

TABLE 1. Animal assignment

Dose (mg/kg bw/day) [a]	0 (Vehicle controls)	200
# Females	23	23

Data taken from text, p. 1, and Table 1, p. 7, MRID 00057099.
a Animals were treated on GD 6-16, inclusive. Dose is expressed in terms of the active ingredient.

4. **Dose selection rationale:** There was no dose selection rationale provided.

5. **Dosage preparation and analysis:** The preparation method of the test material-vehicle mixture was not described in the study report. The study report stated that the treated group received 200 mg active ingredient/kg body weight; it is therefore assumed that the concentration of the test material in the vehicle was adjusted to account for purity.

   There was no mention of the frequency of preparation or storage conditions of mixtures of test substance with the vehicle. There was also no mention of evaluation of the stability of the test substance in the vehicle or concentration and homogeneity of the test mixtures.

   There were no analytical data provided to indicate whether the mixing procedure was adequate or that the variance between nominal and actual dosage to the study animals was acceptable.

6. **Dosage administration:** All doses were administered once daily by gavage, on gestation days 6 through 16, inclusive. Vehicle control animals were given a volume of 10 mL/kg of body weight/day; however, the volume given to the treated group was not reported. The study report did not state whether dosing was based on the body weight from the most recent body weight determination; however, this is assumed to be the case as the animals were weighed daily during gestation.

C. OBSERVATIONS:

1. **Maternal observations and evaluations:** The animals were checked once daily for mortality or clinical signs. Body weights were recorded daily throughout gestation but were reported only for GDs 0, 6, 16, and 20. Food consumption was not measured. Dams were sacrificed on GD 20. Examinations at sacrifice consisted of recording numbers of corpora lutea, implantations, live fetuses, dead fetuses, early or late resorptions, and externally abnormal fetuses. There was no mention of further gross necropsies of the dams.

2. **Fetal evaluations:** All fetuses were weighed and subjected to external examination. Approximately one-third of the fetuses from each litter were subjected to soft tissue examination of the organs of the neck, thorax, and abdomen by gross dissection; one-third were examined by the Wilson method of serial free-hand sectioning; and one-third were

subjected to skeletal examination after staining by the Staples and Schnells Alizarin red S method. External, visceral, and skeletal anomalies were not further characterized as variations and malformations. Litter incidences of fetal morphological data were not reported and fetal individual morphological data were not provided.

D. DATA ANALYSIS:

1. **Statistical analyses:** The following data were analyzed using ANOVA: maternal body weight gains during dosing and gestation, numbers of corpora lutea, implantations, live fetuses, and normal fetuses, litter weights, and weights of normal fetuses. Litter incidences of external, visceral, and skeletal anomalies were not reported, and fetal incidences of these endpoints were not compared statistically.

   "Standard errors" were reported for some of the data. The term "standard error" can either be a synonym of "standard deviation"or a separate statistical measure of variance that can be converted to standard deviation by multiplying by the square root of n. As it was unknown which was the case in the study report, the data were summarized as reported.

2. **Indices:** The following index was calculated from cesarean section records of animals in the study:

   Implant/corpora lutea ratio = The number of implantations/the number of corpora lutea

   The reviewer calculated pre- and postimplantation loss indices as follows:

   Preimplantation loss (%) = (total number of corpora lutea minus total number of implantations/total number of corpora lutea) x 100.

   Postimplantation loss (%) = (total number of implantations minus total number of live fetuses/number of implantations) x 100.

3. **Historical control data:** Historical control data (cumulative control data) were provided for fetal external, visceral, and skeletal anomalies to allow comparison with concurrent controls. However, these data were provided only in aggregate form rather than by individual study with appropriate descriptive statistics, and there was no information provided regarding the number of studies included, dates of the studies, source of the animals, or the vehicle(s) and route(s) of administration.

II. RESULTS:

A. MATERNAL TOXICITY

1. **Mortality and clinical observations:** Clinical observations were not reported. Two dams in the treated group died during the study: one on GD 16, and the other on GD 17. In the absence of clinical signs or necropsy data, it is not possible to rule out the possibility that these deaths were treatment-related.

2. **Body weight** - Body weight data are summarized in Table 2. Mean absolute body weights of the control and treated groups were similar throughout the study. The study report included mean maternal weight gain during dosing and gestation; however, it was unclear how these means were calculated (see deficiency section). Mean body weight changes during the pre-dosing, dosing, and post-dosing intervals were estimated by the reviewer using mean absolute body weight data. The estimated mean body weight gain of the treated group was 12% less than controls for the GD 6-16 (dosing) interval and was 29% greater than controls for the post-dosing interval, and these differences are considered treatment-related. Corrected body weights and body weight gains were estimated by the reviewer as noted in the table footnotes, and there were no remarkable differences between the corrected body weights and body weight gains of the treated and control groups.

TABLE 2. Mean maternal body weights and body weight gains (g)		
Gestation Day	Dose in mg/kg bw/day (# of Dams)	
	Vehicle controls (23)	200 (21)
Absolute body weights [a]		
0	193.91	198.81
6	213.26	218.33
16	261.74	261.19
20	301.52	312.38
Corrected body weight [b]	268.1	275.01
Body weight changes		
0-6 (Pretreatment) [c]	19.35	19.52
6-16 (Treatment) [c]	48.48	42.86 (88) [d]
16-20 (Post-treatment) [c]	39.78	51.19 (129)
0-20 [e]	107.61±5.56	113.57±4.43
Corrected BW Gain [f]	74.19	76.2

Data taken from Tables 1, 5, and 8, pp. 7, 12-13, and 17-18, respectively, MRID 00057099.

a Data reported as means, standard deviations or standard errors were not provided.

b Estimated by reviewer as Corrected body weight = Mean GD 20 weight minus mean litter weight. Mean litter weights for vehicle controls and treated groups were 33.42 and 37.37 g, respectively.

c Calculated by reviewer using mean absolute body weight data. Not subjected to statistical analysis.

d Number in parentheses is percent of vehicle control; calculated by reviewer.

e Data given as Mean ± standard error; no statistically significant intergroup differences.

f Estimated by reviewer as Corrected BW Gain = Corrected body weight minus GD 0 body weight.

3. **Food consumption**: Food consumption was not measured.

4. **Gross pathology**: There were no gross pathology data reported.

5. **Cesarean section data**: Data collected at cesarean section are summarized in Table 3. There were no remarkable differences between pregnancy rates, pre- and postimplantation losses, early and late resorptions per dam, mean live litter size, mean fetal weights, or fetal sex ratios of the treated and control groups. There were no abortions, early deliveries, or total litter resorptions in either group.

TABLE 3. Cesarean section observations [a]

Observation	Dose in mg/kg bw/day	
	Vehicle Controls	200
# Animals Assigned (Mated)	23	23
# Animals Pregnant	23	21[b]
Pregnancy Rate (%)	100	100 [b]
# Nonpregnant	0	0 [b]
Maternal Wastage		
# Died	0	2
# Aborted	0	0
# Premature Delivery	0	0
Total # Corpora Lutea	237	239
Corpora Lutea/Dam	10.30±0.29	11.38±0.55
Total # Implantations	213	213
(Implantations/Dam)	9.26±0.46	10.14±0.44
Total # Litters	23	21
Total # Live Fetuses	206	207
(Live Fetuses/Dam)	8.96	9.86
Total # Dead Fetuses	0	0
(Dead Fetuses/Dam)	0	0
Total # Resorptions	7	6
Early (complete)	7	6
Late (partial)	0	0
Resorptions/Dam	0.3	0.29
Early (complete)	0.3	0.29
Late (partial)	0	0
Litters with Total Resorptions	0	0
Mean Fetal Weight (g) [c]	3.75	3.81
Sex Ratio (% Male)	46.6	50
Preimplantation Loss (%) [d]	10.13	10.88
Postimplantation Loss (%) [e]	3.29	2.82

Data taken from Tables 6, 7, and 8, pp. 14-15, 16, and 17-18, respectively, MRID 00057099.

a Data expressed as mean ± standard error where appropriate; however, standard errors were not provided and/or legible for all means.

b Two dams that died during the study are excluded because their pregnancy status was not reported.

c Excluded one grossly malformed fetus in the treated group. Mean fetal weight was reported only for combined sexes.

d Calculated by reviewer as Preimplantation Loss = (Total corpora lutea minus total implantations/total corpora lutea) x 100.

e Calculated by reviewer as Postimplantation Loss = (Total implantations minus total live fetuses/Total implantations) x 100.

B. DEVELOPMENTAL TOXICITY: The total numbers of fetuses (litters) in the vehicle control and treated groups were 206 (23) and 207 (21), respectively. The study report did not include litter incidences of fetal structural alterations, and it was unclear whether all findings, both major and minor were reported. Selected fetal morphological observations are given in Table 4.

1. **External examination:** A single fetus in the treated group had multiple external abnormalities, which included the following: shortening of all four limbs; only four digits on each paw; slightly protruding tongue; no developed ridging of palate; and slight edema of the neck.

2. **Visceral examination:** Reported visceral abnormalities were limited to the following: esophageal dilatation in a single fetus from the treated group; convoluted ureters in 12 and 14 fetuses from the vehicle control and treated groups, respectively; and unilateral or bilateral dilatation of the renal pelvis and calyces in 8 and 2 fetuses from the vehicle control and treated groups respectively.

3. **Skeletal examination:** A single fetus in the treated group (presumed to be the same fetus that had external abnormalites, although this was not stated in the study report) had multiple skeletal abnormalities, which included the following: narrowed skull; fused and pointed lower jaw; clavicle shorter on one side; shortened, malformed limbs; malformed pelvic girdle; one or more poorly ossified cervical vertebra(e); and one or more unossified sternebra(e). All observations from the remaining fetal skeletal examinations were found at similar incidences in the treated and vehicle control groups. However, it must be noted that the reported observations concerned only the axial skeleton; it is therefore unknown whether the appendicular skeleton was routinely examined. Sternebral ossification was reported as the number and percent of fetuses with either one or more unossified sternebra(e) or one or more poorly ossified sternebra(e), and ossification of the cervical vertebrae was reported only as the number and percent of fetuses with one or more poorly ossified cervical vertebral centra.

TABLE 4. Fetal morphological observations [a]		
Observations [b]	Dose in mg/kg bw/day (number of litters)	
	0 (23)	200 (21)
External Abnormalities		
Number of fetuses examined	206	207
Number of fetuses affected	0	1
Multiple external abnormalities [c]	0	1
Visceral Abnormalities - Open Dissection		
Number of fetuses examined	69	69
Convoluted ureter(s)	12	14
Visceral Abnormalities - Serial Sections		
Number of fetuses examined	69	69
Dilated renal pelvis and calyces - unilateral or bilateral	8	2
Dilated esophagus	0	1
Skeletal Abnormalities		
Number of fetuses examined	68	69
Multiple severe skeletal abnormalities [d]	0	1
Occipitals poorly or irregularly ossified	46	34
Interparietals poorly or irregularly ossified	37	33

Data taken from Tables 3 and 4a-4c, pp. 9 and 10-11, respectively, MRID 00057099.

a Data are given as the number of fetuses with the specified abnormality. Litter incidences were not reported, and individual data for fetal morphological endpoints were not included in the study report. Abnormalities were not further categorized as malformations/variations or any other classifications of anomalies.

b Some observations may be grouped together.

c Included the following: shortening of all four limbs; only four digits on each paw; slightly protruding tongue; no developed ridging of palate; and slight edema of the neck.

d Included the following: narrowed skull; fused and pointed lower jaw; clavicle shorter on one side; shortened, malformed limbs; malformed pelvic girdle; one or more poorly ossified cervical vertebra(e); and one or more unossified sternebra(e).

III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS:** The study author concluded that there were no effects on maternal weight or body weight gain during gestation, or numbers of implantations, live fetuses, malformations, or fetal deaths. The study author did not identify LOAELs or NOAELS.

B. REVIEWER COMMENTS:

1. **Maternal toxicity:** It is unknown whether there were any treatment-related clinical signs or whether the two deaths in the treated group were treatment-related. However, maternal toxicity was evident as decreased maternal body weight gain during dosing, followed by compensatory increased body weight gain during the post-dosing interval.

   Therefore, the maternal toxicity LOAEL for 21Z73 in Wistar rats is 200 mg/kg/day, based on decreased maternal body weight gain during dosing; NOAEL is not identified.

   This differs from the study author's conclusions, and the source of the difference is as follows. The reviewer compared the weight differences for the GD 6-16 interval both by subtraction of the group means for these days and by finding the means of the individual weight gains of the animals in each group for these days. [These were all the body weight data reported.] It is unclear from the study report exactly which days were considered part of the "dosing" interval in the calculation of the mean body weight gains for "dosing," and the reported mean body weight gains were markedly different from those calculated by the reviewer.

2. **Developmental toxicity:**

   a. **Deaths/resorptions:** Maternal treatment did not result in an increase in fetal deaths or resorptions.

   b. **Altered growth:** Maternal treatment did not result in decreased fetal weights. It was not possible to adequately evaluate fetal ossification rates due to insufficient data.

   c. **Developmental abnormalities:** Treatment with the test article did not result in an increased incidence of fetal structural alterations.

   Therefore, the developmental toxicity LOAEL for 21Z73 in Wistar rats is not identified, and the developmental toxicity NOAEL is greater than or equal to 200 mg/kg bw/day.

C. STUDY DEFICIENCIES: There were numerous deficiencies noted in the conduct of this study.

Major deficiencies include the following:

- The study had a single dose level and a concurrent control group.
- The study report did not include full identification of the test material (chemical identification, percentage active, batch or lot number, physical properties, purity/impurities, expiration date). It is also unknown whether the technical form of the active ingredient was used.
- There were no analyses for test material stability, homogeneity and concentration in the dosing medium.

- Individual daily clinical observations were not included in the study report; nor were clinical observations reported in summary form.
- Gross necropsies of the dams were not conducted.
- Individual and group mean body weights were only reported for GDs 0, 6, 16, and 20 instead of the 3-day intervals during the dosing period specified in the guideline. Also, it was unclear how the reported group mean body weight gains during dosing were derived; the reviewer was unable to obtain similar numbers by subtracting the GD 6 group mean body weight from the GD 16 mean body weight or by finding the means of the individual body weight gains during the GD 6-16 interval.
- Gravid uterine weights and body weight changes adjusted for gravid uterine weights were not reported.
- Food consumption was not measured.
- Litter incidences were not included for fetal morphological observations, and the litter was not considered the basic unit of analysis in the evaluation of fetal structural alterations. Individual data for these endpoints were also not provided; therefore the reviewer was unable to calculate numbers and percent of litters with structural alterations.

The following deficiencies were considered minor and did not affect the classification of the study:

- The test substance was administered during GD 6-16 (inclusive) rather than through the day prior to cesarean section as specified in the guideline. However, the dosing interval included the period of major organogenesis in the rat; this is only considered a minor deficiency and does not affect the classification of the study.
- It is unknown whether sibling matings were avoided.
- Mean fetal body weights were only reported for combined sexes.
- Approximately one-third of the fetuses were examined for skeletal alterations rather than one-half as specified by the guideline.

DATA FOR ENTRY INTO ISIS

evelopmental Study - rats (870.3700a)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
109701	57099	developmental	rats	GD 6-16	oral	gavage	200	0, 200	none	200	decr body wt gain	Maternal
109701	57099	developmental	rats	GD 6-16	oral	gavage	200	0, 200	≥200	n/a	none	Developmental

DATA EVALUATION RECORD

PERMETHRIN (21Z)

STUDY TYPE: CHRONIC TOXICITY/CARCINOGENICITY FEEDING - RAT [OPPTS 870.4300 (83-5)] MRID 97441 & 77054

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

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Toxicology and Risk Analysis Section
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Task Order 01-89

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Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract DE-AC05-00OR22725

EPA Reviewer: Yung Yang, Ph.D. **Signature:** Yung G. Yang
Reregistration Branch 2, Health Effects Division (7509C) **Date** 11/21/2006
EPA Work Assignment Manager: J. Stewart, Ph.D. **Signature:** Joycelyn Stewart
Toxicology Branch, Health Effects Division (7509C) **Date** 1/30/2007

TXR # 0050649

DATA EVALUATION RECORD

This is an updated DER of HED Doc. No. 008163. The classification has been changed.

STUDY TYPE: Combined chronic toxicity/carcinogenicity - diet - Rat; OPPTS 870.4300 [§83-5]

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin, technical grade, purity not stated, cis:trasn 25:75

SYNONYMS: 21Z; 3-phenoxybenzyl(1RS)-*cis, trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-carboxylate (IUPAC)

CITATION: McSheehy, T, J. Finn. (1980). 21Z: Potential toxicity and oncogenicity in dietary administration to rats for a period of 104 weeks. December 19, 1980. (MRID 54465). Final report prepared by Life Science Research, at Elm Farm Laboratories, Occold, Nr. Eye, Suffolk and Stock, Essex, CM4 9PE. . Laboratory report number 80/82L003/2831, and submitted by Burroughs Wellcome Co., RTP, July 2, 1980. MRID 97441. Unpublished.

Piercy, D. (Letter dated June 25, 1981). Report HEFG 81-C045: 21Z Rat carcinogenicity study (summary tables of tissue examination and neoplasms). MRID 77054. Unpublished.

SPONSOR: Wellcome Research Laboratories, Berkhamsted Hill, Berkhamsted, Hertfordshire, HP4 2QE

EXECUTIVE SUMMARY: In a combined chronic toxicity/carcinogenicity study (MRID 97441), permethrin (technical grade, purity not specified, Batch No. 533/17/x) was administered to groups of Wistar strain rats (specific-pathogen free) (60/sex/group) at dietary concentrations delivering doses of 0, 10, 50, or 250 mg/kg/day for up to 104 weeks. Additional groups of 15 male and female rats were included for clinical pathology studies (satellite study).

No treatment-related or biologically significant effects were observed on body weight, weight gain, food consumption, food efficiency, hematology, clinical chemistry, urinalysis parameters, eyes, organ weights (females only), or gross lesions in male and female rats fed permethrin at doses up to 250 mg/kg/day. The only noteworthy clinical sign was tremors observed in ten males and five females in the high dose group for a 2-week period after week 90. The mortality rates in male rats at study termination were 58%,

78% ($p<0.05$), 67%, and 80% ($p<0.01$) at 0, 10, 50, and 250 mg/kg/day. The lack of a clear dose-related trend and treatment-related cause of death indicate that the increased mortality may not be treatment related. No treatment-related mortality was observed in females. The absolute liver weight of high-dose male rats was elevated by 19% ($p<0.05$) compared with the controls, and the relative liver weight was also slightly increased. Mid- and high-dose male and female rats had significantly increased incidences of periacinar hepatocyte hypertrophy in the liver. The incidence of hepatocyte fatty vacuolation in the liver (all locations combined) was 9/59, 16/56 ($p=0.07$), 17/58 ($p<0.05$), and 22/52 ($p<0.01$) for the control, low-, mid-, and high-dose male rats, respectively. In addition, 9/52 ($p<0.05$) high-dose male rats had hyperplasia of the pelvic epithelium in the kidney compared with 2/59 for controls and 6/52 ($p<0.05$) high-dose male rats had erythrocytes and erythrophagocytosis in the sinus of the thymic lymph nodes compared with 1/59 control. High-dose females had no other lesions that occurred with statistically significant increased incidences compared with the control incidences. The HED HIARC evaluated the toxicology database of permethrin and determined that the increased liver weight and hypertrophy observed in the liver are adaptive and reversible effects and are not considered adverse effects.

The LOAEL for permethrin is 250 mg/kg/day based on tremors observed in males and females; the NOAEL is 50 mg/kg/day.

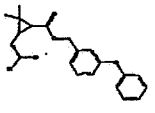
There were no treatment related increases in tumor incidences at any dose of the test material compared with control incidences. Dosing was considered adequate based on the increased incidence of hepatocyte fatty vacuolation and periacinar hepatocyte hypertrophy at the mid- and high-dose levels.

This chronic/carcinogenicity study in the rat is **Unacceptable/Guideline (upgradeable)**. The study may be upgraded upon submission of data listing on the study deficiencies section. It should be noted that this study was conducted before Subdivision F or OPPTS 870.4300 guidelines were established.

COMPLIANCE: Assigned and dated Quality Assurance statement was provided. GLP, Data Confidentiality, and Flagging statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1.	**Test material:**	21Z (permethrin)
	Description:	Technical grade
	Lot/Batch #:	533/17/x (a total of 40 batches were provided by the sponsor in the form of premixes)
	Purity:	Not reported
	Compound Stability:	Not reported
	CAS # for TGAI:	52645-53-1
	Structure:	

2. **Vehicle and/or positive control:** The test material was administered in the diet; no positive control was used in this study.

3. **Test animals:**

Species:	Rat	
Strain:	Wistar (specific-pathogen free)	
Age/weight at study initiation:	~5-6 weeks of age; 90-110 g both sexes: mean weight by sex: males: 108-109 g; females 104-105 g	
Source:	Charles River, U.K., Ltd., Margate, Kent	
Housing:	Housed five/cage of like sex in polypropylene cages (52 × 35 × 18 cm) with stainless steel floors and lids	
Diet:	Ground Spratts Laboratory Diet No. 2, *ad libitum*	
Water:	Tap water, *ad libitum*	
Environmental conditions:	**Temperature:**	22°C ± 3°C
	Humidity:	55% ± 15%
	Air changes:	15/hr
	Photoperiod:	12 hours dark/12 hours light
Acclimation period:	1 week	

B. STUDY DESIGN:

1. **In life dates** - Start: April 25, 1977; End: May 7, 1979

2. **Animal Assignment/Dose Levels:** Animals were assigned randomly to the test groups noted in Table 1 based on body weight so that the mean body weights for the four groups per sex differed by no more than 1 g. Dietary concentrations of test material were adjusted weekly during the first and last 26 weeks and every 2 weeks during weeks 27-78 to maintain the constant doses listed in Table 1.

TABLE 1: STUDY DESIGN

Test Group	Dose to animal[a] (mg/kg/day)	Main Study 24 months		Satellite Study[b] 24 months	
		Male	Female	Male	Female
Control	0	60	60	15	15
Low (LDT)	10	60	60	15	15
Mid (MDT)	50	60	60	15	15
High (HDT)	250	60	60	15	15

Data taken from page 15 of the study report, MRID 97441.

[a]The concentration of test material in the diet was adjusted during the study to maintain a constant weight-normalized dose for each group

[b]Satellite animals were used for clinical pathology (hematology, clinical chemistry, urinalysis) and were necropsied at study termination.

3. **Dose Selection:**

The dose selection rationale was not reported.

4. **Diet preparation and analysis:**

Diets were prepared weekly for the first and last 26 weeks and every 2 weeks during weeks 27-78. The sponsor provided the testing laboratory with 5% premixes prepared in ground Spratts Laboratory Diet No. 2. The premixes were mixed with appropriate amounts of feed for 20 minutes in a food mixer equipped with a horizontal Archimedes screw to achieve calculated concentrations. Data on homogeneity and stability of test material in the diet were not provided in the report.

Results -
Homogeneity Analysis: no data

Stability Analysis: no data

Concentration Analysis: Verification of target concentrations were not reported

Analytical data were not provided; therefore the mixing procedure and variance between nominal and actual dosage could not be evaluated.

5. Statistics

Blood parameters and absolute and relative (to body weight) organ weights were analyzed by a series of Student's t-tests using a pooled within-treatment variance. Mortality of male rats was analyzed by chi-square test with Yates correction or by Fisher's Exact Probability Test. A computer program was used for trend and homogeneity analysis of proportions and for life-table analysis. Similar analyses were used for latency analysis of mammary fibroepithelial tumors. Incidence data were analyzed by Fisher's Exact Probability Test (two-tailed). Level of statistical significance was not reported but assumed to be $P \leq 0.05$.

C. METHODS:

1. Observations:

a. **Cageside Observations:** Animals were inspected two times a day for the first 2 weeks and once a day thereafter for signs of toxicity and mortality.

b. **Physical Examinations:** Each animal was given a physical examination with palpation once a week.

c. **Neurological Evaluations:** A neurological evaluation was not performed on these animals. This study was conducted between 1977 and 1979, before neurological evaluations were required for chronic toxicity/carcinogenicity studies.

2. **Body weight:** Animals were weighed at study initiation, once a week for the first 26 weeks, every 2 weeks from weeks 27-78, and once a week from weeks 79-103.

3. **Food consumption and compound intake:** Food consumption for each cage of animals was measured weekly for the duration of the study and calculated as g food/rat/week. Overall food efficiency was calculated by the reviewer from data on total food consumed and overall body weight gain. Doses calculated as mg/kg body weight/day were determined on the same schedule as body weight measurements.

4. **Ophthalmoscopic examination:** Eyes of all rats on study were examined using a binocular ophthalmoscope before initiating treatment and the eyes of the control and 250-mg/kg/day group were examined during weeks 7, 26, 51, 78, and 103. A mydriatic solution (0.5% Tropicamide) was instilled in the eye to dilate the pupils before the examinations.

5. **Hematology & Clinical Chemistry:** Blood was collected from the orbital sinus of ten nonfasted rats per sex per group for hematology and clinical chemistry analysis. Scheduled times for blood collection were as follows: week 6 (all groups), week 26 (control and high-dose groups), week 51 (control and high-dose groups), 78 (control and high-dose groups), and 103 (all groups). Additional samples were drawn 1 to 3 weeks after scheduled bleeding to verify results or to extend the analysis to other groups. Satellite animals were used for hematology and clinical chemistry analysis. Main study-animals were used as needed for blood collection when satellite animals died during the study. The CHECKED (X) parameters were examined.

a. Hematology:

X	Hematocrit (HCT)*	X	Leukocyte differential count*
X	Hemoglobin (HGB)*	X	Mean corpuscular HGB (MCH)*
X	Erythrocyte count (RBC)*	X	Mean corpusc. HGB conc.(MCHC)*
X	Leukocyte count (WBC)*	X	Mean corpusc. volume (MCV)*
X	Platelet count*	X	Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Clotting time)		
X	(Prothrombin time)		

* Recommended for combined chronic/carcinogenicity studies based on Guideline 870.4300.

b. Clinical Chemistry:

	ELECTROLYTES		OTHER
	Calcium*		Albumin*
	Chloride*		Creatinine*
	Magnesium*	X	Urea nitrogen*
	Phosphorus*		Total Cholesterol*
X	Potassium*		Globulins*
X	Sodium*	X	Glucose (nonfasting)*
	ENZYMES (more than 2 hepatic enzymes)*		Total bilirubin
X	Alkaline phosphatase (ALK)*	X	Total protein (TP)*
	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase	X	Serum protein electrophoresis
	Lactic acid dehydrogenase (LDH)		
X	Alanine amino-transferase (ALT/ SGPT)*		
X	Aspartate amino-transferase (AST/ SGOT)*		
	Gamma glutamyl transferase (GGT)*		
	Sorbitol		
	Glutamate dehydrogenase*		

* Recommended for combined chronic and carcinogenicity studies based on Guideline 870.4300.

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6. **Urinalysis:** Urine was collected overnight during weeks 6, 26, 52, 78, and 103 from the same 10 rats per sex per group as used for blood collection. The rats were deprived of water during collection. The CHECKED (X) parameters were examined.

	Appearance*	X	Glucose*
X	Volume*	X	Ketones*
X	Specific gravity / osmolality*	X	Bilirubin*
X	pH*	X	Blood/ red blood cells*
X	Sediment (microscopic)	X	Nitrate
X	Protein*	X	Urobilinogen

* Recommended for combined chronic toxicity and carcinogenicity studies based on Guideline 870.4300.

7. **Sacrifice and Pathology:** All main study and satellite animals that died on study and those sacrificed *in extremis* or on schedule by carbon dioxide asphyxiation were subjected to a detailed gross pathological examination. Bone marrow smears were collected from all animals sacrificed *in extremis* or on schedule, but were not examined. The CHECKED (X) tissues were collected for microscopic examination. All tissues from all animals dying or sacrificed before study termination and all tissues from control and high-dose group rats surviving to study termination were examined microscopically. In addition, the liver and thyroid gland from the low- and mid-dose groups were examined microscopically. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
X	Tongue	X	Aorta, thoracic*	XX	Brain (multiple sections)*+
X	Salivary glands*	XX	Heart*+	X	Periph.nerve*
X	Esophagus*		Bone marrow*		Spinal cord (3 levels)*
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*+	X	Eyes (retina, optic nerve)*
	Jejunum*	X	Thymus		GLANDULAR
X	Ileum*			XX	Adrenal gland*+
	Cecum*		UROGENITAL		Lacrimal gland
X	Colon*	XX	Kidneys*+	X	Mammary gland
	Rectum*	X	Urinary bladder*		Parathyroids*
XX	Liver*+	XX	Testes*+	XX	Thyroids*
	Gall bladder* (not rat)		Epididymides*+		OTHER
	Bile duct (rat)	X	Prostate*	X	Bone (sternum and/or femur)
X	Pancreas*	X	Seminal vesicle*	X	Skeletal muscle
	RESPIRATORY	XX	Ovaries*+	X	Skin*
X	Trachea*	XX	Uterus*+	X	All gross lesions and masses*
XX	Lung*+	X	Mammary gland*		
	Nose*				
	Pharynx*				
	Larynx*				

* Required for combined chronic/carcinogenicity studies based on Guideline 870.4300.
+Organ weight required in combined chronic/carcinogenicity studies.

II. RESULTS

A. OBSERVATIONS:

1. **Clinical signs of toxicity:** Clinical signs of toxicity were not summarized in tabular form. According to the text, body tremors were observed in two high-dose males and one high-dose female before week 90 of the study. After week 90 of the study, body tremors were observed in ten males and five females in the high-dose groups. The tremors occurred daily or intermittently over a 2-week period. Convulsions were observed in male and female rats at all dose levels, including controls and are not considered treatment related.

2. **Mortality:** No effect was observed on the mortality rate in female rats fed the test material for up to 104 weeks. At the end of the treatment period (104 week) 22, 20, 18, and 20 females in the 0-, 10-, 50-, and 250-mg/kg/day groups had died during the study. Therefore, 38 (63%), 40 (67%), 42 (70%), and 40 (70%) female rats survived to study termination.

   Mortality in male rats is summarized in Table 2. Larger numbers of male rats in all groups fed the test material died during the study than in the control group. A total of 1, 4, 1, and 8 male rats died during the first year of the study, 10, 9, 7, and 21 male rats died during the first 18 months, and 35, 47, 40, and 48 male rats died during the entire study in the 0-, 10-, 50-, and 250-mg/kg/day groups, respectively. Therefore, only 25 (42%), 13 (22%), 20 (33%), and 12 (20%) male rats survived to study termination. Survival rates in two groups (10 and 250 ppm) were below the minimal requirement according to current guidelines. The greatest excess deaths in the treated groups occurred during weeks 41-50, 61-70, and 91-100, when a total of 30 deaths occurred among high-dose rats compared with only 11 for the controls.

TABLE 2. Cumulative mortality in male rats

Weeks on Study	Dose (mg/kg/day)			
	0	10	50	250
Week 0	0	0	0	0
Week 52	1	4	1	8
Week 78	10	9	7	21
Week 103	35 (58%)	47* (78%)	40 (67%)	48** (80)
Survival at study termination	25/60 (42%)	13/60 (22%)	20/60 (33%)	12/60 (20%)

Data taken from Tables 2A and 2B (pp. 49-51) of the study report, MRID 97441.
*p<0.05, **p<0.01, statistically significant treated group compared with the control.

3. **Neurological Evaluations**

   Neurological evaluations were not conducted in this study and were not required at the time this study was conducted.

B. BODY WEIGHT

Mean body weights are summarized in Table 3. No treatment-related effects were observed on body weight in male or female rats fed any dose of the test material. Absolute body weights were similar throughout the study, and weight gain was similar, except intermediate- and high-dose group male rats lost less weight than controls during the last 26 weeks of the study and high-dose females gained more weight than controls during the same time period.

TABLE 3: Mean body weights and body weight gains

Week of Study	Dose (mg/kg/day)							
	0	10	50	250	0	10	50	250
Body Weight (g)	Males				Females			
Week 0	109	109	108	108	104	105	104	105
Week 52	736	742	730	732	375	387	383	384
Week 78	779	778	777	780	459	476	462	450
Week 103/104	685	658	714	720	460	477	471	472
Weight Gain (g)								
Weeks 0-52	627	633	622	624	271	282	279	279
Weeks 52-78	43	36	47	48	84	89	79	66
Weeks 78-103/104	-94	-120	-63	-60	1	1	9	22
Weeks 0-103/104	576	549	606	612	356	372	367	367
Food Consumption (g) Weeks 0-103/104	18,510	18,227	18,275	19,014	13,606	13,950	13,918	14,128
Food Efficiency[a]	3.11	3.01	3.32	3.22	2.62	2.67	2.64	2.60

Data obtained from Table 3, pages 52-54 and 57 in the study report, MRID 97441.
[a]Food efficiency = (total weight gain (g)/total food consumed (g) × 100)

C. FOOD CONSUMPTION AND COMPOUND INTAKE

1. **Food consumption**: Food consumption was similar among all groups of male and female rats. Total food consumed is summarized in Table 3.

2. **Compound consumption (time-weighted average)**: The concentration of test material in the diet was adjusted weekly or biweekly to maintain the same dosage throughout the study.

3. **Food efficiency:** Food efficiency was similar among all groups of male and female rats. Food efficiency values are summarized in Table 3.

D. **OPHTHALMOSCOPIC EXAMINATION:** The only notable abnormality observed during the eye examinations was retinal pallor in five high-dose females at 77 weeks and in four at 101 weeks compared with only one control at each time point.

E. BLOOD ANALYSES:

1. Hematology: Generally, statistically significant changes in hematologic parameters did not show a trend with time and did not exceed ±6% compared with controls except as noted. Prothrombin time was increased in high-dose male rats at 6 (+16%), 51 (+24%), and 53 weeks (+7%), but not at 26, 78 or 102 weeks. The lack of a sustained effect on prothrombin time suggests that the changes are not related to treatment with the test material. At study termination, platelet counts were 22% greater than controls in high-dose males, but were 13% and 16% less than controls in mid- and high-dose females, respectively. Also at study termination, the white blood cell count in high-dose males was elevated by 43% probably due to the 76% and 29% elevations in nucleophil and lymphocyte counts, respectively.

2. Clinical Chemistry: Statistically significant changes were observed in serum chemistry parameters, but the changes were not consistent with dose or duration of treatment. In addition, the values were within the normal range for Wistar rats and are, therefore, not considered related to treatment with the test material.

F. URINALYSIS: Urine protein levels were elevated in high-dose males after 5 weeks; the levels were within normal range and were not considered related to treatment with the test material. The remaining parameters were not affected by treatment with the test material.

G. SACRIFICE AND PATHOLOGY:

1. Organ weight: Selected organ weights are presented in Table 4. After feeding the test material for 103 weeks high-dose male rats had mean absolute and relative (to body weight) liver weights significantly greater than those of controls. The absolute and relative adrenal weight of high-dose males exceeded that of controls by about 10-fold because two rats had adrenal neoplasms. Excluding these animals resulted in adrenal weights being similar to those of the control group. Absolute lung and kidney weights of high-dose females were 16% and 8% less, respectively, than those of controls; the relative weights also were slightly less than those of the controls.

TABLE 4: Organ weights in rats fed permethrin for 2 years

Organ	Dose (mg/kg/day)			
	0	10	50	250
MALES				
Terminal body weight (g)	674 ± 87	653 ± 66	706 ± 93	717 ± 94
Liver (g)	27.5 ± 2.7	27.3 ± 3.5	29.2 ± 3.7	32.8 ± 4.6* (119)[a]
Liver (% body weight)	4.1 ± 0.6	4.2 ± 0.7	4.2 ± 0.6	4.6 ± 1.5** (112)
Adrenal (g × 1000)	84 ± 42	78 ± 22	76 ± 19	78 ± 14[b]
Adrenal (% body weight × 1000)	12.6 ± 5.9	12.2 ± 4.0	11.0 ± 2.9	11.1 ± 2.1[b]
FEMALES				
Terminal body weight (g)	458 ± 57	473 ± 76	463 ± 67	470 ± 72
Lungs (g)	2.5 ± 1.6	2.2 ± 0.9	2.2 ± 0.6	2.1 ± 0.3* (84)
Lungs (% body weight)	0.57 ± 0.40	0.47 ± 0.19	0.49 ± 0.18	0.46 ± 0.10* (81)
Kidneys (g)	3.6 ± 0.8	3.6 ± 0.5	3.4 ± 0.6	3.3 ± 0.5** (92)
Kidney (% body weight)	0.82 ± 0.29	0.78 ± 0.16	0.75 ± 0.17	0.70 ± 0.07* (85)

Data taken from Tables 10A and 10B, pages 135-138 of the study report, MRID 97441.
[a]Numbers in parenthesis are the percent of controls calculated by the reviewer.
[b]Absolute and relative weights before excluding two animals that had adrenal tumors were 850 g ± 1903** g and 115.5% ± 269.2%**, respectively.
*p<0.05, **p<0.01, statistically significant treated group compared with controls.

2. **Gross pathology** : The liver was swollen in 4 of 8 male rats in the high-dose group and 1/1 in the mid-dose group that died or were killed *in extremis* during the first 52 weeks of the study. The liver was not swollen in four low-dose or the one control that died during this time. Foci (unspecified), dark foci, or pale foci were observed on the adrenal glands of 6/58, 11/58, 7/65, and 13/60 female rats in the control, low-, mid-, and high-dose groups, respectively. No other notable gross lesions were observed in either male or female rats in the main study.

3. **Microscopic pathology:**

a. **Non-neoplastic**: Among the eight high-dose male rats that died during the first 52 weeks of the study, two had periacinar hepatocyte hypertrophy and three had periacinar necrosis. The single mid-dose male that died also had periacinar hypertrophy. These lesions were not found in the single control and the four low-dose male rats that died during the first 52 weeks.

Notable lesions in rats surviving more than 52 weeks are summarized in Table 5. The most notable finding in rats that survived more than 52 weeks was periacinar hepatocyte hypertrophy; the incidence was significantly increased in the mid- and high-dose group male and female rats. Mid- and high-dose male rats also had a significantly increased incidence of hepatocyte fatty vacuolation for all locations in the liver combined. The incidence of hepatocyte vacuolation was increased slightly but not significantly in females. High-dose male rats had significantly increased incidences of hyperplasia of the pelvic epithelium of the kidney, erythrocytes and erythrophagocytosis in the sinus of the thymic lymph nodes, and hypercellularity in the spleen. The study author did not report the number of male and female rats examined at each anatomical site. The reviewer assumed that the total number of animals of each sex examined was equal to the number examined at each site.

The study authors did present an inventory of the combined number of males and females examined at each site (MRID 77054).

Other findings showed significantly decreased incidences and/or severity in high-dose rats compared with the controls including nephropathy (decreased severity) in males and females, arteritis (decreased incidence) in females, parathyroid hyperplasia (decreased incidence) in both sexes, especially females, hypercellularity in the spleen (decreased incidence) of females, and hemorrhage in the thymus (decreased incidence) of males. The incidence of findings in the low- and mid-dose groups occasionally achieved statistical significance with no corresponding statistical increase in the high-dose groups. The lack of a dose-related trend suggests that these lesions are not treatment related, and, therefore, they are not listed in Table 5.

TABLE 5: Notable nonneoplastic lesions in male and female rats fed Z12

Organ/lesion	Dose (mg/kg/day)			
	0	10	50	250
Males – All rats surviving >52 weeks				
No. animals examined	59	56	58	52
Kidney				
Hyperplasia of the pelvic epithelium	2	2	1	9*
Liver				
Hepatocyte fatty vacuolation (any location)	9	16	17*	22**
Periacinar hepatocyte hypertrophy	3	2	11*	19**
Lymph nodes (thymic)				
Erythrocytes and erythropagocytosis in sinuses	1	5	2	6*
Spleen				
Hypercellular	9	19*	16	19**
Females – All rats surviving >52 weeks				
No. animals examined	58	58	60	60
Liver				
Hepatocyte fatty vacuolation (any location)	26	7	11	31
Periacinar hepatocyte hypertrophy	3	2	17*	38**

Data taken from Tables 11C (pp. 143-166), 11E (pp. 170-180), 11G (pp. 184-188) of the study report, MRID 97441.

b. Neoplastic: There were no statistically significant increases in the incidences of neoplasms at any anatomical site that could be attributed to treatment with any dose of the test material. Common neoplasms occurring with high incidence in all groups included pituitary adenoma/carcinoma in male and female rats, thyroid follicular/parafollicular cell adenomas/carcinomas in males, and mammary fibroepithelial carcinomas in females.

III. DISCUSSION and CONCLUSIONS

A. INVESTIGATORS' CONCLUSIONS: The study authors concluded that feeding of 21Z (permethrin) caused a decrease in survival of male rats at 250 mg/kg/day, hepatocyte hypertrophy in males and females at 50 and 250 mg/kg/day, and no effect at 10 mg/kg/day.

B. REVIEWER COMMENTS: Male rats fed 10- and 250-mg/kg/day of the test material had significantly decreased survival rates at the end of the study compared with the controls. Both groups had fewer than the 25% minimum number of survivors at study termination. The study authors did not report the cause of death for animals in any group; therefore, any association of the deaths with treatment with the test material is equivocal. Further there is no explanation for the high mortality rate in the low-dose group. Notable clinical signs were restricted to tremors observed in high-dose male and female rats during a 2-week period late in the study. Body weights, body weight gain, and food consumption were similar in treated and control groups throughout the study and overall food efficiency was also similar in the treated and control groups. Except for retinal pallor in a few high-dose female rats, no noteworthy eye abnormalities were observed.

Hematologic and clinical chemistry evaluations revealed statistically significant changes that were transient, small in magnitude, or lacked a dose-related trend. Any statistical changes noted were not associated with any other pathological findings. No treatment-related changes were observed in urinalysis parameters. Postmortem evaluations showed significantly elevated absolute and relative liver weights in high-dose male rats that corresponded to an increased hepatocyte hypertrophy observed microscopically. In addition, hepatocyte hypertrophy was observed in mid-dose males and mid- and high-dose females. The high-dose group male rats had significantly elevated absolute and relative adrenal weights when two rats with adrenal neoplasms were included. Excluding these two animals resulted in high-dose male rats having adrenal weights similar to those of controls. High-dose female rats had significantly reduced absolute and relative lung and kidney weights. Nephropathy was observed in a large percentage of male and female rats; however, the severity was decreased in both high-dose males and females compared with the controls. The decreased severity in high-dose females may have been related to the decreased weight of the kidney in females and is not considered an adverse finding. The decreased lung weight was not associated with any pathologic findings; therefore, the weight change is not considered toxicologically significant. Other treatment-related microscopic changes observed in the high-dose group male rats included increased incidences of hyperplasia of the pelvic epithelium, hepatocyte fatty vacuolation, and erythrocytes and erythrophagocytosis in the thymic lymph nodes. The incidence of hepatocyte fatty vacuolation was also significantly increased in the mid-dose group male rats and marginally ($p=0.07$) increased at the low-dose. The study authors did not consider hepatocyte fatty vacuolation to be a treatment-related finding, but a degenerative change common to the Wistar rat. Because the incidence was significantly increased at the mid and high doses and marginally increased at the low dose, the reviewer considers this finding to be related to treatment with the test material. There was no corresponding statistically significant increase in the incidence of hepatocyte fatty vacuolation in female rats. The significantly increased incidence of hypercellularity in the spleen is not considered treatment related because of the lack of a dose-related trend. On April 18, 2002, the HIARC evaluated the toxicology database of permethrin and determined that the increased liver weight and hypertrophy observed in the liver are adaptive and reversible effects and are not considered adverse effects.

Under the condition of this study, the lowest-observed-adverse-effect level (LOAEL) for permethrin (21Z) in rats is 250 mg/kg/day based on tremors observed in males and females; the corresponding no-observed-adverse-effect level (NOAEL) is 50 mg/kg/day.

No treatment-related increased incidences of neoplasms were observed in male or female rats receiving any dose of the test material. Common neoplastic findings included pituitary adenomas/carcinomas in both sexes, thyroid follicular/parafollicular adenomas/carcinomas in males, and mammary fibroepithelial carcinomas in females. Based on the significantly increased incidence of hepatocyte fatty vacuolation and periacinar hepatocyte hypertrophy in the liver of both male and female rats it appears that these animals were adequately dosed. In addition, mortality in the groups of male rats was high due to unknown causes. The study authors did not provide a dose selection rationale to judge the basis for the dose selection.

C. **STUDY DEFICIENCIES:** The following items were not provided:
   a. physical description, purity, and stability of the test material,
   b. data on homogeneity, stability, and concentration verification of test material in the diet,
   c. a dose selection rationale,
   c. blood samples from low- and mid-dose animals for hematologic and clinical chemistry evaluations except for the first and last sampling periods, and
   d. a tissue inventory for microscopic lesions.

Tox Review # 50649

Page ___ is not included in this copy.

Pages 305 through 309 are not included in this copy.

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DATA EVALUATION REPORT

PERMETHRIN/109701
(21Z73)
STUDY TYPE: RODENT DOMINANT LETHAL ASSAY IN MICE
[OPPTS 870.5450 (§84-2)]
MRID 00070583
Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 02-05

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Disclaimer

This review may have been altered subsequent to the contractor's signature above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725

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Date 8/8/2002
Signature: Joycelyn Stewart
Date

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Rodent dominant lethal assay in mice [OPPTS 870.5450 (§84-2)]

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 21Z73 (Permethrin, purity not provided)

SYNONYMS: 3-phenoxybenzyl dl, cis, trans 2,2-dimethyl-3-(2,2-dichlorovinyl) cyclopropane-1-carboxylate

CITATION: Chesher, B.C., J.C. Malone, M.J. Parker (1975) 21Z73, dominant lethal study in male mice. Research and Development (V & A), Wellcome Research Laboratories, Berkhamsted. Dec. Code: HEFG 75-10, Lab Ref. No.: T.L. 37-75. November 27, 1975. MRID 00070583. Unpublished

SPONSOR: FMC Co.

EXECUTIVE SUMMARY: In a mouse dominant lethal assay (MRID 00070583), 10 male CD-1 mice were treated once daily via the oral route with 21Z73 at a dose of 452 mg/kg body weight per day for five consecutive days. The test agent was mixed at 40% w/v in corn oil. Starting immediately after the final dosing, each male was mated with 3 untreated virgin females per week for six weeks. The females were sacrificed 14 days after the midweek of their mating period and the pregnancy rate and the number of living and dead implants determined.

21Z73 was tested at 1/5 LD_{50} daily for 5 days (a total final dose equal to the acute LD_{50} value). The mice showed signs of hypersenitivity (which is consistent with other pyrethroids) lasting for about three hours following the fourth and fifth doses but no other clinical signs were noted. All treated mice survived until the end of the study. The trimethylphosphate positive control reduced the pregnancy rate and increased the percentage of dead implants compared to the corn oil solvent control in weeks one and two. The differences were statistically significant. The solvent control values were appropriate. **There was no statistically significant difference between the control group and the 21Z73 treated group with respect to fertilization rate or the number of living and dead implantations in any week.**

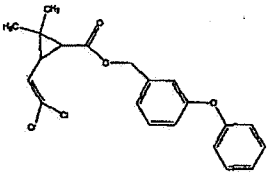
This study is classified as **Unacceptable/guideline (upgradeable).** It is upgradeable if the data on purity of the test material is provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test material: 21Z73, permethrin

Description:	Not provided
Lot/Batch #:	ZB
Purity:	Not provided
CAS # of TGAI:	52645-53-1
Solvent Used:	Corn oil



2. Control materials:

Negative:	None
Solvent / final volume / route of administration:	Corn oil / 0.2 mL / oral gavage
Positive / final dose(s) / route of administration:	Trimethylphosphate / 40% w/v in corn oil / oral gavage

3. Test compound administration:

Volume:	0.2 mL/mouse
Route of administration:	Oral gavage
Dose level used:	452 mg/kg/day for 5 consecutive days

4. Test animals:

Species:	Mouse
Strain:	CD-1
Age/weight at study initiation:	Males: not provided Females: not provided
Source:	Charles River, UK
No. animals used per dose:	10 males mated to 3 untreated females/week for 6 weeks
Properly Maintained?	Iinformation not provided

B. TEST PERFORMANCE:

1. Treatment:

a. Test compound and solvent control
Dosing: __ once __ twice (24 hr apart)
_x_ other (describe): once daily for 5 consecutive days

2. **Mating:** Immediately after the last treatment each male was caged with three untreated females. The females remained with the male for one week at which time they were removed and replaced with three new females. This process was repeated for six weeks. A six-week mating period in mice allows sampling of sperm treated at all germ cell stages.

3. **Caesarian procedures:** Not described

4. **Evaluation criteria:** Pregnancy rates and the numbers of living and dead implants were determined for each animal and each treatment group. Differences between groups were statistically evaluated for significance.

5. **Statistical methods:** Statistical tests were two-tailed and performed at the 5% level. Pregnancy rates in the solvent control group and the positive control and the test material treated groups were compared using Fisher's exact test. Preimplantation losses were determined by using implant data from all pregnant females to calculate the average number of implants per male with one set of data for each test group. The two-sample permutation test was used to compare the solvent control group with the positive control group and with the test material group. A Chi-square test was used for an intragroup comparison of the proportion of dead implants attributable to each male. If statistically significant differences in the proportion of dead implants were seen between males in the same group, then the F-test was used to evaluate intergroup variation. If no statistically significant differences were seen in the intragroup comparison, then a Chi-square test was used for the intergroup analysis.

II. REPORTED RESULTS:

A. PRELIMINARY TOXICITY ASSAY: No preliminary toxicity assay was reported.

B. DOMINANT LETHAL ASSAY: Slight hypersensitivity, lasting about three hours, was seen in treated males following the fourth and fifth doses of 21Z73. No mortality or other clinical signs were noted. Statistically significant decreases in the pregnancy rate were seen in the positive control group in week 1 and 2 but not at later mating intervals. The pregnancy rate was statistically higher at week 1 in the 21Z73 group (96.67%) compared to the solvent control group at week 1 (63.33%); however, the solvent control value was particularly low. No statistically significant decreases in pregnancy rate were seen at any time in the 21Z73 group.

A summary of the number of living and dead implants is given in Table 1.

TABLE 1. Summary of the number of living and dead implants (group totals)

Week	Solvent Control		Positive Control		21Z73	
	Living	Dead	Living	Dead	Living	Dead
1	208	10	0	2	309	18
2	305	10	81	55	326	23
3	300	13	216	17	332	21
4	300	12	255	9	273	14
5	243	29	212	11	245	14
6	312	24	263	5	356	13

A statistically significant decrease in the mean number of implants, compared to the solvent control group, was seen in the positive control group in weeks 1, 2 and 5. A summary of the mean number of implants for the solvent and 21Z73 treated groups is given in Table 2.

TABLE 2. Mean implant comparison between solvent control and 21Z73 groups				
Week	Mean Number of Implants		Difference Between Means	Probability
	Solvent Control	21Z73		
1	11.35	11.25	0.1	0.824
2	12.73	12.58	0.15	0.856
3	12.12	12.58	-0.47	0.324
4	11.03	11	0.03	0.988
5	12.41	10.25	2.16	0.002
6	11.6	12.3	-0.7	0.23

Statistically significant increases in the percent of dead implants compared to the solvent control values were seen in the positive control group at weeks 1 and 2. A comparison between the solvent control and 21Z73 groups is shown in Table 3. No statistically significant increase in the percent dead implants was seen at any mating interval in the 21Z73 treated group.

TABLE 3. Percentage of dead implant comparison between solvent control and 21Z73 groups				
Week	Percent Dead Implants		Between Groups Chi-square or F-ratio	Probability Value
	Solvent Control	21Z73		
1	4.59	5.5	F(1,18) = 0.0744	0.7882
2	3.17	6.59	F(1,25) = 0.5915	0.449
3	4.15	5.95	C-S (1) = 1.1042	0.29
4	3.85	4.88	F(1,26) = 0.2042	0.6551
5	10.66	5.41	C-S (1) = 4.9254	0.03
6	7.14	3.52	F(1,27) = 1.2881	0.2664

III. REVIEWER'S DISCUSSION/CONCLUSIONS:

A. **DISCUSSION**: 21Z73 did not induce dominant lethal mutations in the germ cells of male mice as tested in this study. The test material was administered at a dose said to be one-fifth the acute LD_{50} per day for five consecutive days, proper experimental protocol was followed and the solvent and positive control values were appropriate. The study is classified as unacceptable/guideline because not all acceptance criteria were met as noted in the Study Deficiencies section.

B. **STUDY DEFICIENCIES**: No signed and dated GLP, Data Confidentiality or Quality Assurance Statements were provided and the test material purity and physical description were not given. The mice were said to be sexually mature but the actual age and weights were not provided. The study does provide useful information although not all acceptance criteria were met.

DATA EVALUATION RECORD

PERMETHRIN
(21Z73)

STUDY TYPE: SUBCHRONIC ORAL TOXICITY - RAT
[OPPTS 870.3100 (§82-1a)]
MRID 00025914

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-89

Primary Reviewer:
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Date: MAR 28 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

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PERMETHRIN/PC Code 109701

Subchronic (90-day) Oral Toxicity Study (rodent) / 1
[OPPTS 870.3100/OECD 408 (§82-1a)]

EPA Reviewer: Linnea Hansen, Ph.D.
Toxicology Branch (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch (7509C)

Signature: Linnea Hansen
Date: January 30, 2002
Signature: Joycelyn Stewart
Date: Feb 15, 2002

TXR#: 0050649

DATA EVALUATION RECORD

STUDY TYPE: Subchronic Oral Toxicity Study - Rat [OPPTS 870.3100 (§82-1a)]

P.C. CODE: 109701

DP BARCODE: D269531
SUBMISSION CODE: S504352

TEST MATERIAL (PURITY): 21Z73 (purity not reported)

SYNONYMS: Permethrin; 3-phenoxybenzyl dl, cis, trans 2,2-dimethyl-3-(2,2-dichlorovinyl) cyclopropane-1-carboxylate

CITATION: Williams, L.M., Thompson, P.M., Clampitt, R.B., and Malone, J.C. (1976) 21Z73, Rat oral 90 day study. Wellcome Research Laboratories, Berkhamsted and Beckenham. Lab. Ref. No. TL. 3-76 (Document Code HEFG76-1). February 25, 1976. MRID 00025914. Unpublished.

SPONSOR: Wellcome Research Laboratories, Berkhamsted and Beckenham.

EXECUTIVE SUMMARY: In a 90-day dietary toxicity study (MRID 00025914), groups of 18 C.S.F. Wistar rats/sex/dose were given 21Z73 (a.i. not reported, Lot/BatchWP) administered in feed at 0, 200, 600, 2000 or 4000 ppm (equivalent to 17.0, 49.9, 179.6 or 357.4 mg/kg/day for males and 0, 18.5, 56.2, 176.5 or 356.7 mg/kg/day for females). Six/sex/group were continued without test compound for an additional 36 days during the recovery period.

At 4000 ppm, all male and female animals showed signs of hypersensitivity beginning on day 1. By week 3, hypersensitivity abated from males but persisted in females throughout the study period. Hypersensitivity in 4000 ppm females disappeared within 3 days after the test substance was removed from the diet. Mean body weight/weight gain were statistically decreased for 4000 ppm males during the study (data illegible-estimated from study graph at -7.7%/-5%; however, initial 4000 ppm mean body weights may be low due to problems with watering system during first weeks of study), but improved during the recovery period. Increased liver weights were observed in 4000 ppm males (relative 43% above controls) and females (absolute/relative increases of 22%/14%) during the study period, but returned toward control values during the recovery period. In addition, absolute and relative thyroid weights were reportedly decreased in 4000 ppm males and females (data not legible). No treatment-related deaths were reported. There were no treatment-related changes in food consumption, food efficiency, hematology, clinical chemistry, urinalysis, histopathology or estrus cycle parameters. **The LOAEL is 4000**

ppm (equivalent to 357.4 mg/kg/day, males and 356.7 mg/kg/day, females) based on decreased body weight/weight gain in males and hypersensitivity reactions in both sexes. The NOAEL is 2000 ppm (equivalent to 179.6 mg/kg/day, males and 176.5 mg/kg/day, females).

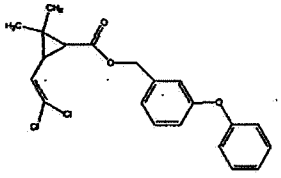
This study is classified as **Unacceptable/Guideline (upgradable) [OPPTS 870.3100/OECD 408 (§82-1a)]** and does not satisfy the Subdivision F guideline requirements for a subchronic oral study in the rodent. It can be upgraded to Acceptable/Guideline upon submission of (1) information on the purity of the test material; (2) information on the diets' homogeneity and test material concentration analyses; (3) submission of a legible copy of the study to verify investigators' conclusions and (4) individual animal clinical examination data. However, a new study is not required because this data requirement may be satisfied with the chronic toxicity/carcinogenicity study in the rodent (OPPTS 870.4300/OECD 453 [§83-5(a)]).

COMPLIANCE: Signed and dated GLP, Quality Assurance, Flagging statement and Data Confidentiality statements were not provided. This study was conducted prior to the promulgation of the current US EPA GLP guidelines.

I. MATERIALS AND METHODS

A. MATERIALS

1. **Test material:** 21Z73
   Description: not reported.
   Lot/Batch # : WP
   Purity: a.i. not reported.
   Stability of compound: reported as 30 days in feed (data not provided).
   CAS #: 52645-53-1
   Structure:



2. **Vehicle and/or positive control:** None; the test material was administered in the feed.

3. **Test animals**
   Species: rat.
   Strain: C.S.F. Wistar
   Age/weight at study initiation: weanling males and females, exact age not reported; 64 - 166 g.
   Source: Charles River (facility not indicated).
   Housing: 6/cage/sex/dose (cage type not indicated).
   Diet: Diet 41B meal ex Heygates, *ad libitum.*
   Water: tap, *ad libitum.*
   Environmental conditions: 21°C ± 2°C
   Light/dark cycle: 12 hours.
   Acclimation period: not reported.

B. STUDY DESIGN

1. **In life dates:** Not indicated. The dates of study conduct were given as Start: 3/75; End: 10/75

2. **Animal assignment**: Animals were assigned to one of 5 groups (Table 1). Eighteen rats/sex/dose were given dietary 21Z73 at target concentrations of 0, 200, 600, 2000 or 4000 ppm for 90 days. Ten rats/sex/dose were killed after the 90 day study period (main group) and used for data collection. Six rats/sex/dose were fed diet without the test substance for an additional 36 days and then killed after the recovery period (satellite group). The fate of the other 2 rats/sex/group was not reported but they were apparently removed from the study.

TABLE 1. Study design			
Target dose (ppm)	Number of animals		Actual mean dose (mg/kg/day)
Males			
	Main Group (days 0-90)	Satellite Group (days 0-126)	
0	12	6	0
200	12	6	17.0
600	12	6	49.9
2000	12	6	179.6
4000	12	6	357.4
Females			
0	12	6	0
200	12	6	18.5
600	12	6	56.2
2000	12	6	176.5
4000	12	6	356.7

Data taken from pp. 163 and 169; MRID 00025914.

3. **Dose selection rationale:** Not reported.

4. **Test diet preparation and analysis:** A 10% premix of 21Z73 in diet was prepared every 2-3 weeks, and the diets in the animal cages were changed twice weekly. Diets were reported to be stable for 30 days but actual data were not provided. Information on homogeneity and concentration were not provided. It was therefore not established whether the mixing procedure was adequate or the variance between nominal and actual dosage to the animals was acceptable.

5. **Statistics:** Statistical comparisons were done using the t-test for body weight, estrus cycle, hematology and clinical chemistry.

A. METHODS

1. **Observations:** Animals were observed daily for mortality and moribundity.

2. **Body weight:** Animals were weighed weekly throughout the study, apparently beginning on day 0.

3. **Food consumption, compound intake and food efficiency:** Food consumption (calculated as g/kg/day for the 0-90 day and 91-126 day periods) and compound intake (mg/kg/day) were recorded weekly. Food efficiency was also determined (calculated as g food consumed/g body weight/week). 5

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4. **Blood was collected** from 6/sex/satellite dose group animals at 14, 28, 56 and 90 days for hematology and clinical biochemical analysis from the orbital plexus under light ether anesthesia. At 90 days, 10 rats/sex/main dose group were orbitally bled prior to killing. At 126 days, the 6 rats/sex/satellite group dose were killed after the orbital bleed. The animals appeared to have been fasted, based on the statement that animals were fasted prior to the urine collection on days 90 and 126, but this was not specifically stated in the report. The CHECKED (X) parameters were examined.

a. **Hematology**

X	Hematocrit (HCT)*	X	Leukocyte differential*
X	Hemoglobin (HGB)*	X	Mean corpuscular HGB (MCH)
X	Leukocyte count (WBC)*	X	Mean corpusc. HGB conc. (MCHC)
X	Erythrocyte count (RBC)*	X	Mean corpusc. volume (MCV)
	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Required for subchronic studies based on Subdivision F Guidelines

b. **Clinical chemistry**

	ELECTROLYTES		OTHER
	Calcium*	X	Albumin*
	Chloride*		Creatinine*
	Magnesium	X	Blood urea nitrogen*
	Phosphorus*		Total Cholesterol
X	Potassium*		Globulins
X	Sodium*	X	Glucose*
			Total bilirubin
	ENZYMES	X	Total serum protein (TP)*
X	Alkaline phosphatase (ALK)		Triglycerides
	Cholinesterase (ChE)		A/G ratio
	Creatine phosphokinase (CPK)		
	Lactic acid dehydrogenase (LDH)		
X	Alanine aminotransferase (ALT)*		
X	Aspartate aminotransferase (AST)*		
	Gamma glutamyl transferase (GGT)		
	Glutamate dehydrogenase (GDH)		

* Required for subchronic studies based on Subdivision F Guidelines

5. **Urinalysis**

Urinalysis was done using composite samples for each dose level and sex collected overnight from 6/sex/dose group animals (fasted, water deprived) on days 90 or 126. The CHECKED parameters were examined. ✓

	Volume*	X	Protein*
	Specific gravity*	X	Glucose*
	Appearance*	X	Ketones*
X	Sediment*		Urobilinogen
X	pH*		Bilirubin*
			Nitrites
		X	Blood*
			Leukocytes

* Required for subchronic studies based on Subdivision F Guidelines

D. ESTRUS CYCLE

Vaginal smears were prepared daily from main group female animals during days 60 - 90.

E. OPHTHALMOLOGIC EXAMINATION

Ophthalmologic examinations were not done.

F. SACRIFICE AND PATHOLOGY

After 90 days, 10 rats/sex/dose (main group) were sacrificed under CO_2 anesthesia by exsanguination. No information was provided about the remaining two animals/sex/dose group from the main group or on what basis animals to be examined were selected. All satellite group animals were killed in the same manner after 126 days. Necropsies and histologic examination were done on all main group animals. Only necropsies were done on the satellite group animals. The CHECKED (X) tissues were collected for histological examination. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVAS./HEMAT.		NEUROLOGIC
X	Tongue	X	Aorta*	XX	Brain*
X	Salivary glands*	XX	Heart*	X	Periph. nerve*
#	Esophagus*	#	Bone marrow*		Spinal cord (3 levels)[T]
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*	X	Eyes (optic n.)
#	Jejunum*	XX	Thymus*		
#	Ileum*				GLANDULAR
#	Cecum*		UROGENITAL	XX	Adrenal gland*
X	Colon*	XX	Kidneys*+		Lacrimal gland[T]
	Rectum*	X	Urinary bladder*	#	Mammary gland[T]
XX	Liver*+	XX	Testes*+		Parathyroids*
	Gall bladder*	#	Epididymides	X	Thyroids+
X	Pancreas*	X	Prostate		
		X	Seminal vesicle		OTHER
	RESPIRATORY	XX	Ovaries	#	Bone
#	Trachea*	XX	Uterus*	X	Skeletal muscle
XX	Lung*	X	vagina	#	Skin
	Nose			#	All gross lesions and masses*
	Pharynx				
	Larynx				

* Required for subchronic studies based on Subdivision F Guidelines
\+ Organ weight required in subchronic studies.
[T] Required only when toxicity or target organ
\# Tissue preserved but not examined

II. RESULTS

A. OBSERVATIONS

1. **Toxicity:** All 4000 ppm male and female animals showed signs of hypersensitivity (details not specifically described) beginning on day 1. By week 3, hypersensitivity abated in males but persisted in females throughout the 90 day study period. Hypersensitivity disappeared within 3 days after the test substance was removed from the diet of 4000 ppm females. Individual animal clinical finding data were not provided.

2. **Mortality:** Three male rats (one 600 and two 2000 ppm) died during the study, however none of the deaths were treatment related.

B. **BODY WEIGHT AND WEIGHT GAIN:** Body weight data were summarized in two figures (pp. 167-8 of MRID 00025914). Individual and mean data values for days 0 - 90 were illegible in the text tables, however recovery period values were legible (Appendix, Tables 1 and 1A, pp. 184-7 of MRID 00025914). Depiction of these data in figures provided with the study (pp 167, 167A, 168 and 168A of MRID 00025914) provided an estimate of comparative weight gain, but statistical analysis could not be provided by the reviewer. The study authors conclude that body weight was significantly decreased for

4000 ppm males only. Based on examination of the graphical presentation of the data, approximate mean body weights for males at the end of the dosing period (day 90) were estimated to be (control to high dose, respectively) 452, 440, 461, 455 and 417 g (high dose -7.7% less than controls). Weight gain of the high dose males during treatment, also estimated from this graph, was approximately only -5% less than controls; however, this may be artificially low due to the problems with the watering system that occurred in the treated animal groups during the first weeks. The study authors reported that some small differences in body weights of 200 ppm females, 2000 ppm males and 4000 ppm males occurred due to a problem with the watering system from day 0 - 14 of the study. After these problems were resolved, body weights of the 200 ppm female and 2000 ppm male groups were similar to controls. On day 91 (first day of recovery for the satellite recovery group), mean weights for males were (control to high dose, respectively) 436, 447, 424, 454 and 391 (high dose -10.3% less than controls). Mean body weights of females did not show significant differences. At day 90, the approximate mean body weights for females were 256, 260, 252, 266 and 252.

In the recovery groups, body weights improved towards control values during the recovery period. At the end of the recovery period, mean body weights of males were 460, 492, 468, 511 and 446 g. Mean body weights of females were 295, 286, 298, 303 and 276 g.

C. FOOD CONSUMPTION, COMPOUND INTAKE

1. **Food consumption:** No differences were reported between treatment groups during the study period. (Table 2). During the recovery period, food consumption was reduced 16% for 2000 ppm males (statistical analysis was not included by the authors and could not be done by the reviewer given the data presentation.) This decrease is relatively small, showed no dose-response and is not considered treatment-related.

TABLE 2. Mean food consumption (g/kg/day) of animals fed 21Z73 (Permethrin) for 90 days

Sex	Treatment group (ppm)				
	0	200	600	2000	4000
Days 0 - 90 (study period)					
Males	82.8	83.2	83.2	82.1	83.8
Females	89.7	92.2	93.6	88.2	89.2
Days 91 - 126 (recovery period)					
Males	81.2	81.0	76.0	68.2 (-16)	88.8
Females	93.0	95.4	98.2	95.8	103.4

Data taken from Tables 2 and 2A, pp. 188; MRID 00025914.
Numbers in parentheses are percent difference from control.

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2. **Compound consumption:** Compound consumption is shown in Table 1. Both males and females consumed comparable amounts of test substance during the study.

3. **Food efficiency:** Food conversion was not different for any treated group versus control during the study period (Table 3). Data from weeks 1-12 of the treatment period indicated no consistent pattern of treatment-related food conversion changes. Similarly, during recovery, there was no consistent dose-related changes in food conversion. During Week 14 (recovery), all groups of males including controls showed weight loss or at 4000 ppm, very low conversion, while females showed weight loss at 200 ppm and low conversion at 4000 ppm (but not at later times). During Week 18 of the recovery period, food conversion of 2000 ppm females was increased 75% and was also increased by 113% in males at 4000 ppm. This observation is likely not toxicologically relevant, however. (Statistical analysis was not reported by the authors and could not be done by the reviewer given the data presentation.)

TABLE 3. Mean food conversion[a] of animals fed 21Z73 (Permethrin) for 90 days at selected time points

Sex	Treatment group (ppm)				
	0	200	600	2000	4000
Week 13 (end of study period)					
Males	12.7	10.6	11.9	10.1	12.0
Females	28.5	20.7	26.7	20.3	27.9
Week 18 (end of recovery period)					
Males	3.1	1.6	2.6	2.4	6.6
Females	4.1	3.9	3.4	7.2 (+75)	4.1

Data taken from Tables 4 and 4A, pp 190; MRID 00025914.
[a] Food conversion = g food consumed/ g body weight increase over 7 day period.
Numbers in parentheses are percent difference from control.

D. BLOOD WORK

1. **Hematology:** The study report provided a summary table (Tables 1-3, p. 172-174 of MRID 00025914) showing increases and decreases of hematology parameters relative to controls. In the individual animal tables, means and standard deviations were also provided for each group; however, parts of the data table were illegible (Tables 6, 6A, 7 and 7A, pp. 192-210 of study report). Small, sporadic, statistically significant variations in some hematologic parameters were reported during the study, however there were no toxicologically relevant dose-related effects. A slight, transient leukopenia (data now shown) was reported at days 14 and 28 in 4000 ppm males and females, but these decreases were generally not statistically significant and were not present at the 90-day terminal bleed. The latter finding could not be confirmed due to illegibility of the data tables in the Appendix.

TABLE 4. Selected mean ± SE hematology parameters of rats fed 21Z73 for 90 days. Terminal Values at 90 days					
Parameter	Treatment Group (ppm)				
	0	200	600	2000	4000
MALES					
MCH (pg)	20.1 ± 0.3	19.9 ± 0.3	20.0 ± 0.3	19.7 ± 0.3	18.9 ± 0.2*(-6)
FEMALES					
HCT (%)	46.3 ± 0.4	45.0 ± 0.5*(-3)	45.3 ± 0.2*(-2)	44.8 ± 0.4*(-2)	45.2 ± 0.6
MCV (fL)	62.0 ± 1.2	60.4 ± 1.3	66.7 ± 1.4*(+8)	64.9 ± 0.4*(+5)	61.6 ± 1.0
MCH (pg)	21.2 ± 0.5	20.8 ± 0.5	23.3 ± 0.6**(+10)	21.7 ± 0.3	21.1 ± 0.2
RBC (10^6/mm^3)	7.5 ± 0.1	7.5 ± 0.1	6.8 ± 0.1**(-9)	6.9 ± 0.1**(-8)	7.4 ± 0.1

Data taken from Table 6A, pp. 198, MRID 00025914.
* Statistically significant versus control, $p \leq 0.05$.
** Statistically significant versus control, $p \leq 0.01$.
Numbers in parentheses are percent difference from control.

2. **Clinical chemistry:** Clinical chemistry data were shown in Table 5, pp.175 of MRID 00025914 not as data values, but as "decrease, increase and/or statistically significant." Actual data values were presented in the Appendix, Table 9A, pp. 221 of MRID 00025914, but most were unreadable even with magnification. The study authors report that no toxicologically relevant, dose-related changes in clinical chemistry parameters occurred during the study. Based on the actual data values that could be deciphered, the reviewer agrees with their conclusion but all of the parameters could not be verified.

E. **URINALYSIS:** The samples analyzed for each sex and dose group represented one composite sample; therefore, no individual animal data were available. No treatment-related observations were reported. Small quantities of blood observed in the collected urine of 600 ppm male rats were likely due to a cut on the foot of one of the animals.

F. **ESTRUS CYCLE:** No treatment-related findings were reported in the number of estrus cycles in treated versus control groups.

G. **SACRIFICE AND PATHOLOGY:**

1. **Organ weight:** Organ weight data, shown in the text on page 176 of MRID 00025914, were not expressed as actual data values but as "increase, decrease or significant" compared to controls. Individual and mean relative and absolute organ weight values were included in the Appendix (pp. 224 - 231, MRID 00025914), but many of the values were unreadable, even with magnification.

The study authors report no consistent, dose-related changes in absolute or relative kidney, spleen, lung, heart, brain, thymus, gonads, adrenals or pituitary weights during

either the study or recovery period. Absolute liver weights in 4000 ppm females were increased by about 22% relative to controls (statistically significant), but no increase was seen in males. However, relative liver weights were significantly increased in both sexes (males 43% and females 14% above controls). The absolute mean liver weights for each group (shown as control to high dose, respectively) were difficult to read but appeared to be 12.43, 11.24, 13.98, 12.77 and 13.19 g, males and 6.83, 7.17, 6.84, 8.01 and 8.30 g, females. Relative liver weight values were also difficult to read but appeared to be 2.46, 2.45, 2.95, 3.24 and 3.53%, males and 3.15, 3.29, 3.19, 3.53 and 3.59%, females. Liver weights returned to control levels at the end of the recovery period, however. In addition, absolute and relative thyroid weights in 4000 ppm males and females were decreased at the end of the study period (actual data values were not legible). These organ weight changes in liver and thyroid were not accompanied by microscopic changes or clinical laboratory changes and are therefore not considered to be of toxicologic significance.

2. **Gross pathology:** No treatment-related findings were reported.

3. **Microscopic pathology:** Histologic examinations of control and 4000 ppm animals were conducted. No treatment-related abnormalities were reported. (Actual histology data were not legible in the text).

III. DISCUSSION

A. **INVESTIGATORS' CONCLUSIONS:** Dietary administration of 0, 200, 600, 2000 or 4000 ppm 21Z73 to male and female rats for 90 days resulted in increased liver weight and hypersensitivity reactions from male and female animals given the 4000 ppm dose. In addition, body weights of 4000 ppm males decreased during the study. Both sexes showed a slight leukopenia only during the first weeks of treatment. A decrease in thyroid weights was also observed at 4000 ppm. No treatment-related toxicologic changes were observed in any other parameter tested during the study and/or recovery period. All of these changes observed during the study treatment period resolved toward control levels when the test substance was removed from the diet. There were no treatment-related alterations observed in either sex at 2000 ppm or below.

B. **REVIEWER'S COMMENTS:** The reviewer tentatively agreed with the conclusions of the study authors, which could not always be verified due to illegibility of some data tables. Hypersensitivity, observed in both sexes (transiently in males, more persistently in females but disappearing soon after cessation of treatment) and decreased body weight gain in males are considered to be treatment-related adverse effects. Although the increased liver weights in both sexes are considered to be related to treatment, in the absence of correlated alterations in clinical laboratory findings or liver histopathology, the increases in this study are considered to be adaptive changes rather than adverse effects. Similarly, the decreases in thyroid weight were not accompanied by microscopic findings. The transient decreases in WBC count in the early weeks of the study were not considered biologically significant and also did not persist throughout treatment.

The LOAEL for permethrin in male and female Wistar rats is 4000 ppm (equivalent to 357.4 mg/kg/day in males and 356.7 mg/kg/day in females), based on hypersensitivity (males and females) and decreased body weight gain (males). The NOAEL is 2000 ppm (equivalent to 179.6 mg/kg/day in males and 176.5 mg/kg/day in females). The study is classified as **Unacceptable/guideline (upgradable)**. It may be upgraded to Acceptable/Guideline upon submission of (1) information on the purity of the test material; (2) information on the diets' homogeneity and test material concentration analyses; (3) a legible copy of the study report so that all data tables may be examined and (4) individual animal data for clinical observations. However, a new study is not required because this data requirement may be satisfied with the chronic toxicity/carcinogenicity study in the rodent (OPPTS 870.4300/OECD 453 [§83-5(a)]).

C. **STUDY DEFICIENCIES:** This study was conducted prior to promulgation of the EPA GLP guidelines. The purity of the test material was not indicated in the study report, although a lot no. was given. There were no analyses provided for homogeneity and concentration of the diet preparations. A number of parameters required for subchronic studies based on Subdivision F guidelines were not presented in this study. Measurements not present included: platelet counts and blood clotting measurements; serum calcium, chloride, phosphorus and creatinine; urine volume, specific gravity, appearance and bilirubin; histologic examination of esophagus, rectum and trachea. Individual animal findings for clinical observations were not provided. In addition, illegible data in this microfiche copy of the study (the only copy currently available within the Agency) made interpretation of this study especially difficult.

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DATA FOR ENTRY INTO ISIS

ubchronic (90-day) Oral Study - rodents (870.3100)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
109701	00025914	subchronic	rat	90 days	oral	dietary	17.0-357.4	17.0, 49.9, 179.5, 357.4 ♂ 18.5, 56.2; 176.5, 356.7 ♀	176.5	356.7	neuromuscular system, body wt. decr.	Toxicity

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DATA EVALUATION REPORT

PERMETHRIN/109701
(27Z75)
STUDY TYPE: RODENT DOMINANT LETHAL ASSAY IN MICE
[OPPTS 870.5450 (§84-2)]
MRID 00029829

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 02-05

Primary Reviewer:
B.L. Whitfield, Ph.D.

Signature: B L Whitfield
Date: MAR 2 6 2002

Secondary Reviewers:
Cheryl B. Bast, Ph.D., D.A.B.T

Signature: Cheryl B Bast
Date: MAR 2 6 2002

Robert H. Ross, M.S., Group Leader

Signature: Robert H. Ross
Date: MAR 2 6 2002

Quality Assurance:
LeeAnn Wilson, M.A.

Signature: L. A. Wilson
Date: MAR 2 6 2002

Disclaimer

This review may have been altered subsequent to the contractor's signature above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle, LLC, for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

EPA Reviewer: Yung G. Yang, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 8/13/2002
Signature: Joycelyn Stewart
Date 8/13/2002

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Rodent dominant lethal assay in mice; OPPTS 870.5450 [§84-2]

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 27Z75 (Permethrin, purity not provided)

SYNONYMS: 3-phenoxybenzyl dl, cis, trans 2,2-dimethyl-3-(2,2-dichlorovinyl) cyclopropane-1-carboxylate

CITATION: Chesher, B.C., J.C. Malone, M.J. Parker (1975) 27Z75, dominant lethal study in male mice. Research and Development (V & A), Wellcome Research Laboratories, Berkhamsted. Dec. Code: HEFG 76-2, Lab Ref. No.: T.L. 5-76. February 17, 1976. MRID 00029829. Unpublished

SPONSOR: Burroughs Wellcome Co.

EXECUTIVE SUMMARY: In a CD-1 mouse dominant lethal assay (MRID 00029829), 10 male mice were treated once daily via the oral route with 27Z75 at a dose of 285 mg/kg/day for five consecutive days. The test agent was mixed at 40% w/v in corn oil. Starting immediately after the final dosing, each male was mated with 3 untreated virgin females per week for six weeks. The females were sacrificed 14 days after the midweek of their mating period and the pregnancy rate and the number of living and dead implants determined.

27Z75 was tested at a total final dose (1425 mg/kg) equal to the acute LD_{50} value administered over a five-day period. The mice showed signs of slight hypersensitivity (which is consistent with other pyrethroids) lasting for 2 - 3 hours following the third, fourth and fifth doses but no other clinical signs were noted. All treated mice survived until the end of the study. The trimethylphosphate positive control reduced the pregnancy rate and increased the percentage of dead implants compared to the corn oil solvent control in weeks one and two. The differences were statistically significant. The solvent control values were appropriate. **There was no statistically significant difference between the control group and the 27Z75 treated group with respect to fertilization rate or the number of living and dead implantations in any week.**

This study is classified as Unacceptable/Guideline. It does not satisfy the requirement for FIFRA Test Guideline [OPPTS 870.5450 (§84-2)] for rodent dominant lethal data because no signed and

dated GLP or Quality Assurance Statements were provided and the purity of the test material was not given.

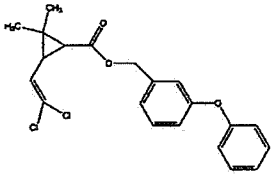
COMPLIANCE: No signed and dated GLP, Data Confidentiality or Quality Assurance Statements were provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test material: 27Z75

Description:	Not provided
Lot/Batch #:	C 6699-65
Purity:	Not provided
CAS # of TGAI:	52645-53-1
Solvent Used:	Corn oil



2. Control materials:

Negative:	None
Solvent / final volume / route of administration:	Corn oil / 0.2 mL / oral gavage
Positive / final dose(s) / route of administration:	Trimethylphosphate / 679 mg/kg (40% w/v in corn oil) / oral gavage

3. Test compound administration:

Volume:	0.2 mL/mouse
Route of administration:	Oral gavage
Dose level used:	285 mg/kg/day for 5 consecutive days

4. Test animals:

Species:	Mouse
Strain:	CD-1
Age/weight at study initiation:	Males: not provided Females: not provided
Source:	Charles River, UK
No. animals used per dose:	10 males mated to 3 untreated females/week for 6 weeks
Properly Maintained?	Information not provided

B. TEST PERFORMANCE:

1. Treatment:

a. Test compound and solvent control
Dosing: __ once __ twice (24 hr apart)
__x__ other: once daily for 5 consecutive days

2. **Mating:** Immediately after the last treatment each male was caged with three untreated females. The females remained with the male for one week at which time they were removed and replaced with three new females. This process was repeated for six weeks. A six-week mating period in mice allows sampling of sperm treated at all germ cell stages.

3. **Caesarian procedures:** Not described

4. **Evaluation criteria:** Pregnancy rates and the numbers of living and dead implants were determined for each animal and each treatment group. Differences between groups were statistically evaluated for significance.

5. **Statistical methods:** Statistical tests were two-tailed and performed at the 5% level. Pregnancy rates in the solvent control group and the positive control and the test material treated groups were compared using Fisher's exact test. Preimplantation losses were determined by using implant data from all pregnant females to calculate the average number of implants per male with one set of data for each test group. The two-sample permutation test was used to compare the solvent control group with the positive control group and with the test material group. A Chi-square test was used for an intragroup comparison of the proportion of dead implants attributable to each male. If statistically significant differences in the proportion of dead implants were seen between males in the same group, then the F-test was used to evaluate intergroup variation. If no statistically significant differences were seen in the intragroup comparison, then a Chi-square test was used for the intergroup analysis.

II. REPORTED RESULTS:

A. Preliminary toxicity assay: No preliminary toxicity assay was reported.

B. Dominant lethal assay: Slight hypersensitivity, lasting 2 - 3 hours, was seen in treated males following the third, fourth and fifth doses of 27Z75. No mortality or other clinical signs were noted. Statistically significant decreases in the pregnancy rate were seen in the positive control group in week 1 and 2 but not at later mating intervals. No statistically significant decreases in pregnancy rate were seen at any time in the 27Z75 group.

A summary of the number of living and dead implants is given in Table 1.

TABLE 1. Summary of the number of living and dead implants (group totals)

Week	Solvent Control		Positive Control		27Z75	
	Living	Dead	Living	Dead	Living	Dead
1	208	10	0	2	212	10
2	305	10	81	55	311	23
3	300	13	216	17	296	19
4	300	12	255	9	325	21
5	243	29	212	11	230	17
6	312	24	263	5	322	21

A statistically significant decrease in the mean number of implants, compared to the solvent control group, was seen in the positive control group in weeks 1, 2 and 5. A summary of the mean number of implants for the solvent and 27Z75 treated groups is given in Table 2. No statistically significant decrease in the mean number of implants was seen in the test material treated group at any mating period.

TABLE 2. Mean implant comparison between solvent control and 27Z75 groups

Week	Mean Number of Implants		Difference Between Means	Probability
	Solvent Control	27Z75		
1	11.35	10.32	1.04	0.377
2	12.73	11.95	0.78	0.275
3	12.12	11.7	0.42	0.393
4	11.03	13.35	-2.32	0
5	12.41	11.87	0.53	0.374
6	11.6	12.62	-1.02	0.077

Statistically significant increases in the percent of dead implants compared to the solvent control values were seen in the positive control group at weeks 1 and 2. A comparison between the solvent control and 27Z75 groups is shown in Table 3. No statistically significant increase in the percent dead implants was seen at any mating interval in the 27Z75 treated group.

TABLE 3. Percentage of dead implant comparison between solvent control and 27Z75 groups

Week	Percent Dead Implants		Between Groups Chi-square or F-ratio	Probability Value
	Solvent Control	27Z75		
1	4.59	4.5	F(1,18) = 0.0010	0.9747
2	3.17	6.89	F(1,25) = 2.6434	0.1165
3	4.15	6.03	F(1,26) = 0.2170	0.6452
4	3.85	6.07	F(1,26) = 0.8500	0.365
5	10.66	6.88	F(1,24) = 0.8650	0.3616
6	7.14	6.12	F(1,27) = 0.0667	0.7981

III. REVIEWER'S DISCUSSION/CONCLUSIONS:

A. DISCUSSION: 27Z75 did not induce dominant lethal mutations in the germ cells of male mice as tested in this study. The test material was administered at a dose said to be one-fifth the acute LD_{50} per day for five consecutive days; however, the total dose (1425 mg/kg) is lower than the limit dose (5000 mg/kg) for compounds without clear testing of the MTD. The study is classified as Unacceptable/Guideline and does not satisfy guideline requirements for a dominant lethal assay.

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B. **STUDY DEFICIENCIES:** No signed and dated GLP, Data Confidentiality or Quality Assurance Statements were provided and the test material purity and physical description were not given. The mice were said to be sexually mature but the actual age and weights were not provided. The deficiencies noted were data presentation deficiencies rather than experimental deficiencies. The study does provide useful information although not all acceptance criteria were met.

DATA EVALUATION RECORD

PERMETHRIN

STUDY TYPE: SUBCHRONIC INHALATION TOXICITY - RAT (OPPTS 870.3465/OECD 413) MRID 00096713

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37830
Task Order No. 02-04

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	Date:	NOV 28 2001
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	Date:	NOV 28 2001
Robert H. Ross, M.S., Group Leader	Signature:	Robert H. Ross
	Date:	NOV 28 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature:	L. A. Wilson
	Date:	NOV 28 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

EPA Reviewer: Yung G. Yang, Ph.D.
Reregistration Branch 2, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 6/6/2002
Signature: Joycelyn Stewart
Date 6/13/2002

TXR#: 0050649

DATA EVALUATION RECORD

STUDY TYPE: Subchronic Inhalation Toxicity - rat; OPPTS 870.3465 [§82-4]; OECD 413.

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 21Z73 (Permethrin, 94.7% a.i.)

SYNONYMS: None provided

CITATION: Alexander, D.J., Clark, G.C., Jackson, .C. et al. (1980) Permethrin technical inhalation study in rats 15 x 6 hour exposures over a 3 week period. Huntingdon Research Center, Huntingdon, Cambridgeshire, England. WLC 34/80323, November 11, 1980. MRID 00096713. Unpublished.

SPONSOR: Wellcome Co., Berkhamsted, England

EXECUTIVE SUMMARY: In a 15-day inhalation toxicity study (MRID00096713) Permethrin (94.7% a.i., Lot # ZJ) was administered to groups of 5 male and 5 female Charles River rats/concentration by dynamic whole-body inhalation exposure at concentrations of 0, 6.1, 42.2, or 583 mg/m^3 (0.0061, 0.042, or 0.583 mg/L) for 15 exposures (6 hours/day for 2 days during week 1, 5 days during weeks 2 and 3, and 3 days during week 4).

There was no test material-related effect on mortality, body weight or weight gain, food consumption, hematology, organ weights, or gross pathology. Weight gain was actually greater in all treated groups than in the respective control groups. Clinical signs were observed in the treated groups. Two female rats in the 0.0061 mg/l group were observed to have slightly labored breathing 30 minutes into the first exposure but not subsequently. In the 0.042 mg/l (MCT) group, licking of the inside of the mouths became more extensive then in the low-treatment group and involved most of the rats. All 5 females were observed to have slightly labored breathing during the first exposure but not subsequently. Labored breathing was not observed in male rats in either the 0.0061 or 0.042 mg/L groups. All rats in the 0.042 mg/L group appeared more alert than in the control and low-dose groups and adopted a hunched posture with open eyes during the early part of some exposures. The 0.583 mg/L group (HCT) demonstrated less activity, greater response to auditory or touch stimuli, and more extensive licking behavior than the other groups. Body tremors were observed in this group beginning with 3 females during the last hour of the first exposure and in 3 males during the second exposure. In both instances, tremors continued post exposure. The tremors reached a peak incidence, 5 males and 4 females,

during the 5th exposure (3rd day of the second week) and declined thereafter, with only 1 male and 1 female showing tremors on exposure day 15 (2nd exposure of week 4). Slightly labored breathing was recorded in 1 male and 1 female in this group.

The hypersensitivity to noise or touch became evident in the 0.583 mg/L (HCT) group following the second exposure and involved 5 males and 5 females. This sign tapered off with continued exposures, but was still displayed by 3 females following the 7th exposure. Rales, poor grooming, and crusty brown staining around the nose were observed occasionally in the 0.583 mg/L group, with incidences higher in females than in males. Microscopic pathology on the lungs showed focal to diffuse pneumonitis and perivascular inflammation - although to some degree more severe in the treated groups, could not be clearly distinguished from the respiratory infection present in all animals.

On April 18, 2002, the HIARC determined that the dose/endpoint can be used for risk assessment purpose because the clinical signs of neurotoxicity were observed in the first day of exposure. **The LOAEL is 0.583 mg/L in male and female rats based on body tremors and hypersensitivity to noise. The NOAEL is 0.042 mg/L.** This 15-day inhalation toxicity study in the rat is classified **acceptable/non-guideline.** This study does not satisfy the guideline requirement for a subchronic inhalation study OPPTS 870.3465

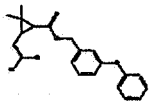
COMPLIANCE: Signed and dated GLP and Quality Assurance statements were provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test material:

Description:	Brown, viscous liquid
Lot/Batch #:	Lot ZJ
Purity:	94.7% a.i. (25.2% w/w *cis* isomer, 69.5% w/w *trans* isomer)
Compound Stability:	Not provided
CAS # of TGAI:	52645-53-1
Structure:	



2. Vehicle and/or positive control: None. Compressed air to generate aerosol.

3. Test animals:

Species:	Rat
Strain:	Charles River CD (Sprague-Dawley)
Age/weight at study initiation:	Age not provided Male group weight means of 122-124 g; female group mean weights of 122-125 g
Source:	Charles River, Kent, U.K.
Housing:	5/cage in suspended polypropylene cages with stainless steel, wire mesh tops and floors.
Diet:	Spratt's Laboratory Diet (weighed amount)

Water:	Tap water, available *ad libitum*	
Environmental conditions:	**Temperature:**	21.0-26.4°C
	Humidity:	41.3%
	Air changes:	Not provided; air flow of 11 L/minute
	Photoperiod:	12 hr light/12 hr dark
Acclimation period:	1 week	

B. STUDY DESIGN:

1. **In life dates**: Start: approximately January 17, 1980; End: approximately February 17, 1980

2. **Animal assignment**: Animals were assigned to the test groups in Table 1 by means of a computerized randomization procedure so that each group contained the same number of animals by sex and so that group weights were approximately equal.

TABLE 1: Study design

Test group	Nominal Concentration (mg/L)	Analytical Concentration (mg/L)[a]	MMAD µm	GSD	Rats/sex
Control	0 (No aerosol)	0	Not applicable	Not applicable	5
Low (LCT)	0.0067	0.0061±0.0048	1.7	(0.45-1.7µm)[b]	5
Mid (MCT)	0.0460	0.0422±0.0097	1.7	(0.45-1.7µm)	5
High (HCT)	0.627	0.583±0.0841	0.65	(0.45-1.7µm)	5

[a] Corrected for purity of 94.7%.
[b] Range based on collection on stages of impactor; includes ~98% of collected particles.
Data taken from pp. 4, 28-29, MRID 00096713.

3. **Dose selection rationale**: The report stated that target concentrations of 5, 50, and 500 mg/m^3 were based on the sponsor's request. Previously, 500 mg/m^3 was shown to be the highest concentration attainable with Permethrin technical. Details of the earlier study were not provided.

4. **Generation of the test atmosphere / chamber description**: Four identical 130 L Perspex chambers were used for the exposures. Each chamber contained 10 individual animal compartments. Permethrin was delivered via a syringe to a glass atomizer (A.T.M. Wingent Ltd., Cambridge) mounted below each cage. The test material was introduced to the center jet of the atomizer with an infusion pump. Dry, oil-free compressed air delivered to the outer ring of the atomizer at a rate of 25 L/minute was used to generate the aerosol. The control group was exposed to clean air; there was no infusion pump below the atomizer.

   Time to equilibrium was 24 minutes (theoretical).
   Analytical Chemistry consisted of gas-liquid chromatography.

   Test atmosphere concentration: The sampling port was at the level of the animals'

breathing zone. A sample of the chamber air was drawn through a glass microfiber filter at the rate of 10 L/minute. Volumes of air for the low, intermediate and high-dose groups were 200, 100, and 20 L, respectively (measured by a wet-type gas meter). Sampling occurred at least three times/exposure. Following gravimetric determination, the deposit was dissolved in chloroform and quantified by gas-liquid chromatography. Results are in Table 1 above.

Particle size determination: Particle size distribution was measured once/exposure. Samples were collected by passing the chamber atmosphere through a Cascade multi-stage impactor (Casella Cascade Impactor, Casella Ltd., London) at the rate of 17.5 L/minute. The deposits on the four stages of the impactor and on the filter were dissolved in chloroform and the amount of Permethrin was quantified by gas-liquid chromatography. Volumes of 35 (high-dose) to 350 L (low dose) were withdrawn at 3-4 hours into the exposures.

5. Statistics: Males and females were analyzed separately. For all parameters except organ weight, a one-way analysis of variance (ANOVA) was carried out, and the treated groups were compared with the respective control groups using the Student's "t" test. Organ weights were analyzed separately. Following adjustment for final body weight as a covariate or where the linear relationship between organ weight and body weight were significantly different from zero at the 10% level, an ANOVA was carried out. If variances were significant by Bartlett's test, a logarithmic transformation was performed. Group means were compared using the Willliams' test for contrasting increasing dose levels with the control. Significance was flagged at the 5% and 1% levels.

C. METHODS:

1. Observations:

1a. Cageside Observations: Animals were observed twice/day (morning and evening) for clinical signs and appearance.

1b. Clinical Examinations: Animals were observed at regular intervals during the exposures and observations were recorded every 30 minutes. The alertness of the animals was tested by knocking on the side of the cage. Animals were also examined for clinical signs during transfer from the exposure cages to the home cages.

1c. Neurological Evaluations: No neurological evaluations were performed. Neurologic evaluations other than those performed as part of the clinical observations are not required according to OPPTS 870.3465 guidelines.

2. Body weight: Animals were weighed individually on the day following arrival and weekly thereafter including the day of sacrifice.

3. Food consumption: Cagewise (5 animals/cage) food consumption was assessed weekly beginning with the day after arrival and ending with the day of sacrifice. Food efficiency was not calculated.

4. **Ophthalmoscopic examination:** Eyes were not examined as part of this study.

5. **Hematology & Clinical Chemistry:** Blood was collected from the orbital sinus of all rats prior to the 14th exposure for hematology and clinical analysis. Rats were lightly anaesthetized with ether prior to sampling. The animals were food-fasted but allowed access to water for 1 hour prior to sampling. The CHECKED (X) parameters were examined.

a. Hematology

X	Hematocrit (HCT)*	X	Leukocyte differential count*
X	Hemoglobin (HGB)*	X	Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*	X	Mean corpuscular HGB concentration (MCHC)*
X	Erythrocyte count (RBC)*	X	Mean corpuscular volume (MCV)*
X	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
X	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Recommended for subchronic inhalation studies based on Guideline 870.3465

b. Clinical chemistry

	Electrolytes		Other
X	Calcium	X	Albumin*
X	Chloride	X	Creatinine*
	Magnesium	X	Urea nitrogen*
X	Phosphorus	X	Total Cholesterol*
X	Potassium*		Globulins
X	Sodium*	X	Glucose*
	Enzymes (More than 2 Hepatic Enzymes Eg., *)		Total bilirubin
X	Alkaline phosphatase*	X	Total serum protein (TP)*
	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase		Serum protein electrophores
	Lactic acid dehydrogenase (LDH)		
X	Alanine amino-transferase (ALT/also SGPT)*		
	Aspartate amino-transferase (AST/also SGOT)*		
	Sorbitol dehydrogenase*		
	Gamma glutamyl transferase (GGT)*		
	Glutamate dehydrogenase		

* Recommended for subchronic inhalation studies based on Guideline 870.3465

6. **Urinalysis***Urine was collected from overnight food- and water-fasted animals between the 13th and 14th exposures during week 4. The CHECKED (X) parameters were examined.

	Appearance*	X	Glucose*
X	Volume*	X	Ketones
X	Specific gravity / osmolality*		Bilirubin
X	pH*	X	Blood / blood cells*

X	Sediment (microscopic)	X	Nitrate
X	Protein*	X	Urobilinogen

* Optional for inhalation toxicity studies

7. Sacrifice and pathology

All animals that died and those sacrificed on schedule were subjected to gross pathological examination and the CHECKED (X) tissues were collected for histological examination. All tissues from the control and high dose animals were examined. The larynx, lungs, nasal passages, and trachea from all groups were examined. The (XX) organs, in addition, were weighed.

	Digestive System		Cardiovasc./Hemat.		Neurologic
	Tongue		Aorta, thoracic*	XX	Brain*+
	Salivary glands*	XX	Heart*+	X	Peripheral nerve*
X	Esophagus*		Bone marrow*		Spinal cord (3 levels)*
X	Stomach*		Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*+	X	Eyes (optic nerve)*
	Jejunum*	XX	Thymus*+		**Glandular**
X	Ileum*			XX	Adrenal gland*+
X	Cecum*		**Urogenital**		Lacrimal gland
X	Colon*	XX	Kidneys*+	X	Parathyroid*
	Rectum*	X	Urinary bladder*	XX	Thyroid*
XX	Liver*+	XX	Testes*+		**Other**
	Gall bladder* (not rat)		Epididymides*+		Bone (sternum and/or femur)
	Bile duct* (rat)	XX	Prostate*		Skeletal muscle
X	Pancreas*		Seminal vesicles*	X	Skin
	Respiratory	XX	Ovaries*+		All gross lesions and masses*
X	Trachea*	XX	Uterus*+		
XX	Lung*		Mammary gland*		
X	Nose* (Nasal passages)				
	Pharynx*				
X	Larynx*				

* Recommended for subchronic rodent studies based on Guideline 870.3465
+ Organ weights required

II. RESULTS

A. OBSERVATIONS:

1. Clinical signs of toxicity: The behavior of the control group which consisted of activity for 30 minutes followed by a sleeping posture was considered normal by the study authors. The 0.0061 mg/L (LCT) treatment group behaved similarly but were observed to lick the insides of their mouths, a behavior frequently observed during inhalation exposures. Two female rats in this group were observed to have slightly labored breathing 30 minutes into the first exposure but not subsequently. In the 0.042 mg/L (MCT) group, licking of the inside of the

mouths became more extensive then in the low-treatment group and involved most of the rats. All 5 females were observed to have slightly labored breathing during the first exposure but not subsequently. Labored breathing was not observed in male rats in either the 0.0061 or 0.042 mg/L groups. All rats in the 0.042 mg/L group appeared more alert than in the control and low-dose groups and adopted a hunched posture with open eyes during the early part of some exposures. Following the 14th exposure, the fur of these animals was observed to be slightly oily, presumably due to deposition of the test material.

The 0.583 mg/L group (HCT) demonstrated less activity, greater response to auditory or touch stimuli, and more extensive licking behavior than the other groups. Body tremors were observed in this group beginning with 3 females during the last hour of the first exposure and in 3 males during the second exposure. In both instances, tremors continued postexposure. The tremors reached a peak incidence, 5 males and 4 females, during the 5th exposure (3rd day of the second week) and declined thereafter, with only 1 male and 1 female showing tremors on exposure day 15 (2nd exposure of week 4). Slightly labored breathing was recorded in 1 male and 1 female in this group.

The hypersensitivity to noise or touch became evident in the 0.583 mg/L (HCT) group following the second exposure and involved 5 males and 5 females. This sign tapered off with continued exposures, but was still displayed by 3 females following the 7th exposure. Rales, poor grooming, and crusty brown staining around the nose were observed occasionally in the 0.583 mg/L group, with incidences higher in females than in males.

2. **Mortality**: There were no deaths during the study.

3. **Neurological evaluations**: Other than the observations of tremors and increased reactivity to noise and touch stimuli, no other neurological evaluations were reported.

B. **BODY WEIGHT AND WEIGHT GAIN**: There were no effects of treatment on body weight or body weight gain (Table 2). Treated rats of both sexes gained more weight than their respective control groups.

TABLE 2. Average body weights and body weight gains during 4 weeks of treatment.

Analytical Concentration (mg/L)	Body Weights (g±SD)					Total Weight Gain	
	Week -1	Week 1	Week 2	Week 3	Week 4	g	% of control
Male							
0	123	176	227	269	288	165	
LCT (0.0061)	125	176	263	288	310	185	112
MCT (0.0422)	122	191	233	277	295	173	105
HCT (0.5830)	124	176	231	280	298	174	106

Female							
0	122	154	174	230	209	47	
LCT (0.0061)	123	159	145	206	211	88	187
MCT (0.0422)	124	160	179	198	208	84	179
HCT (0.5830)	125	159	174	193	203	78	166

[a] Data obtained from page 30 in the study report (standard deviations were not reported).

C. **FOOD CONSUMPTION:**

1. **Food consumption**: Food consumption did not differ among the groups for either sex. For males, total food consumption values in the control, low, mid-, and high-concentration groups were 603, 663, 603, and 620 g, respectively. Respective values for females were 486, 476, 439, and 475 g.

2. **Food efficiency**: Food efficiency was not calculated.

D. **OPHTHALMOSCOPIC EXAMINATION** - Ophthalmoscopic examinations were not performed.

E. **BLOOD ANALYSES:**

1. **Hematology**: There were no treatment related effects on hematology parameters. Statistically significant changes were either not dose related or, in the case of thromboplastin time, not biologically significant. The thromboplastin time of males in the high-concentration group (0.583 mg/L) was slightly increased over the control value, 30.0 seconds vs 24.2 seconds in the male control group, but was slightly decreased in the mid- and high-concentration groups for females, 18.8 and 18.4 seconds vs 21.6 seconds in the female control group.

2. **Clinical chemistry**: Plasma glucose was significantly reduced in males in the high-dose group ($p<0.001$) and in females in the mid- ($p<0.05$) and high-dose groups ($p<0.01$). In males, cholesterol was increased in a concentration-related manner, but none of the values attained significance compared with the control value. The cholesterol effect was not observed in female rats.

F. **URINALYSIS**: Protein was present in the urine of all groups of male rats with a significant increase over the control value in the 0.583 mg/L group ($p<0.01$). However, there was no clear concentration-response relationship. In the absence of a concentration-related trend, this effect was not considered treatment related. A significant increase in the specific gravity of urine of females in the 0.583 mg/L group ($p<0.05$) was also not clearly dose related.

G. **SACRIFICE AND PATHOLOGY:** 469

1. **Organ weight**: Liver weights of males and females in the 0.583 mg/L dose group were significantly increased over respective control weights by 12 and 11%, respectively (males, $p<0.01$; females, $p<0.05$). These increases are a normal adaptation to chemical treatment and, furthermore, are not biologically significant. The mean pituitary weight in males in the 0.583 mg/L group was significantly reduced compared with the control group (68%, $p<0.01$) and the reduction in all groups showed a concentration-related trend. However, for females in the 0.583 mg/L group, the value was higher than the control group value (by 19%). The significance of the reduced pituitary weights in male rats in the high-concentration group is unknown. All other organ weight changes were not dose-related.

2. **Gross pathology**: No treatment-related differences in incidences of gross pathology observations were observed.

3. **Microscopic pathology**: Inflammatory changes in the lungs consisting of interstitial pneumonitis, perivascular and peribronchial lymphoid "cuffing," and occasional macrophage aggregation, were observed in rats of all groups. The study authors stated that the frequency and severity of these effects were more pronounced in all treated groups than in the control groups. The reviewer noted that severity/frequency of some of these effects were more often noted or described as occasional and small whereas the descriptors minimal and moderate were more often applied to both sexes in the 0.583 mg/L dose group. However, in the absence of numeric values for the several descriptors and without a clear tally of incidences, the reviewer generally agrees with but could not make a clear judgement as to the accuracy of the study authors' comments. Rhinitis was observed in all treated groups of male rats (incidences in the control through high-concentration groups of 0, 3, 2, and 4, respectively). However, in female rats incidences (2/5 to 3/5) were similar among control and treated groups). Summary tables were not provided for microscopic observations.

III. DISCUSSION and CONCLUSIONS

A. **INVESTIGATORS' CONCLUSIONS:** The investigators concluded that inhalation of permethrin technical "for 6 hours a day for 15 exposures caused only minimal physical, biochemical and histological changes." Tremors, persisting up to 24 hours after exposure, and hyperreactivity to noise and touch were observed only in the 0.583 mg/L group, with female rats more severely affected than male rats. There was no affect of treatment on body weight, food consumption, hematology parameters, urinalysis, or macroscopic pathology. Glucose was significantly reduced in males in the high-concentration group and in females in the mid- and high-concentration groups. The increase in cholesterol in the high-concentration group of male rats did not attain significance, but was considered treatment related. Microscopically, inflammatory changes in the lungs were more frequent and more severe in all treated groups compared with the control groups. Likewise, rhinitis was more frequently observed in the nasal turbinates of treated rats compared with the control groups. However, differences in inflammatory changes and rhinitis were not apparent among the treatment groups.

B. **REVIEWER COMMENTS:** The reviewer generally agrees with the conclusions of the

study authors. There was no test material-related effect on mortality, body weight or weight gain, food consumption, hematology, organ weights, or gross pathology. Weight gain was actually greater in all treated groups than in the respective control groups. Clinical signs were observed in the treated groups. Two female rats in the 0.0061 mg/l group were observed to have slightly labored breathing 30 minutes into the first exposure but not subsequently. In the 0.042 mg/l (MCT) group, licking of the inside of the mouths became more extensive then in the low-treatment group and involved most of the rats. All 5 females were observed to have slightly labored breathing during the first exposure but not subsequently. Labored breathing was not observed in male rats in either the 0.0061 or 0.042 mg/L groups. All rats in the 0.042 mg/L group appeared more alert than in the control and low-dose groups and adopted a hunched posture with open eyes during the early part of some exposures. The 0.583 mg/L group (HCT) demonstrated less activity, greater response to auditory or touch stimuli, and more extensive licking behavior than the other groups. Body tremors were observed in this group beginning with 3 females during the last hour of the first exposure and in 3 males during the second exposure. In both instances, tremors continued post exposure. The tremors reached a peak incidence, 5 males and 4 females, during the 5th exposure (3rd day of the second week) and declined thereafter, with only 1 male and 1 female showing tremors on exposure day 15 (2nd exposure of week 4). Slightly labored breathing was recorded in 1 male and 1 female in this group.

The hypersensitivity to noise or touch became evident in the 0.583 mg/L (HCT) group following the second exposure and involved 5 males and 5 females. This sign tapered off with continued exposures, but was still displayed by 3 females following the 7th exposure. Rales, poor grooming, and crusty brown staining around the nose were observed occasionally in the 0.583 mg/L group, with incidences higher in females than in males. Respiratory infections were present in animals of all groups and the observation of slightly labored breathing in females lacked a clear dose-response relationship (incidences of 0/5, 2/5, 5/5, and 1/5 in the control through high-dose group). It is possible that slightly labored breathing was overshadowed by the more frank effects of body tremors and hypersensitivity to noise in females in the high-dose group. Microscopic pathology on the lungs showed focal to diffuse pneumonitis and perivascular inflammation - although to some degree more severe in the treated groups, could not be clearly distinguished from the respiratory infection present in all animals.

On April 18, 2002, the HIARC determined that the dose/endpoint can be used for risk assessment purpose because the clinical signs of neurotoxicity were observed in the first day of exposure. **The LOAEL is 0.583 mg/L in male and female rats based on body tremors and hypersensitivity to noise. The NOAEL is 0.042 mg/L.** This 15-day inhalation toxicity study in the rat is classified **acceptable/non-guideline.** This study does not satisfy the guideline requirement for a subchronic inhalation study OPPTS 870.3465

C. **STUDY DEFICIENCIES:** This study was initiated before 1996 OPPTS 870.3465 subchronic guidelines were in place and does not fulfill many of the subchronic guidelines. This 4-week, 15-day study is suitable as a range finding study to set exposure concentrations for a subchronic study.

Major deficiencies include:

1. Inadequate exposure period (15 days over 4 weeks rather than 90 days).
2. Animals were not healthy.
3. Inadequate number of animals (5 animals/sex rather than 20 animals/sex).

Minor deficiencies include:

4. Ophthalmological examinations were not conducted.
5. Some organs and tissues were not examined microscopically. In light of no significant changes in hematology and clinical chemistry parameters (except reduced glucose in both sexes) this deficiency is not major.
6. Some of the data tables were unreadable.

DATA FOR ENTRY INTO ISIS

ıbchronic Inhalation Study - rodents (870.3465)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/L	Doses tested mg/L	NOAEL mg/L	LOAEL mg/L	Target organ(s)	Comments
09701	00096713	subchronic	rat	15 days over 4 weeks	inhalation	inhalation	0.0061-0.583	0, 0061, 0.0422, 0.583	0.042 (♂&♀)	0.583 (♂&♀)	lungs, cns	

4189

DATA EVALUATION RECORD

PERMETHRIN/109701
STUDY TYPE: METABOLISM AND PHARMACOKINETICS - RAT
[OPPTS: 870.7485 (§85-1)]
MRID 00102185

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 02-05

Primary Reviewer: Robert A. Young, Ph.D., D.A.B.T.	Signature: Date:	Robert A. Young MAR 2 6 2002
Secondary Reviewers: H.T.Borges, Ph.D., MT (ASCP), D.A.B.T.	Signature: Date:	H.T. Borges MAR 2 6 2002
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross MAR 2 6 2002
Quality Assurance: Lee Ann Wilson, M.S.	Signature: Date:	L. A. Wilson MAR 2 6 2002

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

EPA Reviewer: Yang Yung, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/23/2002.
Signature: Joycelyn Stewart
Date 6/3/2002.

DATA EVALUATION RECORD

TXR#: 0050649

STUDY TYPE: Metabolism - rat [OPPTS 870.7485 (§85-1)] OECD 417.

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin, purity not reported; cis:trans 40:60

SYNONYMS: PP557; [3-phenoxybenzyl (±) cis:trans -2,2-dimethyl-3'(2,2-dichlorovinyl)-cyclopropane-1-carboxylate]

CITATION: Bewick, D.W., Leahey, J.P. 1978. The analysis of the permethrin metabolite 3-(2,2-dichlorovinyl)-1-methylcyclopropane-1,2-dicarboxylic acid in the excreta of rats given a single oral dose of ^{14}C-permethrin. RJ0019B. ICI America, Inc., Plant Protection Division. 27 April 1978, unpublished. MRID 00102185.

SPONSOR: ICI Americas, Inc.

EXECUTIVE SUMMARY: In a metabolism study (MRID 00102185), male Wistar-derived rats were given a single low dose (2.0 mg/rat) or single high dose (20 mg/rat) of permethrin ([^{14}C-cyclopropane]permethrin, 40:60 cis-trans ratio and non-labeled permethrin, 38.2:59.3 cis-trans ratio; no purity or lot/batch nos. for either) intragastrically. Feces and urine collected one day prior to dosing and for three days postdose were analyzed for radioactivity and metabolites.

These experiments provided an initial and cursory effort at identification and quantitation of major metabolites in the urine and feces of rats following single oral doses (2 or 20 mg/rat) of [^{14}C-cyclopropane]permethrin. Approximately 78.5% of the administered radioactivity was recovered over the 3-day experimental period (dose group not specified). A conjugated metabolite, 3-(2,2-dichlorovinyl)-1-methylcyclopropane-1,2-dicarboxylic acid, was identified in both the urine and feces that reportedly accounted for approximately 2.2% of the administered dose. No additional data were provided regarding characterization of the remaining recovered radioactivity.

This metabolism study in the rat is classified **Unacceptable/Guideline** and does not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD 417] in rats. The unacceptability is the result of deficiencies in level of detail provided which prevent verification/ validation of findings (e.g., insufficient data regarding characterization of recovered radioactivity, no dose confirmation, no lot/batch numbers for the test article).

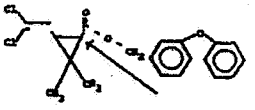
COMPLIANCE: The studies were conducted prior to implementation of GLP guidelines and, therefore, there was no claim regarding GLP compliance. Certification of Access to Raw Data and Data Confidentiality statements were signed subsequent to completion of the studies provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test compound:

Radiolabelled test material:	[^{14}C-cyclopropane]permethrin *cis:trans* (40:60)
Radiochemical purity	Not provided
Specific Activity	50 mCi/mM
Lot/Batch #:	Not provided
Non-Radiolabelled Test Material:	Permethrin *cis:trans* (38.2:59.3)
Description:	Not provided
Lot/Batch #:	Not provided
Purity:	Not provided
Contaminants:	None reported
CAS # of TGAI:	52645-53-1
Structure:	



Position of radiolabelling in 14$_C$-cyclopropane labeled permethrin

2. Vehicle and/or positive control: The test article was administered in commercially available corn oil.

3. Test animals:

Species:	Rat, male	
Strain:	Wistar-derived SPF	
Age/weight at study initiation:	Adults (195-205 g)	
Source:	Alderley Park ICI	
Housing:	Housed individually in metabolism cages	
Diet:	Stock rat diet *ad libitum* (Oakes, LTD., Congleton, Cheshire)	
Water:	Ad libitum	
Environmental conditions:	**Temperature:**	22±2°C
	Humidity:	50±5% r.h.
	Air changes:	Not specified
	Photoperiod:	12 hrs/12 hrs
Acclimation period:	3 days	

4. **Preparation of dosing solutions:** The low dose (2.0 mg; 25 µCi/rat) was prepared by mixing 80 µCi [^{14}C-cyclopropane]permethrin with 5.77 ml non-labeled permethrin (1 mg/ml). The mixture was brought to 100 ml with a non-specific solvent. A 6.5 ml aliquot was removed for specific activity analysis and the solvent evaporated from the remaining stock solution. The resulting residue was dissolved in 3.0 ml of corn oil. The high dose (20 mg; 25 µCi/rat) was prepared similarly but used 80 µCi [^{14}C-cyclopropane]permethrin in 63.4 ml non-labeled permethrin (1 mg/ml). Solubility of permethrin in corn oil was reportedly verified by assessing radioactivity in 10 µl samples of the corn oil dosing solution.

B. STUDY DESIGN AND METHODS:

1. **Group arrangements**: The test groups for MRID 000102185 shown in Table 1. No information was provided regarding selection procedures for the establishing the groups.

TABLE 1: Dosing groups for metabolism identification studies for permethrin.

Test Group	Dose	Number/sex	Remarks
Low dose	1 ml(2.0 mg)	2♂	Metabolite analysis; urine and feces collected prior to and for 3 days following single intragastric dose
High dose	1 ml(20 mg)	2♂	Metabolite analysis; urine and feces collected prior to and for 3 days following single intragastric dose

Data taken from p. 1, MRID 00102185.

2. **Dosing and sample collection:** The test article was administered intragastrically. Total dosing volume for both dose groups was 1.0 ml. In addition to radioactivity analysis by liquid scintillation counting (LSC; quench-corrected and efficiency determined by use of an internal standard), preparative layer chromatography (PLC), thin-layer chromatography (TLC), and gas-liquid chromatography were used for metabolite analysis.

Expired air: Expired air was not collected.

Urine: Urine was collected daily from Day-1 through Day 3. Samples were maintained at -20°C until analysis. Filtered samples were mixed with scintillation fluid and radioactivity analyzed in triplicate in most cases by LSC. Filtered sediment was combusted and counted separately. Aliquots of urine extracts (5.0 ml) were also prepared for GC-MS. The procedure included, evaporation of the acetone and extraction with diethyl ether (3 x 20 ml). The acid-soluble fraction and ether-soluble fractions were further extracted and analyzed by GC-MS.

Feces: Feces were collected daily from Day-1 through Day 3. Samples were maintained at -20°C until analysis. Fecal samples were homogenized and extracted three times with acetone, centrifuged, and analyzed for radioactivity. Following combustion in a sample oxidizer, LSC was performed as previously described. Fecal extracts (50 ml) were also prepared for GC-MS similar to the procedures for urine samples.

Blood - Blood samples were not collected. 487

Tissues - No tissues were collected.

a. **Pharmacokinetic studies:** Absorption/elimination kinetics were not protocol elements.

b. **Metabolite characterization studies:** Metabolite analysis was performed by mass spectrometry in multiple ion detection mode. An analytical flow chart for preparation of urine and fecal samples was provided in the study report.

3. Statistics: Description of statistical analyses were limited to those applied to LSC analyses.

II. RESULTS

A. **PHARMACOKINETIC STUDIES:** Recovery of radioactivity was limited to that found in the collected urine and fecal samples. Based upon total administered radioactivity (24.1 µCi/rat as determined by dpm/unit of dose solution), recovery of administered radioactivity over the 3-day experimental period was approximately 78% (Table 2). (The study report is unclear what does the single rat received.)

TABLE 2. Recovery of administered radioactivity (expressed in µCi [% of dose]) in urine and feces following a single oral dose of [^{14}C]-permethrin to a rat[a].

Sample	Day 1	Day 2	Day 3
Fecal extract	5.07 [21%]	1.51 [6%]	0.40 [2%]
Solid residue- feces post extraction	2.36 [10%]	0.71 [3%]	0.11 [0.5%]
Urine filtrate	4.89 [20%]	2.62 [11%]	1.31 [5%]
Suspended solids in urine	<0.01	<0.01	<0.01
Total	12.32 [51%]	4.84 [20%]	1.82 [7.5%]

Data taken from Table 2, p. 11, MRID 000102185.
[a]The study report is unclear what dose the single rat received.

1. **Absorption:** Absorption, implied from urinary excretion data over three days, was at least 36%. In the absence of biliary excretion analysis, it is not possible to determine what portion of radioactivity excreted in the feces represented absorbed test article. Assessment of absorption, distribution and excretion were not protocol elements for MRID 00102185.

2. **Tissue distribution:** Tissue distribution was not a protocol element for MRID 00102185.

3. **Excretion:** Quantitative assessment of excretion was not a protocol element for MRID 00102185.

B. **METABOLITE CHARACTERIZATION STUDIES:** The study authors noted that only water soluble fractions converted to ether-soluble fractions by acid hydrolysis were analyzed due to contaminant interferences encountered with the organo-soluble fractions. Metabolite IIa, 3-(2,2-dichlorovinyl)-1-methylcyclopropane-1,2-dicarboxylic acid, was identified in both

matrices. This metabolite accounted for 0.82% of the total radioactivity in the urine and 1.4% of the total activity in the feces.

III. DISCUSSION and CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS**: In a metabolism study (MRID 00102185), male Wistar-derived rats were given a single low dose (2.0 mg/rat) or single high dose (20 mg/rat) of permethrin ([^{14}C-cyclopropane]permethrin, 40:60 cis-trans ratio and non-labeled permethrin, 38.2:59.3 cis-trans ratio; no purity or lot/batch nos. for either) intragastrically. Feces and urine collected one day prior to dosing and for three days postdose were analyzed for radioactivity and metabolites.

Recovery of radioactivity in the feces and urine over the 3-day experiment period accounted for 78.5% of the administered dose, most being eliminated during the first day. Following extraction procedures and GC-MS analysis, Metabolite IIa (3-(2,2-dichlorovinyl)-1-methylcyclopropane-1,2-dicarboxylic acid) was identified in extracts of both urine and feces. It was reported that this metabolite in the represented 0.82% of radioactivity in the excreted in the urine and 1.4% of the radioactivity excreted in the feces. Due to extraction/preparative related difficulties, only the conjugated form of the metabolite was detected.

B: **REVIEWER COMMENTS:** These experiments provided an initial and cursory effort at identification and quantitation of major metabolites in the urine and feces of rats following a single oral doses (2 or 20 mg/rat) of [^{14}C-cyclopropane]permethrin. Approximately 78.5% of the administered radioactivity was recovered over the 3-day experimental period. Although accounting of radioactivity inventory was not necessarily an experiment protocol item, this value appears to be based upon only one rat and it is unclear what dose the rat received. The study authors identified and characterized a conjugate metabolite (3-(2,2-dichlorovinyl)-1-methylcyclopropane-1,2-dicarboxylic acid) in both the urine and feces that reportedly accounted for approximately 2.2% of the administered dose. However, no data were provided that allowed for verification of this quantitative assessment. No other data were provided regarding characterization of the remaining recovered radioactivity.

This metabolism study in the rat is classified **Unacceptable/NonGuideline** and does not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD 417] in rats. The unacceptability is the result of deficiencies in level of detail provided which prevent verification/validation of findings (e.g., insufficient data regarding characterization of recovered radioactivity, no dose confirmation, no lot/batch numbers for the test article). Although the study provided data affirming a conjugated metabolite in the urine and feces of the rat, the study was very cursory.

C. **STUDY DEFICIENCIES:** The study report (MRID 00102185) lacked details regarding the test article (no purity data or lot/batch nos.) and no dose confirmation data were provided. The extraction/preparation procedures were such that only a conjugated metabolite could be characterized. Additionally, quantitative data were lacking that prohibited validation of the presented data and an overall assessment of the metabolism of the test article. This study was conducted prior to implementation of GLP guidelines.

swelling are adaptive and reversible effects and are not considered adverse effects (HED Doc# 0050731). **Therefore, the systemic toxicity LOAEL is 1000 mg/kg/day based on clinical neurotoxic signs and decreased body weight gain and food consumption. The NOAEL is 100 mg/kg/day.**

This one-year dog study is classified **Acceptable/Guideline** and satisfies the guideline requirement for a chronic toxicity study in dogs.

H

DATA EVALUATION RECORD

PERMETHRIN/109701
STUDY TYPE: DERMAL PENETRATION STUDY - MOUSE
[OPPTS: 870.7600 (§85-3)]
MRID 00153970

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 02-05

Primary Reviewer: H.T.Borges, Ph.D., MT (ASCP), D.A.B.T.	Signature: Date:	H T Borges APR 10 2002
Secondary Reviewers: Robert A. Young, Ph.D., D.A.B.T.	Signature: Date:	Robert A. Young APR 10 2002
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross APR 10 2002
Quality Assurance: LeeAnn Wilson, M.S.	Signature: Date:	L. A. Wilson APR 10 2002

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

EPA Reviewer: Yung Yang, Ph.D. **Signature:** Yung G. Yang
Toxicology Branch, Health Effects Division (7509C) **Date** 5/16/2002
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. **Signature:** Joycelyn Stewart
Work Assignment Manager, Health Effects Division (7509C) **Date** 6/3/2002

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Rodent *In Vivo* Dermal Penetration Study - Mouse OPPTS 870.7600 [§85-1]

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin (purity not reported, radiopurity >98%)

SYNONYMS: None reported

CITATION: Shah, P.V., Monroe, R.J., Guthrie, F.E. (1981) Comparative rates of dermal penetration of insecticides in mice. Toxicology Program/Dept. Entomology, North Carolina State Univ., Raleigh, NC. Laboratory report number not provided. MRID 00153970. Toxicology and Applied Pharmacology, 59: 414-423.

SPONSOR: Not provided.

EXECUTIVE SUMMARY: In a dermal penetration study (MRID 00153970), permethrin (1 mg/kg. containing 1 μCi of activity, lot number not reported, specific activity ≥98%) was applied to a 1 cm^2 area on the shaved backs of three female Duplin ICR mice/group. The mice were killed 1, 5, and 15 minutes, and 1, 8, and 48 hours after application and the rate of dermal penetration and tissue distribution determined.

This is a journal article published in Toxicology and Applied Pharmacology. Permethrin is one of the 14 insecticides used to compare the rates of dermal penetration of pesticides in mice. The results indicated that dermal penetration by permethrin was rapid (T_{50} = 5.9 minutes), was extensively absorbed (~88% in 8 hours), once absorbed had a wide volume of distribution, and was rapidly metabolized primarily in the liver and excreted.

This study is classified as **Acceptable/Non-guideline** and does not satisfy the guideline requirement for a dermal penetration study (870.7600). It does provide supportive data on the rate of absorption, distribution, and excretion of permethrin in mice.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided. The study did not follow OPPTS 870.7600 guidelines; these included: **not reporting the lot number or purity of the test material, the use of mice when rats are required, the use of acetone as the delivery vehicle, verification of the doses applied, the use of a single dose when multiple doses are suggested, the use of three animals per treatment duration group.**

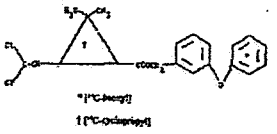
instead of four, not washing the application site following treatment, the limited dose application area, protection of the dose site following treatment, and the reporting requirements

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test material: Permethrin, cis isomer

Description:	Not reported
Lot/Batch #:	Not reported
Purity:	Not reported
Compound Stability:	Not reported
CAS # for TGAI:	52645-53-1
Vehicle/Solvent used:	Acetone
Radiolabeling:	^{14}C-label on cyclopropyl alcohol
Specific Activity:	55.9 mCi/mol
Radiochemical Purity:	>98%
Source:	FMC Corporation, Middleport, NY
Structure:	



2. Relevance of Test Material to Proposed Formulation(s):

Not applicable

3. Test animals:

Species:	Female mice
Strain:	Duplin ICR
Age/weight at study initiation:	7-8 weeks
Source:	Flow Laboratories, Duplin, VA
Housing:	individually in glass metabolism cages
Diet:	*ad libitum*
Water:	*ad libitum*
Environmental Conditions	
Temperature:	Not reported
Humidity:	Not reported
Air changes:	Not reported
Photoperiod:	12 hrs light/dark
Acclimation period:	48 hours

B. STUDY DESIGN

Most guideline requirements in OPPTS 870.7600 were not followed.

1. **Dose**

No dose selection rationale was provided.

Actual Dose: 1 mg/kg bdy wt. containing 1 μCi of activity

Dose volume: adjusted to contain 1 mg/kg bdy wt.

Duration of exposures: 1, 5, and 15 minutes; 1, 8, and 48 hours

Termination periods (time from dose to sacrifice): 1, 5, and 15 minutes; 1, 8, and 48 hours

Number of animals/group: 3 mice/time interval for dermal penetration. All mice were dosed as described below.

2. **Animal Preparation**

Seventy-two hours before dosing, a 3-4 cm^2 on the upper shoulder of the animal was shaved. Care was taken not to abrade the skin.

3. **Dose Preparation, Administration and Quantification:**

Preparation: Dose suspensions were prepared in acetone so that 0.1 mL contained 1 mg/kg bdy wt. test material and 1.0 μCi of radiolabel.

Application: The dosing solution was applied by syringe to an area of 1 cm^2 on the shaved upper shoulder. The acetone was allowed to dry before the mouse was placed into a glass metabolism chamber. The journal article does not mention whether the site was protected.

Quantification: Not reported.

4. **Skin Wash (Pre-Sacrifice)**

Not done.

5. **Sample Collection**

To determine the rate of permethrin absorption and distribution, three mice/group were killed 1, 5, and 15 minutes, and 1, 8, and 48 hours after test material application. Following death by ether asphyxiation, a 3-4 cm^2 area of skin at the application site was removed to determine the quantity of unabsorbed and laterally diffused radiolabeled permethrin. Urine was collected by gently rubbing the area of the bladder while holding the animal over a collection device. This urine was added to any collected in the metabolism chamber including rinsate. Blood was collected by cardiac puncture. In addition, the following tissues and organs were collected: heart, lungs, brain, kidney, bladder, fat, ear, spleen, bone marrow, muscle, stomach, intestine, liver, blood, feces, exhaled CO_2, and residual carcass.

6. Sample Preparation and Analysis

Storage of the samples was not described. In most cases, urine, whole organs or aliquots of homogenates were combusted in a Harvey Biological Oxidizer equipped with a CO_2 trapping device containing 15 mL scintillation fluid (Harvey Instruments Corp., Hillsdale, NJ). The residual carcass was homogenized in water and aliquots combusted for collection of CO_2 as previously described.

Sample radioactivity was determined using a Packard Tri-Carb scintillation counter. Quenching was corrected by internal standardization and oxidized samples corrected to total percent recovery of radioactivity for each time interval.

II. RESULTS:

A. SIGNS AND SYMPTOMS OF TOXICITY:

Clinical signs of toxicity were not reported.

B. TOTAL ABSORBED DOSE

The rate of permethrin penetration is shown in Table 1. The study authors reported that overall recovery of the radiolabel was ≥90% and no lateral diffusion occurred. The $T_{0.5}$, or time to 50% penetration, was 5.9 ± 1.3 minutes. By 8 hours, ~88% of the applied dose had been absorbed.

TABLE 1. Geometric Mean of Permethrin Penetration Rate[a,b]

Permethrin	Percent Penetration				
	1 min.	5 min.	15 min.	1 hr.	8 hr.
	36.2	40.9	63.1	79.7	88.1

[a]From Table 1, page 417 of MRID 00153970
[b]Percent of penetrated radiolabel not determined at 48 hours.

The distribution of the radiolabel 5, 15, and 60 minutes after treatment is shown in Table 2. The data shows that although ~40% of the permethrin was absorbed within 5 minutes of application, the limited amount found in the blood suggests a wide distribution volume.

TABLE 2. Percent of Permethrin Radiolabel Recovered in Various Fractions 5, 15, and 60 Minutes after Dermal Application [a]

	Blood			Liver			Fat			Excreta [b]			Carcass		
Minutes	5	15	60	5	15	60	5	15	60	5	15	60	5	15	60
Permethrin	<0.1	1.0	5.8	0.1	0.7	5.0	<0.1	0.1	0.6	<0.1	0.2	7.4	40.8	60.4	60.9

[a]From Table 2, page 418 of MRID 00153970
[b]Includes urine, CO_2 and feces

Table 3 shows the distribution of the radiolabel 8 hours after dermal treatment. The data shows that the greatest portion was found in the excreta. The highest organ concentration was found in the intestine followed by the liver, suggesting that metabolism by the liver was the primary elimination route. As shown in Table 2, the disparity between the blood and carcass data indicates permethrin has a wide volume of distribution.

TABLE 3. Average percent of radiolabel recovered in tissues and excreta 8 hours after treatment [a,b].

Tissue	Lungs	Kidney	Ear	Stomach	Intestine	Liver	Blood	Excreta[c]	Carcass
Permethrin	0.2	0.6	0.2	0.7	6.3	2.6	2.4	63.8	10.4

[a] Data from Table 3 page 420, MRID 00153970
[b] Percent of radiolabel found in heart, brain, bladder, fat, spleen, bone marrow, and muscle not shown as all were ≤0.1%
[c] Primarily found in feces.

III. DISCUSSION and CONCLUSIONS

A. INVESTIGATORS' CONCLUSIONS:

Other than stating that permethrin dermal penetration is rapid, no specific conclusions were made by the study authors. This study was provided as a journal article done on 14 different pesticides to compare rates of dermal penetration resulting from class and structural activity relationships.

B. REVIEWER COMMENTS:

As noted above, this MRID was provided as a journal article from Toxicology and Applied Pharmacology that compared the rates of dermal penetration by 14 of pesticides (carbamates, organophosphates, botanical types, and chlorinated hydrocarbons). It is for this reason that the standard dermal template was not used. As reported in the article, dermal penetration by permethrin was rapid (T_{50} = 5.9 minutes), was extensively absorbed (~88% in 8 hours), once absorbed had a wide volume of distribution, and was rapidly metabolized primarily in the liver and excreted. Of the 14 pesticides tested, permethrin was one of the most rapidly absorbed and excreted.

C. STUDY DEFICIENCIES:

As a point of reference this study followed essentially none of the guidelines of OPPTS 870.7600. The deficiencies include: not reporting the lot number or purity of the test material, the use of mice when rats are required, the use of acetone as the delivery vehicle, verification of the doses applied, the use of a single dose when multiple doses are suggested, the use of three animals per duration group instead of four, not washing the application site following treatment, the limited dose application area, protection of the dose site following treatment, and the stipulated reporting requirements. Nonetheless, the study is considered **Acceptable/Nonguideline and provides supportive data regarding the absorption and distribution of permethrin in mice.**

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I

DATA EVALUATION RECORD

PERMETHRIN
(PP557)

STUDY TYPE: SUBCHRONIC ORAL TOXICITY - DOG [OPPTS 870.3150 (§82-1b)]

MRID 00048493
71951

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task No.01-89

Primary Reviewer: Gary A. Sega, Ph.D.	Signature: Date:	Gary Sega AUG 0 8 2001
Secondary Reviewer: H. Tim Borges, M.T.(A.S.C.P.), Ph.D. D.A.B.T.	Signature: Date:	H T Borges AUG 0 8 2001
Robert H. Ross, Group Leader	Signature: Date:	Robert H. Ross AUG 0 8 2001
Quality Assurance: Eric Lewis, M.S.	Signature: Date:	Eric B. Lewis AUG 0 8 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed and operated by UT-Battelle, LLC, for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725

PERMETHRIN/PC Code 109701

Subchronic (90 Day) Oral Study-Nonrodent / 1
OPPTS 870.3150/OECD 409 (§82-1b)

EPA Reviewer: Linnea Hansen, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang for
Date: 6/6/2002
Signature: Joycelyn B Stewart
Date: 6/6/2002

TXR#: 0050649

DATA EVALUATION RECORD

STUDY TYPE: Subchronic Oral Toxicity- Dog [OPPTS 870.3150 (§82-1b)]

P.C. CODE: 109701

DP BARCODE: D269531
SUBMISSION CODE: S504352

TEST MATERIAL: Permethrin, technical; purity 89.4 -98.8%

SYNONYMS: PP557; 3-phenoxybenzyl dl, *cis, trans* 2,2-dimethyl-3-(2,2-dichlorovinyl) cyclopropane-1-carboxylate

CITATION: Edwards, D.B., *et al.* (1976) Toxicity study in beagle dogs (Oral administration for 3 months). Inveresk Research International, Edinburgh, EH21 7UB, Scotland. IRI Report No. 462 (Project No. 404641), February, 1976. MRID 00071951. Unpublished.

SPONSORS: Imperial Chemical Industries Ltd., Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire SK10 4TJ.

EXECUTIVE SUMMARY: In a subchronic oral toxicity study (MRID 00071951), Permethrin (purity 89.4 - 98.8%; batch nos. P23, P28, P32, P35, P37 and P38) was administered daily in capsules to four beagle dogs/sex/dose at doses of 0, 10, 100 or 2000 mg/kg/day for 3 months.

At 2000 mg/kg/day, mild body tremors 1-2 hours after dosing, lasting about 1 hour, were seen throughout the study (number of animals affected and frequency not indicated). The mean weight gain of females was -31% less than controls. This decrease was not statistically significant, primarily because one female in this group (181♀) showed most of the reduced gain, but mean body weight gain was still slightly reduced (-11.5% less than controls) when 181♀ was excluded from calculation of gain. No significant weight changes were observed in males. Mean total food consumption and food efficiency were reduced in females (-12% and -26%, respectively) but showed no significant changes in males. The decrease in females appeared to be largely due to female 181♀ but food consumption/efficiency per animal could not be calculated due to housing animals 2 per cage. The decreased body weight gain, food consumption and food efficiency in females are considered possible treatment-related effects. Mean liver/body weight ratios were increased in both males (+23%) and females (+12%); absolute liver weights were only slightly increased (+11% males, female data partly illegible). Increased liver weight was not considered an adverse effect due to lack of corresponding clinical

laboratory alterations or liver histopathology. There were no treatment-related deaths and no effects on ophthalmological parameters, clinical laboratory parameters (hematology, clinical chemistry, urinalysis), gross pathology or microscopic pathology. **Under the conditions of this study, the subchronic toxicity LOAEL is 2000 mg/kg/day based on clinical signs in males and females and a possible body weight decrease in females. The NOAEL is 100 mg/kg/day.**

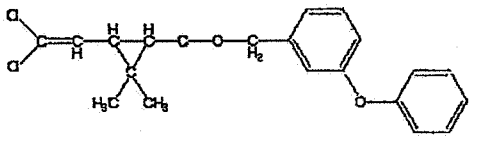
This subchronic toxicity study is classified as **Unacceptable/Guideline (upgradable)** and does not satisfy the guideline requirement for a 90-day oral toxicity study in non-rodents [OPPTS 870.3150 (§82-1b)]. It may be upgraded to Acceptable/Guideline upon submission of the following confirmatory data: (1) individual animal data for clinical signs of toxicity including number of animals affected and frequency/severity of finding for each animal; (2) weekly body weight data for weeks 2-12 of the study and (3) a legible copy of the study report to verify the findings in this study that could not be clearly read in the only available microfiche copy available to the Agency. This data requirement for OPPTS 870.3150 may be satisfied by chronic toxicity study in dogs on permethrin.

COMPLIANCE: Signed and dated Quality Assurance, GLP, Data Confidentiality and Flagging statements were not provided. This study was conducted prior to promulgation of the US EPA GLP guidelines.

I. MATERIALS AND METHODS

A. MATERIALS

1. **Test material:** Permethrin
   Description: oily liquid at room temperature; 40:60 cis:trans isomer ratio
   Lot/Batch #: Batches P23, P28, P32, P35, P37 and P38
   Purity: 89.4 - 98.8%, depending on Batch
   Stability of compound: not provided (compound was stored at 4°C)
   CAS No.: [52645-53-1]
   Structure:



Permethrin
(PP557)

   impurities: unknown

2. **Vehicle and/or positive control:** The test substance was administered in gelatin capsules. The control animals received the same number and size of empty gelatin capsules as used for the treated animals.

3. **Test animals:**
   Species/strain: beagle
   Age and weight at study initiation: ages not provided; males 4.9-9.3 kg, females 4.2-7.9 kg
   Source: not provided
   Housing: housed in pairs according to sex
   Diet: Complete dry diet (Spratts dog diet), 400g/day/animal (800 g/kennel/day)
   Water: *ad libitum*, source not provided
   Environmental conditions
   Temperature: not provided
   Humidity: not provided
   Air changes: not provided
   Photoperiod: not provided
   Acclimation period: 2 weeks

B. STUDY DESIGN

1. **In life dates:** Start: October 15, 1975; end: January 21, 1976

2. **Animal assignment:** Dogs were assigned to the test groups in Table 1. The method used for assignments was not provided.

TABLE 1: Study design		
Test group and dose level(mg/kg/day)	Number of animals/sex	
	Male	Female
Control 0	4	4
Low 10	4	4
Mid 100	4	4
High 2000	4	4

Data taken from text table, p. 8, MRID 00040493.

3. **Rationale for dose selection:** Doses were based on the results of an earlier dose-ranging study in which doses up to 8000 mg/kg/day were used with no serious adverse effects. The duration of this range-finding study and other details were not provided in the study report.

4. **Dose preparation and analysis:** Animals were weighed once a week and the doses to be administered during the subsequent 7 days were prepared on the same day on the basis of bodyweight. The animals were given single daily oral doses using gelatin capsules. No dose analysis was performed since the test material was used as received.

   Results –
   Stability: Stability of the test material was not provided.
   Homogeneity: Homogeneity testing was not performed since the doses were capsulated.
   Concentration: Test material was described as 89.4 - 98.8% pure, depending on the Batch No. No corrections for purity were mentioned in determining the daily doses.
   Conclusions: It is assumed that doses administered to animals contained the nominal amount of the test material.

5. **Statistics:** When necessary, statistical evaluation of the results was undertaken using Analysis of Variance followed by Student's t-test. Treated groups were compared with controls by sex. Body/organ weights, hematological, biochemical and urinalysis parameters were evaluated using a 95% confidence level ($p \leq 0.05$) for significance.

C. METHODS

1. **Observations:** The animals were observed at intervals throughout the working day for mortality or any signs of ill-health or reaction to dosing.

2. **Body weight:** Dogs were weighed weekly, beginning two weeks prior to dosing, at the start of the exposure period (day 0) and continuing through week 13.

3. **Food consumption and food efficiency:** Food residues were measured daily for each kennel and consumption was expressed as g uneaten food/dog/week. Each animal was administered 400 g food per day (800 per cage per day), for a total of 2800 g food/animal/week or 36,400 g/animal/study. Food efficiency was calculated by the reviewer from group means, using the equation (g body wt. gain ÷ g food consumed) × 100.

4. **Ophthalmoscopic examination:** Ophthalmoscopic examinations were conducted using indirect ophthalmoscopy on each dog before dosing and again after 4 and 12 weeks of dosing. During the dosing period, the eyes were examined prior to dosing.

5. **Blood:** Blood samples were drawn from all dogs before the initiation of treatment, and after 4 and 12 weeks of dosing. Samples were collected after fasting the dogs overnight. The CHECKED (X) parameters were examined.

   a. **Hematology**

X	Hematocrit (HCT)* (packed cell vol.)	X	Leukocyte differential count*
X	Hemoglobin (HGB)*	X	Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*	X	Mean corpusc. HGB Conc.(MCHC)*
X	Erythrocyte count (RBC)*	X	Mean corpusc. volume (MCV)*
X	Platelet count*	X	Reticulocyte count
	Blood clotting Measurements*(thromboplastin time) (prothrombin time)		

Data taken from p. 10, MRID 00040493.
* Recommended for subchronic studies based on OPPTS 870.3150 Guidelines

b. Clinical chemistry

	ELECTROLYTES		OTHER
	Calcium*	X	Albumin*
X	Chloride*		Blood creatinine*
	Magnesium	X	Blood urea nitrogen*
	Phosphorus*	X	Total Cholesterol*
X	Potassium*		Albumin/Globulin ratio
X	Sodium*	X	Glucose*
		X	Total serum protein (TP)*
	ENZYMES	X	Protein electrophoresis
	Alkaline phosphatase (ALK)*		
	Gamma glutamyl transferase (GGT)*		
X	Serum alanine aminotransferase (ALT)*		
X	Serum aspartate aminotransferase (AST)*		
X	Leucine aminopeptidase (LAP)		
X	Lactate dehydrogenase (LDH)		
X	Alpha hydroxy butyric dehydrogenase (HBDH)		

Data taken from pp. 10-11, MRID 00040493.
* Recommended/suggested for subchronic studies based on OPPTS 870.3150 Guidelines

6. **Urinalysis** was conducted on all dogs before treatment, and after 4 and 12 weeks of treatment. Urine was collected over 16 hours of a 21-hour period of water deprivation. The CHECKED (X) parameters were examined.

X	Appearance*	X	Albumin
X	Volume*	X	Glucose*
X	Specific Gravity*	X	Ketones
X	pH*	X	Blood*
X	Sediment (microscopic)	X	Bile Pigments
X	Protein*	X	Urobilinogen
	Osmolality*		

Data taken from pp. 10-11, MRID 00040493.
* Recommended for subchronic studies based on OPPTS 870.3150 Guidelines

7. **Sacrifice and pathology:** At the end of the dosing period each dog was given an anesthetic dose of pentobarbitone, exsanguinated and necropsied. Gross morphological examinations were conducted on all dogs and major tissues and organs. Organs checked (X) were sectioned and examined microscopically for all animals. In addition, the checked (XX) organs were weighed.

X	DIGESTIVE SYSTEM	X	CARDIOVASC./HEMAT.	X	NEUROLOGIC
X	Tongue	X	Aorta*	XX	Brain*+
X	Salivary glands*	XX	Heart*	XX	Pituitary
X	Esophagus*	X	Bone marrow*	X	Nerve* (sciatic)
X	Stomach*	X	Lymph nodes*		
X	Duodenum*	XX	Spleen*		**GLANDULAR**
X	Jejunum*	XX	Thymus*+		
X	Ileum*			XX	Adrenal gland*+
X	Colon* (upper and		**UROGENITAL**		Harderian glands
X	lower)			X	Mammary gland*
X	Caecum*	XX	Kidneys*+	XX	Thyroids* (including parathyroid,
X	Rectum*	X	Urinary bladder*		when present)
XX	Liver*	XX	Testes*		
X	Gall bladder*	X	Epididymides*		**OTHER**
XX	Pancreas*	XX	Seminal vesicle(s)*Prostate*		
		XX	Ovaries*+	X	Skeletal muscle
	RESPIRATORY	XX	Uterus*+	X	All gross lesions and masses*
				X	Skin*
	Nose*			X	Bone
X	Trachea*				
	Pharynx*				
	Larynx*				
XX	Lung*				

Data taken from pp. 12-13, MRID 00040493.
* Required for subchronic studies based on OPPTS 870.3150 Guidelines
+ Organ weight required in subchronic and chronic studies.

II. RESULTS

A. OBSERVATIONS

1. **Toxicity:** Those animals receiving 2000 mg/kg/day exhibited mild body tremors 1-2 hours after dosing. The tremors lasted for about 1 hour and were seen from day 1 throughout the study. No other clinical signs were seen. Summary or individual animal data were not presented to show the frequency of this finding.

2. **Mortality:** All dogs survived to scheduled necroscopy.

B. BODY WEIGHT

Group mean body weights and body weight gains are shown in Table 2. Only values for initial and final body weight data (and cumulative gain) were presented in the data tables of this study report, although individual animal and mean values for all of the study weeks were presented in graphical form. In addition, mean body weight values for males and females were calculated by the reviewer because only a combined mean was provided in the study report body weight data table (however, separate mean body weight gains for males and females were calculated in the study report). Females receiving 2000

mg/kg/day gained less weight than control females (-31%) although the decrease was not reported to be statistically significant. One female accounted for most of the reduced weight gain in this group (181♀), gaining only 350 g over the 13 week dosing period, compared to 2400 to 5200 g for the other 3 females. This female also appeared to have reduced food consumption, based on per cage food consumption data (see below). This female was relatively small compared to other females (4250 g at study start) and the pre-treatment food consumption in this cage was slightly reduced compared to other groups, suggesting that the animal may have been ill or otherwise compromised prior to dosing. When mean body weight/weight gain were calculated for 2000 mg/kg/day females excluding this animal, values were as follows: initial body weight 6583 g; final body weight 9967 g and gain 3383 g (-11.5% below controls; not analyzed statistically).

The study authors concluded that there were no treatment-related effects on body weight and attributed the lower weights in 2000 mg/kg/day females to 181♀. The reviewer considers the decrease in females to be a possible treatment-related effect because a slight decrease in cumulative gain was also observed when gain was calculated for the remaining three females of this group and because an effect of treatment on 181♀ cannot be absolutely ruled out.

TABLE 2. Mean body weights (kg) and body weight gains (kg) of dogs treated with Permethrin for 13 weeks.[a]

Study week	Exposure concentration (mg/kg/day)			
	0	10	100	2000
Males, Mean Body Weight (n=4/group) ± SD				
Initial	7.288 ± 1.673	6.175 ± 0.750	6.500 ± 1.074	6.875 ± 1.697
13	11.325 ± 1.803	11.275 ± 1.907	12.450 ± 2.666	10.800 ± 1.930
Males, Mean Body Weight Gain (Week 13 -Week 0)				
Weight Gain	4.038 ± 1.580	5.100 ± 1.201(126)[b]	5.950 ± 2.175(147)	3.925 ± 1.274 (-3)
Females, Mean Body Weight (n=4/group)				
Initial	6.150 ± 1.276	6.575 ± 0.497	6.300 ± 1.073	6.000 ± 1.399
13	9.975 ± 2.307	10.375 ± 2.656	9.925 ± 1.063	8.625 ± 3.292 (-13.5)
Females, Mean Body Weight Gain (Week 13 -Week 0)				
Weight Gain	3.825 ± 1.451	3.800 ± 2.529	3.625 ± 0.698 (-5.2)	2.625 ± 2.011 (-31)

Data taken from Table 1, p 20, *MRID 00040493.*

a Mean body weights for males and females were calculated by the reviewer because only combined mean weights were provided in the study report.

b Number in parenthesis represents percent of control, calculated by reviewer.

C. FOOD CONSUMPTION and FOOD EFFICIENCY

1. **Food consumption and food efficiency:** Food consumption values shown in Table 3, below, have been converted by the reviewer to g consumed/animal/time period from the g uneaten/animal/time period shown in the study report. The study authors drew conclusions about food consumption from combined mean male and female values (although separate data for each sex were available); separate calculations for males and females were done by the reviewer. Food consumption and food efficiency in treated males were similar to controls (Table 3). Food consumption of high dose females was reduced in 10 of the 13 weeks of dosing, being about 12% lower than controls for the total 13-week study period. Food efficiency for females in the high dose group was reduced by about 26% below the control value overall. Most of the decrease in food consumption and efficiency in females was due to reduced consumption in one cage (animal nos. 181♀ and 182♀) and most probably to 181♀, which also showed markedly reduced weight gain during the study. In the second cage of females, there was no treatment-related effect on food consumption or efficiency. The study authors concluded that there were no treatment-related effects in either males or females and that at 2000 mg/kg/day, they consumed 93% of the total food offered (mean for males and females combined). However, as discussed above, separate calculation of food consumption showed that males consumed about 95% of their food, while females consumed about 88% and while efficiency was not altered in males, it was reduced in females (by about 26%). The reviewer therefore concludes that there may have been a slight treatment-related effect on food consumption and food efficiency. Because the animals were doubly housed, individual animal effects on food consumption/efficiency could not be sorted out.

TABLE 3. Mean food consumption (g/animal/wk), mean total food consumed (g/animal/dosing period) and mean food efficiency [(g weight gain/g food consumed) x 100] of dogs treated with Permethrin for 13-weeks

Study week	Exposure concentration (mg/kg/day)			
	0	10	100	2000
Males, mean g food consumption/wk (Max.=2800g)				
1	2800	2750	2587 (92.3)[b]	2400 (85.7)
Males, mean g total food consumption over 13 weeks (Max.=36,400g)				
1-13	35,656	36,225	36,093	34,475 (94.7)
Males, mean body weight gain (g) over 13 weeks				
Weight Gain 1-13 wks (g)	4038	5100	5950	3925
Food Efficiency[a] (%)	11.32	14.07	16.48	11.38
Females, mean g food consumption/wk (Max.=2800g)				
1	2593	2618	2462	2050 (73.2)
6	2800	2800	2800	2575 (92.0)
13	2800	2800	2750	2250 (80.4)
Females, mean g total food consumption over 13 weeks (Max.=36,400g)				
1-13	34,312	35,918	34,915	31,943 (87.8)
Females, mean body weight gain (g) over 13 weeks				
Weight Gain 1-13 wks (g)	3825	3800	3625	2625 (68.6)
Food Efficiency (%)	11.15	10.58	10.38	8.21 (73.6)

Data taken from Table 1 and Appendix 1, p. 20 and 30, respectively, MRID 00040493. N = 4, each group
[a]Food efficiency calculated by reviewer as: (mean body weight gain/mean food consumption) x 100
[b]Numbers in parentheses indicate percent of control value, calculated by reviewer.

D. WATER INTAKE

Water was provided *ad libitum*. The amount of water consumed/kennel was assessed by visual inspection of the graduated water troughs, a more accurate measurement being made if an effect related to dosing was seen. No between group differences were noted.

E. OPHTHALMOSCOPIC EXAMINATION

Examination of the eyes before dosing commenced and after 4 and 12 weeks of dosing revealed no abnormalities.

F. BLOOD WORK

1. **Hematology:** Hematological parameters for all dosed animals were within normal limits initially and after 4 and 12 weeks of dosing.

2. **Clinical chemistry:** All clinical chemistry parameters for all dosed animals were within normal limits initially and after 4 and 12 weeks of dosing. It was noted that after 4 weeks of dosing those animals receiving 100 and 2000 mg/kg/day showed a slight depression of plasma glucose compared with the controls (see Table 4). However, the decrease was not statistically or biologically significant.

TABLE 4. Plasma glucose levels (mg/100 mL) in dogs treated with Permethrin, for 13 weeks.

Dose Levels (mg/kg/day)	0	10	100	2000
Males				
0 weeks (before dosing)	87.5 ± 10.2[a]	89.0 ± 4.1	87.3 ± 6.1	75.3 ± 12.1 (86.1)[b]
4 weeks	75.8 ± 11.7	82.8 ± 2.6	79.6 ± 5.7	63.3 ± 17.2 (83.5)
12 weeks	76.0 ± 5.0	70.2 + 4.9	62.0 ± 5.9 (81.6)	71.0 ± 12.7 (93.4)
Females				
0 weeks	84.0 ± 11.3	85.8 ± 15.8	79.0 ± 8.4 (94.0)	77.8 ± 5.1 (92.6)
4 weeks	79.5 ± 2.4	84.0 ± 5.6	59.6 ± 8.2 (75.0)	61.2 ± 18.7 (77.0)
12 weeks	62.2 ± 6.2	64.0 ± 6.9	66.0 ± 5.8 (106)	67.0 ± 9.3 (108)

Data taken from Appendices 5-7, pp. 37-42, MRID 00040493. N = 4, all groups.
[a]SD (standard deviation)
[b]Percent of control value, calculated by reviewer.

G. URINALYSIS

No treatment-related changes in urinalysis values were found.

H. SACRIFICE AND PATHOLOGY

1. **Organ weight:** One male in the low dose group, and one male and one female in the high dose group, had liver weights that exceeded 4% of the body weight. The study report noted that the group mean relative (to body weight) liver weights were higher than the controls (Table 5, below). Statistical analysis showed that this increase was only significant for the high dose group. This study report only calculated combined male and female organ weights. When mean liver weights were evaluated for each sex independently using the individual animal data, at 2000 mg/kg/day males showed slightly greater increases in liver weights (absolute liver weights +11%; female data was partly illegible but appeared to be an increase of less than +5%. Relative liver weights in males increased by +23%, vs. +12%, females. Evaluation of absolute liver weights for males and females separately at 10 and 100 mg/kg/day (and females at

2000 mg/kg/day) was not possible because some values were illegible. However, since the increases in liver weights were not correlated with any clinical laboratory or histopathological changes, the reviewer agreed with the study authors that it was not toxicologically significant.

TABLE 5. Absolute and relative liver weights among dogs fed Permethrin for 13 weeks.

Observation	Dose Levels (mg/kg/day)			
	0	10	100	2000
Males + Females combined (N = 8, all groups)				
Absolute Liver wt.±SD (g)	327.05 ±8.2	359.32 ±7.4 (+9.8)[a]	393.46 ±0.9 (+20.3)	350.29 ±2.4 (+7.1)
Relative Liver wt.±SD (%)	3.11 ±0.44	3.35 ±0.52	3.54 ±0.38 (13.8)	3.67 ±0.50* (+18)

Data taken from Appendix 11a/b, pp. 49-52, MRID 00040493.
* Significantly different from control, $p \leq 0.05$.
a Values in parentheses indicate percent increase relative to controls.

2. **Gross pathology:** No treatment-related macroscopic changes were seen at terminal sacrifice. Parasitic granulomata were seen in the kidneys of one male and one female dog in the high dose group, but this finding was only confirmed histologically in the male dog. Small localized areas (2-3 mm dia.) of submucosal congestion and/or hemorrhage were seen in the jejunum, ileum and upper colon of one male and one female dog in the control group and in one male from the high dose group. These findings were only confirmed histologically in the control male. Autopsy findings for dogs from the low and intermediate dose groups were limited to small localized areas of submucosal congestion and/or hemorrhage in the jejunum, ileum and colon, with three males and three females in the low dose group and one male in the intermediate dose group showing this effect.

3. **Microscopic pathology:** No lesions were identified which could be attributed to the test chemical. Evidence of larval ascarid migration through tissues, particularly liver, lung and kidney were seen in three males and three females from the high dose group and in one male and two females from the control group.

   There was some variation in the maturation of the testes in both the controls and high dose groups. This variation reflects normal differences in the time of onset of puberty in these animals.

III. DISCUSSION

A. **INVESTIGATORS' CONCLUSIONS:** The study authors determined that the only significant treatment-related effect observed in this study was mild body tremors, observed within hours of dosing in animals treated with 2000 mg/kg/day permethrin,

persisting for about an hour and occurring throughout the study. No significant effects were reported at 10 or 100 mg/kg/day. The decrease in mean body weight gain and food consumption observed at 2000 mg/kg/day was determined to be primarily due to one female, was not statistically significant and was therefore not considered a treatment-related effect. Decreases in serum glucose in males and females at week 4 and males at weeks 4 and 12 in the 2000 mg/kg/day groups (and at week 4 at 100 mg/kg/day) were not considered toxicologically significant because they were within normal limits, did not persist in females and in males, the mean initial glucose level on day 0 was lower than the other dose groups. Although dose-dependent increases in absolute and relative liver weights (combined for both sexes) were observed and were related to treatment, they were not considered an adverse effect in the absence of associated liver histopathology.

B. **REVIEWER'S COMMENTS:** Permethrin was administered orally at dose levels of 0, 10, 100 or 2000 mg/kg/day to male and female beagle dogs (4/sex/group) for 13 weeks. All animals survived to terminal sacrifice.

The reviewer agreed with the conclusions of the study author, with the exception of body weight and food consumption findings, discussed below. The only clinical observation noted was mild body tremors 1-2 hours after dosing in the high dose group. These lasted about 1 hour and were seen from day 1 throughout the study. Transient tremors is a consistently reported finding in animal studies on permethrin.

Both mean body weight and body weight gain were reduced in females from the high-dose group. The mean weight gain of this group was 69% of the control value although this decrease was not statistically significant, primarily because just one female in this group showed most of the weight reduction. No other significant weight changes were noted in any other group.

Mean total food consumption was slightly lower for males from the high dose group compared to controls (98.4% overall) and for the high dose females (96.7% of controls, overall). However, none of these reductions reached statistical significance and food efficiencies for males from all treated groups were similar or higher than the control males, although none of the increases were statistically significant. With females, food efficiencies for the dosed animals decreased in a dose-dependent manner, dropping to 71% of the control's food efficiency for the high dose females. However, none of these decreases were statistically significant. The cage with 181♀, the female with low wieght gain, also showed reduced food consumption.

The decreases in body weight, food consumption and food efficiency in high dose females are largely due to unusually low gain and food consumption in one female. This female may have had compromised health prior to dosing based on lower initial body weight and slightly reduced food consumption relative to other cages in the pretreatment weeks. The other cage of two females showed no treatment-related changes in body weight or food consumption. Although these effects in females are questionable, the

reviewer considers the overall decrease in body weight gain of females to be a possible effect of treatment because mean body weight gains were still -11.5% below controls if that animal were excluded from calculations and because treatment-related effects on animals 181♀ (and possibly 182♀) cannot be absolutely ruled out. Because the animals were not housed individually, the food consumption could not be evaluated for individual animals.

All hematological parameters were within normal limits for all dose groups throughout the study. Clinical chemistry studies showed a slight decrease in glucose levels in the mid- and high-dose groups. For males, the decrease was 79 and 83% of the controls, respectively, at 4 weeks; and 82 and 93%, respectively, at 12 weeks. However, it is noted that the day 0 glucose level was only 86% of controls as well. For females, the decrease was 75 and 77% of the controls, respectively, at 4 weeks; there was no glucose decrease in females at 12 weeks. None of the glucose decreases were statistically or biologically significant.

The only notable change in organ weights was seen in the liver at 2000 mg/kg/day. The absolute mean liver weights for males and females combined (calculated by study authors) were slightly higher than the controls for all dose groups although this effect was not dose-related. In addition, the mean liver/body weight ratios were slightly higher in all treated groups compared to controls and this ratio did appear to be dose-related. However, the increase in the relative liver weights was only statistically significant in the high-dose animals. Only some calculations of male and female absolute and relative liver weights could be performed due to illegible data tables, but when calculated separately, males appeared to have a more pronounced effect than females on liver weight. This finding is considered to be an "adaptive" response and not an adverse effect in the absence of related clinical laboratory alterations or liver histopathology.

Histopathological examination found no lesions that could be attributed to treatment with the test chemical. Localized areas of submucosal congestion and/or hemorrhage were observed in the jejunum, ileum and upper colon in some animals from all dose groups and the control. The authors suggested that this may have been caused by the dogs ingesting wood chips from their bedding. The incidence of this observation was not dose-related. In addition, this finding had been observed previously in other undosed dogs and was therefore not considered to be treatment-related.

Under the conditions of this study, the subchronic toxicity LOAEL is 2000 mg/kg/day based on tremors (males and females) and a possible decrease in body weight gain in females. The NOAEL is 100 mg/kg/day.

This subchronic toxicity study is classified as **Unacceptable / Guideline (upgradable)** and does not satisfy the guideline requirement for a 90-day oral toxicity study in non-rodents [OPPTS 870.3150 (§82-1b)]. It may be upgraded to Acceptable/Guideline upon submission of the following confirmatory data: (1) individual animal data for

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clinical signs of toxicity including number of animals affected and frequency/severity of finding for each animal; (2) weekly body weight data for weeks 2-12 of the study and (3) a legible copy of the study report to provide verification of the findings in this study that could not be clearly read in the only available microfiche copy available to the Agency. This data requirement for OPPTS 870.3150 may be satisfied by chronic toxicity study in dogs on permethrin.

C. **STUDY DEFICIENCIES:** (1) The animals in this study could have tolerated a higher dose of the test article; higher doses may have produced more clear-cut effects. (2) Neither a summary nor individual animal data table for clinical observations was provided. (3) The body weight data table only presented initial and week 13 body weights, although weekly weights were measured. Graphical presentation of the mean and individual animal body weights throughout the study provided some information, however. (4) Also, some mean data values were calculated by combining male and female data, for example body weight and liver weight.

This study was carried out more than 24 years ago, when many of the current EPA regulations/guidelines for toxicity testing had not been promulgated. Also, much of the data in the only copy of the report available to the Agency is, or is almost, illegible due to poor microfiche quality.

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DATA FOR ENTRY INTO ISIS

ıbchronic Oral Study - non-rodents (870.3150)

?C code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
09701	00071951	subchronic	dog	13 weeks	oral	capsule	10-2000	0, 10, 100, 2000 mg/kg/day	100	2000	neuromuscular system, possible decr. body wt.	Toxicity

4

DATA EVALUATION REPORT

PERMETHRIN/109701
(PP557)
STUDY TYPE: RODENT DOMINANT LETHAL ASSAY IN MICE
[OPPTS 870.5450 (§84-2)]
MRID 00043730

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 02-05

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Disclaimer

This review may have been altered subsequent to the contractor's signature above.

Oak Ridge National Laboratory, Managed and Operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

EPA Reviewer: Yung Yang, Ph.D.
Toxicology Branch, Health Effects Division (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 5/17/2002
Signature: Joycelyn Stewart
Date 5/21/2002

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Rodent dominant lethal assay in mice; [OPPTS 870.5450 (§84-2)]

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): PP557 (Permethrin, 95.3% a.i., cis/trans ratio - 37.5/57.8)

SYNONYMS: 3-phenoxybenzyl dl, cis, trans 2,2-dimethyl-3-(2,2-dichlorovinyl) cyclopropane-1-carboxylate

CITATION: McGregor, D.B. and G.A. de S. Wickramaratne (1976) Dominant lethal study in mice of ICI-PP557. Inveresk Research International, Edinburgh, EH21 7UB, Scotland. IRI Project No. 406722. November, 1976. MRID 00043730. Unpublished

SPONSOR: Imperial Chemical Industries, Alderley Park, Macclesfield, Cheshire.

EXECUTIVE SUMMARY: In a dominant lethal assay (MRID 00043730), male CD-1 mice (15/dose) were treated once daily via the oral route with PP557 in corn oil at a dose of 15, 48 or 150 mg/kg/day for five consecutive days. Starting immediately after the final dosing, each male was mated with 2 untreated virgin females per week for eight weeks. The females were sacrificed 13 days after the midweek of their mating period and the pregnancy rate and the number of living and dead implants determined.

PP557 was tested to an upper dose determined by a preliminary toxicity test in which two of six male mice given 150 mg/kg for five consecutive days died by day seven following start of treatment. Four mice, two each from the 15 and 48 mg/kg groups, died during the dominant lethal study but the cause of death was not given. There were no statistically or biologically significant differences in the pregnancy rate or number of living and dead implants in PP557 treated groups when compared with the solvent control group. The solvent and positive control values were appropriate. **There was no statistically significant difference between the control group and the PP557 treated groups with respect to fertilization rate or the number of living and dead implantations in any week.**

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This study is classified as **Acceptable/Guideline**. It satisfy the requirement for FIFRA Test Guideline OPPTS 870.5450 (84-2) for rodent dominant lethal data.

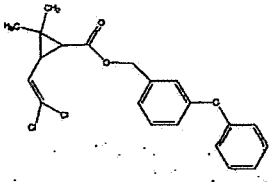
COMPLIANCE: No signed and dated GLP, Data Confidentiality or Quality Assurance Statements were provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test material: PP557

Description:	Straw-colored solid at room temperature
Lot/Batch #:	25
Purity:	95.3% a.i. (cis/trans ratio - 37.5/57.8)
CAS # of TGAI:	*52645-53-1*
Solvent Used:	Corn oil



2. Control materials:

Negative:	None
Solvent / final volume / route of administration:	Corn oil /10 mL/kg/day / oral gavage
Positive / final dose(s) / route of administration:	Ethylmethanesulfonate / 100 mg/kg/day / oral gavage

3. Test compound administration:

Volume:	10 mL/kg/day
Route of administration:	Oral gavage
Dose level used:	15, 48 or 150 mg/kg/day for 5 consecutive days

4. Test animals:

Species:	Mouse
Strain:	CD-1
Age/weight at study initiation:	Males: 10 - 12 weeks/ Females: 8 - 10 weeks/
Source:	Charles River
No. animals used per dose:	15 males mated to 2 untreated females/week for 8 weeks
Properly Maintained?	Iinformation not provided

B. TEST PERFORMANCE:

1. Treatment:

a. Test compound and solvent control
Dosing: __ once ___ twice (24 hr apart)
_x__ other (describe): once daily for 5 consecutive days

2. **Mating:** Immediately after the last treatment each male was caged with two untreated females. The females remained with the male for one week at which time they were removed and replaced with two new females. This process was repeated for eight weeks. An eight-week mating period in mice allows sampling of sperm treated at all germ cell stages. Females were killed 13 days after midweek of the mating period.

3. **Caesarian procedures:** Not described

4. **Evaluation criteria:** The uteri were examined for live implantations and early and late deaths.

5. **Statistical methods:** A week by week analysis of variance was applied to the results and the data were analyzed using the Genstat program from the Rothamsted Experimental Station, Hertfordshire, England.

II. REPORTED RESULTS

A. **Preliminary toxicity assay:** Groups of six male mice were treated with PP557 at doses of 18.8, 37.5, 75, 150, 300 or 600 mg/kg/day for five consecutive days and survival recorded on day seven. Two of the six mice in each of the top three dose groups were dead by day seven. A dose of 150 mg/kg/day for five days was chosen as the top dose for the dominant lethal assay.

B. **Dominant lethal**: Fifteen male mice per group were treated with PP557 at doses of 15, 48 or 150 mg/kg/day for five consecutive days and mated with two virgin females each per week for eight weeks. The females were killed 15 or 16 days after being placed with a male mouse. Fertilization was assumed to have occurred two or three days after the female was placed with the male. No discussion of deaths or clinical signs in the PP557 treated mice was presented; however, from the individual animal data tables, two males in the 48 mg/kg group died, one during the second week and one during the sixth week. Two males in the 15 mg/kg group died during the fourth week. The cause(s) of death were not given. No deaths occurred in the 150 mg/kg group.

There were no statistically significant differences in the pregnancy rate of female mice mated to PP557 treated males during any mating interval when compared to the respective solvent control group. The pregnancy rate in the high dose group in week 1, the mating interval most likely to reveal a treatment related effect on pregnancy rate, was 86.7% while that of the solvent control group in week 1 was 80.0%. The EMS positive control did not affect the pregnancy rate. No statistically significant differences between groups was seen in the mean numbers of implantations per female in any mating week except weeks three and seven. The differences ($p < 0.05$) seen in weeks three and seven were small and due mainly to the low dose group. Because no dose-response relationship was seen, the differences were not considered biologically significant. The total implantation results are summarized in Table 1.

TABLE 1. Estimated means of total implantations per pregnancy in the dominant lethal mutation test of PP557

Week	Group					[1]s.e.d.	[2]F
	1	2	3	4	5		
1	11.19	11.8	11.5	11.71	11.04	0.56	[3]0.?4 ns
2	12.37	13.31	12.89	12.89	12.4	0.54	0.84 ns
3	13.13	12.32	13.62	11.63	12.61	0.42	2.63 *
4	13.23	12.89	12.11	12.88	12.77	0.58	1.06 ns
5	12.54	12.7	13.04	12.88	13.03	0.53	0.28 ns
6	12.1	12.1	13.33	12.5	12.5	0.47	1.76 ns
7	12.43	12.53	11.83	11.5	12.9	0.43	2.74 *
8	12.07	11.81	12.17	12.42	12.5	0.57	[3]0.?? ns

[1]s.e.d. = standard error of the difference of the means.
[2]F = variance ratio.
[3] ? = illegible entry on Table 2 from MRID 43730, p. 14.
* = statistically significant at $p < 0.05$.
ns = not statistically significant.
Group: 1 = solvent control; 2 = 150 mg/kg PP557; 3 = 48 mg/kg PP557; 4 = 15 mg/kg PP557; 5 = EMS positive control.

No statistically significant increases in early deaths compared to the solvent control values were seen in PP557 treated groups. The study authors describe four methods of analyzing early deaths and express the results of the study in each of the four methods. The results are expressed as:

1. the number of pregnancies with one or more early deaths and as the number of pregnancies with two or more early deaths;
2. the number of live implants and late deaths per pregnancy;
3. the percentage of implants recorded as early deaths;
4. the number of early deaths per pregnancy.

Results expressed as described in options 1 - 3 are summarized in Appendix Tables 1 through 4 (MRID 43730, pp. 15 - 18). The results expressed as the number of early deaths per pregnancy are shown in Table 2 (reproduced from Table 7 of MRID 43730, p. 19).

TABLE 2. *Transformed* ($\sqrt{Y} + \sqrt{Y+1}$) early deaths/pregnancy in the dominant lethal mutation test of PP557.

Week	Group					[1]s.e.d.	[2]F
Week	1	2	3	4	5		
1	1.59	1.9	1.43	1.74	3.05	0.23	12.94***
2	1.54	2	1.15	1.51	2.83	0.26	15.62***
3	1.79	2.13	2	1.62	2	0.27	1.29 ns
4	1.71	1.73	1.99	2.08	1.66	0.23	0.94 ns
5	1.98	1.8	1.74	1.47	1.56	0.24	1.54 ns
6	1.72	1.52	1.75	1.77	1.77	0.22	0.37 ns
7	1.97	1.85	1.5	1.65	1.09	0.22	5.92 ***
8	1.35	1.52	1.53	1.46	1.66	0.2	0.51 ns

[1]s.e.d. = standard error of the difference of the means.
[2]F = variance ratio.
*** = statistically significant at $p < 0.001$.
ns = not statistically significant.
Group: 1 = solvent control; 2 = 150 mg/kg PP557; 3 = 48 mg/kg PP557; 4 = 15 mg/kg PP557; 5 = EMS positive control.

III. REVIEWER'S DISCUSSION/CONCLUSIONS:

A. The study is classified as **Acceptable/Guideline**. PP557 was tested to an acceptably high dose as based on the preliminary toxicity test, proper experimental protocol was followed and the solvent and positive control values were appropriate. There was no evidence that PP557 induced dominant lethal mutations in the germ cells of male mice.

B. STUDY DEFICIENCIES: No deficiencies with respect to study protocol were identified; however, the authors did not provide information on any clinical signs seen in treated mice and did not give or speculate on the cause of death of the four male mice that died during the study. No signed and dated GLP or Quality Assurance Statements were provided.

APPENDIX

(MRID 00043730)

THE FOLLOWING ATTACHMENTS ARE NOT AVAILABLE ELECTRONICALLY. SEE THE FILE COPY.

Tox Review # 50849

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_____ A draft product label.

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DATA EVALUATION RECORD

PERMETHRIN

STUDY TYPE: SUBCHRONIC INHALATION TOXICITY - RAT (OPPTS 870.3465/OECD 413) MRID 00096710

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
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Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37830
Task Order No. 02-04

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	Date:	NOV 28 2001
Robert H. Ross, M.S., Group Leader	Signature:	Robert H. Ross
	Date:	NOV 28 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature:	L. A. Wilson
	Date:	NOV 28 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

EPA Reviewer: Yung Yang, Ph.D.
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, HED (7509C)

Signature: Yung C. Yang
Date 12/18/2001
Signature: Joycelyn Stewart
Date 1/28/2002

TXR#: 0050649

DATA EVALUATION RECORD

STUDY TYPE: 4-Day Inhalation Toxicity - rat; OPPTS 870.3465 [§82-4]; OECD 413.

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): 21Z73 (Permethrin, purity not provided)

FORMULATION: Permethrin (1%, a.i.) in arcton (99%)

SYNONYMS: None provided

CITATION: Harper, D.W., Piercy, D.W.T., James, J.A., Thomson, P.M. (1977) 21Z73 - 4 day rat inhalation study. Wellcome Co., Berkhamsted, England. A09, October 21, 1977. MRID 00096710. Unpublished.

SPONSOR: Not provided.

EXECUTIVE SUMMARY: In a 4-day inhalation toxicity study (MRID00096710) Permethrin (Ref. No. #P2610)] was administered to groups of 6 female Sprague-Dawley rats/concentration by dynamic whole- body exposure at concentrations of 0 and approximately 0.02 mg/L continuously for 4 days. The test material (1%) was delivered from pressurized containers containing Arcton 12 (99%) as the carrier.

There was no reported test material-related effect on mortality, hematology, clinical chemistry, organ weights, or gross and histologic pathology. The treated animals huddled in the exposure chamber and groomed the excess test material on their fur. As a consequence of this behavior, food intake was lower than that of the control group and the treated group lost weight whereas the control group gained weight over the 4-day period. Effects on the lungs in the treated group - congestion and macrophage infiltration - could not be clearly distinguished from the respiratory infection present in all animals. Other than weight loss in the treated group, a LOAEL for clinical findings could not be established. **A LOAEL or NOAEL could not be established because of the respiratory infection in both the control and treated groups.**

This 4-day inhalation toxicity study in the rat is **Unacceptable/Non-Guideline** and does not satisfy the guideline requirement for a subchronic inhalation study OPPTS 870.3465; OECD 413 in the rat. The study is not upgradable due to the short (4 day) exposure duration (subchronic

studies require 90 day) and lack of aerosol particle size determination. The study can be used as a range-finding study.

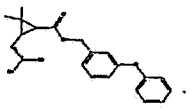
COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test material:

Description:	Formulation (1%, a.i.) provided in aerosol cans with metered valve; no other information provided
Lot/Batch No.:	Ref. No. P2610
Purity:	Not provided
Compound Stability:	Not provided
CAS No. of TGAI:	Not provided



2. Vehicle and/or positive control: Propellant was Arcton 12 (99%)

3. Test animals:

Species:	Rat	
Strain:	Sprague-Dawley	
Age/weight at study initiation:	Age not provided 200-230 g	
Source:	OLAC 1976 Ltd.	
Housing:	Plastic cages with wire mesh floor (control group and treatment group prior to exposure)	
Diet:	Heygates diet 41B meal, *ad libitum*	
Water:	Not described, available *ad libitum*	
Environmental conditions:	**Temperature:**	Not provided
	Humidity:	Not provided
	Air changes:	Not provided; air flow of 11 L/minute
	Photoperiod:	Not provided
Acclimation period:	Not provided	

B. STUDY DESIGN:

1. In life dates - Start: Not provided; End: Not provided

2. Animal assignment: the animal assignment technique was not explained; the control and treatment groups consisted of 6 females each as noted in Table 1.

TABLE 1: Study design					
Test group	Nominal Conc.[a] (mg/L)	Analytical Conc.[b] (mg/L)	MMAD µm	GSD	Rats/sex
Control	0				6 females
Treatment	0.02	≥0.02	Not provided	Not provided	6 females

[a]Based on air flow and weight discharged.
[b]Based on collections on cascade impactor.

3. **Dose selection rationale** - Information on dose level selection was not provided. Based on the amount of active ingredient (0.02 mg/L or approximately 1%) and propellant (1.98 mg/L or approximately 99%), the total exposure of active ingredient plus Arcton 12 carrier was approximately 2 mg/L.

4. **Generation of the test atmosphere / chamber description:** The group of 6 female rats was exposed continuously in an 80 L exposure chamber for 4 days. The test atmosphere was generated by discharging the test material from the pressure packs via a 200 µL electronically-activated valve at 1-minute intervals. The control group may have remained in the plastic home cages.

   Time to equilibrium was not provided.
   Analytical Chemistry was not provided.

   Test atmosphere concentration - Aerial samples were taken at the side of the exposure chamber opposite the inlet, an area with the potentially lowest concentration. Samples were collected on a cascade impactor. The analyzed aerial concentration was given as ≥0.02 mg/L, but the method of analysis was not stated. Therefore, the analyzed concentration is most likely a gravimetric concentration. Results are in Table 1 above.

   Particle size determination - Not reported. Aerosol droplets were visible on the chamber walls and on the test animals.

5. **Statistics** - The analysis methods were not described.

C. METHODS:

1. Observations:

1a. **Cageside observations:** Animals were inspected at undefined "regular intervals" for signs of toxicity and mortality.

1b. **Clinical examinations**: Clinical examinations were conducted at undefined "regular intervals."

1c. **Neurological Evaluations**: No neurological evaluations were performed. Neurologic evaluations other than those performed as part of the clinical observations are not recommended according to OPPTS 870.3465 guidelines.

2. **Body weight**: Animals were weighed on days -3, 0, and 4.

3. **Food consumption:** Food consumption for each animal was assessed for days -3 to 0 and 0 to 4 and mean daily diet consumption was calculated over these two periods as g food/day. Food efficiency was not calculated. Compound intake could not be calculated.

4. **Ophthalmoscopic examination:** Eyes were not examined as part of this study.

5. **Hematology & Clinical chemistry:** Blood was collected from the orbital sinus of all rats on days -3 and +4 for hematology and clinical analysis. Rats were lightly anaesthetized with ether on day -3 and with pentobartitone sodium on day +4. The animals were not fasted. The CHECKED (X) parameters were examined.

a. **Hematology**

X	Hematocrit (HCT)*		Leukocyte differential count*
X	Hemoglobin (HGB)*	X	Mean corpuscular HGB (MCH)*
X	Leukocyte count (WBC)*	X	Mean corpuscular HGB concentration (MCHC)*
X	Erythrocyte count (RBC)*	X	Mean corpuscular volume (MCV)*
	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Recommended for subchronic inhalation studies based on Guideline 870.3465

b. **Clinical chemistry**

	Electrolytes		Other
	Calcium		Albumin*
	Chloride		Creatinine*
	Magnesium	X	Urea nitrogen*
	Phosphorus		Total Cholesterol*
X	Potassium*		Globulins
X	Sodium*	X	Glucose*
	ENZYMES (more than 2 hepatic enzymes eg., *)		Total bilirubin
	Alkaline phosphatase*		Total serum protein (TP)*
	Cholinesterase (ChE)		Triglycerides
X	Creatine phosphokinase		Serum protein electrophores
	Lactic acid dehydrogenase (LDH)		
	Alanine amino-transferase (ALT/also SGPT)*		
X	Aspartate amino-transferase (AST/also SGOT)*		
	Sorbitol dehydrogenase*		
	Gamma glutamyl transferase (GGT)*		

X	Glutamate dehydrogenase		

* Recommended for subchronic inhalation studies based on Guideline 870.3465

6. **Urinalysis***

Urine was not collected.

* Optional for inhalation toxicity studies

7. **Sacrifice and pathology** : All animals that died and those sacrificed on schedule were subjected to gross pathological examination and the CHECKED (X) tissues were collected and examined microscopically. The (XX) organs, in addition, were weighed.

	Digestive System		Cardiovasc./Hemat.		Neurologic
	Tongue		Aorta, thoracic*	XX	Brain*+
	Salivary glands*	XX	Heart*+		Peripheral nerve*
	Esophagus*		Bone marrow*		Spinal cord (3 levels)*
	Stomach*		Lymph nodes*		Pituitary*
	Duodenum*		Spleen*+		Eyes (optic nerve)*
	Jejunum*		Thymus*+		**Glandular**
	Ileum*				Adrenal gland*+
	Cecum*		**Urogenital**		Lacrimal gland
	Colon*	XX	Kidneys*+		Parathyroid*
	Rectum*		Urinary bladder*		Thyroid*
XX	Liver*+		Testes*+		**Other**
	Gall bladder* (not rat)		Epididymides*+		Bone (sternum and/or femur)
	Bile duct* (rat)		Prostate*		Skeletal muscle
	Pancreas*		Seminal vesicles*		Skin
	Respiratory		Ovaries*+		All gross lesions and masses*
	Trachea*		Uterus*+		
XX	Lung*		Mammary gland*		
X	Nose* (Nasal cavities)				
	Pharynx*				
	Larynx*				

* Recommended for subchronic rodent studies based on Guideline 870.3465
+ Organ weights required

II. RESULTS

A. OBSERVATIONS :

1. **Clinical signs of toxicity**: The rats huddled in the corner of the exposure chamber furthest from the test material inlet. When roused, they appeared normal, but groomed themselves "more frequently and vigorously than usual." No other observations were recorded.

2. **Mortality**: There were no deaths during the exposure. All rats were sacrificed immediately after the 4-day exposure.

3. **Neurological evaluations**: There were no neurological evaluations.

B. BODY WEIGHT AND WEIGHT GAIN:

TABLE 2. Average body weights and body weight gains during 4 days of treatment are listed in Table 2. The treated group lost weight. The percent of control weight gain was not calculated.

Gravimetric concentration (mg/L)	Body weights (g±SD)		Total weight gain	
	Day 0	Day +4	g	% of control
0	227.9±2.9	232.5±3.5	4.6	
0.02	224.5±5.0	215.2±4.0*	-9.3**	Not determined

[a] Data obtained from page 008 in the study report.
* Significantly different ($p < 0.05$) from the control.
** Significantly different ($p < 0.01$) from the control.

C. FOOD CONSUMPTION:

1. **Food consumption**: Mean food intakes (g/rat/day) in the control and treatment groups over the 0 to day 4 treatment period were 19.5 and 14.1.

2. **Food efficiency**: Food efficiency was not calculated.

D. OPHTHALMOSCOPIC EXAMINATION: Ophthalmoscopic examinations were not performed.

E. BLOOD ANALYSES:

1. **Hematology**: The authors stated that there were no significant dose-related changes in hematology parameters. The hematology tables were unreadable.

2. **Clinical chemistry**: The study authors stated that there were no significant dose-related changes in clinical chemistry parameters. The clinical chemistry tables were unreadable.

F. URINALYSIS: Urinalysis was not performed.

G. SACRIFICE AND PATHOLOGY:

1. **Organ weight**: Only the brain weight relative to body weight was affected. The relative brain weight of the treated group was increased (112% of the control value; $p<0.01$). As there were no histological correlates, the increased relative brain weight is most likely attributable to the reduced body weight.

2. Gross pathology: All control and treated rats showed signs of pneumonia ranging from "just visible" to "obvious." Both groups had similar numbers of animals with pale lungs, but

congestion was visible in 3/6 of the treated animals vs 0/6 of the control animals. Pale livers were also observed in similar numbers of control and treated animals, but 4/6 treated animals also had obvious mottling of the liver whereas 0/6 animals in the control group had mottled livers.

3. **Microscopic pathology**: There were no treatment related differences between the control and treated groups in lesions of the lungs, kidneys, livers, hearts, or brains. The lungs from both groups showed evidence of respiratory infection; both congestion and macrophages were present in the alveolar spaces of more treated animals (3/6) than control animals (1/6 and 0/6, respectively). There was no histologic correlate for the grossly observed "mottling" of the liver.

III. DISCUSSION AND CONCLUSIONS

A. **INVESTIGATORS' CONCLUSIONS:** The study authors concluded that there were no treatment-related effects on hematology or clinical chemistry parameters, absolute and relative organ weights or incidences of microscopic lesions in selected organs. Continuous exposure of female rats to the test material for 4 days resulted in less food intake than that of controls which resulted in weight loss. Deposition of the test material on the fur elicited a more frequent and vigorous grooming response.

B. **REVIEWER COMMENTS:** The reviewer agrees with the conclusions of the study authors with the caveat that exposure to the test material may have exacerbated the respiratory infection in the treated animals. The presence of the respiratory infection in all animals invalidates the observations of lesions in the lungs. Due to the nature of the continuous exposure, the rats huddled and did not eat and, therefore, lost weight. The weight loss does not appear to be attributable to the toxicity of the test material. Invalidation is based on the deficiencies listed below.

This 4-day inhalation toxicity study in the rat is **Unacceptable/Non-Guideline** and does not satisfy the guideline requirement for a subchronic inhalation study OPPTS 870.3465; OECD 413 in the rat. The control and treated rats showed signs of pneumonia, and therefore a direct effect of the test material on the lungs could not be ascertained. The study is not upgradable because of multiple guideline deficiencies, including a greatly shortened exposure period and failure to provide the MMAD and GSD. The study can be used as a range-finding study.

C. **STUDY DEFICIENCIES:**

This study was initiated before 1996 OPPTS 870.3465 subchronic guidelines were in place and does not fulfill many of the subchronic guidelines. This 4-day study is more suitable for the 4-hour acute inhalation toxicity guideline (OPPTS 870.1300), but also does not fulfill the acute guidelines. The study also does not fulfill acute guidelines (OPPTS 870.1300) due to lack of healthy animals, failure to determine/report aerosol particle size, and lack of a post-exposure observation period of 14 days. In spite of the respiratory infection observed in all animals, the study can be used as a range-finding study.

Major deficiencies include:

1. Inadequate treatment period (4 continuous days rather than 6 hours/day, 5 days/week for 90 days).
2. Animals were not young and healthy.
3. Inadequate number of animals tested (10/sex/group recommended).
4. Only one sex tested.
5. Oral exposure was not minimized (the animals were observed to groom excess test material).
6. Aerosol particle size, MMAD±GSD, was not determined.

Minor deficiencies include:

7. Treatment of the control group was not clearly described.
8. Inadequate description of monitoring/analyses method.
9. Failure to perform all hematology and clinical chemistry analysis.
10. Several organs and tissues not examined microscopically.
11. Some of the data tables were unreadable.

DATA FOR ENTRY INTO ISIS

-Day Inhalation Study - rodents (870.3465)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/L	Doses tested mg/L	NOAEL mg/L	LOAEL mg/L	Target organ(s)	Comments
109701	00096710	subchronic	rat	4 days (continuous)	inhalation	inhalation	0-0.02	0, 0.02	not attained	0.02	body weight decrease*	

The body weight decrease is attributable to the exposure methodology rather than the toxicity of the test material.

Permethrin **Subchronic Oral Toxicity Study (Dog)**

EPA Reviewer: Yung Yang, Ph.D.
Toxicology Branch, HED (7509C)
EPA Secondary Reviewer: Alberto Protzel, Ph.D.
Toxicology Branch, HED (7509C)

Signature: Yung G. Yang
Date 4/4/2002
Signature:
Date 4/4/2002

TXR# 0050649

DATA EVALUATION RECORD

This is an updated executive summary of HED Doc. No. 008163.

STUDY TYPE: Subchronic Oral Toxicity (Capsule) - Dogs OPPTS 870.4100

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352
TEST MATERIAL (PURITY): Permethrin (94.5 %, a.i., cis/trans: 25/75)
SYNONYMS: 3-phenoxybenzyl (±) *cis:trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-1-carboxylate
CITATION: Reynolds, J. et al. (1978) Permethrin Oral Administration to Dogs for 6 months. Wellcome Foundation, Study No. HEFG 78-14, December 1, 1978. MRID 00029832, Unpublished.
SPONSOR: Burroughs Wellcome, Co.

EXECUTIVE SUMMARY: In a subchronic oral toxicity study (MRID 00029832), permethrin (94.5%, a.i., cis/trans 25/75) was administered to beagle dogs (4/sex/group) by gelatin capsule at dose levels of 0, 10, 50, or 250 mg/kg/day for six months.

No treatment-related effects were observed in clinical signs, body weight, food consumption, clinical chemistry, hematology, opthalmoscopy, electrocardiography, plasma antipyrine level, and urinalysis. For females, there is a significant increase in relative liver weight at 50 and 250 mg/kg/day (+22% and +24% compared to the control, respectively). For males, there is a significant increase in relative liver weight at 10 mg/kg/day (+17% compared to the control), but no dose-response relationship was seen (+11% and +9% for 50 and 250 mg/kg/day groups, respectively). Gross necropsy and histopathological examination did not reveal treatment-related effects. A special neuropathology analysis of the dorsal root ganglia, trigeminal ganglion and proximal root, peripheral nerves, spinal cord and brain also did not show evidence of chemical-related pathological effects. It was considered that with only four animals per sex per group, and in the absence of pathological changes in the liver, this study did not demonstrate definite effects on the liver. Therefore, **the NOAEL is established at 250 mg/kg/day (HDT).**

This subchronic dog study is classified **Acceptable/Guideline** and satisfies the guideline requirement for a subchronic toxicity study in dogs.

Table. Summary of the relative liver weight for permethrin treated dogs.

Dose Group	Males			Females		
	Individual weight (g) [1]	Mean ± SD	% [2]	Individual weight (g) [1]	Mean ± SD	% [2]
Control	2.647 2.709 2.833 2.681	2.713 ± 0.084	--	2.910 2.191 2.458 2.639	2.550 ± 0.303	--
10 mg/kg/day	3.843 3.102 2.286 3.253	3.180 ± 0.674	+17%	2.237 2.640 2.905 2.635	2.594 ± 0.336	--
50 mg/kg/day	3.180 3.074 2.679 3.103	3.009 ± 0.138	+11%	3.019 2.935 3.240 3.167	3.090 ± 0.138	+22%
250 mg/kg/day	2.782 3.460 2.689 2.908	2.960 ±0.345	+9%	3.314 3.511 2.988 2.859	3.168 ± 0.298	+24%

[1] Individual liver weights relative to body weight.
[2] % increase over the control group.

M1

DATA EVALUATION RECORD

PERMETHRIN

STUDY TYPE: CARCINOGENICITY - MOUSE
(OPPTS 870.4200b/OECD 451)
MRID 00102110

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37830
Task Order No. 02-04

Primary Reviewer:		Robert H. Ross
Andrew A. Francis, M.S., D.A.B.T.	Signature:	for A.A. Francis
	Date:	DEC 19 2001
Secondary Reviewers:		Sylvia S. Talmage
Sylvia S. Talmage, Ph.D., D.A.B.T.	Signature:	
	Date:	DEC 19 2001
Robert H. Ross, M.S., Group Leader	Signature:	Robert H. Ross
	Date:	DEC 19 2001
Quality Assurance:		L. A. Wilson
Lee Ann Wilson, M.A.	Signature:	
	Date:	DEC 19 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

[PERMETHRIN/109701]

Carcinogenicity Study (mice) (1977)
OPPT 870.4200b/ OECD 451

EPA Reviewer: Yung G. Yang, Ph.D.
Reregistration Branch 2, HED (7509C)
Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 1/8/2002
Signature: Joycelyn Stewart
Date 1/30/2002

TXR# 0050649

DATA EVALUATION RECORD

This is an updated DER and supplement to HED Doc. #007408 and #004204
The NOAEL/LOAEL has been changed.

STUDY TYPE: Carcinogenicity - mice, feeding; OPPTS 870.4200b [§83-2b]; OECD 451.

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): PP557 (Permethrin)(94.0-98.9%)

SYNONYMS: **3-phenoxybenzyl (±) *cis:trans*-3-(2,2-dichlorovinyl)-2,2-dimethyl-cyclopropane-1-carboxylate; FMC 33297**

CITATION: Hart, D. P.B. Banham, I.S. Chart, et al. (1977) PP557: Whole life feeding study in mice. ICI Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire SK10 4TJ, UK. CTL report numbers: CTL/P/358 and CTL/P359, December 28, 1977. MRID 00102110. Unpublished.

Guttmann, E.M. (1990) Permethrin (PP557): Whole life feeding study in mice, phase 3 summary of MRIDs 69703, 69704 and 102110. ICI Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire SK10 4TJ, UK. CTL study number: PM0034, April 27, 1990. MRID 92142032. Unpublished.

SPONSOR: ICI Americas Inc.

EXECUTIVE SUMMARY: In a carcinogenicity study (MRID 00102110, 92142032) PP557 (94.0-98.9 % a.i., batch/lot #'s P24, P34, P35, P36, P44, P52, BX4, and BX6; cis:trans 40:60) was administered to pathogen free Alderley Park mice (70/sex/dose) in the diet at dose levels of 0, 250, 1000, or 2500 ppm (equivalent to 0, 26.9, 110.5, or 287.2 mg/kg/day for males and 0, 29.8, 124.2, or 316.1 mg/kg bw/day for females) for up to 98 weeks. Ten males and females per group were set aside for each of 26- and 52-week interim studies during which necropsies were done and hematology, and clinical chemistry parameters were measured.

No significant compound-related effects on mortality or clinical signs were noted. Transient decreases occurred in body weight gain in high-dose males and high-dose females, but at study termination (98 weeks), the final body weight and body weight gain for male mice in the high-

dose group were reduced by only 5 and 12%, respectively and the final body weight and body weight gain in females in the high-dose group were unaffected. Food consumption was decreased in the high-dose groups relative to controls during the first week of the study, but was increased at most time points thereafter. No treatment-related changes were seen in hematology or clinical chemistry parameters. Increases of 31 to 48% were seen in liver weights and liver weights corrected for body weight in high-dose males and females compared to the controls. Centrilobular hepatocellular eosinophilia was increased in high-dose males and high-dose females at 52 and 98 weeks compared to the controls. Other liver effects included smooth endoplasmic reticulum proliferation, increased nuclear microbodies, and increased aminopyrine-N-demethylase activity in high-dose animals of both sexes compared to the respective controls. Kidney weights were decreased by 21% in high-dose males, but were slightly increased in high-dose females. Proximal tubular epithelium vacuolation was decreased in number and incidence in high-dose males. The HED HIARC evaluated the toxicology database of permethrin and determined that the increased liver weight and other effects observed in the liver are adaptive and reversible effects and are not considered adverse effects.

Under the conditions of this study, the NOAEL for Permethrin is 2500ppm (287.2 mg/kg/day for males and 316.1 mg/kg/day for females). The LOAEL is not established.

At the doses tested, there was no evidence compared to controls of a significant increase in unusual tumor types or in tumor bearing animals. A non-significant increase in lung adenomas in male mice and in lung adenomas plus carcinomas in female mice at the highest dose (2500 ppm in the diet) was not considered evidence of a carcinogenic effect in light of the high incidences in the control groups of both sexes. In addition to the lungs, major organs examined included liver, kidney, testes, ovary, bladder, brain, and thyroid. The dosing based on toxic response was marginal in both males and females. However, the dosing is considered adequate because higher doses would have resulted in a significant weight deficit in male mice.

This carcinogenicity study in mice is classified **Acceptable/Guideline** and satisfies the guideline requirement for a carcinogenicity study [OPPTS 870.4200b; OECD 451] in mice.

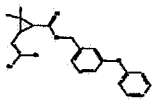
COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements were not provided in the original study (MRID 00102110). The study was completed prior to the implementation of GLP compliance. Statements on current GLP and Data Confidentiality were included in the study review (MRID 92142032).

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test material: PP557

Description:	Technical; nominal isomeric ratio = 40 cis/60 trans
Lot/Batch #:	P24, P34, P35, P36, P44, P52, BX4, and BX6
Purity:	94.0-98.9 % a.i.
Compound Stability:	Stable for 4 weeks
CAS # of TGAI:	52645-53-1
Structure:	



2. Vehicle and/or positive control: Test substance was mixed with food.

3 Test animals:

Species:	Mice	
Strain:	Alderley Park, specific pathogen free	
Age/weight at study initiation:	5-6 weeks/males: 29.5-29.8g; females: 23.2-23.4g (group mean weights)	
Source:	Not supplied	
Housing:	Five mice/galvanized wire mesh cages over trays lined with absorbent paper. Barrier maintained.	
Diet:	77 Parts Stock diet (Oakes Ltd., Congleton, Cheshire, U.K.), 18 parts malt extract, 5 parts corn oil, *ad libitum*	
Water:	Tap water, *ad libitum*	
Environmental conditions:	**Temperature:**	21-24°C
	Humidity:	mean 58%; range 30-80%
	Air changes:	not supplied
	Photoperiod:	12 hrs dark/12 hrs light
Acclimation period:	6-10 days	

B. STUDY DESIGN:

1. In life dates - Start: Nov. 17, 1975; End: ~July 25, 1977

2. Animal Assignment/Dose Levels: Animals were assigned randomly to the test groups noted in Table 1.

TABLE 1: STUDY DESIGN							
Test Group	Concentration in Diet (ppm)	Dose[a] (mg/kg/day)		Main Study 98 weeks		Interim Sacrifice 26 and 52 weeks[b]	
		Male	Female	Male	Female	Male	Female
Control	0	0	0	50	50	20	20
Low (LDT)	250	26.9	29.8	50	50	20	20
Mid (MDT)	1000	110.5	124.2	50	50	20	20
High (HDT)	2500	287.2	316.1	50	50	20	20

[a]Calculated by the reviewer from the food intake and body weight data on pages 434-441 and pages 446-449 of the study document.
[b]Ten mice/dose/sex were designated for interim sacrifices at both 26 and 52 weeks, but animals that died were not replaced from the main study.

3. **Dose selection:** The dose levels were selected based on the results from a previous 28-day study (ICI Central Toxicology Laboratory Report No. CTL/P/356; see Appendix 1). In this study groups of 20 pathogen free Alderly Park mice/sex/dose were fed diets containing 0, 80, 200, 400, 1000, 2000, or 4000 ppm PP557 for 28 days. The 80 ppm concentration was increased to 10,000 ppm for weeks 3 and 4. Decreased final body weights were seen at 10,000 ppm (5%, both sexes), and increased absolute and relative (to body weight) liver weights were seen at 10,000 and 2000 ppm in both sexes compared to the controls (the 4000 ppm group was not examined). Increased centrilobular hepatocellular eosinophilia was seen in all animals at 10,000 ppm and in 2/5 females at 2000 ppm. Dietary concentrations of 250, 1000, and 2500 ppm were chosen for the chronic study in mice.

4. **Diet preparation and analysis**: Diet was prepared every 2 weeks by mixing appropriate amounts of test substance adjusted for the purity of the batch used with the modified stock diet (Oakes Ltd., Congleton, Cheshire, U.K.). The dietary ingredients were mechanically mixed for 10 minutes then extruded into 1 cm x 3-5 cm pellets. The pellets were dried in a vacuum oven at 40°C or less or air dried in open trays. Stability of the dietary mixtures over a 4-week period was confirmed in a previous study [Bradbrook, C., *et al.* (1977) ICI Central Toxicology Laboratory Report No. CTL/P/354]. Homogeneity of the mixtures was apparently not measured directly. Storage conditions were not specified. During the study, samples of treated food were analyzed prior to study initiation and at weeks 31, 65, 77, 80, 87, and 89 for stability and concentration.

   Results - Homogeneity analysis: No data were presented on homogeneity.

   Stability analysis: The test substance was said to be stable over a 4-week period, but the test data were not included in the study.

   Concentration analysis: The range of values for the 250 ppm dietary mixture was 225-295 ppm (mean 251 ppm); the 1000 ppm mixture ranged from 885-1105 ppm (mean 988 ppm); and the 2500 ppm mixture ranged from 2200-2805 ppm (mean 2527 ppm).

The concentration analyses showed a maximum deviation of +18% from the target value (250 ppm) in only one incidence; the remaining tested concentrations were all within ±12% of the nominal value for all the dietary mixtures. Although no data was presented specifically testing the homogeneity of the dietary mixtures, the consistency of the concentration analyses and duplicate sample deviation provided evidence that the diets were adequately mixed. The variance between nominal and actual dosage to the animals was acceptable.

5. Statistics - Mean body weight gains and food consumption were analyzed by analysis of variance and student's t-test. Analysis of variance and analysis of covariance were utilized for organ weights and body weights. Mortality was analyzed by the logrank test. The animals in the interim study groups were included in the calculations up to the weeks the surviving mice were killed as scheduled. The tumor incidences were analyzed by Fisher's exact test and by the logrank test.

   The Reviewer considers the analyses used to be appropriate except that tumor analysis should have been performed on main study animals only.

C. METHODS:

1. **Observations**: Animals were inspected daily for signs of toxicity and mortality.

2. **Body weight:** Animals were weighed at study initiation, weekly for the first 12 weeks, and every 2 weeks for the remainder of the study.

3. **Food consumption and compound intake:** Food consumption for each cage of 5 mice was determined weekly for the first 12 weeks and for weeks 16, 21-24, 33-36, 45-48, 57-60, 69-72 81-84, and 93-96, and mean daily diet consumption was calculated as g food/mouse/week. Food utilization (food consumption in g per unit time/body weight gain in g) was calculated for the first 4 and 12 weeks of the study as time-weighted averages from the consumption and body weight gain data. The compound intake (mg/kg bw/day) was not calculated by the study authors.

4. **Hematology & Clinical chemistry**: Blood was collected by heart puncture from surviving animals in the 26- and 52-week interim groups containing 10 or less mice/group for hematology and clinical chemistry analysis. The period of fasting was not specified in the study. The CHECKED (X) parameters were examined.

a. Hematology

X	Hematocrit (HCT)	X	Leukocyte differential count*
X	Hemoglobin (HGB)		Mean corpuscular HGB (MCH)
X	Leukocyte count (WBC)	X	Mean corpusc. HGB conc.(MCHC)
	Erythrocyte count (RBC)		Mean corpusc. volume (MCV)
X	Platelet count		Reticulocyte count

	Blood clotting measurements		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

*Minimum required for carcinogenicity studies (Control and HDT unless effects are observed) based on Guideline 870.4200 & OECD 451

b. Clinical chemistry*

	ELECTROLYTES		OTHER
	Calcium		Albumin
	Chloride		Creatinine
	Magnesium	X	Urea nitrogen
	Phosphorus		Total Cholesterol
	Potassium		Globulins
	Sodium	X	Glucose
	ENZYMES		Total bilirubin
	Alkaline phosphatase (ALK)		Total protein (TP)
	Cholinesterase (ChE)		Triglycerides
	Creatine phosphokinase		Serum protein electrophoresis
	Lactic acid dehydrogenase (LDH)		
X	Alanine amino-transferase (ALT/also SGPT)		
X	Aspartate amino-transferase (AST/also SGOT)		
	Gamma glutamyl transferase (GGT)		
	Glutamate dehydrogenase		

* Not required for carcinogenicity studies based on Guideline 870.4200 & OECD 451

5. Urinalysis*

Pooled 24-hour urine samples were collected from two cages of male and female mice at 25 and 51 weeks. Fasting was not specified in the study. The CHECKED (X) parameters were examined.

	Appearance	X	Glucose
X	Volume		Ketones
X	Specific gravity	X	Bilirubin
X	pH		Blood/blood cells
	Sediment (microscopic)		Nitrate
X	Protein		Urobilinogen

* Not required for carcinogenicity studies based on Guideline 870.4200 & OECD 451

6. Sacrifice and pathology

All animals that died and those sacrificed on schedule were subjected to gross pathological examination and the CHECKED (X) tissues from all animals were collected and subjected to histological examination. Bone marrow smears were performed only on the 26- and 52-week interim kill animals. Sections of liver and sciatic nerve from 5 mice/sex/group were also examined by electron microscopy (only 3 group-4 females were available). Liver

samples were taken from 3 to 5 mice/sex/group at 26 and 52 weeks for aminopyrine-N-demethylase activity measurements. The (XX) organs from half of the surviving animals at 26 and 52 weeks and from 10 males and 11 females at 98 weeks, in addition, were weighed. The thymus was weighed at 52 weeks only.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue	X	Aorta, thoracic*	XX	Brain (multiple sections)*+
X	Salivary glands*	XX	Heart*+	X	Peripheral nerve*
	Esophagus*	X	Bone marrow*	X	Spinal cord (levels not given)*
X	Stomach*	X	Lymph nodes*	X	Pituitary*
X	Duodenum*	XX	Spleen*+	X	Eyes (retina, optic nerve)*
X	Jejunum*	XX	Thymus		GLANDULAR
X	Ileum*			X	Adrenal gland*+
X	Cecum*		UROGENITAL		Lacrimal gland
X	Colon*	XX	Kidneys*+		Parathyroids*
	Rectum*	X	Urinary bladder*	X	Thyroids*
XX	Liver*+	XX	Testes*+		OTHER
	Gall bladder* (not rat)	X	Epididymides*+		Bone (sternum and/or femur)
	Bile duct* (rat)	X	Prostate*	X	Skeletal muscle
X	Pancreas*	X	Seminal vesicle*		Skin*
	RESPIRATORY	X	Ovaries*+	X	All gross lesions and masses*
	Trachea*	X	Uterus*+		
XX	Lung*++	X	Mammary gland*		
	Nose*				
	Pharynx*				
	Larynx*				

* Required for carcinogenicity studies based on Guideline 870.4200.
+Organ weight required in carcinogenicity studies.
++Organ weight required if inhalation route.

II. RESULTS:

A. OBSERVATIONS:

1. **Clinical signs of toxicity:** No treatment-related clinical signs were reported in the study.

2. Mortality: The percent mortality of each study group at selected times throughout the study is shown in Table 2. The percent mortality was calculated utilizing all 70 mice/dose/sex up to the scheduled interim sacrifices at 26 and 52 weeks. In high-dose males, the percent mortality was consistently higher than the control during the second year of the study, and higher in high-dose females throughout the study. Mortality was also higher than the control in mid-dose females from weeks 32 through 56. These differences, although consistent in high-dose animals, were not statistically significant.

TABLE 2: Group percent mortality[a]				
Gender/Week	0 ppm	250 ppm	1000 ppm	2500 ppm
MALES				
@ 52 Weeks (%)	23.7	20.2	18.0	25.1
@ 76 Weeks (%)	44.9	35.8	38.5	57.5
@ 98 Weeks (%)	74.6	72.8	71.3	79.8
FEMALES				
@ 52 Weeks (%)	9.4	6.7	16.3	17.1
@ 76 Weeks (%)	37.0	32.5	35.7	44.0
@ 98 Weeks (%)	76.4	82.1	76.6	85.5

[a] Data obtained from pages 432-433 in the study report.

B. BODY WEIGHT

Body weights and body weight gains at selected time periods in the study are shown in Table 3. Final body weights for males and females in the high-dose group were 95 and 108% of the respective control values. The group mean body weight gains were slightly lower in high-dose males and females throughout most of the study, but the cumulative differences from the control were statistically significant over only three time periods in males (weeks 0-16, 36, and 92) and once in females (weeks 0-16). The mean overall weight gains at study termination (98 weeks) were lower in high-dose males but higher in high-dose females compared to respective control values (88 and 115% of the control values for males and females, respectively). Therefore, treatment with PP557 was considered to have only a transient effect on body weights for both sexes. Statistical calculations were problematic toward the end of the study since large percentages of some study groups died between weeks 92 and 98. For example, only 7 mice survived to study termination in the female high-dose group.

TABLE 3: Mean bodyweights (BW) and bodyweight gains (BWG)*				
Gender/Week	0 ppm	250 ppm	1000 ppm	2500 ppm
MALES Initial BW [±SD]	29.8 [±2.6]	29.5 [±2.8]	29.6 [±2.8]	29.7 [±2.6]
Final BW [±SD] (% C)	52.0 [±6.3]	50.1 [±4.6]	49.5 [±7.9]	49.3 [±6.2] (95)
BWG Wk 1 (% C)	5.8	6.1 (105)	5.8 (100)	5.6 (97)
BWG Wk 0-16 (%C)	21.6	20.3 (94)	20.2 (94)	19.7 (91)*
BWG Wk 0-92 (% C)	27.4	27.7 (101)	24.5(89)	20.5 (75)*
BWG Wk 16-52 (% C)	9.1	9.9 (109)	9.2 (101)	7.9 (87)
BWG Wk 52-92 (% C)	-3.6	-6.4 (-178)	-6.3 (-175)	-7.0 (-194)
Overall BWG Wk 0-98 (% C)	22.2	20.6	19.9	19.6 (88)
FEMALES Initial BW [±SD]	23.2 [±2.0]	23.4 [±1.9]	23.4 [±1.9]	23.3 [±1.8]
Final BW [±SD] (% C)	47.3 [±8.2]	53.5 [±8.6]	50.3 [±5.1]	51.0 [±12.3] (108)
BWG Wk 1 (% C)	3.8	3.9 (103)	3.9 (103)	3.8 (100)
BWG Wk 0-16 (% C)	21.1	19.5 (92)	19.4 (92)	19.0 (90)*
BWG Wk 0-92 (% C)	27.7	29.8 (108)	29.2 (105)	24.2 (87)
BWG Wk 16-52 (% C)	12.1	16.4 (136)	9.6 (79)	10.9 (90)
BWG Wk 52-92 (% C)	-5.6	-6.1 (109)	-1.8 (32)	-6.4 (114)
Overall BWG Wk 0-98 (% C)	24.1	30.1	26.9	27.7 (115)

% C = percent of control weight.
* Data obtained or calculated from pages 434, 437, 438, 441, and 442-445 in the study report.
* Statistically different ($p < 0.05$) from the control.

C. FOOD CONSUMPTION AND COMPOUND INTAKE:

1. **Food consumption**: Food consumption for high-dose males and high-dose females was slightly lower than that of respective controls during the first week of treatment. Thereafter, food consumption was consistently higher in high- and mid-dose males, and sporadically higher in low-dose males from treatment weeks 2 through12 of the study (Table 4). Statistically significant differences ($p < 0.01$) from the control were seen in 7/12 measurements in high- and mid-dose males and in 3/12 measurements in low-dose males. The significant increases in food consumption in treated males ranged about 4% to 15% higher than the control group. In females, statistical significance ($p < 0.01$) was reached in only 3/12 measurements at the high dose, 4/12 measurements at the mid dose, and 3 at the low dose. The food consumption was decreased about 5% in high-dose females compared to the control during weeks 5 ($p < 0.05$) and 6 ($p < 0.01$). High-dose males and females consumed

equal (±0.1g) or greater weekly amounts of food than the control group at most measurements through weeks 12 - 96 of the study (males: equal or greater food consumption in 21/29 measurements and females in 19/29 measurements). However, the increases in food consumption in high-dose animals compared to the control groups were statistically significant in only two measurements in high-dose males and females from weeks 12 through 96.

TABLE 4: TWA food consumption (g/mouse)[a] and food efficiency[b]

Parameter	0 ppm	250 ppm	1000 ppm	2500 ppm
MALES				
Total food cons. wks 1-12	480.8	492.6	508.7	504.4
Total food cons. wks 13-98	3538.6	3510.4	3542.8	4126.3
Mean daily food cons.	5.86	5.84	5.91	6.01
Food Efficiency wks 1-4[b]	8.27	8.28	8.10	7.91
Food Efficiency wks 1-12[b]	3.75	3.77	3.71	3.56
FEMALES				
Total food cons. wks 1-12	424.2	432.6	438.6	433.5
Total food cons. wks 13-98	3699.2	3734.7	3714.7	3741.1
Mean daily food cons.	6.01	6.07	6.15	6.09
Food efficiency wks 1-4	6.81	7.07	7.10	6.76
Food efficiency wks 1-12	3.75	3.81	3.66	3.57

[a] Calculated from data obtained from pages 446 - 449 in the study report.
[b] Calculated from data obtained from page 450 in the study report.

2. **Compound consumption (time-weighted average):** The compound consumption was not calculated by the study authors; however, the body weight and food consumption data were present enabling the calculation of the time-weighted average compound consumption values by the reviewer. These estimated values are included in Table 1.

3. Food efficiency: The food efficiency over the first 4 and 12 weeks of the study is shown in Table 4. The study authors calculated the food utilization (g food consumed/g bodyweight gained), which can easily be converted to food efficiency (reciprocal of food utilization x 100). The food efficiency over the first 12 weeks of the study was decreased in high-dose males and females by about 5% compared to the control group. The study authors found that the differences in food utilization between the high-dose animals and the controls were not statistically significant.

D. BLOOD ANALYSES:

1. **Hematology**: A slight increase in platelet count was seen in high-dose males compared to the control group at 26 weeks ($p < 0.05$) and at 52 weeks (NS) (Table 5). The platelet count in high-dose females was also slightly increased at 52 weeks, but the increase was not statistically different from the control group. Slight increases in total leucocyte counts were seen in high-dose males ($p < 0.05$) and females (NS) at 52 weeks compared to the control groups. Lymphocytes were also slightly elevated in high-dose males at 52 weeks ($p < 0.05$). Although the leucocyte and lymphocyte counts were elevated in high-dose males compared to the controls, the counts were not higher than the normal range for mice of this age. All other variations seen in hematology parameters were not dose related and were within the normal range.

TABLE 5: Values for selected hematology parameters

Hematology parameter (±95% confidence limits)	0 ppm	250 ppm	1000 ppm	2500 ppm
MALES				
Total Leucocytes, 52 wks. ($x10^9$/L) (±1.0)	5.8	4.2	3.8	7.7*
Lymphocytes, 52 wks. ($x10^9$/L) (±1.0)	4.9	3.4	2.7	6.3*
Platelets, 26 wks. ($x10^9$/L) (±15.0)	134.0	138.0	138.0	164.0*
Platelets, 52 wks. ($x10^9$/L) (±24.0)	163.0	162.0	176.0	192.0
FEMALES				
Total Leucocytes, 52 wks. ($x10^9$/L) (±1.0)	6.0	4.9	4.0*	7.1
Lymphocytes ($x10^9$/L) (±1.9)	4.7	3.9	2.8	5.6
Platelets, 26 wks. ($x10^9$/L) (±14.0)	132.0	123.0	134.0	134.0
Platelets, 52 wks. ($x10^9$/L) (±23.0)	144.0	144.0	155.0	161.0

* Data obtained from pages 367 - 371 in the study report.
* Statistically different ($p < 0.05$) from the control.

2. **Clinical chemistry**: There were no changes in clinical chemistry parameters attributable to treatment. Although there were some sporadic values among treated groups that were significantly different from the control group, the differences were not dose-related and were within the normal range.

3. **Urinalysis**: There were no treatment-related changes seen in urinalysis.

E. SACRIFICE AND PATHOLOGY:

1. **Organ weight**: Selected group mean organ weights and organ weights adjusted for body weights are shown in Table 6. The group mean liver weights in high-dose males at 26 and 52 weeks were increased by about 39% ($p < 0.01$) and 41% ($p < 0.05$), respectively, compared to

the control values after adjusting the numbers for body weights. After 98 weeks of treatment, the liver weights of males were not significantly affected compared to the control group. The mean kidney weights in males were significantly ($p < 0.01$) decreased by 18% to 21% in all treated groups compared to the control group. After adjusting for body weights, the differences between the treated and control groups were less, but still significant ($p < 0.01$) especially for the mid- and high-dose groups (decreases of 18% and 17%, respectively). Group mean brain weights were slightly (8%, $p < 0.01$) decreased in high-dose males and slightly (+11%, $p < 0.01$) increased in high-dose females compared to the control groups. The brain weights remained decreased by 7% and increased by 11% ($p < 0.01$) in high-dose males and females, respectively, after adjusting the values to body weights.

Group mean liver weights in high-dose females were increased by 32%, 28%, ($p < 0.05$) and 39% ($p < 0.01$) after 26, 52, and 98 weeks of treatment, respectively, compared to the control group. The liver weights of females after adjusting for body weights were increased by 27% ($p < 0.05$), 31%, and 38% ($p < 0.01$) at the same observation times in high-dose animals. The group mean heart weight in high-dose females both before and after adjusting for body weights was increased by 28% to 29% ($p < 0.05$) compared to the control group. Kidney weights in high-dose females were slightly increased compared to the controls, but the changes were not statistically significant.

Additional organ weight changes were recorded that were statistically significant, but were not included in Table 6 because of the lack of any dose relationship or wide individual variations. Wide variations were seen in lung and spleen weights, which made statistical comparisons difficult and misleading. After omitting two outlying values in low-dose females and one in high-dose females, a trend toward increased lung weight was seen in high-dose females. The unadjusted spleen weights were decreased in males at both the mid and high doses and were increased in mid- and high-dose females. However, omitting outlying spleen weight values resulted in significantly decreased spleen weight only in the mid-dose males (~ -49%, $p < 0.05$).

TABLE 6: Selected group mean organ weights at 26, 52, and 98 weeks [a]				
Organ, weeks., adjustment (±95% confidence limit)	0 ppm	250 ppm	1000 ppm	2500 ppm
MALES				
Liver, 26 wks. (±0.80)	2.60	2.53	3.05	3.57
Liver, 26 wks., adjusted for body wt. (±0.33)	2.62	2.50	2.98	3.64**
Liver, 52 wks., (±0.66)	2.51	3.58*	2.93	3.72*
Liver, 52 wks., adjusted for body wt. (±0.61)	2.61	3.41	3.05	3.67*
Liver, 98 wks. (±1.40)	3.41	3.92	2.89	3.31
Liver, 98 wks. adjusted for body wt. (±1.39)	3.27	3.97	2.84	3.44
Kidney, 52 wks. (±0.196)	0.840	0.920	0.856	0.820
Kidney, 52 wks., adjusted for body wt. (±0.194)	0.864	0.879	0.884	0.808
Kidney, 98 wks. (±0.103)	1.101	0.908**	0.893**	0.874**
Kidney, 98 wks., adjusted for body wt. (±0.089)	1.077	0.916*	0.884**	0.898**
Heart, 98 wks., adjusted for body wt. (±0.031)	0.294	0.271	0.277	0.284
Brain, 98 wks., (±0.016)	0.445	0.441	0.417*	0.410**
Brain, 98 wks., adjusted for body wt. (0.016)	0.443	0.442	0.416*	0.412**
FEMALES				
Liver, 26 wks. (±0.50)	2.37	2.82	2.43	3.13*
Liver, 26 wks., adjusted for body wt. (±0.34)	2.45	2.53	2.58	3.12*
Liver, 52 wks., (±0.37)	2.50	2.60	3.23*	3.21*
Liver, 52 wks., adjusted for body wt. (±0.30)	2.49	2.60	3.20**	3.25**
Liver, 98 wks. (±0.49)	2.69	3.29	3.15	3.74**
Liver, 98 wks., adjusted for body wt. (±0.48)	2.73	3.28	3.10	3.78**
Kidney, 52 wks. (±0.068)	0.554	0.582	0.731**	0.612
Kidney, 52 wks., adjusted for body wt. (±0.070)	0.554	0.582	0.729**	0.615
Kidney, 98 wks., adj. for body wt. (±0.113)	0.731	0.738	0.710	0.861
Heart, 98 wks. (±0.035)	0.247	0.296*	0.270	0.318**
Heart, 98 wks., adjusted for body wt. (±0.033)	0.251	0.295	0.265	0.321**
Brain, 98 wks., (±0.025)	0.428	0.443	0.441	0.476**
Brain, 98 wks., adjusted for body wt. (0.023)	0.431	0.442	0.437	0.479**

[a] Data obtained from pages 384-390 and 451 - 455 in the study report.
* Statistically different ($p < 0.05$) from the control.
** Statisitcally different ($p < 0.01$) from the control.

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2. **Gross pathology**: No treatment-related findings were reported on gross necropsy.

3. **Microscopic pathology**:

a. **Non-neoplastic**: Microscopic findings in liver and kidney and aminopyrine-N-demethylase (APDM) activity in liver tissue are summarized in Table 7. The most consistent non-neoplastic effects of PP557 treatment were seen in the liver of both sexes. Eosinophilia of the centrilobular hepatocytes was not seen in the controls or low-dose groups of either sex, but was seen in 40% to 70 % of all high-dose animals tested at 52 or 98 weeks. The liver APDM activity was increased in mid- and high-dose males at 26 weeks by 108% and 247% ($p < 0.01$), respectively, compared to the control group, and the increases appeared to be dose-related. At 52 weeks, the APDM activity was slightly increased in all treated male groups, but the enzyme activity in the control group was also higher and the differences were not dose-related or statistically significant. The liver APDM activity in high-dose females was slightly higher than the control, but the difference was not statistically significant. The examination of liver cells using the electron microscope revealed smooth endoplasmic reticulum proliferation in both sexes at the high dose after 26 weeks of treatment, and a marked increase in nuclear microbodies in high-dose animals after 52 weeks. The electron microscope examinations were done in only 1 to 5 animals making statistical comparisons difficult; however, the results are consistent with increased metabolic activity in the livers of treated animals at the high dose, and with liver hypertrophy in response to chemical treatment.

Decreased incidences and amounts of kidney proximal tubular epithelium vacuolation were seen in all treated males at 26 weeks and in high-dose males at 98 weeks compared to the controls. Tubular epithelium vacuolation was seen in 100% to 50% of control males at 26 and 52 weeks, respectively, but was not seen in high-dose males or in control or treated female mice.

The microscopic changes observed in the liver and kidney in high-dose animals supported the observed organ weight differences as treatment-related observations. No other microscopic treatment-related findings were reported.

TABLE 7: Microscopic findings at 52 and 98 weeks [a]				
Organ, time period, finding	0 ppm	250 ppm	1000 ppm	2500 ppm
MALES				
Liver, 52 wks., centrilobular hepatocellular eosinophilia	0/9[b]	0/7	0/8	6/10++
Liver, 98 wks., centrilobular hepatocellular eosinophilia	0/12	0/14	3/14	7/10++
Liver, 26 wks., SER[c] proliferation	0/4	not done	1/4	2/2
Liver, 52 wks., increased nuclear microbodies	0/5	not done	0/4	4/5+
Liver, 26 wks., APDM[d] activity (µM aminophenazone/g/hr) (±0.057)[e]	0.105	0.086	0.218**	0.364**
Liver, 52 wks., APDM[c] activity (µM aminophenazone/g/hr) (±0.225)	0.329	0.561	0.451	0.522
Kidney, 52 wks., proximal tubular epithelium vacuolation graded moderate or marked	9/9	2/7++	2/8++	0/10++
Kidney, 98 wks., proximal tubular epithelium vacuolation graded moderate or marked	6/12	7/14	7/14	0/10+
FEMALES				
Liver, 52 wks., centrilobular hepatocellular eosinophilia	0/9	0/9	2/7	4/10
Liver, 98 wks., centrilobular hepatocellular eosinophilia	0/12	0/14	1/13	4/7++
Liver, 26 wks., SER[b] proliferation	0/4	not done	1/4	3/4
Liver, 52 wks., increased nuclear microbodies	0/4	0/5	0/3	4/5+
Liver, 26 wks., APDM[c] activity (µM aminophenazone/g/hr) (±0.067)	0.192	0.197	0.178	0.219
Liver, 52 wks., APDM[c] activity (µM aminophenazone/g/hr) (±0.028)	0.060	0.075	0.075	0.091
Kidney, 52 wks., proximal tubular epithelium vacuolation, any grade of severity	0/9	0/9	0/7	0/10
Kidney, 98 wks., proximal tubular epithelium vacuolation, any grade of severity	0/12	0/9	0/13	0/7

[a] Data obtained from pages 383, 392, 398, 463, and 464 in the study report.
[b] Incidence of finding/number of animals examined
[c] SER (smooth endoplasmic reticulum)
[d] APDM (aminopyrine-N-demethylase)
[e] ±95% confidence limit
** Statistically different ($p < 0.01$) from the control; t-test by the study authors.
+ Statistically different ($p < 0.05$) from the control; Fisher's exact test by the reviewer.
++ Statistically different ($p < 0.01$) from the control; Fisher's exact test by the reviewer.

b. Neoplastic: The study authors compared neoplastic findings among groups based on total animals (70/sex) rather than on main study animals (50/sex). Utilizing the Fisher's exact test, the incidences of pulmonary adenomas in high-dose males were not significantly different at the $p< 0.05$ level from the control group at any time point examined in the study. The study authors found that the incidence of lung adenomas in high-dose males compared to the control group was statistically significant using the logrank test, which takes into account the survival rates. The incidence of pulmonary adenomas was slightly increased in high-dose females, especially in animals that died during weeks 79 to 97 (9/20), compared to the control group (4/19). The lung adenoma incidences in high-dose females compared to that of the control group were not significantly different at any time period examined or for the entire study. Total tumor-bearing males in the control through high-dose groups were 15, 16, 28, and 30. For females respective values were 44, 41, 39, and 33. Based on total animals (70/sex), the study authors concluded that PP557 had no effect on the incidence of tumor-bearing animals.

The reviewer evaluated lung tumor incidences of main study animals, omitting the data from the 26- and 52-week interim sacrifices (Table 8). Incidences of total tumors (adenomas) in the control and high-dose males were 10/36 and 17/37 ($p=0.09$, Fisher's exact test). For females, incidences of lung adenomas in the control and high-dose groups were 11/46 and 15/40 ($p=0.13$, Fisher's exact test). Adenocarcinomas were also reported for females. Total incidences of adenomas plus adenocarcinomas in the control and high-dose females were 11/46 and 16/40 ($p=0.09$, Fisher's exact test). The differences between the control and high-dose groups for both sexes are non-significant, but border on being marginally statistically significant. One lung adenoma was reported in control males up to 52 weeks (incidence of 1/34) and two adenomas were reported in low-dose females up to 52 weeks (incidence of 2/23). It could not be ascertained whether the adenomas were present at the interim sacrifice or were present in main-study females that died.

TABLE 8: Neoplastic findings in the lungs -main study animals[a]				
Organ: treatment period	**0 ppm**	**250 ppm**	**1000 ppm**	**2500 ppm**
MALES				
Lung adenoma: 53-78 wks.	1/11[b]	0/8	1/12	5/17
Lung adenoma: 79 -97 wks.	4/13	3/19	7/14	7/10
Lung adenoma: @ 98 wks.	5/12	3/14	5/14	5/10
Lung adenoma: total	10/36	6/41	13/40	17/37
FEMALES				
Lung adenoma: 53-78 wks.	5/15	0/16	2/14	2/13
Lung adenoma: 79 -97 wks.	4/19	4/22	5/16	9/20
Lung adenoma: @ 98 wks.	2/12	2/9	3/13	4/7
Lung adenocarcinoma: total	0	1	1	1
Lung adenomas plus adenocarcinoma: total	11/46	7/47	11/43	16/40

[a] Data obtained from pages 474- 486 in the study report.
[b] Number of mice with tumor/number of mice examined.

III. DISCUSSION and CONCLUSIONS

A. INVESTIGATORS' CONCLUSIONS: The investigators concluded that a toxic effect was obtained in this study based on decreased body weight gain in high-dose animals compared to the control groups. The increased liver aminopyrine-N-demethylase activity and the microscopic liver effects including increased hepatocellular eosinophilia, smooth endoplasmic reticulum proliferation and increased nuclear microbodies seen in high-dose animals compared to the control were adaptive processes by the liver in response to the chemical agent and were not adverse effects. The decrease in kidney proximal tubular epithelium vacuolation seen in high-dose males was also considered to be an adaptive response to treatment. The toxicological no effect level was 1000 ppm in both sexes.

B. REVIEWER COMMENTS: The percent mortality was slightly higher in high-dose animals compared to the controls after 52, 76, and 98 weeks of treatment, but the differences were not found to be statistically significant. The body weight and body weight gain were slightly less in high-dose animals than in the control groups especially during the first year of the study. The differences were only sporadically significant statistically, and the final group mean body weight of high-dose females at week 98 was slightly higher than that of the control group. However, the decreased body weight gain in high-dose animals occurred with an increase in food consumption in both sexes and a slight decrease in food efficiency

measured over the first 12 weeks of the study, which is consistent with a toxic effect of PP557 at the high dose.

Increases in total leucocyte, lymphocyte and platelet counts were seen in high-dose animals compared to the control groups. The increases were statistically significant in males, but not in females. The changes in hematological parameters are of doubtful toxicological significance since the leucocyte counts were within the normal range, and only the platelet counts appeared to be dose-dependent. There were also no changes seen in bone marrow.

Group mean absolute liver weights and liver weights adjusted for body weight were increased in high-dose animals of both sexes at 26 weeks and 52 weeks and in high-dose females at 98 weeks. The absolute and adjusted liver weights were also increased in mid-dose females at 52 weeks. Microscopic examination revealed increased centrilobular hepatocellular eosinophilia in high-dose animals at 52 and 98 weeks compared to the control groups. Increased smooth endoplasmic reticulum and increased nuclear microbodies were seen in high-dose animals with electron microscopy. Liver amino-N-pyrine demethylase activity was increased in high-dose males and females at 26 and 52 weeks, and in mid-dose males at 26 weeks. These observations are indicative of increased metabolic activity in the liver as a response to the chemical challenge. No treatment-related changes were seen in plasma alanine transaminase or plasma aspartate transaminase. On April 18, 2002, the HED HIARC evaluated the toxicology database of permethrin and determined that the increased liver weight and other effects observed in the liver are adaptive and reversible effects and are not considered adverse effects.

Absolute kidney weights and kidney weights adjusted for body weight were significantly decreased in mid- and high-dose males at 98 weeks and increased in mid- and high-dose females at 52 weeks. The kidney weight changes were not dose-related in females or in males when adjusted for body weight. A decrease in proximal tubular epithelium vacuolation was seen at all dose levels in males compared to the control group after 26 weeks of treatment and in high-dose males after 98 weeks of treatment. Vacuolation of the tubule epithelium was not seen in the female control or any female dose group. The kidney effects appear to be adaptive. No changes were seen in treated animals in urinalysis or in clinical chemistry measurements that would indicate an adverse kidney effect.

Other organ weight changes seen in the study that were statistically significant, especially the heart and brain weight changes, were not supported by any physiological or microscopic findings and are probably not toxicologically significant.

Under the conditions of this study, a NOAEL for Permethrin is 2500ppm (287.2 mg/kg/day for males and 316.1 mg/kg/day for females). The LOAEL is not established.

Although the study authors found statistically increased lung adenomas in male mice using the logrank test (11/70, 6/70, 13/70, and 17/70 for groups 1, 2, 3, and 4, respectively; $p<0.05$), the statistical significance was only marginal ($p=0.09$) when only the main study

animals were considered and when the Fisher test was applied. The study authors also found a slight increase in the incidence of lung adenomas in high-dose females compared to the controls, but the increase was not statistically significant (11/70, 8/70, 10/70, and 15/70 for groups 1, 2, 3, and 4, respectively). Using only the main study animals and applying the Fisher exact test, the incidences of adenomas and adenomas plus carcinomas were not statistically significant in high-dose female mice ($p=0.13$ and $p=0.09$, respectively). One lung adenocarcinoma was found in each of the female treated groups; none were seen in males. There was no evidence of an increase in the progression of pulmonary adenoma to carcinoma with PP557 treatment, and no adverse non-neoplastic treatment-related pulmonary effects were seen. The reviewer agrees with the study authors that this evidence is insufficient to establish PP557 as a carcinogen. Historical control data on lung adenomas in this strain of mice might yield a more definitive conclusion on this subject.

Based on the absence of systemic effects, it is questionable that dosing was adequate in this study. Higher food consumption and lower body weights early in the study for both males and females indicates a toxic action of the test material. Although the final body weight and body weight gain in males in the high-dose group were decreased by only 5 and 12%, respectively, at 98 weeks compared with the control values (females were unaffected), these decreases coupled with the nonsignificant increases in mortality for both sexes at the 72-week observation indicate that the high-dose was a threshold for more serious effects.

C. STUDY DEFICIENCIES:

Several pages of the study including summary tables were unreadable.

There were some deviations from the guidelines that are relatively minor, but should be noted:

1. Hematology parameters were measured at 26 and 52 weeks instead of 18 months and at study termination. In some cases, fewer than 10 animals per group were available. The measurements that were done, however, included clinical chemistries in excess of guideline requirements.
2. Bone marrow was examined at 52 weeks instead of at study termination and less than 10 animals per group were tested.
3. According to MRID 92142032, tissues not specifically listed under histological evaluations were thought to be examined with accompanying organs, for example: parathyroids, trachea, gall bladder, skin.
4. Homogeneity of the dietary mixture was not specifically measured, but duplicate samples and concentration measurements of the dietary mixtures were within acceptable limits.
5. Several different batches of the test material were used throughout the study.
6. The study authors based their carcinogenicity evaluation on tumors in the total number of animals instead of main study animals. However, data were provided to enable the reviewer to evaluate carcinogenicity on the basis of main study animals.

The following problems are more serious, but are not likely to result in any change in the outcome of the study:

1. Some determinations were made using too few animals for meaningful statistics. Animals designated for interim studies were not replaced from the main study if they died, therefore, only 7 low-dose males and 7 mid-dose females were available at 52 weeks. Organ weights at 52 weeks were done on as few as 2 animals/group; liver weights were done on 3-5 animals/group; APDM activity was measured on 3-5 animals/group and the electron microscopic studies were done on 2-5 animals/group.
2. An inadvertent mix up of the diets resulted in group 2 females being fed the high-dose diet and vice versa for weeks 24-28. Since no irreversible effects were seen at the high dose, any effects on the group-2 females were likely reversed quickly resulting in no changes in the study outcome.
3. There was no overall summary table for non-neoplastic findings that included the animals that died or were killed at unscheduled times during the study. Summary tables for tumor incidences for animals that comprised the main study were not provided (or were unreadable).
4. Historical data that show the normal ranges of lung adenomas in Alderley Park mice should be provided.

DATA FOR ENTRY INTO ISIS

Carcinogenicity Study - mice (870.4200b)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
109701	00102110, 92142032	carcinogenicity	mice	98 weeks	oral	diet	27-316	male:27, 111, 287; female: 30, 124, 316	males: 287; females: 316	Not established		

APPENDIX 1

PP557: 28 DAY FEEDING STUDY IN MICE

MRID Not provided

DOSE SELECTION STUDY IN MICE

MRID No: Not provided
Study type: 28-day dose range finding study for a lifetime feeding study in mice
Test material: PP557 (permethrin) (94.7%, batch no. 24)
Report no: CTL/P/356
Submitted by: Imperial Chemical Industries Limited
Sponsored by:
Testing facility: Central Toxicology Laboratory, Imperial Chemical Industries Limited
Citation: Clapp, M., et al. (1977) PP557: 28 day feeding study in mice. Central Toxicology Laboratory, Imperial Chemical Industries Limited. Dec. 19, 1977. Unpublished.

METHODS:

Test animals: Specific pathogen free Alderley Park mice, 5-6 weeks old, weighing: males, 28.10-29.40 g; females, 25.3-25.9 g at study initiation.
Housing: 5 mice/wire mesh cage over collection tray; barrier maintained; temperature: 22°C ±1°C; about 10 air changes/hour; 12 hours light, 12 hours light.
Group size: 20 males, 20 females
Test concentrations: 0, 80, 200, 400, 1000, 2000, 4000 ppm in the diet. After 2 weeks, the 80 ppm concentration was increased to 10,000 ppm for the remainder of the study.
Substance intake:0; 8 (1st 2 weeks), 1000 (2nd 2 weeks); 20; 40; 100; 200; 400 mg/kg/day (calculated assuming a 50 g mouse eats 5 g food/day)
Duration: 28 days

RESULTS:

Clinical signs: No clinical signs were observed.
Mortality: No animals died during the study.
Body weight: The body weight gain was decreased by about 5% in males and 13% and 6% in females compared to the control group in the 2 weeks after the 80 ppm concentration was increased to 10,000 ppm.
Food consumption: The total food consumption per cage was increased 9-15% in all male treated groups and 4-10% in females at 80/10,000, 100, 200, and 400 ppm compared to the controls. Food efficiency was decreased from 7.25 and 5.92 in the control to 4.65 and 4.45 in the 80/10,000 ppm group for males and females, respectively.
Clinical pathology: Hematology parameters were not measured.
Organ weights: Absolute liver weights increased by about 18% ($p < 0.05$) and 19% in males and by 25% and 6% in females at 2000 ppm and 80/10,000 ppm, respectively (organs of the 4000 ppm groups were not weighed). The liver weights relative to body weights increased by about 19% and 31% ($p < 0.01$) in males and by 20% and 9% ($p < 0.05$) in females at 2000 ppm and 80/10,000 ppm, respectively. The absolute and relative kidney weights were slightly decreased in males and increased in females at 2000 ppm.

Histopathology: Increased incidences of eosinophilia of centrilobular hepatocytes were seen in the livers of males and females at 80/10,000 ppm (males: control, 0/5; 80/10,000 ppm, 5/5; females: control, 1/5; 80/10,000 ppm, 5/5). An increased incidence of regenerating kidney tubule epithelium was seen in males at 80/10,000 ppm (control, 0/5; 80/10,000, 2/5).

CONCLUSIONS:

The recommended dietary concentrations for a lifetime study in mice were 250, 1000, and 2500 ppm. The lowest-observed-adverse-effect-level (LOAEL) in this study was 2000 ppm based on increased liver weights and microscopic changes. The no-observed-adverse-effect-level (NOAEL) was 1000 ppm.

This study is classified as **Acceptable/Nonguideline** and was used as a dose range-finding study.

EPA Reviewer: Yung G. Yang, Ph.D.
Reregistration Branch 2, Health Effects Division (7509C)
Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, Health Effects Division (7509C)

Signature: Yung G. Yang
Date 2/6/2002
Signature: Joycelyn Stewart
Date 2/6/2002

TXR#: 0050649

DATA EVALUATION RECORD

This is an updated DER for HED Doc. #001437 and #004204. The final conclusion has not been changed.

STUDY TYPE: Carcinogenicity - mice, feeding OPPTS 870.4200b [§83-2b]; OECD 451.

PC CODE: 109701 **DP BARCODE:** D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): FMC 33297 (Permethrin)(technical grade, purity not specified)

SYNONYMS: 3-phenoxybenzyl (±) *cis:trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-1-carboxylate; PP557

CITATION: Ellison, T. (1979) Analysis of physical observations, twenty-four month oral carcinogenicity study of FMC 33297 in mice. Bio/dynamics, Inc., Mettlers Road, East Millstone, New Jersey 08873. Bio/dynamics Study Number: 76-1695, FMC Study Number: ACT 115.35, October 9, 1979. MRID 00062806. Unpublished

Nye, D. (1990) Phase 3 summary of MRID: 61901, 62806. Twenty-four month carcinogenicity study with FMC 33297 in mice. Study numbers ACT 115.35 (FMC) and 76-1695 (Bio/dynamics). Summary by FMC Corporation, March 13, 1990. MRID 92142033. Unpublished

No author (1977) Appendix L - A twenty-four month oral carcinogenicity study of FMC 33297 in mice, Bio/Dynamics Laboratory, November 30, 1977, Fat Analyses for parent FMC 33297. MRID 00066528. Unpublished.

SPONSOR: FMC Corporation, Toxicology Division, P.O. Box 8, Princeton, New Jersey 08540

EXECUTIVE SUMMARY: In a carcinogenicity study (MRID 00062806, 92142033) FMC 33297 (permethrin, % a.i. not specified, Lot #s MR176 and MR807) was administered to Charles River CD-1 mice (75/sex/dose) in the diet at dose levels of 0, 20, 500, or 2000 ppm for males (equivalent to 0, 3, 71, or 286 mg/kg/day, respectively) and 0, 20, 2500, or 5000 ppm for females (equivalent to 0, 3, 357, or 714 mg/kg/day, respectively) for 24 months.

Mortality was significantly increased in high-dose males after 75 weeks of treatment, but was not significantly different from the control group after 104 weeks. Clinical signs consisting of distended abdomens, ano-genital staining, and alopecia were increased in treated males compared to the control during the first year of treatment, but were not dose-related at 24 months. Insufficient data were provided on body weights (with the exception of final body weights for females), body weight gains, organ weights (with the exception of brain weights of females at study termination), hematology parameters, and gross and microscopic changes for the reviewer to evaluate. An 8% increase in final female body weight was not considered a biologically significant effect. Although difficult to evaluate in the absence of summary data, the effects listed by the study author - transient increased body weights, decreased leucocyte counts and liver and kidney inflammatory changes - do not appear to be toxicologically significant.

A NOAEL and LOAEL for FMC 33297 (permethrin) in mice could not be determined in this study due to major study deficiencies including failure to include summaries of numbers of animals with clinical signs and data on body weights, body weight gains, organ weights, hematology parameters, and gross and microscopic necropsy findings.

A joint FDA-EPA audit of this study conducted in late 1980 at Bio/Dynamics and FMC facilities did not reveal any inadequacies in the conduct or reporting of this study serious enough to compromise the usefulness of these study results for oncogenic evaluation. However, the audit concluded that this study was not useful for assessment of chronic toxicity (HED Doc. #004204).

On December 12, 1988 the HED Cancer Peer Review Committee reviewed the study and concluded that there were statistically significant increases in liver adenoma at all doses for males and at mid- and high-doses for females with a significant dose-related trend in both sexes. Combined liver adenoma/carcinoma also showed statistically significant increases at mid- and high-doses for male and female mice. Statistically significant increases in lung adenomas and combined adenoma/carcinoma at all doses were observed in females only. Carcinoma were increased at all doses but only at HDT that the increase was statistically significant. The incidences of adenoma and carcinoma at mid- and high-doses were outside historical control ranges. There were also significant dose-related trends for lung adenomas, carcinomas and combined adenoma/carcinomas in females. The incidences of lung tumors in male mice (adenoma or carcinoma, or combined) were not statistically significant at any dose, nor was there a dose-related trend for any of them.

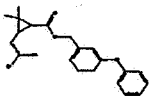
This carcinogenicity study in mice is classified as **Acceptable/Guideline (OPPT 870.4200b; §832b)** for evaluation of carcinogenicity. However, this study may not be used for regulatory purpose on assessment of chronic toxicity.

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements were not provided in the original study (MRID 00062806). The study was completed prior to the implementation of GLP compliance. Statements on current GLP and Data Confidentiality were included in the study review (MRID 92142033).

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: FMC 33297

Description:	Permethrin Technical
Lot/Batch #:	MR176 and MR807
Purity:	not specified
Compound Stability:	not specified
CAS # of TGAI:	52645-53-1
Structure	

2. Vehicle and/or positive control: Test substance was mixed with food.

3 Test animals:

Species:	Mice	
Strain:	CD-1 (COBS)	
Age/weight at study initiation:	Not specified	
Source:	Charles River	
Housing:	Mice were housed individually in stainless steel hanging cages in racks containing 60 cages per side.	
Diet:	Purina Laboratory Chow, *ad libitum*	
Water:	Tap water, *ad libitum*	
Environmental conditions:	**Temperature:**	Not specified
	Humidity:	Not specified
	Air changes:	Not specified
	Photoperiod:	Not specified
Acclimation period:	Not specified	

B. STUDY DESIGN:

1. **In life dates** - Start: Dec. 3, 1976 End: Dec. 10, 1978

2. **Animal Assignment/Dose Levels:** Animals were assigned randomly to the test groups noted in Table 1.

TABLE 1: STUDY DESIGN

Test Group	Concentration in Diet (ppm)		Dose[a] mg/kg/day		Main Study 98 weeks	
	Male	Female	Male	Female	Male	Female
Control	0	0	0	0	75	75
Low (LDT)	100/20[b]	100/20[b]	15/3	15/3	75	75
Mid (MDT)	2500/500[c]	2500	375/71	357	75	75
High (HDT)	5000/2000[d]	5000	286	714	75	75

Data taken from p. 6, MRID 00062806.
[a] Calculated by the reviewer assuming a 20 g mouse consumes 3 g food/day.
[b] Low-dose male and female mice were fed diets containing 100 ppm FMC 33297 for the first 33 days; the concentration was reduced to 20 ppm for the remainder of the study.
[c] Mid-dose male mice were fed diets containing 2500 ppm FMC 33297 for the first 33 days; the concentration was reduced to 500 ppm for the remainder of the study.
[d] High-dose male mice were fed diets containing 5000 ppm FMC 33297 for the first 33 days; the concentration was reduced to 2000 ppm for the remainder of the study.

3. **Dose Selection:** The rationale for dose level selection in this study was not included in the report.

4. **Diet preparation and analysis:** Diet preparation procedures were not included in the report. Stability of the dietary mixtures was measured after storage for 42-45 months at 0°C. Analyses for homogeneity and concentration of the dietary mixtures were carried out during the study. Dietary samples were taken during treatment weeks 1, 7, 10, 12, 30, 41, 43, 45, 56, 69, 81, 95, and 105 for analysis of FMC 33297 concentration.

Results - Homogeneity Analysis: No data were presented on homogeneity.

Stability Analysis: The data presented showed an overall average decrease in concentration of about 5% over 42 to 45 months storage at 0°F; however, the differences ranged from +1% in a 500 ppm sample to -12% in a 100 ppm and 2000 ppm sample.

Concentration Analysis: The range of values for FMC 33297 dietary concentrations generally ran low, but were usually within 85% of the target concentration. However, deviations did occur including those greater than 20% (100 ppm, week 1; 2000 ppm, week 44) and greater than 15% (2000 ppm, week 95). These deviations are understandable, but the 500 ppm sample from week 43 tested at about 10% of the target value and the 20 ppm sample from week 44 tested about 600% of the target value. The possible reasons for these aberrant values were investigated, but were apparently never resolved. There were also samples of the 100 ppm diet for treatment week 7 analyzed for FMC 22397 content, but the study protocol stated that the 100 ppm diet was replaced with the 20 ppm diet after week 4.

Although no data was presented specifically testing the homogeneity of the dietary mixtures, the consistency of the concentration analyses provided evidence that the diets were

adequately mixed. The variance between nominal and actual dosage to the animals was not acceptable in several cases, and with the data incomplete and scattered among numerous laboratory reports, the acceptability of the diet preparation procedures is questionable.

5. **Statistics** - The neoplastic findings were analyzed using the Chi-squared test and the Armitage-Cochran and Yates corrected Chi-squared test (obtained from the study summary, MRID 92142033, pp. 7-8). Any additional statistical procedures used in the study were not included in the document.

C. METHODS:

1. **Observations** - Animals were inspected daily for signs of toxicity and mortality for the first 47 days and twice daily for the remainder of the study. Each animal was given a detailed physical examination once each week throughout the study.

2. **Body weight** - Weighing intervals were not given in the document.

3. **Food consumption and compound intake** - Food consumption measurement procedures were not given in the document.

4. **Hematology & Clinical Chemistry** - Blood was collected from surviving animals at study termination for hematology measurements The time period of fasting and the method of blood collection were not specified in the study. The CHECKED (X) parameters were examined.

a. Hematology

	Hematocrit (HCT)	X	Leukocyte differential count*
	Hemoglobin (HGB)		Mean corpuscular HGB (MCH)
X	Leukocyte count (WBC)		Mean corpuscular HGB conc.(MCHC)
	Erythrocyte count (RBC)		Mean corpuscular volume (MCV)
	Platelet count		Reticulocyte count
	Blood clotting measurements	X	Erythrocyte morphology
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Minimum required for carcinogenicity studies (Cont. and HDT unless effects are observed) based on Guideline 870.4200 & OECD 451

b. Clinical Chemistry*Clinical chemistry parameters were not measured.
*Clinical chemistry studies are not required for carcinogenicity studies based on Guideline 870.4200b & OECD 451.

5. **Sacrifice and Pathology**: All animals that died and those sacrificed on schedule were subjected to individual necropsy examinations; the CHECKED (X) tissues were collected

and examined microscopically. With the exception of brain and liver weights in females, organs that were weighed (XX) were not discussed in the document.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue		Aorta, thoracic*	XX	Brain (multiple sections)*+
X	Salivary glands*	X	Heart*+	X	Peripheral nerve*
X	Esophagus*	X	Bone marrow*	X	Spinal cord (3 levels)*
X	Stomach*	X	Lymph nodes*	X	Pituitary*
X	Duodenum*	X	Spleen*+	X	Eyes (retina, optic nerve)*
X	Jejunum*	X	Thymus		GLANDULAR
X	Ileum*			X	Adrenal gland*+
X	Cecum*		UROGENITAL		Lacrimal gland
X	Colon*	X	Kidneys*+	X	Parathyroids*
X	Rectum*	X	Urinary bladder*	X	Thyroids*
XX	Liver*+	X	Testes*+		OTHER
X	Gall bladder* (not rat)	X	Epididymides*+	X	Bone (sternum and/or femur)
	Bile duct* (rat)	X	Prostate*	X	Skeletal muscle
X	Pancreas*	X	Seminal vesicle*	X	Skin*
	RESPIRATORY	X	Ovaries*+	X	All gross lesions and masses*
X	Trachea*	X	Uterus*+		
X	Lung*++	X	Mammary gland*		
X	Nose*				
	Pharynx*				
	Larynx*				

* Required for carcinogenicity studies based on Guideline 870.4200.
+Organ weight required in carcinogenicity studies.
++Organ weight required if inhalation route.

II RESULTS:

A. OBSERVATIONS:

1. **Clinical signs of toxicity** - Selected clinical signs observed during the first and second year of the study are shown in Table 2. The observed frequency of observations of distended abdomens, yellow staining of the ano-genital area, and alopecia were increased in males in a dose-related manner especially during the first year of the study. However, the numbers of animals displaying these signs were not provided. Distended abdomens and yellow staining were also seen more frequently in high-dose males compared to the control group during the second year of treatment, but the dose effect was not as clear largely due to increased frequency of observations in the control and low dose groups. Although these signs were seen more frequently in treated females compared to the control group, there was no clear dose effect and the signs were not seen as frequently as in males.

TABLE 2: Selected clinical signs, number of observations / average number of survivors[a]

Sign / weeks	Control	20 ppm	500 ppm	2000 ppm
MALES				
Distended abdomen / wks. 0-52	68/73[b]	89/69	123/69	191/69
Distended abdomen / wks. 52-104	140/45	148/46	363/49	234/32
Yellow staining / wks. 0-52	259/73	304/69	372/69	460/69
Yellow staining / wks. 52-104	402/45	618/46	634/49	409/32
Alopecia / wks. 0-52	38/73	36/69	66/69	74/69
Alopecia / wks. 52-104	162/45	213/46	216/49	158/32
Sign / weeks	**Control**	**20 ppm**	**2500 ppm**	**5000 ppm**
FEMALES				
Distended abdomen / wks. 0-52	12/73	124/72	53/70	39/71
Distended abdomen / wks. 52-104	27/49	94/52	56/47	66/47
Yellow staining / wks. 0-52	12/73	5/72	3/70	19/71
Yellow staining / wks. 52-104	54/49	82/52	81/47	166/47
Alopecia / wks. 0-52	14/73	23/72	47/70	37/71
Alopecia / wks. 52-104	90/49	84/52	131/47	80/47

[a] Data calculated and summarized from pages 23-109 in the study report (MRID 00062806).
[b] Number of observations/TWA no. animals alive during time period.

2. <u>Mortality</u> - The percent mortality of each study group at selected times throughout the study is shown in Table 3. In high-dose males, the percent mortality was consistently higher than the control during the first year of the study, and was significantly higher ($p<0.01$) at 75 weeks (calculated by the reviewer). By week 104, the percent mortality in high-dose males was only 10% (NS) higher than in the control group. The increase in mortality in high-dose males was not dose-related. Mortality was slightly higher in high-dose females at 52 weeks compared to the control group, but the difference was not statistically significant and mortality in high-dose females was comparable to the control group at 104 weeks.

TABLE 3: Group percent mortality[a]

Treatment period	Control	20 ppm	500 ppm	2000 ppm
MALES				
@ 52 Weeks	8%	16%	13%	19%
@ 75 Weeks	29%	25%	24%	53%
@ 104 Weeks	75%	67%	65%	85%
Treatment period	**Control**	**20 ppm**	**2500 ppm**	**5000 ppm**
FEMALES				
@ 52 Weeks	7%	13%	16%	12%
@ 75 Weeks	Illegible	24%	29%	29%
@ 104 Weeks	72%	57%	68%	71%

[a] Data summarized by the reviewer from pages 23-109 in the study report (MRID 00062806).

B. BODY WEIGHT

The average body weight of treated females increased slightly compared to the control group [control, 28.227 g; 20 ppm, 28.882 g; 2500 ppm 30.292 g; 5000 ppm, 30.545 g (Data were obtained from p. 355 in the study document)]. At study termination, the increase in the female high-dose group compared to the control group was 8%. No further numerical data for body weights and body weight gains were presented in the study document (body weights of males were not provided). In the review of the study (MRID 92142033, p. 6), the author stated that the body weights of high-dose males and females were significantly increased during the first 6 months of the study, but were comparable to the controls by the end of the study. The food consumption of both sexes was stated to be slightly higher in treated animals generally than that of the control groups.

C. FOOD CONSUMPTION AND COMPOUND INTAKE

1. **Food consumption** - The food consumption numerical data were not included in the study. A slight increase in food consumption in all treated groups compared to the respective control groups was reported in the study review (MRID 92142033, p. 6).

2. **Compound consumption (time-weighted average)** - The compound consumption was not calculated by the study author, but was estimated by the reviewer assuming a 20 kg mouse consumes 3 g food per day. The values are included in Table 1.

3. Food efficiency - No data on food efficiency was presented in the study document.

D. BLOOD ANALYSES:

1. **Hematology** - No numerical data on hematology parameters were presented in the document. Slightly decreased differential leucocyte counts were reported in mid-dose females and in high-dose males and females compared to the control groups. Slightly higher mature (segmented) neutrophil counts were reported in high-dose males compared to the controls (These statements appeared on p. 7 of MRID 92142033).

2. **Clinical Chemistry** - Clinical chemistry parameters were not measured in the study.

E. SACRIFICE AND PATHOLOGY:

1. **Organ weight** - The absolute brain weight of females was slightly decreased at the high dose (control, 0.480 g; high dose, 0.473 g), but the difference was not statistically significant. The brain weight relative to body weight, however, was significantly decreased (control, 1.7325; high dose, 1.5675, $p<0.05$). The difference was attributed to increased mean body weight in treated females (control, 28.227 g; high dose, 30.545 g). An increase in the mean liver weight of 0.63 g was reported for high-dose females compared to the control group. Data were obtained from p. 355 in the study document. No additional data were presented on organ weights

2. **Gross pathology** - No data on gross pathology were presented in the study document.

3. **Microscopic pathology** -

a) **Non-neoplastic** - Minimal to mild inflammatory changes characterized by mononuclear leucocyte infiltrates or more complex chronic inflammatory changes were reported in the liver and kidneys from animals in all groups. Amyloidosis was also seen in many mice from all groups. No treatment-related histopathology findings were seen in the brain. The non-neoplastic histologic changes were considered incidental to treatment by the author of the study review (MRID 92142033, p. 9). Raw data on lesions were provided, but there were no data summaries.

b) **Neoplastic** - Significant increases in the incidences of hepatomas and bronchioalveolar adenomas in the liver and lungs of both mid- and high-dose females compared to the control group were seen in the study (Table 4). The incidence of hepatocellular carcinoma was significantly increased in males at the mid-dose compared to the control group, but the carcinoma incidence at the high dose was comparable to the control. The significance of the increased incidences of hepatomas in treated females was related to the low incidence of this neoplasm in the female control group compared to the male control group. The effect of removing animals from the main study as stated in Appendix L, MRID 00066528 (different numbers of animals from the different groups) at the 16-month sacrifice was not accounted for by the study author.

TABLE 4: Selected neoplastic findings, total incidence / number of mice examined

Organ: neoplastic finding	Control	20 ppm	500 ppm	2000 ppm
MALES				
Liver: hepatoma[a]	16/75	20/75	18/75	17/73
Liver: hepatocellular carcinoma[a]	4/75 (5%)	6/75	13/75* (17%)	5/73
Lung: bronchioalveolar adenoma[a]	18/75	19/75	20/75	17/73
Lymphatics: lymphosarcoma[b]	8/75	6/75	8/75	11/73
Mice with 1 or more neoplasms[b]	43/75	44/75	53/75	43/73
Organ: neoplastic finding	**Control**	**20 ppm**	**2500 ppm**	**5000 ppm**
FEMALES				
Liver: hepatoma[b]	3/72 (4%)	2/75 (3%)	15/74* (20%)	17/73** (23)
Liver: hepatocellular carcinoma[b]	0/72	2/75	3/74	0/73
Lung: bronchioalveolar adenoma[b]	12/72 (17%)	14/75 (19%)	28/74* (38%)	26/73* (36%)
Lymphatics: lymphosarcoma[b]	13/72	15/75	15/74	14/73
Mice with 1 or more neoplasms[b]	35/72	42/75	53/74	52/73

[a] Data obtained from pages 126 and 127 in the study report (MRID 00062806).
[b] Data obtained from pages 7 and 8 of the summary report (MRID 92142033).
* Significantly different ($p<0.05$) from the control by the Chi-square test and the Armitage-Cochran and Yates corrected Chi-square test.
** Significantly different ($p< 0.01$) from the control by the Armitage-Cochran and Yates corrected Chi-square test.

III. DISCUSSION and CONCLUSIONS

A. <u>INVESTIGATOR'S CONCLUSIONS</u>: The conclusions expressed in the original study are apparently unavailable. The study document under review (MRID 00062806) primarily deals with the clinical signs observed during treatment with FMC 33297 and a possible effect of caging on tumor incidence and mortality. The author of report MRID 00062806 concluded that the male and female mice in the treated groups exhibited a higher incidence of abdominal distention or hardened stomachs than the mice in the control groups, but there was no apparent dose relationship. The incidence rate of yellow staining around the ano-genital area was increased in high-dose males. The incidences of alopecia and lacrimation exhibited no apparent treatment-related effects. There were no definitive effects of caging on tumor incidence or mortality. Conclusions expressed in a review of the original study, MRID 92142033, included the determination of a systemic no-observed-effect-level (NOEL) in males of 500 ppm and 20 ppm in females. The increased incidences of hepatoma in the mid- and high-dose female groups compared to the control group appeared to be treatment-related. The increases in bronchoalveolar adenoma in mid- and high-dose females were not sufficiently large to define an oncogenic relationship. The author also concluded that an

MTD was reached based on body weight differences and histopathological lesions in treated compared to control animals.

B. **REVIEWER COMMENTS:** A significant increase in mortality was seen in high-dose males compared to the controls after 75 weeks of treatment (control, 29%; high-dose, 53%, $p<0.01$), but not at study termination when the difference was only about 10%. The increase in mortality in males did not show a clear dose effect, and there was no effect in females. It is not clear why the study author made a distinction between spontaneous deaths and moribund deaths. Furthermore, the author did not account for the 16-month interim sacrifice (Appendix L, MRID 00066528) in which various numbers of animals from the different groups were sacrificed in order to analyze fat for the test material (in response to a feed mixup). These animals are not accounted for in the mortality tables of MRID 00062806.

The clinical signs of distended abdomens and yellow ano-genital staining seen throughout the study were reported to show a treatment-related effect when examined for the first 52 weeks of the study. Total observations rather than number of animals observed with the clinical sign were reported, making it difficult for the reviewer to evaluate the significance of the observations. Because these signs were also observed in the control groups, some historical control information for this sign in this strain of mice is needed.

Insufficient data were provided on body weights (with the exception of final body weights for females), body weight gains, organ weights (with the exception of brain weights of females at study termination), leucocyte counts, segmented neutrophil counts, and inflammatory changes in the liver and kidney for the reviewer to evaluate. The reviewer does not consider an 8% increase in final female body weight a biologically significant effect. Although difficult to evaluate in the absence of summary data, the effects listed by the study author - decreased leucocyte counts and liver and kidney inflammatory changes - do not appear to be toxicologically significant.

In the absence of summary data on clinical signs, body weights and body weight changes throughout the study; organ weights (except brain weight of females); and summary data on gross and microscopic observations, a NOAEL and LOAEL for FMC 33297 in mice could not be set in this study.

On December 12, 1988 the HED Cancer Peer Review Committee reviewed the study and determined that the following neoplastic lesions were significant findings:

Statistically significant increases in liver adenoma in male mice at all doses (and outside historical control range at all doses) with a significant dose-related trend; statistically significant increases in combined liver adenoma/carcinoma at mid- and high-doses, with a significant dose-related trend.

Statistically significant increase in liver adenoma in female mice at the mid-and high-dose (both were outside historical control range) with a significant dose-related trend; statistically

significant increases in combined liver adenoma/carcinoma at mid- and high-dose, with a significant dose-related trend.

Statistically significant increases in lung adenoma and combined adenoma/carcinoma at all doses in females; carcinoma were significantly increased at HDT only, but were increased at all doses. The incidences of adenoma and carcinoma were outside historical controls at mid- and high-dose (carcinomas only slightly so at mid-dose). There were also significant dose-related trends for lung adenomas, carcinomas and combined adenoma/carcinomas in females.

The incidence of lung tumors in male mice (adenoma or carcinoma, or combined) were not statistically significant at any dose, nor was there a dose-related trend for any of them.

C. STUDY DEFICIENCIES:

The study author based the MTD in this study on body weight differences and histopathological lesions seen in animals treated at the highest dose, but the body weight increased slightly in treated animals and the histopathological lesions seen were said to be non-treatment related. These findings are not sufficient as a basis for an MTD.

The study lacked numerical data on body weights and body weight gains, food consumption, hematology, organ weights, and gross and microscopic non-neoplastic findings. The study also lacked experimental details on husbandry, etc.

The source of the animals sacrificed for the 16-month fat analysis needs to be explained. If these animals were part of the main study, then total numbers of animals in the raw data tables need to be reduced at the appropriate time.

There also were a number of inconsistencies in the study that were not likely to significantly change the conclusions of the study; however, they do undermine the confidence in how carefully the study was conducted or the data transposed. Some of these inconsistencies include:

According to the protocol, the low concentration of 100 ppm was reduced to 20 ppm after 33 days in both sexes and the middle and high concentrations for males were reduced from 2500 ppm to 500 ppm and from 5000 ppm to 2000 ppm, respectively. The middle and high concentrations of 2500 ppm and 5000 ppm were not changed for females. These changes were said to begin on 2/5/1977 with sponsor authorization on 2/9/1977. Complicating these dates further, is a laboratory report of dietary concentrations for a diet containing 100 ppm from treatment week 7, which was 3 weeks past the last time this concentration was supposed to be used.

The mortality of animals in this study was listed several times in the document and none of them agreed on how many died or survived. The reviewer went to the incidence tables for

this information since it should be closest to the original data and found several cases where deaths were listed but not tabulated in the "no. survivors" column.

The dietary mixtures, which were stored frozen and analyzed after being fed, were found to be incorrect at least 2 times by over 100%, once by a factor of 10. Considerable effort was expended trying to explain the discrepancy, but it was apparently never resolved.

The control diet was found to contain small amounts of the test substance in several cases. This could have arisen from contamination of mixing equipment or cross contamination during shipping or storage. The latter seemed to be the most likely case, but it was never completely resolved.

A joint FDA-EPA audit of this study conducted in late 1980 at Bio/Dynamics and FMC facilities did not reveal any inadequacies in the conduct or reporting of this study serious enough to compromise the usefulness of these study results for oncogenic evaluation. However, the audit concluded that this study was not useful for assessment of chronic toxicity (HED Doc. #004204).

DATA FOR ENTRY INTO ISIS

Carcinogenicity Study - mice (870.4200b)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Target organ(s)	Comments
109701	00062806, 92142033	carcinogenicity	mice	24 months	oral	diet	3-714	Males: 0, 3, 71, 286 Females: 0, 3, 357, 714	Not set	Not set	liver: hepatoma lungs: bronchoalveolar adenoma	

DATA EVALUATION RECORD

PERMETHRIN

STUDY TYPE: CARCINOGENICITY - MOUSE
(OPPTS 870.4200b/OECD 451)
MRID 00062806, MRID 92142033, 00066528

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37830
Task Order No. 02-04

Primary Reviewer: Andrew A. Francis, M.S., D.A.B.T.	Signature: Date:	R H Ross for A. A. Francis JAN 0 3 2001
Secondary Reviewers: Sylvia S. Talmage, Ph.D., D.A.B.T.	Signature: Date:	Sylvia S. Talmage JAN 0 3 2001
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross JAN 0 3 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson JAN 0 3 2001

Disclaimer

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Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

DATA EVALUATION RECORD

PERMETHRIN
(272Z75)

STUDY TYPE: SUBCHRONIC ORAL TOXICITY - RAT
[OPPTS 870.3100 (§82-1a)]
MRID 00050199

Prepared for

Health Effects Division
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U.S. Environmental Protection Agency
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Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
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Oak Ridge, TN 37831
Task Order No. 02-05

Primary Reviewer: Melissa D. Halpern, Ph.D.	Signature: Robert N. Ross for M.D. Halpern Date: DEC 22 2001
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Robert H. Ross, M.S., Group Leader	Signature: Robert N. Ross Date: DEC 22 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature: L.A. Wilson Date: DEC 22 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

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PERMETHRIN **Subchronic Oral Toxicity Study (OPPTS 870.3100)**

EPA Reviewer: Yung G. Yang, Ph.D.
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Toxicology Branch, HED (7509C)

Yung G. Yang Date 1/23/2002
Joycelyn Stewart Date 1/24/2002

TXR # 0050649

DATA EVALUATION RECORD

STUDY TYPE: Subchronic Oral Toxicity Study - Rat [OPPTS 870.3100 (§82-1a)]

DP BARCODE: D269531
P.C. CODE: 109701

SUBMISSION CODE: S504352
TOX. CHEM. NO.: 652BB

TEST MATERIAL (PURITY): 27Z75, Permethrin (a.i. % not reported)

SYNONYMS: Permethrin

CITATION: Williams, L.M., Clampitt, R.B., James, J.A., Reynolds, J. and Woods, N.C. (1976) 27Z75, Rat oral 90 day toxicity study. Wellcome Research Laboratories, Berkhamsted. Lab. Ref. No. TL.6-76. February 19, 1976. MRID 00050199. Unpublished.

SPONSOR: Wellcome Research Laboratories.

EXECUTIVE SUMMARY: In a 90-day dietary toxicity study (MRID 00050199), Wistar rats (18/sex/group) were given 27Z75 (permethrin, a.i. % not reported, Lot/Batch 551/3/1) administered in feed at 0, 60, 200, 600 or 2000 ppm (equivalent to 0, 5, 18, 54 or 180 mg/kg/day for males and 0, 5, 18, 54 or 176 mg/kg/day for females). Ten males and 10 females from each group were killed at 90 days. The remaining animals (8/sex/group) were continued without test compound for an additional 29 days in the recovery period.

No treatment-related deaths, clinical signs, changes in body weight, food consumption or food efficiency were reported. There were no treatment-related changes in hematologic, clinical chemistry, urinalysis or estrus cycle parameters tested.

Males dosed with 600 and 2000 ppm had increased fat content in the renal cortex without accompanying morphological alterations. These changes were not considered toxicologically relevant.

Small, statistically significant decreased relative and absolute adrenal weights in 2000 ppm females were reported, however the percent changes are incalculable because of illegible data. Statistically significant, moderately increased absolute and relative lung and spleen weight in 2000 ppm males (percent changes incalculable because of illegible data) were observed during the recovery period only. In addition, slight, statistically significant increased relative and/or absolute pituitary gland weights were observed in all female dosed groups during both the study and recovery periods (percent changes incalculable).

Given the illegibility of the organ weight data, a NOAEL and LOAEL cannot be determined. If readable data were provided, a NOAEL and LOAEL could possibly be assigned.

This study is classified as **Unacceptable/Guideline (upgradeable)** and does not satisfy guideline requirement for a subchronic oral toxicity study.

COMPLIANCE: Signed and dated GLP, Quality Assurance, Flagging statement and Data Confidentiality statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test material: 27Z75

   Description: not reported.
   Lot/Batch # : 551/3/1
   Purity: a.i. not reported.
   Stability of compound: not reported.
   CAS #: not reported.
   Structure: not provided

2. Vehicle and/or positive control

   None; the test material was administered in the feed.

3. Test animals

   Species: rat.
   Strain: Wistar.
   Age/weight at study initiation: males and females: age not reported, 100 - 162 g.
   Source: Charles River.
   Housing: 6/cage, same sex.
   Diet: Diet 41B meal ex Heygates.
   Water: tap, *ad libitum*.
   Environmental conditions: not reported.
   Acclimation period: not reported.

B. STUDY DESIGN

1. In life dates

   Start: 05/75; End: 09/75

2 Animal assignment

Animals were assigned to one of 5 groups (Table 1). Eighteen rats/sex/dose were given 27Z75 at target concentrations of 0, 60, 200, 600 and 2000 ppm in the diet for 90 days. Ten rats/sex/dose were killed after 90 days (study period). The remaining 8/sex/dose were fed diet without the test substance for an additional 29 days and then killed (recovery period).

TABLE 1. Study design

Target dose (ppm)	Number of animals	Actual mean dose (mg/kg/day)
MALES		
0	18	0
60	18	5
200	18	18
600	18*	54
2000	18	180
FEMALES		
0	18	0
60	18	5
200	18	18
600	18	54
2000	18	176

Data taken from pp. 3 and 6; MRID 00050199.
* One animal from this group was missexed and removed from the study on day 35.

3. Dose selection rationale

Dose selection rationale was not reported. However, the objective of the study was stated as ". . . to evaluate the cumulative toxicity of 27Z75 in rats with a view to determining the 'no effect' level."

4. Test diet preparation and analysis

A 10% premix of 27Z75 in diet was prepared every 14 days, and diets were changed twice weekly. Information on homogeneity, stability and concentration were not provided.

Results –
Homogeneity: Not reported.

Stability: Not reported.

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Concentration: Not reported.

The lack of any analytical data did not establish that the mixing procedure was adequate, or that the variance between nominal and actual dosage to the animals was acceptable.

5. Statistics

Data were analyzed using the t-test. Statistical significance was achieved at $p \leq 0.05$.

C. METHODS

1. Observations

Animals were observed daily for mortality and moribundity.

2. Body weight

Animals were weighed weekly throughout the study.

3. Food consumption and compound intake

Food consumption and compound intake were recorded weekly.

4. Blood was collected from the orbital plexus under light ether anesthesia from 6/sex/group at days 14, 28, 56, 90 and 119 for hematology and clinical biochemical analysis. At 90 days, 10/sex/group were orbitally bled prior to killing. The remaining 8/sex/dose were continued without test substance for an additional 29 days (recovery period) and bled prior to killing at that time. The CHECKED (X) parameters were examined.

a. Hematology

X	Hematocrit (HCT)*	X	Leukocyte differential count*
X	Hemoglobin (HGB)*	X	Mean corpuscular HGB (MCH)
X	Leukocyte count (WBC)*	X	Mean corpusc. HGB conc. (MCHC)
X	Erythrocyte count (RBC)*	X	Mean corpusc. volume (MCV)
	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Required for subchronic studies based on Subdivision F Guidelines

b. Clinical chemistry

	ELECTROLYTES		OTHER
	Calcium*		Albumin*
	Chloride*		Creatinine*
	Magnesium	X	Blood urea nitrogen*
	Phosphorus*		Total Cholesterol
X	Potassium*		Globulins
X	Sodium*	X	Glucose*
			Total bilirubin
	ENZYMES	X	Total serum protein (TP)*
X	Alkaline phosphatase (ALK)		Triglycerides
	Cholinesterase (ChE)		A/G ratio
	Creatine phosphokinase (CPK)		
	Lactic acid dehydrogenase (LDH)		
X	Alanine aminotransferase (ALT)*		
X	Aspartate aminotransferase (AST)*		
	Gamma glutamyl transferase (GGT)		
	Glutamate dehydrogenase (GDH)		

* Required for subchronic studies based on Subdivision F Guidelines

5. Urinalysis

Urinalysis was done using composite samples collected overnight from fasted, water deprived animals. The CHECKED parameters were examined.

	Volume*	X	Protein*
	Specific gravity*	X	Glucose*
	Appearance*	X	Ketones*
	Sediment*		Urobilinogen
X	pH*		Bilirubin*
			Nitrites
		X	Blood*
			Leukocytes

* Required for subchronic studies based on Subdivision F Guidelines

D. ESTRUS CYCLE

Vaginal smears were taken daily from all female rats during the final 30 days of the study to determine the number of estrus cycles.

E. OPHTHALMOLOGIC EXAMINATION

Ophthalmologic examinations were not done.

F. NEUROTOXICITY SCREENING

Neurotoxicity screening was not done.

G. SACRIFICE AND PATHOLOGY

After 90 days, 10/sex/dose were sacrificed via exsanguination under anesthesia. The remaining 8/sex/dose were killed in the same manner after 119 days. Necropsies were done on all 0 and 2000 ppm animals. The CHECKED (X) tissues were collected for histological examination. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVAS./HEMAT.		NEUROLOGIC
X	Tongue	X	Aorta*	XX	Brain*
X	Salivary glands*	XX	Heart*	X	Periph. nerve*
	Esophagus*	#	Bone marrow*		Spinal cord (3 levels)[T]
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*	X	Eyes (optic n.)
#	Jejunum*	XX	Thymus*		
X	Ileum*				GLANDULAR
#	Cecum*		UROGENITAL	XX	Adrenal gland*
X	Colon*	XX	Kidneys*+		Lacrimal gland[T]
	Rectum*	X	Urinary bladder*	#	Mammary gland[T]
XX	Liver*+	XX	Testes*+		Parathyroids*
	Gall bladder*	#	Epididymides	XX	Thyroids+
X	Pancreas*	XX	Prostate		
		X	Seminal vesicle		OTHER
	RESPIRATORY	XX	Ovaries	X	Bone
X	Trachea*	X	Uterus*	X	Skeletal muscle
XX	Lung*	#	Vagina	X	Skin
	Nose				All gross lesions and masses*
	Pharynx				
	Larynx				

* Required for subchronic studies based on Subdivision F Guidelines
\+ Organ weight required for subchronic studies.
[T] Required only when toxicity or target organ
\# Tissue preserved but not examined

II. RESULTS

A. OBSERVATIONS

1. Toxicity – No treatment related toxicity was observed. The 2000 ppm male that died on day 61 (see below) exhibited weight loss from day 49 until death.

2. Mortality – Three rats died during the study, however none of the deaths were treatment related. A male 200 ppm animal was found dead on day 61; death was attributed to staphylococcal infection of the kidney. In addition, a 2000 ppm male and 600 ppm female died under anesthesia while being bled.

B. BODY WEIGHT AND WEIGHT GAIN

No statistically significant changes in body weight were observed in any treatment group, as reported by the study authors. (These data could not be presented by the reviewer in tabular form or checked for statistical relevance because the results in the text were illegible. Graphical presentation of body weight data on page 7 of MRID 00050199 appear to show no statistically significant changes, however.)

C. FOOD CONSUMPTION, COMPOUND INTAKE

1. Food consumption

No differences were observed between groups (Table 2). Statistical analysis was not provided and could not be checked by the reviewer because individual data values were unreadable even with magnification.

Table 2. Mean food consumption in animals fed 27Z75 (Permethrin) for 90 days

Sex	Treatment group (ppm)				
	0	60	200	600	2000
Weeks 1 - 13 (study period)					
Males	86	88	89	90	90
Females	94	86	89	90	88
Weeks 14 - 17 (recovery period)					
Males	63	63	64	66	61
Females	85	74	79	81	85

Data taken from Table 2, pp. 27; MRID 00050199.

2. Compound consumption

Compound consumption is shown in Table 1. Both males and females consumed comparable amounts of test substance during the study.

3. Food efficiency

Food conversion was not different for any treated group versus control during the study or recovery period (Table 3) as reported by the study authors. However, values were decreased in all females dosed groups during the study and recovery periods. Statistical evaluation by the reviewer could not be done because individual data values were not legible, even with magnification.

Table 3. Mean food conversion[a] in animals fed 27Z75 (Permethrin) for 90 days					
Sex	Treatment group (ppm)				
	0	60	200	600	2000
Week 13 (end of study period)					
Males	7.5	7.5	7.7	7.5	7.5
Females	15.2	12.7	13.2	13.5	14.1
Week 17 (end of recovery period)					
Males	8.9	10.2	8.9	9.2	8.7
Females	18.5	13.4	13.9	15.3	15.9

Data taken from Table 4, pp 29; MRID 00050199.
[a] Food conversion = g food consumed/ g body weight increase over 7 day period.

D. BLOOD WORK

1. Hematology

Hematology data, shown in the text on pages 9 and 10 of MRID 00050199, were not expressed as mean hematologic values but as "not affected, fluctuating, increase, decrease (slight, moderate or severe) or statistically significant" compared to controls. Individual hematologic values were included in the Appendix (Tables 6A-G, pp. 31-50, MRID 00050199), but the results were illegible, even with magnification. Given the presentation, it is difficult for the reviewer to accurately assess these data.

The study authors report no toxicologically significant changes in any hematologic parameters during the study. While accurate hematology data assessment by the reviewer was not possible, the reviewer was comfortable accepting the study authors' conclusions for these parameters because they included a statement that any statistically significant changes remained within the normal range for the species.

2. Clinical chemistry

Clinical chemistry data, shown in the text on page 10, were not expressed as mean clinical chemistry values but as "not affected, increase, decrease (slight, moderate or severe) or statistically significant" compared to controls. Individual and mean clinical chemistry values were included in the Appendix (Tables 7A-C, pp.51 - 61, MRID 00050199), but the values were unreadable, even with magnification. Given the presentation, it is difficult for the reviewer to accurately assess these data.

The study authors report no toxicologically significant changes in any clinical chemistry parameters during the study. Statistically significant decreased glucose levels observed in male and female dosed groups were reported to be within the normal range for this species, and the reviewer was comfortable accepting the study authors conclusions for this parameter.

E. URINALYSIS

No treatment-related effects were reported. Large quantities of blood observed in the collected urine of 600 ppm male rats were likely due to a cut on the foot of one animal.

F. ESTRUS CYCLE

No treatment-related findings were reported in the number of estrus cycles of treated versus control groups.

G. SACRIFICE AND PATHOLOGY

1. Organ weight

Organ weight data, shown in the text on pages 15 and 16 of MRID 00050199, were not expressed as actual data values but as "increase, decrease (slight or moderate) or statistically significant" compared to controls. Individual and mean relative and absolute organ weight values were included in the Appendix (Tables 9A - D, pp. 63 - 71, MRID 00050199), but the values were unreadable, even with magnification. Given the presentation, it is difficult for the reviewer to accurately assess these data.

Slight, statistically decreased relative and absolute adrenal weight were reported in 2000 ppm females during the study period. Moderate, statistically significant increased absolute and relative lung and spleen weight in 2000 ppm males were observed at the end of the recovery period only. In addition, the table on page 15 of MRID 00050199 shows slight, statistically significant increased relative and/or absolute pituitary gland weight in 60, 200, 600 and 2000 ppm females at the end of the study period. However, the authors did not discuss these data in their conclusions. The reviewer has no means to determine the extent of a "slight" increase in pituitary gland weight. Further, "slight" changes in other organ weight parameters appear to have been utilized by the authors to determine the NOAEL. The reviewer was troubled by the apparent disregard of pituitary gland data, and feels unable to accept the authors' conclusions on organ weight without being able to see the actual data results.

2. Gross pathology

No treatment-related findings were reported.

3. Microscopic pathology

Slightly increased fat content of the renal cortex tubules was observed in 600 and 2000 ppm males. No morphologic alterations accompanied the increased fat content, however. (Actual histology data were not readable in the text)

III. DISCUSSION

A. DISCUSSION

Dietary administration of 0, 60, 200, 600 or 2000 ppm 27Z75 to male and female rats for 90 days resulted in changes in organ weights. However, these changes could not be accurately evaluated by the reviewer.

No treatment-related deaths, clinical signs, changes in body weight, food consumption or food efficiency were reported. There were no treatment-related changes in hematology, clinical chemistry, urinalysis or estrus cycle parameters tested.

Statistically significant decreased relative and absolute adrenal weights in 2000 ppm females during the study period, and increased absolute and relative lung and spleen weight in 2000 ppm recovery males were reported. In addition, statistically significant increases in relative and absolute pituitary gland weight were observed in all female dosed groups during the study period. However, these changes were impossible to assess by the reviewer because the actual data values were illegible. (Changes reported in hematologic and clinical chemistry parameters were unable to be fully evaluated because of the manner in which the data were reported. However, the study authors stated that any small statistically significant changes were still within the normal range for this species. The reviewer was comfortable accepting this statement even without actual data values.)

The study authors concluded in the summary (p.2 of MRID 00050199) that the no affect level for 27Z75 is > 600 ppm and < 2000 ppm, but the effects at 2000 ppm are considered to be minimal. The reviewer is not certain upon what data this conclusion was based. The authors' do not apparently address the increased pituitary weights in 60, 200, 600 and 2000 ppm females. Given the lack of readable data in the study, it is not feasible to assign a LOAEL and/or NOAEL.

B. STUDY DEFICIENCIES

Study deficiencies fall into two categories. The first are omissions in requirements that may not have been applicable in 1975 when the study was conducted. These include the lack of analytical information on the test material, and missing hematologic, clinical chemistry and urinalysis parameters. These deficiencies do not affect the validity of the study. The second is the preponderance of illegible data which makes analysis of this study impossible. This study could likely be upgradable if readable data were produced.

PF

DATA EVALUATION RECORD
Permethrin
(FMC 45801)

STUDY TYPE: SUBCHRONIC ORAL TOXICITY-RAT
OPPTS: 870.3100 (§82- 1a)
MRID 00040491

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 01-89

Primary Reviewer: Nancy B. Munro, Ph.D.	Signature: Date:	Robert H. Ross for Nancy B. Munro APR 0 4 2001
Secondary Reviewers: H. Tim Borges, Ph.D., M.T.(A.S.C.P.), D.A.B.T.	Signature: Date:	HT Borges APR 0 4 2001
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross APR 0 4 2001
Quality Assurance: Gary A. Sega, Ph.D.	Signature: Date:	Gary Sega APR 0 4 2001

Disclaimer

This review may have been altered subsequent to the contractors' signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract DE-AC05-00OR22725

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Permethrin **Subchronic Oral Toxicity [OPPTS:870.3100 (§82-1a)]**

EPA Reviewer: Yung G. Yang, Ph.D. Yung G. Yang Date: 12/18/2001
Reregistration Action Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D. Joycelyn Stewart Date: 1/28/2002
Toxicology Branch, HED (7509C)

DATA EVALUATION RECORD

TXR 0050649

This is an updated DER. The classification has been changed to unacceptable.

STUDY TYPE: Subchronic Oral Dietary Toxicity – Rat [OPPTS: 870.3100 (§)82-1a]

DP BARCODE: D269531 SUBMISSION CODE: S504352
P.C. CODE:109701 TOX. CHEM. NO.: 652BB

TEST MATERIAL: Permethrin

SYNONYMS: Permethrin; FMC 45801; purity not reported.

CITATION: Becci, P. J. and Parent, R. A. (1980) Evaluation of the subchronic toxic effects of FMC 45801 when administered in the diet to Long-Evans rats over a ninety-day period, Food and Drug Research Laboratories, Inc., Waverly Research Center, Waverly, New York, 14892, April 3, 1980. MRID 00040491. Unpublished.

SPONSOR: FMC Corporation, Princeton, NJ 08540

EXECUTIVE SUMMARY: In a repeated dose oral feeding toxicity study (MRID 00040491), Permethrin (Batch No. 79-50-2, purity not given) was administered to groups of 20 male and 20 female Long-Evans rats in the diet at dose levels of 0, 50, 75, 100 or 500 ppm (0, 2.97, 4.31, 5.71, or 29.9 mg/kg in males, and 0, 3.94, 5.76, 7.38, or 37.9 mg/kg in females) for 90 days.

All animals survived to study termination. Clinical observation revealed no treatment-related signs. No biologically or toxicologically significant changes in body weights, weight gains, food consumption, food efficiency, hematology, clinical biochemistry, or urinalysis endpoints were observed. No neurotoxicity testing was done nor were ophthalmological examinations performed. Gross pathology was unremarkable.

Minor increases in absolute and relative liver weights were observed in all male groups except for the 100 ppm males; no histopathological or clinical biochemical correlates were observed. The changes could reflect biological variation or an adaptive response to xenobiotic compound exposure. The liver weight changes were not considered toxicologically significant or adverse. No other treatment-related effects were reported for any organ weight or histopathological endpoint. Thus, no effects were observed upon which to base a LOAEL value for either sex.

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Under the conditions of this study, the subchronic toxicity NOAEL for both sexes is ≥500 ppm (29.9 and 37.9 mg/kg/day for males and females, respectively). It should be noted that the animals could have tolerated a higher dose.

This subchronic oral toxicity study in rats is classified as **Unacceptable/Guideline** and does not satisfy the [OPPTS:870.3100 (§82-1a)] Subdivision F guideline requirements.

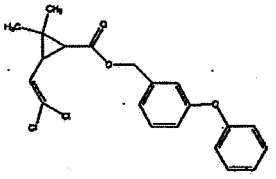
COMPLIANCE: Signed and dated GLP, Flagging, and Data Confidentiality statements were not provided. Animal husbandry conformed to the standards established in DHEW Publication No. 78-23, "Care and Use of Laboratory Animals."

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound: Permethrin

   Description: brown mottled solid
   Batch No.: 79-50-2
   Purity: not given
   Stability of compound: Unspecified
   CAS No: 52645-53-1
   Structure:



2. Vehicle and/or positive control: Corn oil

3. Test animals

   Species: rat
   Strain: Long-Evans
   Age/weight at study initiation: five - six weeks; males: 120-198 g; females: 104-160 g
   Source: Blue Spruce Farms, Altamont, NY
   Housing: Individually in stainless steel wire mesh bottom cages
   Diet: Charles River RHM Lab Diet, *ad libitum*
   Water: Tap water, *ad libitum*
   Environmental conditions:
   Temperature: Not provided
   Humidity: Not provided
   Air changes: Not provided

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Photoperiod: 12-hour light-dark cycle
Acclimation period: One week

B. STUDY DESIGN

1. In life dates: Start: October 17, 1979; end: January 21-25, 1980

2. Animal assignment

Animals were assigned to groups by means of stratified randomization of body weights using a calculator-generated random number sequence.

TABLE 1. Study design

Dose Group	Concentration in Diet (ppm)	Dose (mg/kg/day)[a]		Number of Rats[b]	
		Male	Female	Male	Female
A	0	0	0	30	30
B	50	2.97	3.94	30	30
C	75	4.31	5.76	30	30
D	100	5.71	7.38	30	30
E	500	29.9	37.9	30	30
F	Untreated[c]	0	0	50	50

Data taken from p.4, Text Table, p. 75, and Tables 2 and 7, pp. 21, 23, 68, and 70, MRID 00040491.
[a]Dose estimated by reviewer from nominal concentration in diet, daily food consumption during Week 13, and terminal body weight data.
[b]Thirty rats per group were used for the first six weeks of treatment; 10 animals per group were taken for clinical testing and urinalysis and then killed, leaving 20 rats/group for the remainder of the study.
[c]Animals used for pretrial clinical testing.

3. Dose selection rationale

A dose selection rationale was not provided.

4. Diet preparation and analysis

The test substance was suspended in corn oil. It was mixed into the diet using a Hobart blender. Preliminary sampling was performed for concentration and for homogeneity analysis; no mention was made of stability analysis. For the homogeneity study, five samples from each of the top, middle, and bottom one-third of the feed batches were taken. The diet was freshly prepared weekly; 5-8 vertical core samples were taken and combined from each batch to be sent to the sponsor for concentration analysis.

Results –
Homogeneity analysis: no results are provided.

Stability analysis: no results are provided.

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Concentration analysis: no results are provided.

5. Statistical analysis

Body weight, food consumption, food utilization efficiency, absolute and relative organ weights, and clinical data were evaluated by analysis of variance. Differences among groups were identified using the Least Significant Difference test. Pathology incidence data were evaluated using the Fisher Exact Test, when appropriate.

C. METHODS

1. Observations

The animals were observed daily for clinical signs of toxicity. Mortality checks were performed twice daily.

2. Body weight

Body weights were recorded weekly.

3. Food consumption and compound intake

Food consumption was recorded weekly. No data were presented on compound intake. These values were estimated by the reviewer from weekly food consumption data during Week 13, terminal body weight, and nominal concentrations in the diet. They are presented in Table 1.

4. Ophthalmoscopic examination

Ophthalmic examinations were not performed.

5. Blood was collected from the periorbital plexus prior to study initiation on 15 animals per sex per group (Group F) and after 6 and 13 weeks' feeding from 10 animals randomly selected per sex per treatment group. The animals used prior to study initiation and after 6 weeks' exposure were killed but not necropsied. The CHECKED (X) parameters were examined.

a. Hematology

X		X	
X	Hematocrit (HCT)*	X	Leukocyte differential count*
X	Hemoglobin (HGB)*		Mean corpuscular HGB (MCH)
X	Leukocyte count (WBC)*		Mean corpusc. HGB conc.(MCHC)
X	Erythrocyte count (RBC)*		Mean corpusc. volume (MCV)
X	Platelet count*		Reticulocyte count
	Blood clotting measurements*		Blood cell morphology
	(Activated thromboplastin time)		Sedimentation rate
X	(Clotting time)		
	(Prothrombin time)		

*Required for subchronic studies based on Subdivision F Guidelines.

b. Clinical chemistry

X	ELECTROLYTES	X	OTHER
X	Calcium	X	Albumin*
	Chloride		Albumin/globulin ratio
	Magnesium		Blood creatinine*
	Phosphorus	X	Blood urea nitrogen*
X	Potassium*	X	Total cholesterol*
	Sodium*	X	Globulins
		X	Glucose*
	ENZYMES[a]	X	Total bilirubin
		X	Total serum protein*
X	Alkaline phosphatase (ALK)*		Triglycerides
	Cholinesterase (ChE)		Serum protein electrophoresis
	Creatine phosphokinase		
X	Lactic acid dehydrogenase (LDH)		
X	Alanine aminotransferase (also SGPT)*		
X	Aspartate aminotransferase (also SGOT)*		
	Gamma glutamyl transferase (GGT)		
	Glutamate dehydrogenase		
	Sorbital dehydrogenase		

* Required for subchronic toxicity studies based on Subdivision F Guidelines.
[a]More than two hepatic enzymes should be measured.

6. Urinalysis

Urinalysis, although not required, was carried out prior to study initiation, and after 6 and 13 weeks' dietary exposure on the same animals from which blood was drawn. The CHECKED (X) parameters were examined.

X		X	
X	Appearance	X	Glucose
	Volume	X	Ketones
X	Specific gravity	X	Bilirubin
X	pH	X	Blood
X	Sediment (microscopic)		Nitrites
X	Protein	X	Urobilinogen

7. Sacrifice and pathology

At the end of 13 weeks' exposure, all surviving animals were killed using CO_2 gas and necropsied; the major organs were grossly examined. The CHECKED (X) tissues in the table below were collected for microscopic examination (tissue from large and small intestine were not identified as being from subregions as indicated in the table). All these tissues were microscopically examined from control and high-dose animals. The heart, liver, spleen and grossly abnormal tissues were examined for all dose groups. The (XX) organs, in addition, were weighed.

X	DIGESTIVE SYSTEM	X	CARDIOVASC./HEMAT.	X	NEUROLOGIC
	Tongue (oral cavity)	X	Aorta*	XX	Brain*+
X	Salivary glands*	XX	Heart*+	X	Periph. nerve*
X	Esophagus*	X	Bone marrow*	X	Spinal cord (3 levels)*
X	Stomach*	X	Lymph nodes*	XX	Pituitary*
X	Duodenum*	XX	Spleen*+	X	Eyes (optic n.)*
X	Jejunum*	X	Thymus*+		
X	Ileum*				GLANDULAR
X	Cecum*		UROGENITAL	XX	Adrenal gland*+
X	Colon*	XX	Kidneys*+		Lacrimal gland
	Rectum*	X	Urinary bladder*	X	Mammary gland*
XX	Liver*+	XX	Testes*+	X	Parathyroids*
	Gall bladder*		Epididymides*+	XX	Thyroids*
X	Pancreas*	XX	Prostate*		Coagulation glands
			Seminal vesicle*		
	RESPIRATORY	XX	Ovaries*+		OTHER
X	Trachea*	X	Uterus*+	X	Bone
X	Lung*		Vagina	X	Skeletal muscle
	Nose*			X	Skin*
	Pharynx*			XX	All gross lesions and masses*
	Larynx*				

*Required for subchronic studies based on Subdivision F Guidelines.
+Organ weight required in subchronic and chronic studies.

II. RESULTS

A. OBSERVATIONS

All animals survived to study termination. No treatment-related clinical signs were observed.

B. BODY WEIGHT AND BODY WEIGHT GAIN

No statistically or biologically significant differences from controls were observed for body weights from either males or females at any dose level.

C. FOOD CONSUMPTION AND COMPOUND INTAKE

1. Food Consumption

No biologically significant differences from controls were observed for food consumption from any group at any dose level.

2. Test Compound Intake

Test compound intake was estimated by the reviewer based on nominal diet concentration, food consumption in Week 13, and terminal body weight. The 13-week averages are given in Table 1.

3. Food Efficiency

No treatment-related differences were seen in efficiency of food utilization in either sex at any dose.

D. OPHTHALMOSCOPIC EXAMINATION

Ophthalmoscopic examination was not performed.

E. BLOOD WORK

1. Hematology

No statistically or biologically significant differences from control values were observed for either sex at any dose either at Week 7 or Week 13.

2. Clinical chemistry

A few clinical chemistry results were statistically different from control values but were considered of no biological or toxicological significance as they were within normal historical values for this rat colony, showed no dose-response relationship, and were not consistent over time.

F. URINALYSIS

No treatment-related differences from control rats were reported for any parameter in either sex at any dose.

G. SACRIFICE AND PATHOLOGY

1. Organ weight

Liver weight data are given in Table 2. Absolute liver weights were significantly increased (11-15%) in all treated male groups except the 100 ppm group. The liver-to-body-weight ratio was likewise increased significantly in males at 500 ppm, being 6.4% higher than the control value. The group mean liver-to-brain-weight ratios were compared by the reviewer and these ratios were increased similarly to the absolute liver weights (by 11-16%) over control values, in all but the 100 ppm male group. The absolute liver weights were slightly decreased in all female groups, reaching statistical significance only in the high-dose group (13%); it was also decreased relative to body weight (6.5%) and to brain weight (14%). No treatment-related changes in other absolute or relative organ weights were observed.

TABLE 2. Absolute and relative liver weights for rats administered Permethrin in the diet for 13 weeks[a]

Parameter	Permethrin concentration, ppm				
	0	50	75	100	500
Males					
Terminal body wt (g)	439.5±44.1	464.3 ±45.6	463.1 ±40.6	458.4 ±8.4	469.4 ±8.4
Brain wt (g)	2.1 ±0.1	2.1 ±0.1	2.1 ±0.1	2.1 ±0.1	2.1 ±0.1
Liver: absolute wt. (g)	13.5 ±1.6	15.1* (12)[b] ±2.0	15.0* (11)±1.9	13.8 ±2.1	15.5 ±1.7* (15)
Liver-to-body -wt. ratio (%)	3.1±0.2	3.2 ±0.3	3.2 ±0.3	3.0 ±0.4	3.3 ±0.3* (6.4)
Liver-to-brain-wt. ratio[c]	6.4	7.2 (12)	7.1 (11)	6.6	7.4 (16)
Females					
Terminal body wt. (g)	250.8 ±31.5	237.1 ±22.0	242.3 ±15.7	242.0 ±20.3	238.6 ±22.8
Brain wt. (g)	1.9 ±0.1	1.9 ±0.1	1.9 ±0.1	1.9 ±0.1	1.9 ±0.1
Liver: absolute wt. (g)	7.9 ±1.2	7.5 ±1.0	7.7 ±1.0	7.6 ±0.8	6.9 ±0.9* (13)
Liver-to-body-wt. ratio (%)	3.1 ±0.3	3.2 ±0.3	3.2 ±0.3	3.2 ±0.3	2.9 ±0.3 (6.5)
Liver-to-brain-wt. ratio[c]	4.2	3.9	4.1	4.0	3.6 (14)

Data taken from Tables 7, pp. 68-71, MRID 00040491.
[a] Data are expressed as mean ± standard error with n = 20 for all groups.
*Significantly different from controls, $p \leq 0.05$.
[b] Numbers in parentheses equal percent change from control value, calculated by reviewer.
[c] Ratio of group means, calculated by reviewer.

2. Gross pathology

No treatment-related gross findings were reported at necropsy. One tumor, a renal fibrosarcoma, was observed in a high-dose female rat. The incidence was comparable to those occurring spontaneously and it therefore was not considered treatment-related.

3. Histopathology

No treatment-related histopathologic findings were reported for any organ or tissue.

III. DISCUSSION

A. DISCUSSION

All animals survived to study termination and gross necropsy was unremarkable. No clinical signs related to treatment were observed. No biologically or toxicologically significant changes in body weights, weight gains, food consumption, food efficiency, hematology, clinical biochemistry, or urinalysis endpoints were observed.

Absolute and relative (to body and to brain) liver weights were significantly increased (11 to 15%) in all the male groups except for the 100 ppm males. There were no correlated histopathological or biochemical changes. The increased weights likely reflect an adaptive response to xenobiotic chemical exposure. Liver weights were slightly decreased in the female groups relative to control values, reaching statistical significance only in the high-dose group. The latter change is not considered to be of toxicological significance in the absence of histopathological or clinical biochemical correlates. No other treatment-related histopathological changes were observed.

No changes were observed in this study upon which to base LOAEL values for either male or female animals.

Under the conditions of this study, the NOAEL for both sexes is ≥ 500 ppm (29.9 and 37.9 mg/kg/day for males and females, respectively). It should be noted that the animals could have tolerated a higher dose.

This subchronic oral toxicity study in rats is classified as **Unacceptable/Guideline** and does not satisfy the [OPPTS:870.3100 (§82-1a)] Subdivision F guideline requirements.

B. STUDY DEFICIENCIES

This study has several deficiencies, likely due in part to its age. A major deficiency is that no results of analyses for stability, homogeneity and concentration were reported although samples were provided to the sponsor for homogeneity and concentration analyses. The test diets were prepared weekly, so stability may not have been a problem. The lack of concentration analyses leaves the estimated compound intakes uncertain. This is a particular concern in the absence of effects on which to base a LOAEL.

Minor deficiencies in the clinical chemistry panel included the absence of sodium and blood creatinine assays. The following required tissues for histopathological examination were not included: rectum, epididymides, and seminal vesicles. No ophthalmoscopic examinations were performed on the animals. Signed and dated GLP, Flagging, and Data Confidentiality statements were not provided, likely due to the age of the study. The interpretation of results is not affected by these deficiencies.

DATA EVALUATION REPORT

PERMETHRIN
(PP 557)

STUDY TYPE: ACUTE DELAYED NEUROTOXICITY – HEN
[OPPTS: 870.6100 (§81-7)]
MRID 00097426

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
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Task Order No. 01-87

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Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross JAN 3 0 2001
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson JAN 3 0 2001

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UT-Battelle, LLC, for the U.S. Dept. of Energy under contract DE-AC05-00OR22725

Permethrin

Delayed Neurotoxicity[870.6100(§81-7)]

EPA Reviewer: Yung G. Yang, Ph.D.
Yung G. Yang, Date 11/6/2001
Reregistration Branch 2, HED (7509C)
EPA Work Assignment Manager: Joycelyn Stewart, Ph.D.
Joycelyn Stewart, Date 1/28/2002
Toxicology Branch, HED (7509C)

DATA EVALUATION RECORD

TXR# 0050649

STUDY TYPE: Acute Delayed Neurotoxicity – Hen; OPPTS 870.6100 (§81-7)

DP BARCODE: D269531
SUBMISSION CODE: S504352
PC CODE: 109701
TOX CHEM NO: 652BB

TEST MATERIAL: Permethrin (technical, isomer ratio 25 cis:75 trans)

SYNONYMS: 3-phenoxybenzyl(1*RS*)-*cis,trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate; NRDC 143; PP 557

CITATION: Bond, A.L., Wollon, R.M., Dayan, A.D., and James, J.A. (1980) Neurotoxicity of permethrin after oral administration in the hen. Wellcome Research Laboratories, Berkhamsted Hill. Doc. No. HEFG 80-14, 30 June 1980. MRID 00097426, Unpublished.

SPONSOR: Wellcome Foundation, Ltd.

EXECUTIVE SUMMARY: In a delayed neurotoxicity study (MRID 00097426), a group of 10 domestic hens were administered 0, 2000, or 4000 mg/kg of permethrin (Lot No.: ZJ; isomer ratio 25 cis:75 trans) in corn oil by oral gavage. An additional group of 10 birds was given 500 mg TOCP/kg as the positive control. All birds were given a single oral dose on study day 0 and observed for 21 days. Birds in the permethrin and negative control groups were redosed on study day 21 and observed for an additional 21 days. Toxicity assessments were limited to clinical observations, assessment of ataxia, body weight measurements, and microscopic evaluation of the spinal cord and sciatic nerve. Acetylcholinesterase and neurotoxic esterase activities were not measured.

No treatment-related clinical signs of toxicity and no effects on body weights or food consumption were observed in birds administered permethrin. Ataxia was not seen in birds treated with the test article and no treatment-related lesions were observed on microscopic examination of the nervous tissues.

Following treatment with TOCP, clinical signs and neurohistopathological lesions indicative of delayed neuropathy were observed in these birds.

Therefore, under the conditions of this study, oral administration of permethrin up to 4000 mg/kg does not produce delayed neuropathy in the hen.

This study is classified **Acceptable/Guideline** and does satisfy the requirements for a delayed neurotoxicity study [OPPTS 870.6100 (81-7)] in hens. Although a deficiency was that AChE and NTE activities were not measured, the study is considered sufficient for determining the potential of permethrin to produce delayed neurotoxicity in the hen. This study was conducted prior to initiation of current guidelines.

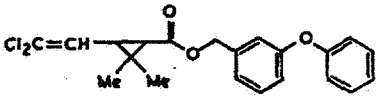
COMPLIANCE: Signed and dated Quality Assurance, Data Confidentiality, Good Laboratory Practice Compliance, and Flagging statements were not provided.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound: permethrin

Description: not given
CAS No.: 52645-53-1
Lot No.: ZJ
Purity: Not stated. isomeric ratio 25 cis:75 trans
Contaminants: none given
Structure:



2. Vehicle and/or positive control

Corn oil was used as the vehicle and negative control. Permethrin was prepared as 40% (w/v) formulation. The positive control used in this study was TOCP.

3. Test animals

Species: domestic hen
Strain: not specified
Age and mean weight at study initiation: adult, age not stated: 1.7-3.1 kg
Source: not stated
Housing: Birds were housed 10/pen in the broiler unit on deep litter in pens measuring 3.07 m x 3.07 m.
Food: Standard layers mash (Heygate and Son, Ltd.) was available *ad libitum*.
Water: Water was available *ad libitum*.
Environmental conditions:
Temperature: 19°±3°C
Humidity: not stated

Air changes: not stated
Photoperiod: 23.5 hours light/ 0.5 hours dark
Acclimation period: not stated

B. STUDY DESIGN

1. In life dates

Start: September 4, 1979; End: October 16, 1979

2. Animal assignment and dosing protocol

Animal assignment and dose selection are listed in Table 1. Hens were ranked according to body weight, separated into blocks, then allocated at random from each block using a computer-generated randomization program.

All birds were given a single oral dose on study day 0 and observed for 21 days. Birds in the permethrin and negative control groups were redosed on study day 21 and observed for an additional 21 days. On study day 13, seven hens in the TOCP group were treated with 10 mg atropine/kg and 50 mg 2-PAM/kg in order to increase their chance of survival.

TABLE 1. Study design

Group	Dose (mg/kg)	Dose Volume (ml/kg)	No. of Animals
Control (corn oil)	0	10	10
Permethrin - low	2000	5	10
Permethrin - high	4000	10	10
Positive Control	500 (TOCP)	0.5	10

Data taken from text table, p. 3, MRID 00097426.

3. Validation of test methods

A positive control group was run concurrently with the test article group to show the ability of the testing facility to detect delayed neuropathy in the hen.

4. Rationale for dose selection

A dose selection rationale was not given in the report. It should be noted that the low dose administered in this study is equivalent to the limit dose for an acute delayed neurotoxicity study in hens.

5. Dose solution preparation and analyses

The test article was administered as a 40% (w/v) technical solution in corn oil, but the method of mixing was not described. The dosing solution was stored at room temperature and used for both dose groups with the volume adjusted to give the proper dose. Concentration was measured initially and stability was analyzed after 1 and 2 months of storage at room temperature. Homogeneity was not analyzed.

Results -
Concentration analysis: The concentration of the test article in the dosing solution was 40.3%.

Homogeneity analysis: not measured

Stability analysis: After 1 and 2 months of storage at room temperature, the concentration of the dosing solution was 39.8% and 39.3%, respectively.

The analytical results show that the actual doses to the animals were acceptable.

6. Statistical analysis

Data were analyzed using the Student t test at the 0.05 level of significance.

C. METHODS

1. Observations

All animals were examined daily for clinical signs of toxicity and mortality.

2. Body weight

Body weights were measured prior to study initiation and on study days 0, 3, 7, 10, 14, 17, 21, 28, 32, 38, and 42.

3. Food consumption and food efficiency

Food consumption and food efficiency were not measured.

4. Neurotoxicity assessment

Abnormalities of gait were assessed on weekdays. The type of motor activity was not described. Ataxia was scored on the following scale:

1. weak grip with toes; slight stagger; reluctant to move
2. just able to raise itself onto toes; reluctant to move; balance unsteady

3. no grip with toes; unable to stand on toes; shuffles around on backs of legs using wings to aid in locomotion
4. paralysis of legs; unable to move; legs splayed

5. Biochemical measurements

Acetylcholinesterase (AChE) and neurotoxic esterase (NTE) activities were not measured in this study.

6. Sacrifice/necropsy/neurohistopathology

Birds in the test article and negative control groups were sacrificed after the second 21-day observation period; birds in the positive control group were sacrificed after the first observation period on day 23. All birds were killed by sodium pentobarbitone injection. All hens that died or that were killed on schedule were subjected to gross necropsy. Three sections of spinal cord and the sciatic nerve were removed and fixed in 10% buffered formalin and processed for microscopic examination.

II. RESULTS

A. CLINICAL OBSERVATIONS AND MORTALITY

One negative control hen and one positive control hen were found dead on days 8 and 21, respectively; necropsy revealed egg peritonitis in both birds. Also in the positive control group, one hen was sacrificed *in extremis* on day 14 due to advanced paralysis and another was found dead on day 23. All hens treated with the test article survived to scheduled sacrifice.

No treatment-related clinical signs of toxicity were observed in hens treated with the test article. One hen in the low-dose group had progressive weight loss after day 35 and necropsy revealed emaciation, peritonitis, and a septicemic carcass. Another low-dose animal appeared lethargic on day 42 but no abnormalities were noted at necropsy.

C. BODY WEIGHTS AND BODY WEIGHT GAINS

Body weight and body weight gain data are given in Table 2. Absolute body weights and body weight changes were similar between the permethrin and negative control groups throughout the study. Weight loss was observed in the positive control animals.

TABLE 2: Body weights and body weight gains (kg) of hens administered permethrin or a positive control[a]

Day of Study	0 mg/kg	2000 mg Permethrin/kg	4000 mg Permethrin/kg	500 mg TOCP/kg
0	2.41	2.44	2.53	2.41
7	2.36	2.53	2.55	2.30
14	2.53	2.53	2.61	2.03
21	2.47	2.52	2.62	1.85
28	2.58	2.51	2.57	-
38	2.54	2.44	2.57	-
42	2.53	2.41	2.59	-
Wt. change 0-21[b]	0.06	0.08	0.09	-0.56
Wt. change 21-42[b]	0.06	-0.11	-0.03	-

Data taken from Appendix 1, p. 8, MRID 00097246.
[a]Some values may be incorrect due to the poor copy quality of the report.
[b]Calculated by reviewer from group means.

D. FOOD CONSUMPTION

Food consumption was not measured.

E. NEUROTOXICITY ASSESSMENT

No signs of ataxia were observed in birds treated with the test article or the vehicle. Hens in the positive control group showed signs of slight ataxia to almost complete paralysis. Signs in these animals were first observed on study day 7 and increased in severity throughout the remainder of the study.

F. BIOCHEMICAL MEASUREMENTS

Acetylcholinesterase (AChE) and neurotoxic esterase (NTE) activities were not measured.

G. NECROPSY

At gross necropsy, no treatment-related abnormalities were seen in any animal in the test article or negative control groups. Hens in the positive control group appeared emaciated and had muscle atrophy in the legs.

H. NEUROPATHOLOGY

No treatment-related lesions were observed in the spinal cord or sciatic nerve from hens given the test article or the vehicle. An occasional degenerating fiber was seen in the sciatic nerve from animals in the control, low-, and high-dose groups and was considered a normal finding.

Degenerative changes were observed in the spinal cord and sciatic nerve of birds in the positive control group.

III. DISCUSSION

A. DISCUSSION

The objective of this study was to determine whether permethrin causes delayed neurotoxicity in the domestic hen. The low-dose was equal to and the high-dose exceeded the limit dose required in Subdivision F guidelines and were repeated for a second 21-day observation period.

No treatment-related mortalities or clinical signs of toxicity were observed. Body weights and body weight gains were not affected by treatment with the test article. Food consumption was not measured.

Ataxia was not seen in birds treated with the test article and no treatment-related lesions were observed on microscopic examination of the nervous tissues.

TOCP was used as a positive control in this study. Clinical signs and neurohistopathological lesions indicative of delayed neuropathy were observed in these birds. Therefore, the testing facility should have been able to detect neuropathy induced by the test article.

Therefore, under the conditions of this study, oral administration of permethrin up to 4000 mg/kg does not produce delayed neuropathy in the hen.

This study is classified **Acceptable/Guideline** and does satisfy the requirements for a delayed neurotoxicity study [OPPTS 870.6100 (81-7)] in hens.

B. STUDY DEFICIENCIES

AChE and NTE activities were not measured and the nervous tissues were not fixed *in situ* by perfusion. However, the high-dose greatly exceeded the limit dose and was administered on two separate occasions without resulting in clinical signs or ataxia. Therefore, the study is considered sufficient for determining the delayed neurotoxicity potential of permethrin.

R1

DATA EVALUATION RECORD

PERMETHRIN/109701
STUDY TYPE: METABOLISM AND PHARMACOKINETICS - RAT
[OPPTS: 870.7485 (§85-1)]
MRID 00089006, 00054719, 92142041, 92142042

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Toxicology and Hazard Evaluation Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 02-05

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Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

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DATA EVALUATION RECORD

TXR#: 0050649

STUDY TYPE: Metabolism - Rat OPPTS 870.7485 [§85-1)] OECD 417.

PC CODE: 109701

DP BARCODE: D269531
SUBMISSION NO.: S504352

TEST MATERIAL (PURITY): Permethrin

SYNONYMS: PP557; [3-phenoxybenzyl (±) cis:trans -2,2-dimethyl-3'(2,2-dichlorovinyl)-cyclopropane-1-carboxylate]

CITATION: 1) Bratt, H., Mills, I.H., and Slade, M. (1977). Permethrin: Tissue retention in the rat. CTL/P/352. ICI Central Toxicology Laboratory, Alderly Park, Macclesfield, Cheshire SK10 4TK, UK. MRID 00089006. Unpublished.

2) Batten, P. (1990). Phase 3 Summary of MRID 00089006 and 54720. Permethrin: Tissue retention in the rat. Report No. CTL/P/352. MRID 92142041. Unpublished.

3) Mills, I., Mullane, M. (1976). PP557: absorption and excretion in the rat. Report No. CTL/P/228, CTL study No. UR0015. MRID 00054719. Unpublished.

4) Batten, P. (1990). Phase 3 Summary of MRID 54719. PP557 (Permethrin): Absorption and excretion in the rat. Report No. CTL/P/228. MRID 92142042. Unpublished.

SPONSOR: ICI Americas, Inc.

EXECUTIVE SUMMARY: In a series of metabolism and disposition experiments (MRID 00089006, 00054719, and MRID 92142041 [summary of MRID 00089006], and MRID 92142042 [summary of MRID 00054719]), male and female Wistar-derived rats were placed on various oral treatment regimens with [^{14}C-alcohol]permethrin ([^{14}C-cyclopropyl]permethrin) or [^{14}C-acid]permethrin ([^{14}C-benzyl]permethrin). For MRID 00054719, [^{14}C-acid]permethrin (>98% purity, 53:47, cis trans ratio; no lot or batch no.) or [^{14}C-alcohol]permethrin (99% purity, 40.5:59.5 cis:trans ratio; no lot or batch no.) were diluted as needed with nonlabeled permethrin (93.6% purity, 40.5:59.5, cis:trans ratio; no lot or batch no.) and given by gavage to two male and two female rats at a dose of 6.5 mg/kg for quantitative and qualitative assessment of excretion. In MRID 00089006, tissue distribution and blood kinetics were assessed in male and

female Wistar-derived rats given repeated or single oral doses of [^{14}C-acid]permethrin (>98% purity; 53:47, cis:trans ratio; no lot or batch no.) or [^{14}C-alcohol]permethrin (99% purity, 38:62 cis:trans ratio; no lot or batch no.)

These studies provided information on the excretion and tissue burdens of permethrin in rats following single or multiple oral doses of either alcohol ([^{14}C-cyclopropyl]permethrin) or acid [^{14}C-benzyl]permethrin). Based upon a limited number of rats, overall recovery was 93.7% to 101% regardless of label position. Following a single oral dose of 6.5 mg/kg, most radioactivity (58-65%) from a single dose of the [^{14}C-alcohol] permethrin was eliminated via the urine over a 7-day period with much of the remainder (29-43%) being excreted in the feces. Urinary excretion of radioactivity following a single dose of [^{14} C-acid] permethrin was slightly less and fecal excretion correspondingly greater. Results of tissue distribution and autoradiographic experiments showed that most radioactivity was associated with adipose tissue and, initially, with the gastrointestinal tract and organs/tissue associated with excretory function. Following oral administration to rats, most permethrin-associated radioactivity appears to be excreted within 48 hours. Following multiple doses, radioactivity in adipose tissue appears to be greater for [^{14}C-alcohol] permethrin than for [^{14}C-acid] permethrin. This is also consistent with blood kinetics data showing lower radioactivity (C_{max}) in the blood of rats receiving [^{14}C-acid] permethrin. Upon cessation of dosing, radioactivity levels in adipose tissues declined. There was no attempt to identify the metabolites in these studies.

This metabolism study in the rat is classified **Unacceptable/Guideline** and does not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD 417] in rats. The unacceptability is the result of deficiencies in level of detail provided which prevent verification/validation of findings (e.g., unreadable data, environmental conditions not reported, no dose confirmation, no lot/batch numbers for the test article).

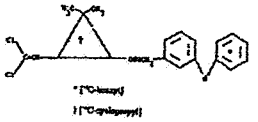
COMPLIANCE: The studies were conducted prior to implementation of GLP guidelines and, therefore, there was no claim regarding GLP compliance. Certification of Access to Raw Data and Data Confidentiality statements were signed subsequent to completion of the studies. provided.

I. MATERIALS AND METHODS:

A. MATERIALS:

1. Test compound:

Radiolabelled test material:	1) [^{14}C-cyclopropyl]permethrin *cis:trans* (40:60) ([^{14}C-alcohol]permethrin) 2) [^{14}C-benzyl]permethrin *cis:trans* (40:60) ([^{14}C-acid]permethrin)
Radiochemical purity	1) not provided 2) 99%
Specific Activity	59.7 mCi/mM
Lot/Batch #:	Not provided
Non-Radiolabelled Test Material:	Permethrin *cis:trans* (38.2:59.3)
Description:	
Lot/Batch #:	Not provided
Purity:	96 % a.i. [determined by GC] in MRID 00089006 93.6% (cis:trans ration of 40.5:59.5) in MRID 00054719
Contaminants:	None reported
CAS # of TGAI:	52645-53-1
Structure:	



2. Vehicle and/or positive control: Both corn oil and propylene glycol were used as vehicles in experiments reported in MRID 00089006. No additional information was provided.

3. Test animals:

Species:	Rat, male	
Strain:	Wistar-derived SPF	
Age/weight at study initiation:	Adults (195-205 g)	
Source:	Alderley Park ICI	
Housing:	Housed in groups of 5 in standard cages	
Diet:	Stock rat diet *ad libitum*	
Water:	Not specified	
Environmental conditions:	**Temperature:**	Not specified
	Humidity:	Not specified
	Air changes:	Not specified
	Photoperiod:	Not specified
Acclimation period:	No information provided	

4. Preparation of dosing solutions: With the exception of the vehicle used, no information was provided in MRID 00089006 regarding preparation of dosing solutions for the various treatment groups.

B. STUDY DESIGN AND METHODS:

1. Group arrangements

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The test groups for MRID 00089006 and 00054719 are shown in Table 1. No information was provided regarding selection procedures for the establishing the groups. These studies provided information on the excretion and tissue burdens of permethrin in rats following single or multiple oral doses of either alcohol ([^{14}C-cyclopropyl]permethrin) or acid [^{14}C-benzyl]permethrin).

TABLE 1: Dosing groups for pharmacokinetic studies for permethrin.

Test Group	Dose (mg/kg)	Number/sex	Remarks
1 preliminary	1.5 0.9	15♀ 15♂	[^{14}C-alcohol]Permethrin; distribution following repeated dosing over 3 wks and up to 2 wks post dose (MRID 00089006) [^{14}C-acid]Permethrin; distribution following repeated dosing over 3 wks and up to 2 wks post dose (MRID 00089006)
2 (tissue kinetics)	1.9	12♂	[^{14}C-alcohol]Permethrin; 12-day repeat dose; 2 rats sacrificed at 1, 5, 8, 15, 20, and 25 days following last dose; $t_{1/2}$ determination in adipose tissue; MRID 00089006
3 (metabolite ID)	5.1	6♂	[^{14}C-alcohol]Permethrin; metabolite identification at 24 hrs (4 rats) following single oral dose and at 4 days (2 rats) following 4 repeated doses; MRID 00089006
4 (tissue burden)	1	60♀	[^{14}C-alcohol]Permethrin; 11-wk daily oral dosing; tissue burdens at weekly intervals over 11 wks and for an additional 7 wks postdose; MRID 00089006.
5 (autoradiography)	6	3♂	[^{14}C-alcohol]Permethrin; autoradiographic analysis at 1, 25, and 96 hrs post dose
6 (blood kinetics)	1010	8♂ 8♂	[^{14}C-alcohol]Permethrin; blood kinetics over 24-hr period; single oral dose [^{14}C-acid]Permethrin; blood kinetics over 24-hr period; single oral dose
7 (absorption/excretion)	6.5 6.5	2♀ 2♂	[^{14}C-alcohol]Permethrin; excreta (expired air, urine, feces) collected at 24-hr intervals for 7 days MRID 00054719. [^{14}C-acid]Permethrin; excreta (expired air, urine, feces) collected at 24-hr intervals for 7 days MRID 00054719.

Data taken from pp. 304, MRID 00089006 and pp. 23-24, MRID 00054719.

2. **Dosing and sample collection:** No details were provided regarding dosing other than route (oral). Both alcohol (^{14}C-cyclopropyl) and acid (^{14}C-benzyl) labeled permethrin were used in most experiments.

Expired air: Expired air was collected every 24 hours over seven days (Exp. 7; MRID 00054719). $^{14}CO_2$ was trapped in 2N sodium hydroxide and aliquots analyzed for radioactivity.

Urine: Urine from rats in Exp. 7 (MRID 00054719) was collected over 24-hour periods for seven days. Aliquots of urine were analyzed for radioactivity by liquid scintillation counting (LSC).

Feces: Feces from rats in Exp. 7 (MRID 00054719) were collected at 24-hour intervals over seven days. Samples were homogenized in acetone, centrifuged, and analyzed for radioactivity.

Blood: Blood samples were collected over a 24-hour period from rats in Experiment Group 6. It appeared that blood was collected at 30 min, 1, 2, 4, 5, 6, 8, 10, 15, and 24 hours (no text designations were provided regarding collection times and graphic displays were unreadable). Blood was mixed with cellulose powder and combusted to $^{14}CO_2$ prior to LSC. Oxidation efficiency was determined and counting efficiency determined by external quench correction.

Tissues: Other tissues examined included adipose, liver, kidneys, and brain (Exp. 1), adipose (Exp. 2), and peri-renal fat, subcutaneous fat, liver, kidneys, and muscle (Exp. 4). Tissues were homogenized and duplicate aliquots combusted prior to radioactivity measurement by LSC. For isolation of residues in fat, adipose tissues were extracted with hexane and separated by thin-layer chromatography (TLC) using silica gel GF plates and hexane: diethyl ether (10:1, v/v) and chloroform:hexane:diethyl ether (8:3:1, v/v/v) solvent systems. Radioactive regions were located using x-ray film.

a. **Pharmacokinetic studies:** Absorption/elimination kinetics were determined for blood over a period of 24 hours. Blood samples were taken via the tail vein at time intervals indicated above.

b. **Metabolite characterization studies:** Metabolite analysis was limited to examining extracts from adipose tissue. Using TLC, comparisons were made to known standards and to ascertain isomer (cis:trans) ratios.

3. **Statistics**: No statistical analyses were described or reported.

II. RESULTS:

A. PHARMACOKINETIC STUDIES:

1. Preliminary experiment:

Experiment no. I (referred to as Preliminary in MRID 00089006) was designed to compare bioavailability and tissue distribution of permethrin with alternate radiolabels (acid vs alcohol). The results of this experiment are summarized in Table 2. [^{14}C- acid] permethrin appeared to exhibit somewhat lower bioavailability as shown by the reduced tissue burdens. Results of the absorption/excretion experiment (MRID 00054719) showed total recovery of administered radioactivity of 93.7% to 10 1 % regardless of label position (alcohol vs acid) although this was based upon only four rats per test article group.

2. **Absorption:** Absorption may be implied from expired air and urinary excretion data obtained from rats in Experiment 7 (MRID 00054719). Based upon these data, at least 50-65% of a single 6.5 mg/kg oral dose was absorbed (Tables 3 and 4). Biliary excretion experiments were not performed and, therefore, the portion of radioactivity recovered in the feces that

represented absorbed material could not be assessed. Limited kinetics data suggested that absorption was relatively rapid.

3. **Tissue distribution:** Experiments in MRID 00089006 focused on radioactivity burdens in various tissues, especially adipose tissue, of rats following various oral administration regimens. However, only a limited array of tissues/organs was examined. Results of Experiment 1 (3-week repeat dosing) are summarized in Table 2. While on treatment, tissue burdens varied slightly but did not decline. Slightly greater levels of radioactivity appeared in tissue of rats given the [^{14}C-alcohol] permethrin than the ^{14}C-acid] permethrin but tissue burdens in both groups declined to levels below detection limits upon removal from treatment. Levels in brain tissue remained low at all time points both during and after treatment. The highest levels of radioactivity among the tissues examined were associated with adipose tissues. During the 11 -week dosing regimen, peri-renal fat exhibited the greatest levels of radioactivity at Week 5 (equivalent to 2.05 µg/g). Levels in subcutaneous fat were similar. Radioactivity in liver, kidneys, and muscle were notably lower than either of the fat tissues (data for the 11-week study are not reproduced in this Data Evaluation Record). Tissue burdens in the 11-week treatment group declined to near detection limits by seven weeks following cessation of treatment.

TABLE 2. Distribution of radioactivity(µg/g tissue) in rat tissue/organs following 3-week administration of [^{14}C-alcohol]permethrin (1.5 mg/kg) or [^{14}C-acid]permethrin (0.9 mg/kg).

Tissue	Radioactivity dose recovered (µg/g tissue)[a]									
	[^{14}C-alcohol] permethrin			[^{14}C-alcohol] permethrin		[^{14}C-acid]permethrin			[^{14}C-acid]permethrin	
	During dosing			Postdose		During dosing			Postdose	
	Wk	Wk 2	Wk 3	Wk 1	Wk 2	Wk 1	Wk 2	Wk 3	Wk 1	Wk 2
Adipose	0.72	0.84	0.94	1.10	0.53	0.33	0.10	0.72	0.27	0.08
Liver	0.17	0.19	0.13	0.08	<0.08	0.22	<0.08	0.08	<0.08	<0.08
Kidney	0.24	0.13	0.12	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08
Brian	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08

[a]Neither percent of dose nor total tissue weights were provided in the study report (MRID 00089006). Data taken from Table 1, p.11, MRID 00089006.

Results of autoradiography experiments (Experiment no. 5, MRID 00089006) showed that most radioactivity up to 24 hours postdose was associated with the gastrointestinal tract although notable levels were observed in the liver, kidneys, skin, fat, and lungs. At 96 hours, all tissue regions showed marked decreases in radioactivity.

4. **Excretion:** Excretion data from MRID 00054719 are summarized in Tables 3 and 4. Approximately 58-65% of a single 6.5 mg/kg oral dose of [^{14}C-alcohol] permethrin was excreted in the urine over the 7-day experimental period, most (~75%) occurring during the first 24 hours (Table 3). There did not appear to be gender-related differences, although the sample size would not allow valid statistical analysis. Approximately 29-43% of the dose was recovered in the feces over seven days. Individual variability prevented meaningful gender comparisons. When compared to the alcohol label, excretion patterns for rats given

[^{14}C-acid] permethrin revealed slightly less urinary excretion and correspondingly greater fecal excretion. Although the study author noted that 1.6% of the administered dose was excreted via expired air, no data were provided to support the contention.

TABLE 3. Time-course of excretion of radioactivity (% of dose) in rats treated with [^{14}C-alcohol]permethrin[a]

Matrix/Time (days)	6.5 mg/kg	
	Males	Females
Urine		
1	48.5	43.1
2	13.1	12.2
3	1.5	1.3
4	0.6	0.4
5	0.6	0.5
6	0.4	0.4
7	0.2	0.4
Total	64.9	58.1
Feces		
1	12.0	27.8
2	10.2	12.9
3	4.6	2.0
4	0.7	0.4
5	0.9	0.1
6	0.4	0.02
7	0.1	0.08
Total	28.8	43.3

[a] Average of two rats/sex
Data taken from Table 1, p. 26, MRID 00054719

TABLE 4. Time-course of excretion of radioactivity (% of dose) in rats treated with [^{14}C-acid]permethrin[a]

Matrix/Time (days)	6.5 mg/kg	
	Males	Females
Urine		
1	41.6	41.6
2	7.3	6.7
3	1.5	0.8
4	0.5	0.4
5	0.05	0.1
6	0	0
7	0	0
Total	51	49.6
Feces		
1	34.8	30.3
2	9.8	17.3
3	2.2	1.3
4	0.7	0.5
5	0.2	0.3
6	0.5	0.3
7	0.1	0.3
Total	48.2	50.2

[a] Average of two rats/sex
Data taken from Table 2, p. 27, MRID 00054719

B. METABOLITE CHARACTERIZATION STUDIES: TLC analysis of adipose tissue extracts revealed that the cis:trans ratio was 1:0.36 versus the ratio of 1:1.50 in the dosing solution. These data were not presented or were possibly available in an unreadable graph in the study report (MRID 00089006) and, therefore, could not be verified.

C. PHARMACOKINETIC STUDIES: Results of Experiment 6 (MRID 00089006) revealed that [^{14}C-alcohol] permethrin (i.e., ^{14}C-cyclopropyl] permethrin) was more rapidly and extensively absorbed into the blood than was [^{14}C-acid] permethrin (i.e., [^{14}C-benzyl] permethrin) following oral administration to rats. Peak concentration (C_{max} = 0.34 µg/g blood) was attained at about 1.5 hour for [^{14}C-alcohol] permethrin. Blood levels declined rapidly to approximately 0.05 µg/g blood at 15 hours. For [^{14}C-acid] permethrin, the C_{max} of ~0.05 /µg/g blood was attained at about 3 hours with only slight fluctuation to 10 hours after which levels remained at approximately 0.03 µg/g to termination at the 24-hour time point.

III. DISCUSSION AND CONCLUSIONS:

A. INVESTIGATORS' CONCLUSIONS: In a series of metabolism and disposition experiments (MRID 00089006, MRID 00054719, MRID 92142041 [summary of MRID 00089006], and MRID 92142042 [summary of MRID 00054719]), male and female Wistar-derived rats were placed on various oral treatment regimens with [^{14}C-alcohol]permethrin ([^{14}C-cyclopropyl]permethrin) or [^{14}C-acid]permethrin ([^{14}C-benzyl]permethrin). For MRID 00054719, [^{14}C-acid]permethrin (>98% purity, 53:47, cis trans ratio; no lot or batch no.) or [^{14}C-alcohol]permethrin (99% purity, 40.5:59.5 cis:trans ratio; no lot or batch no.) were diluted as needed with nonlabeled permethrin (93.6% purity, 40.5:59.5, cis:trans ratio; no lot or batch no.) and given by gavage to two male and two female rats at a dose of 6.5 mg/kg for quantitative and qualitative assessment of excretion. In MRID 00089006, tissue distribution and blood kinetics were assessed in male and female Wistar-derived rats given repeated or single oral doses of [^{14}C-acid]permethrin (>98% purity; 53:47, cis:trans ratio; no lot or batch no.) or [^{14}C-alcohol]permethrin (99% purity, 38:62 cis:trans ratio; no lot or batch no.)

One study (MRID 00054719) reported that urinary and fecal excretion, respectively, accounted for 50-62% and 36-49% of a single 6.5 mg/kg oral dose of permethrin given to rats. Minor amounts (1.6% of dose) were recovered in expired air. The majority of urinary excretion occurred within 24 hours and most fecal excretion occurred during the first 48 hours. There were no significant differences that could be attributed to gender although urinary excretion of [^{14}C- acid] permethrin was slightly less and fecal excretion correspondingly greater than for the [^{14}C- alcohol]permethrin. In the tissue retention study (MRID 00089006), in which rats were given daily1 mg/kg oral doses of either [^{14}C-alcohol]permethrin (11 -week treatment) or [^{14}C-acid]permethrin for 3 weeks, the half-life in adipose tissue was 18 and 7 days, respectively. Maximum concentration of radioactivity in adipose tissue was 2.05 µg eq./g for the alcohol label and 0.72 µg eq./g for the acid label. Administered radioactivity was totally eliminated at 7 weeks (alcohol label) and 2 weeks (acid label). Whole-body autoradiographic experiments affirmed that most radioactivity was associated the gastrointestinal tract and organs/tissue associated with excretion. The study reported that uptake into the blood was rapid (1.5 hours) with a C_{max} of 0.34 ug eq/g and $t_{1/2}$ of 7 hours for the alcohol label. For the acid label, blood concentrations never exceeded 0.05

ug eq/g. Based upon analysis of adipose tissue residues, the study authors noted the occurrence of a biotransformation-mediated shift in cis-trans ratio.

B. **REVIEWER COMMENTS:** These studies provided information on the excretion and tissue burdens of permethrin following single or multiple oral doses to rats. For most experiments, rats were dosed with alcohol (^{14}C-cyclopropyl) or acid (^{14}C-benzyl) labeled permethrin. Based upon a limited number of rats, overall recovery was an acceptable 93.7% to 101 % regardless of label position. Following a single oral dose of 6.5 mg/kg, most radioactivity (58-65%) from a single dose of the [^{14}C-alcohol] permethrin was eliminated via the urine over a 7-day period with much of the remainder (29-43%) being excreted in the feces. Urinary excretion of radioactivity following a single dose of [^{14}C-acid]permethrin was slightly less with fecal excretion correspondingly greater. Results of tissue distribution and autoradiographic experiments showed that most radioactivity was associated with adipose tissue and, initially, with gastrointestinal tract and organs/tissue associated with excretory function. The study author's conclusions and interpretations were consistent with the data provided in the study reports. Following oral administration to rats, permethrin-associated radioactivity appears to be excreted within 48 hours. Following multiple doses, radioactivity in adipose tissue appears to be greater for [^{14}C-alcohol] permethrin than for [^{14}C-acid] permethrin. This is also consistent with blood kinetics data showing lower radioactivity (C_{max}) the blood of rats receiving [^{14}C-acid] permethrin. Upon cessation of dosing, radioactivity levels in adipose tissues declined. Based upon the available data it is not possible to ascertain if the shift in the cis:trans ratio observed in adipose tissue was the result of differential absorption or isomer-specific biotransformation.

This metabolism study in the rat is classified **Unacceptable/Guideline** and does not satisfy the guideline requirement for a metabolism study [OPPTS 870.7485, OECD 417] in rats. The unacceptability is the result of deficiencies in level of detail provided which prevent verification/validation of findings (e.g., unreadable data, environmental conditions not reported, no dose confirmation, no lot/batch numbers for the test article).

C. **STUDY DEFICIENCIES:**

No data were provided to validate excretion via expired air. The studies were not conducted under GLP guidelines. LSC efficiencies were only 70-90% and some data were unreadable and, therefore, not verifiable. TLC data regarding adipose tissue extracts for cis:trans ratio analyses were not presented or were possibly available in an unreadable graph in the study report (MRID 00089006) and, therefore, could not be verified. Examination of tissue/organs was limited to adipose tissue, kidney, liver, and brain.

Additionally, information about the test article source (e.g., batch nos.) were not provided and environmental conditions were not specified. Dose preparation details and dose confirmation data were not provided.

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609