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

SEP 30 1988

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: EPA ID No. 52904-C - Lindane - Review of a Subchronic (Scheduled for 13 Weeks) Dermal Toxicity Study Which Was Terminated After 12 Days Because of High Mortality

TOX Chem No.: 527
TOX Proj. No.: 8-0951
Record No.: 225486

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The Centre International d'Etudes du Lindane (CIEL), through its attorneys, McKenna, Conner and Cuneo, has submitted a report on a 90-day subchronic dermal toxicity study which was conducted at the request of the Agency to help assess for the potential of lindane to affect the kidney. This study was terminated after only 12 days of dosing because of severe dermal lesions which led to death (mostly moribund sacrifices).

The study was reviewed by Toxicology Branch (TB). Refer to the DER attached.

TB has discussed the issue of conducting a 90-day subchronic dermal toxicity study with the registrants (refer to J. Doherty memorandum dated June 16, 1988 summarizing the meeting between TB and CIEL, and a second memorandum also from J. Doherty and dated July 7, 1988), concerning issues related to lindane effects in the kidney and the protocol for

a repeat subchronic dermal toxicity study in rabbits. The CIEL plans to start a second 90-day dermal toxicity study in order to comply with the Agency's request. The second study is expected to be submitted to the Agency in December 1989.

Attachment

Reviewed By: J.D. Doherty
Section I, Toxicology Branch - Insecticide, Rodenticide Support
Secondary Reviewer: E.R. Budd, Section Head
Section I, Toxicology Branch - Insecticide, Rodenticide Support

DATA EVALUATION REPORT

Study Type: 82-3, Subchronic (90-Day) Dermal - Rabbits

TOX Chem. No.: 527

MRID No.:

Accession Number: 406604-01

Test Material: Lindane (≥ 99.5% pure, Batch #DA 433)

Synonyms: gamma-hexachlorocyclohexane

Study Number: 5675-580/3

Sponsor: Centre International d'Etudes du Lindane (CIEL)

Testing Facility: Hazleton UK

Title of Report: Lindane: 13-Week Dermal Toxicity Study (With Interim Kill and Recovery Period) in the Rabbit

Author: D. Brown, D.Phil.

Report Issued: October 2, 1987 (Study Termination Date)

Conclusions:

The study was terminated after 12 days due to severe infections (dermal). More moribund sacrifices were noted in lindane-treated rabbits than in controls. The possible effects of lindane include rapid breathing (at 60 and 400 mg/kg), convulsions (400 mg/kg/day females), weight loss (400 mg/kg/day females), increased liver weight (males and females, 400 mg/kg/day), and increased kidney weight (males, 400 mg/kg/day).

Classification: Core-INVALID

Special Review Criteria (40 CFR 154.70): N/A

Quality Assurance Statement:

No Quality Assurance statement or information was provided. A Quality Assurance statement page was submitted blank.

REVIEW

[Note: Since this study was terminated after 12 days and did not complete the 90-day period required for subchronic testing, this review will address only selected issues.]

The basic design of this study consisted of four groups of 40 male and 40 female rabbits (New Zealand White obtained from Ranch Rabbits Ltd., Crawley) which were dosed with either 0, 10, 60, or 400 mg/kg/day of lindane.

Upon receipt at the Hazleton facility, the rabbits were examined and it was determined that many were not acceptable because of ill health. At least 100 rabbits were sacrificed or replaced prior to the start of the study. The causative agent responsible for the deaths was not determined or at least not reported in the study report dealing with assessing the health of the rabbits prior to initiation. After allocation to treatment groups, the rabbits were housed individually in anodized, stainless steel cages with removable, perforated aluminum flasks. It was stated that "during the study the cage walls, floors and food hoppers were cleaned as often as was necessary to maintain hygienic conditions. Excrement trays beneath the cages were flushed at least once daily."

The test material was applied as a suspension in 5% aqueous carboxymethyl cellulose which was prepared daily. The suspension was applied over an area of approximately 3 inches in diameter, on the back of each rabbit by means of a plastic disposable syringe. The first applications were made with a dosing volume of 4 mL/kg but this was determined to be too large a volume and was changed to 2 mL/kg of higher concentrations of lindane suspension. The test material was administered once daily, 5 consecutive days per week, and kept in contact for 6 hours per day. The control rabbits were dosed with 2 mL/kg of carboxymethyl cellulose. The test and control suspensions were kept in place by a patch assembly consisting of a thin layer of porous gauze, which was heat welded to an aluminum foil backing. In turn, the patch was held in place by a canvas jacket, which covered the upper torso of the rabbit. The area of application was tattooed in order to assure that the test article was applied to the same area every day. The dorsal surface of each rabbit was scheduled to be shaved at least once a week.

One unusual aspect of this study was that the high-dose group rabbits received a single dose of Complan on day 8 of the study. The reason for administration of this substance was not specified. It is considered by TB to be scientifically unsound to administer a substance to the high-dose test group only and not the animals in the other dose groups.

Results:

1. Analysis of Test Formulations - The week 1 preparations of lindane in 5% carboxymethyl cellulose were determined to be 91, 83, and 85 percent of the nominal concentrations of 10, 60, and 400 mg/kg, respectively.

2. Mortality - The study was terminated after 12 days of dosing due to both high mortality in the rabbits and general poor appearance which required sacrificing the rabbits. The following table illustrates the number of deaths or moribund sacrifices. A total of three rabbits were found dead, the others (50) were sacrificed moribund.

Group	Males	Females
Control	1	0
10 mg/kg/day	10	8
60 mg/kg/day	2	12
400 mg/kg/day	11	9

It is evident that the rabbits dosed with lindane had many more deaths than the control rabbits dosed with carboxy-methyl cellulose only. There was, however, no dose-related increase in deaths over the broad range of 10 to 400 mg/kg/day.

Attempts were made to replace the rabbits that died, but four of these rabbits were also sacrificed after developing severe skin lesions.

3. Clinical Signs and Reactions to Treatment - The rabbits from all dosed groups were reported as developing dermatitis and subcutaneous sores and abscesses on their backs. The following table depicts the proportion (as percent) of rabbits with skin lesions. The infections were stated as being "restricted to the clipped area on the rabbits' back."

Group	Males	Females
Control	10	30
10 mg/kg/day	43	60
60 mg/kg/day	23	57
400 mg/kg/day	47	77

The report states that the spread of infections "had been restricted by a more intense washing procedure and the killing of animals carrying heavy infections." This statement is hard for TB to accept because since the study was terminated on day 12, there was only a limited time for recovery or healing to take place.

In addition to the dermal reactions, a few rabbits developed signs of rapid breathing (in the mid- and high-dose groups, males and females) and convulsions (females, high-dose group).

4. Body Weight - At day 7, male body weight was 11 percent below the control group for the high-dose group. Female body weights were -2, -2, and -11 percent for the low-, mid- and high-dose test groups, respectively.

5. Gross Necropsy - Aside from dermal reactions, there were no evident effects of lindane treatment.

6. Organ Weights - Only the liver and kidney were weighed. These organs were reported as being slightly elevated for the high-dose group males and females for relative weights. For example, male relative liver weights were 12.7 percent and females were 11.1 percent. Relative kidney weights were 6.4 percent higher for males and 2.4 percent higher for females.

7. Proof of Absorption - Five male and five female rabbits from each dose group were analyzed for lindane content in the liver, kidney, and adipose tissue. No quantifiable lindane was reported as being found in any of the samples from the control rabbits. The following table (copied from the study report) depicts the results of the analysis of the liver, kidney, and adipose tissue for lindane.

Group Number		Kidney (ug/g) Male	Kidney (ug/g) Female	Liver (ug/g) Male	Liver (ug/g) Female	Adipose(ug/g) Male	Adipose(ug/g) Female
1	Mean	< 0.20	< 0.20	< 0.20	< 0.20	< 1.00	<1.00
	S.D.	-	-	-	-	-	-
2	Mean	< 0.20	< 0.20	0.54	0.34	7.14	6.74
	S.D.	-	-	0.200	0.118	1.704	2.154
3	Mean	0.59	0.42	1.71	0.92	40.5	28.8
	S.D.	0.268	0.166	0.612	0.686	6.54	10.47
4	Mean	2.85	2.78	4.34	2.18	203	193
	S.D.	1.967	1.693	1.376	0.949	80.0	135.7

The above table does not indicate that lindane accumulates in the kidney, a recognized target organ for this chemical. Lindane, however, appears to accumulate in the adipose tissue. It is also noted that the concentration of lindane in the male and female kidneys is approximately equal while the male liver samples contain about twice as much at all three dose levels. This is an interesting finding because the kidneys of males are recognized as being more susceptible to the effects of lindane than the female kidneys.

Conclusion and Discussion:

As a subchronic (90-day) dermal toxicity study, this study is INVALID. The development of dermatitis and skin infections which became of marked incidence by the end of the first week of dosing led to termination of the study after 12 days. Although the study report maintains that lindane treatment alone was not responsible for the dermatitis, there were, however, many more deaths among the lindane-treated rabbits than among the controls. It is possible that lindane treatment (even at dose-levels as low as 10 mg/kg/day) renders the rabbits more susceptible to development of severe lesions due to dermal infection.