(EXCERPT 005/00)

EPA: 68-02-4225
TASK: 029-81
January 14, 1986

DATA EVALUATION RECORD

CYHALOTHRIN

Teratogenicity Study in Rats

STUDY IDENTIFICATION: Killick, M. E. Cyhalothrin: Oral (gavage) teratology study in the rat. (Unpublished study No. RR 0170 and report No. 2661-72/208 prepared by Hazleton Laboratories Europe Ltd., England, for Imperial Chemical Industries Ltd., England; dated June 1981.) Accession No. 073206.

APPROVED BY:

I. Cecil Felkner, Ph.D.
Department Manager
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Date: 1-14-86

1. CHEMICAL: Cyhalothrin; (R,S)α-cyano-3-phenoxybenzyl (±)-cis-3,3 (Z-2-chloro-3,3,3-trifluoroprop-1-en)-2,2-dimethylcyclopropane carboxylate; Grenade.

2. TEST MATERIAL: Cyhalothrin (batch No. 005, ICI code No. Y00102/010/005) was a brown viscous fluid described as a technical grade pyrethroid mixture containing 89.25 percent w/w cyhalothrin.

3. STUDY/ACTION TYPE: Teratogenicity study in rats.

4. STUDY IDENTIFICATION: Killick, M. E. Cyhalothrin: Oral (gavage) teratology study in the rat. (Unpublished study No. RR 0170 and report No. 2661-72/208 prepared by Hazleton Laboratories Europe Ltd., England, for Imperial Chemical Industries Ltd., England; dated June 1981.) Accession No. 073206.

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7. CONCLUSIONS:

A. We assess that the NOEL and LOEL for maternal toxicity are 10 and 15 mg/kg/day, respectively, based on decreases in gestational body weight gains and food consumption reported for the 15 mg/kg/day group. The NOEL for embryolethality is 15 mg/kg/day. The NOEL for fetotoxicity could not be determined due to the presence of minor developmental variations in all dosage groups; therefore, 5 mg/kg/day, the lowest tested dose, is assessed as the LOEL for fetotoxicity.

B. No compound-related teratogenic effects were noted in the presented data; however, the teratogenic potential of cyhalothrin on cardiac and thoracic structures of rat fetuses could not be assessed since the intracardiac injections used in fetal sacrifices may have negatively affected the accuracy of cardiovascular examinations by masking the visualization of cardiac septal defects, valve malformations, pericardial hemorrhages, and various other malformations or lesions in the mediastinum of fetuses.

The registrant should submit data indicating that the method of intracardiac injection used in this study did not affect the findings of the cardiothoracic examinations. In addition, the registrant should submit historical control data (from 1979-1983) on the litter incidence of fetuses with dilated ureters. The classification of this study is pending receipt of the above information.

8. RECOMMENDATIONS: For future studies, we recommend that fetuses be sacrificed by carbon dioxide inhalation or intraperitoneal injection and not by intracardiac injection.

9. BACKGROUND: A range-finding study was conducted at Hazleton Laboratories, Europe (report No. 2586-72/207), to determine dose levels for the teratogenicity study; however, the author did not include details or results from this range-finding study in the teratogenicity study report.

Item 10--see footnote 1.

11. MATERIALS AND METHODS (PROTOCOLS):

A. Materials and Methods: (See Appendix A for details.)

1. Test Material: .Cyhalothrin was described as a brown, viscous fluid consisting of 89.25 percent active ingredient. The test material was supplied by Imperial Chemical Industries Ltd.

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[1] Only items appropriate to this DER have been included.

under the code No. Y00102/010/005. Corn oil was used as the vehicle and control substance. Dosage formulations were prepared once (3 days before the initiation of dosing), divided into daily aliquots, and stored at room temperature until used. The final dosages of 0, 5, 10, and 15 mg/kg/day were achieved by mixtures containing 0.00, 0.56, 1.12, and 16.8 mg of test material (adjusted for purity) per milliliter. Dosing and control volumes were adjusted to 10 mL/kg body weight.

Dosages were based on maternal body weights recorded on gestation day (GD) 6. These dosages were reduced for animals whose body weights decreased below their respective reference level of GD 6, but were not increased to compensate for body weight gains above their reference level.

2. Test Animals and Test System: Specific pathogen-free CD rats were obtained from Charles River Ltd., Kent, England. Animals were examined upon arrival by a veterinarian to assure their suitability for the study. Females were described as being within 227-270 g, and males were reported to be sexually mature. Animals were acclimatized for 17 days and were vaccinated against Sendai virus during this period. Ninety-six females were mated with males on a 2:1 basis; a total of 24 females were assigned to each group. All mated females were dosed from GD 6 through 15 and sacrificed on GD 20.

3. Parameters Measured: Chemical analyses were conducted on samples of dose formulations obtained on the day of preparation and 19 days later.

All animals were observed at least once daily to determine their health status and to record clinical signs of toxicity. Mortality checks were performed twice daily. Maternal body weights were recorded on GD 0, 6 through 15, 18, and 20. Maternal food consumption was recorded on GD 0, 3, 6, 9, 12, 15, 18, and 20. Necropsies were conducted on pregnant animals at GD 20; at this time, gross maternal findings, gravid uterine weight, and number of corpora lutea were recorded. In addition, the number, type, and location of implantations within uteri were recorded.

Fetal body weight, crown-to-rump length, and sex were determined after sacrificing the fetuses with intracardiac injections of Euthatal. Subsequently, all fetuses were examined for gross external abnormalities. Two-thirds of the fetuses from each litter were dissected and examined for visceral abnormalities. Eviscerated fetuses were macerated, stained with Alizarin Red, and examined for skeletal abnormalities. Approximately one-third of the fetuses were fixed in Bouin's fluid and examined by a modification of Wilson's method.

12. REPORTED RESULTS:

A. Test Material: Gas chromatographic analyses were performed at the time dosage formulations were prepared and at the end of the dosing period. Results from these analyses indicate that all formulations were within 104-128 percent of target concentrations and that the test material was stable during the entire dosing period.

B. Maternal Effects: No mortalities were reported for any group. Two animals in the 15 mg/kg/day group exhibited uncoordinated movements of the limbs. No other compound-related clinical findings during pregnancy or gross findings during necropsies were noted.

The author reported that the reduction in mean body weight gain for pregnant animals in the 15 mg/kg/day group was statistically significant, when compared with controls, for the dosing period and for the entire length of gestation. Body weight gains in all other groups were comparable (Table 1). The food consumption of animals in the high-dose group was also significantly reduced (during GD 6-12) compared with controls, while no compound-related effects were noted in the other groups (Table 2).

Data from uterine parameters indicated that the percentage of pregnant animals was comparable for all groups (Table 3), but that the reduction in adjusted body weight gain (calculated by subtracting gravid uterine weight from gestational body weight gain) in the high-dose group was statistically significant (Table 4).

C. Embryonic/Fetal Effects: No compound-related effects were reported for intrauterine deaths. The mean number, body weight, and sex ratio of fetuses from all groups were comparable (Table 5).

Major malformations were noted only in one litter (from the 10 mg/kg/day group); therefore, the study author considered them as incidental (not compound-related) findings. Also considered as incidental was the slight increase in the incidence of minor defects in the high-dose group. Finally, the incidence of skeletal variants was reportedly comparable for all groups (Table 6).

13. STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:

A. The study author concluded that the only maternal effects associated with cyhalothrin were decreases in body weight gains and reductions in food consumption in the high-dose animals. These effects indicated that 15 mg/kg/day elicited maternal toxicity in pregnant rats. However, no compound-related effects resulted in any aspect of fetal development, even at the highest dose tested.

B. A quality assurance statement was signed and dated on July 3, 1981.

TABLE 1. Effects of Cyhalothrin on Maternal Body Weights and Body Weight Gains During Gestation in Rats

Gestation Day	Maternal Body Weight (g) Dose (mg/kg/day) 0	5	10	15
0	249	248	249	251
6	277	273	274	278
7	275	271	271	267
8	282	275	276	271
9	285	280	279	276
10	290	285	284	279
11	298	292	291	284
12	301	296	295	290
13	305	299	302	295
14	311	305	306	302
15	317	312	311	306
18	349	341	346	337
20	351	346	350	341

Study Period	Maternal Body Weight Gain (g) Dose (mg/kg/day) 0	5	10	15
0 - 6 (predosing)	28	25	25	27
6-15 (dosing)	40 [14.4%][a]	39 [14.3%]	37 [13.5%]	28 [10.1%]**
15-20 (post-(dosing)	34	34	39	35
0-20 (gestation)	102 [41.0%][a]	98 [39.5%]	101 [40.6%]	90 [35.9%]*

*Statistically different from control value ($p \leq 0.05$).

**Statistically different from control value ($p \leq 0.01$).

[a][ ], percent change based on body weight at the start of the period.

TABLE 2. Effects of Cyhalothrin on Maternal Food Consumption (g/day) During Gestation in Rats

Gestation Days	Dose (mg/kg/day) 0	5	10	15
0- 3	25.0	23.8	25.0	24.9
3- 6	24.3	23.7	23.9	24.3
6- 9	20.7	18.6	18.7	15.9**
9-12	22.8	21.1	21.6	20.7*[a]
12-15	24.9	22.3	23.5	22.6
15-18	26.1	27.9	26.5	25.9
18-20	16.9	15.3	15.5	15.1

* Statistically different from control value ($p < 0.05$), according to study author's calculations; [a]however, the reviewers did not find this parameter to be different from control by ANOVA and Duncan's test.

** Statistically different from control value ($p < 0.01$).

TABLE 3. Effects of Cyhalothrin on Fertility Incidences in Rats

Parameter	Dose (mg/kg/day) 0	5	10	15
No. mated	24	24	24	24
No. pregnant at GD 20	23	24	24	24
% pregnant at GD 20	96	100	100	100

TABLE 4. Effects of Cyhalothrin on Adjusted[a] Mean Maternal Body Weight and Gravid Uterine Weight in Rats

Parameter	Dose (mg/kg/day) 0	5	10	15
Body weight (g) at GD 20	351	346	350	341
Gravid uterine weight (g)	70	67	74	71
Adjusted body weight (g)[a] at GD 20	281	279	276	270
% adjusted gestational body weight gain	12	13	11	8**

[a] Calculated by subtracting gravid uterine weight from maternal body weight on GD 20.

**Statistically different from control value ($p \leq 0.01$).

TABLE 5. Effects of Cyhalothrin on Group Mean Reproductive Indices in Rats

Parameter	Dose (mg/kg/day)			
	0	5	10	15
No. corpora lutea/female	14.7	15.3	15.3	15.5
No. implantations/litter	13.4	13.0	14.2	13.7
% preimplantation loss	8.8	14.9	7.6	11.8
No. intrauterine deaths/ litter	0.48	0.58	0.25	0.25
% postimplantation loss	3.6	4.5	1.8	1.8
Live fetuses/litter	13.0	12.5	13.9	13.4
Mean fetal weight (g)	3.7	3.7	3.7	3.7
Fetal male/female ratio	0.86	1.03	0.88	0.88

TABLE 6. Effects of Cyhalothrin on the Percentage of Malformations and Variations in Rat Fetuses

Parameter (% Fetuses Affected)	Dose (mg/kg/day) 0	5	10	15
1. External and Visceral Malformations				
Major	0.0	0.0	1.5	0.0
Minor	7.4	14.4	9.0	10.6
2. Skeletal Malformations				
Major	0.0	0.0	1.3	0.0
Minor	15.9	16.6	16.3	20.0
3. Variations	59.9	65.4	54.5	56.4

14. REVIEWERS' DISCUSSION AND INTERPRETATION OF STUDY RESULTS:

A. 1. Maternal Effects: The test material was associated with maternal toxicity (decreased body weight gains during gestation, decreased adjusted body weight gains, and reduced food consumption) at the highest dose tested. Review of the data presented for animals in the other dosage groups revealed that there were no compound-related effects.

2. Embryonic/Fetal Effects: No compound-related effects were noted in the mean group number of pre- and postimplantation losses and in the number, size, weight, and sex ratio of fetuses. However, slight increases (which were not statistically significant) in the fetal and litter incidences of several skeletal and visceral variations (including decreases in skeletal ossification, dilations of ureters, etc.--see Table 7) suggest that the test material may have been fetotoxic even at the lowest dose level tested.

No clear pattern of compound-related malformations was noted in the data presented; however, the methods implemented in this study may have precluded a conclusive examination of cardiac and mediastinal structures in fetuses (see section 14C).

B. The following are differences between the reviewers' and study author's conclusions:

1. We assess that the increases in the incidence of developmental variations noted at all dosage levels are indicative of mild fetotoxic effects, whereas the study author considered these findings as incidental and not compound related.

2. Due to the deficiencies in methodology (see section 14C) we assess that the data in this study are inconclusive; hence, we cannot rule out the possibility that compound-related cardiac and thoracic malformations may have been present, but not noted.

C. The following deficiency in study design and conduct has negatively affected the scientific validity of the study:

The procedure of intracardiac injection is considered unacceptable due to the physical perforation of cardiac structures and to the possible distortion of cardiac and major vessel anatomy produced by the volume of fluid injected into the cardiac chambers. The anatomic disruptions resulting from these procedures may have negatively affected the accuracy of cardiovascular examinations by masking the visualization of cardiac septal defects, valve malformations, pericardial hemorrhages, and various other malformations or lesions in the mediastinum of fetuses.

TABLE 7. Effects of Cyhalothrin on the Incidence of Selected Variations in Fetal Rats

Parameter (% Fetuses Affected)	Dose (mg/kg/day) 0	5	10	15
Fetuses with dilated ureter	1/298	16/299	3/334	12/322
% affected	0.3	5.4	0.9	3.7
Litters with dilated ureter	1/23	4/24	3/24	6/24
% affected	4.2	16.7	12.5	25.0
Fetuses with unossified hyoid	4/207	9/205	15/233	10/220
% affected	1.9	4.4	6.4	4.5
Litters with unossified hyoid	4/23	7/24	7/24	5/24
% affected	17.4	29.2	29.2	20.8

Item 15—see footnote 1.

16. CBI APPENDIX:

Appendix A, Materials and Methods, CBI pp. A4-A22.