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WASHINGTON, D.C. 20460

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HEALTH EFFECTS DIVISION
SCIENTIFIC DATA REVIEWS
EPA SERIES 361

JUL 13 1995

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM:

Subject: 010182-GAO Pirimicarb Technical: Review of Data in Support of Registration of Pirimicarb (MRID Nos.:43496001 to 43496008)

TOX 670A

P.C.#: 106101
Submission #: S479870
Project No.: D210945
EPA ID#: 010182-GAO Pirimicarb Technical

From: Guruva B. Reddy, D.V.M., Ph. D.
Section 4
Toxicology Branch I
Health Effects Division (7509C)

To: Robert Forrest/Beth Edwards
Project Manager 14
Registration Division (7505C)

Thru: John Doherty, Ph.D. 6/22/95
Acting Section Head
Section 4, Toxicology Branch I
Health Effects Division (7509C) 6/20/95

I. **CONCLUSIONS:**

The subacute dermal toxicity study (MRID No.:43496003) in rabbits is **inadequate and not upgradable** because of several deficiencies (refer to DER). The series 81-6 dermal sensitization study (MRID No.:43496002) demonstrated that pirimicarb is a moderate skin sensitizer. Products containing pirimicarb must include a dermal sensitization precautionary statement. The series 81-3 acute inhalation toxicity study (MRID No.: 43496002) was determined to be ACCEPTABLE. The mutagenicity/genotoxicity studies (MRID Nos.: 43496004 to 43496008) were determined to be ACCEPTABLE (refer to memo from Dr. Irv Mauer). An additional mammalian cell gene mutation study is required to fulfill the requirements of genotoxicity battery.

II. **ACTION REQUESTED:**

Zeneca Inc., has submitted acute inhalation (MRID No.:43496001), skin sensitization (MRID No.:43496002), subacute dermal (MRID

No.:43496003) and five mutagenicity studies (MRID Nos.:43496004 to 43496008) in support of registration of technical pirimicarb. This is in response to TB-I evaluation of data base dated May 18, 1994.

III. STUDIES REVIEWED:

STUDY/CLASSIFICATION	TB-I COMMENTS
81-3 Acute inhalation LC_{50} Species: rat Zeneca Central Tox. Lab., UK. HR2247; 10/28/94 MRID No.:43496001 **core-Acceptable** **Tox. Category - III**	In a acute inhalation toxicity study, groups (5/sex/dose) of Wistar-derived male and female rats were exposed nose-only to gravimetric concentrations of 410, 746 or 1205 µg/l pirimicarb with MMAD of 3.02 ± 2.03 to 3.46 ± 1.89 for 4 hours (MRID No.: 43496001; Study No.: HR2247). One high-dose male and three females died during the exposure. Six high-dose and one mid-dose animals were sacrificed in moribund condition. Clinical signs included salivation, slow deep respirations and auditory hypoesthesia. Necropsy of the dead and terminal sacrifice were unremarkable. The calculated LC_{50}, based on the mortality was 948 µg/l for males (95% C.I. 746 to 1047) and 858 µg/l for females (95% C.I. 703 to 1047).
81-6 Dermal sensitization species: guinea pig ICI, UK GG5005 & GG4763; 09/07/90 MRID No.: 43496002 **Acceptable** **Moderate skin sensitizer**	In a dermal sensitization study, thirty male guinea pigs were divided into two groups of 20 treated and 10 controls. During the first induction phase all preparations were administered intradermally in a volume of 0.05 to 0.1ml at clipped sites in the scapular region; the preparations are Freund's complete Adjuvant + corn oil, a 3% (w/v) test substance in corn oil, and a 3% (w/v) test substance in a 1:1 Freund's Complete Adjuvant in corn oil. One week later (second induction), 0.2 to 0.3ml of 75% (w/v) test substance in corn oil was applied to the scapular region of the treatment groups. Animals in the control group received similar intradermal injections/topical application with the exception that no test material was administered. Fourteen days later, both treatment groups received challenge doses of a cutaneous application of 75% (w/v) on the left flank and 30% (w/v) in corn oil right flank. The application sites were scored for erythema 24 and 48 hours later; and expressed as percent response in comparison to control on a scale of 0 to 100%, (0 = not a sensitizer, 1 - 8 weak sensitizer, 9 - 28 = mild sensitizer, 29 - 64 = moderate sensitizer, 65 - 80 = strong sensitizer and 81 - 100 = extreme sensitizer). Formaldehyde in deionized water for intradermal injection and 30% (w/v) dilutions of solution in deionized water were applied for the topical induction and challenge. Challenge with 75% (w/v) and 35% (w/v) of test substance in corn oil resulted scattered mild redness to intense redness and a calculated % response of 32% and 47%, respectively. Positive control formaldehyde elicited an extreme skin sensitization response. **Based on the skin response, pirimicarb was rated as moderate skin sensitizer to guinea pig skin.**
82-2 Dermal-2 week Species: rabbit ICI, UK. CTL/R/308; 05/04/71 MRID No.:43496003 **Supplementary**	In a repeated dose dermal toxicity (14-day), pirimicarb was applied to the clipped backs (≈ area 40 sq. cm.) of four male and female albino rabbits at a dose of 500 mg/kg pirimicarb in a dosage volume of 250 mg/ml in ethylene glycol for 24 hours. The treatments were repeated daily for a total of 14 treatments. Between the treatments, the skin was washed with soap and water, dried and left for 30 minutes. Data were not presented to substantiate the conclusions that repeated dermal application of pirimicarb did not result in clinical toxicity. In addition the effect of ethylene glycol on the bioavailability of pirimicarb was not demonstrated. **The study is classified as core-Supplementary and is not upgradable**

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84-2(a) Gene mutation in bacteria (Ames Assay) ICI, UK. CTL/P/428; 09/22/78 MRID No.:43496004 **Acceptable**	Cultures of Salmonella typhimurium (TA-strains 1535, 1538, 98 and 100) were exposed to graded concentrations of test article for 48 hours in the presence of S-9 activation. No increased frequencies of revertants (reverse gene mutation from his-to-his +) were evident in any strain treated up to the HDT, 2,500 µg/plate.
84-2(a) Reverse Gene mutation in bacteria (Ames Assay) ICI, UK. CTL/P/450; 02/21/80 MRID No.:43496005 **Acceptable**	Replicate cultures of the Ames battery of Salmonella typhimurium mutant strains were exposed in repeat experiments to test article for 48 hours. No increase in reverse mutation was found at doses up to 2,500 µg/plate, with/without S9 activation.
84-2(b) Chromosome damage in human lymphocyte cultures ICI, UK. CTL/P/1655; 03/06/87 MRID No.:43496006 **Acceptable**	Primary lymphocyte cultures from two volunteer blood donors were exposed for 3 hours to test article, with and without S-9 activation. No chromosome aberrations were found at any dose up to the limit of solubility, 500 µg/ml.
84-4 DNA damage/repair in vivo (HPC/UDS) ICI, UK. CTL/P/2824; 03/15/90 MRID No.:43496007 **Acceptable**	Male Sprague Dawley rats were gavaged once at doses of 50, 100 and 250 mg/kg, and hepatocytes (HPC) prepared in culture to sample repair unscheduled DNA synthesis (UDS), measured as increased net nuclear (silver) grain counts (NNGC), at two sampling times (4 and 12 hours post-dose). There was no induced increase in NNGC at any dose up to clinically toxic MTDs (20 - 250 mg/kg).
84-2(b) chromosome damage in vivo (mouse MT) ICI, UK CTL/P/2641; 08/29/89 MRID No.:43496008 **Acceptable**	Male and female mice were orally intubated once at two doses (50% and 80% of the MLD), and bone marrow polychromatic erythrocytes scored for the presence of micronuclei M-PCE, representing the results of chromosome aberrations. No increased M-PCE were induced at dose levels causing cytotoxicity, 43.3 and 69.3 mg/kg.

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

MEMORANDUM

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

SUBJECT: Pirimicarb--Mutagenicity Data Submitted Under MRID Nos. 434960-04, -05, -06, -07, and -08

ID #010182-GAO Pirimicarb Technical

Chemical: 106101 (359C)
RD Record: S479870
HED Project: D210945
(DP Barcode)

FROM: Irving Mauer, PH.D., Geneticist
Toxicology Branch-I
Health Effects Division (7509C)

05-09-95

TO: Guruva Reddy, Ph.D.
Toxicology Branch-I
Health Effects Division (7509C)

THRU: Karl P. Baetcke, Ph.D., Chief
Toxicology Branch-I
Health Effect Division (7509C)

Registrant: Zeneca, Wilmington, DE

Request: Review and evaluate the following five mutagenicity assays, all performed at Zeneca's (successor to ICI) Central Toxicology Laboratory (CTL), Alderley Park, Macclesfield, Cheshire (UK):

1. (84-2a) PIRIMICARB: SHORT-TERM REDICTIVE TESTS FOR CARCINOGENICITY RESULTS FROM THE SALMONELLA MICROSOME REVERSE MUTATION TEST, CTL REPORT NO. CTL/P/428, FINAL REPORT DATED 22 SEP 1978 (MRID 43496004).

2. (84-2a) AN EXAMINATION OF PIRIMICARB FOR POTENTIAL MUTAGENICITY USING THE SALMONELLA/MICROSOME REVERSE MUTATION ASSAY, CTL Report No. CTL/P/540 (CTL Ref. Y00032/001/002), Final Report dated 21 Feb 1980 (MRID 43496005).

3. (84-2b) PIRIMICARB: A CYTOGENETIC STUDY IN HUMAN LYMPHOCYTES IN VITRO, CTL Report No. CTL/P/1655 (CTL Ref. Y00032/001/002), Final Report Dated 6 Mar 1987 (MRID 43496006).

4. (84-4) PIRIMICARB: ASSESSMENT FOR THE INDUCTION OF UNSCHEDULED DNA SYNTHESIS IN RAT HEPATOCYTES IN VIVO, CTL Report No. CTL/P/2824 (Study No. SR0367) Final Report dated 15 Mar 1990 (MRID 43496007).

5. (84-2b PIRIMICARB (TECHNICAL): AN EVALUATION IN THE MOUSE MICRONUCLEUS TEST, CTL Report No. CTL/P/2641, Final Report dated 29 Aug 1989 (MRID 43496008).

TOX I CONCLUSIONS: These studies have been adjudged as follows (detailed reviews are attached):

Study (MRID)	Report Results	TB EVALUATION
1. Gene mutation in bacteria (Ames Assay) (43496004)	Negative for inducing gene mutation in activated Salmonella strains exposed up to 2500 ug/plate.	ACCEPTABLE
2. Reverse gene mutation in bacteria (Ames) (43496005)	Negative for inducing reverse gene mutation in Salmonella typhimurium TA cultures exposed, with/without activation, to doses up to 2500 ug/plate.	ACCEPTABLE
3. Chromosome damage in human lymphocyte cultures (43496006)	Negative for structural chromosome aberrations in primary human lymphocyte cultures exposed, with/without activation, to test article up to the limit of solubility, 500 ug/ml.	ACCEPTABLE
4. DNA damage/repair in vivo (HPC/UDS) (43496007	Negative for inducing unscheduled DNA synthesis in primary rat hepatocytes cultured from rats treated at single oral doses up to a clinically toxic MTD (250 mg/kg).	ACCEPTABLE
5. Chromosome damage in vivo (mouse MT) (43496008)	Negative for inducing micronuclei in polychromatic erythrocytes of mice given single oral doses up to levels producing cytotoxicity (69.3 mg/kg).	ACCEPTABLE

ATTACHMENTS: DERs

Disk 5:Size 3.5:D210945:MAUER:MB

[81-6. Pirimicarb: Skin Sensitization - guinea pig/1990]

Reviewed by: Guruva B. Reddy, D.V.M.,.Ph.D. 5/22/95
Section IV, Tox. Branch I (7509C)
Secondary Reviewer: John Doherty, Ph.D., Acting Section Head
Section IV, Tox. Branch I (7509C) 5/25/95

DATA EVALUATION REPORT

STUDY TYPE: Dermal Sensitization/Guinea pig/81-6

TOX. CHEM. NO.: 359C

P. C. NO.: 106101

MRID NO.: 43496002

TEST MATERIAL: Pirimicarb Technical

SYNONYMS: 2-(dimethylamino)-5,6-dimethyl-4-pyrimidinyl dimethylcarbamate

STUDY/REPORT NUMBERS: GG5005 & GG4763/CTL/P/3087

SPONSOR: Zeneca Inc.
Wilmington, DE 19897

TESTING FACILITY: Imperial chemical Industries, PLC
Alderley Park
Macclesfield, Cheshire
UK

TITLE OF REPORT: Pirimicarb: Skin Sensitization to the Guinea Pig

AUTHOR: N Rattray
A M Leah

STUDY COMPLETION DATE: September 7, 1990

EXECUTIVE SUMMARY: In a dermal sensitization study, thirty male guinea pigs were divided into two groups of 20 treated and 10 controls. During the first induction phase all preparations were administered intradermally in a volume of 0.05 to 0.1ml at clipped sites in the scapular region; the preparations are Freund's complete Adjuvant + corn oil, a 3% (w/v) test substance in corn oil, and a 3% (w/v) test substance in a 1:1 Freund's Complete Adjuvant in corn oil. One week later (second induction), 0.2 to 0.3ml of 75% (w/v) test substance in corn oil was applied to the scapular region of the treatment groups. Animals in the control group received similar intradermal injections/topical application with the exception that no test material was administered. Fourteen days later, both treatment groups received challenge doses of a cutaneous application of 75%

(w/v) on the left flank and 30% (w/v) in corn oil right flank. The application sites were scored for erythema 24 and 48 hours later; and expressed as percent response in comparison to control on a scale of 0 to 100%, (0 = not a sensitizer, 1 - 8 weak sensitizer, 9 - 28 = mild sensitizer, 29 - 64 = moderate sensitizer, 65 - 80 = strong sensitizer and 81 - 100 = extreme sensitizer). Formaldehyde in deionized water for intradermal injection and 30% (w/v) dilutions of solution in deionized water were applied for the topical induction and challenge.

Challenge with 75% (w/v) and 35% (w/v) of test substance in corn oil resulted scattered mild redness to intense redness and a calculated % response of 32% and 47%, respectively. Positive control formaldehyde elicited an extreme skin sensitization response. **Based on the % response, pirimicarb was rated as moderate skin sensitizer to guinea pig skin.**

Classification: ACCEPTABLE. Products containing pirimicarb must include precautionary statement for potential dermal sensitization.

MATERIALS:

1. **Test Compound:** Pirimicarb 97.3% (w/w), Lot # was not given, however codified as sample ref: RS088/E; and described as cream-colored powder, was used in this study. Formaldehyde (40% w/v aqueous solution was used as positive control.

2. **Test Animals:** Species: Guinea pig, Strain: Young adult Porcellus:Dunkin Hartley, Weight: 391 - 479g for the main study and 278 to 418g for the positive control study, Source: Harlan Porcellus, Firgrove Farm, Heathfield, Sussex, UK. The animals were housed in suspended cages and maintained at a temperature of 19°C ± 2°C, a relative humidity of 50 ± 10% and to a 12-hour dark/light cycles. The guinea pigs were acclimated for 6 days to the laboratory environment. Labsure RGP guinea Pig Diet and water was provided ad libitum.

METHODS:

Thirty acclimated male guinea pigs were divided into two groups of twenty treated and ten controls. Doses selected for main study were based on preliminary investigations. Positive controls were run at a different time, but no

details were provided. The sensitizing properties of the test substance were assessed using the method based on the maximization test of Magnusson and Kligman (1970).

Induction:

On Days 1, an area approximately 5cm X 5cm on the scapular region of each animal was clipped free of hair and a row of injections of Freund's Complete Adjuvant in corn oil (1:1), a 3% (w/v) test material in corn oil, and a 3% (w/v) preparation of the test substance in Freund's Complete Adjuvant plus corn oil (1:1), in a volume of 0.05 to 0.1 ml were made on each side of the mid-line. Dermal reactions to the injections were observed for up to 24 hours.

On Day 7, scapular region of each guinea pig was clipped free of hair. A 0.2 to 0.3 ml of 75% (w/v) test material in corn oil was applied on filter paper, placed over the clipped area and kept in place for 48 hours using occlusive dressing. Following removal of dressing, the application site was observed for about 24 hours for dermal reactions.

The control animals were induced by intradermal administration of Freund's Complete Adjuvant in corn oil (1:1), corn oil only and Freund's Complete Adjuvant in corn oil (1:1)using the same procedure described for the test substance induction. For the topical applications (Day 7) only corn oil was applied to the filter paper.

Challenge:

Two weeks after the topical application, an area approximately 15cm X 5cm on both flank areas of all animals was clipped free of hair, An occlusive bandage was prepared by stitching two filter papers to rubber sheeting. A 0.05 to 0.1 ml of 75% (w/v) in corn oil (1:1) was applied to one of the filter papers and 30% (w/v) preparation of test substance in corn oil was applied to other filter paper and was placed on the clipped area so that the 75% preparation was on the left flank and the 30% preparation on the right flank area. After 24 hours, the dressing was cut open and observed for 24 and 48 hours after removal of dressing for dermal reactions on a 4 point scale (0 = no reaction, 1 = scattered mild redness, 2 = moderate diffuse redness, and 3 = intense redness and swelling).

The sensitivity response was expressed as percentage of the test animals that gave response greater than the maximum

seen in control animals. Following is the evaluation criteria:

% net response	description
0	not a sensitizer
1-8	weak sensitizer
9-28	mild sensitizer
29-64	moderate sensitizer
65-80	strong sensitizer
81-100	extreme sensitizer

A 0.3% (w/v) dilution of formaldehyde in deionized water was used for the intradermal injunction induction and 30% (w/v) dilutions of the formaldehyde solution in deionized water were applied for topical induction and the challenge.

QUALITY ASSURANCE:

A statement Quality Assurance Unit, a statement of GLP Compliance and a statement of Confidentiality of Data were attached.

RESULTS:

Two test animals died prior to challenge; and the deaths were considered not to be compound-related. Following challenge with 75% and 30% (w/v) preparation of test substance resulted in a scattered mild redness to intense redness and swelling in 6/19 (32%) and 9/19 (47%) animals, respectively. Controls did not show reaction to the challenge dose. Positive control formaldehyde elicited scattered mild redness to moderate diffuse redness in 17/18 (4%; extreme sensitizer) animals. Based on the % response of 32 and 47, the chemical was classified as moderate skin sensitizer to the guinea pig skin. We concur with the authors conclusions.

DISCUSSION:

The data reporting was thorough and the summary means were supported by individual animal data. Positive control study was run at a different time than the main study; the dates of the study was not provided. Although the studies were run at different times, the outcome of the study was not compromised since the test material elicited positive response to challenge. The challenge with 75% (w/v) and 30% (w/v) preparations of test substance resulted in a scattered mild redness to intense redness and swelling in 6/19 (32%) and 9/19 (47%) animals, respectively. Based on the percent response the chemical was classified as moderate sensitizer

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to the guinea pig skin.

This study is classified as **core-Acceptable** and **satisfies** the requirements, for a § 81-6 dermal sensitization study in guinea pig.

GReddy/Pirimicarb/8D210945/4-21-95

[82-2. Pirimicarb. Repeated dose dermal toxicity - rabbit/1971]

Reviewed by: Guruva B. Reddy, D.V.M., Ph.D. 5/22/95
Section IV, Tox. Branch I (7509C)
Secondary Reviewer: John Doherty, Ph.D., Acting Section Head
Section IV, Tox. Branch I (7509C) 5/24/95

DATA EVALUATION REPORT

STUDY TYPE: Repeated dose dermal toxicity/Rabbit/82-2

TOX. CHEM. NO.: 359C

P. C. NO.: 106101

MRID NO.: 43496003

TEST MATERIAL: Pirimicarb tech.

SYNONYMS: 2-(Dimethylamino)-5,6-dimethyl-4-pyrimidinyl dimethylcarbamate

REPORT NO.: CTL/R/308

SPONSOR: Zeneca Inc.
Zeneca Ag Products
Wilmington, DE 19897

TESTING FACILITY: Imperial Chemical Industries, PLC
Alderley Park
Macclesfield, Cheshire
UK

TITLE OF REPORT: Pirimicarb (PP 062): Subacute Toxicity on Rabbit Skin

AUTHORS: K. Fletcher

REPORT ISSUED: May 4, 1971

EXECUTIVE SUMMARY: In a repeated dose dermal toxicity (14-day), pirimicarb was applied to the clipped backs (≈ area 40 sq. cm.) of four male and female albino rabbits at a dose of 500 mg/kg pirimicarb in a dosage volume of 250 mg/ml in ethylene glycol for 24 hours. The treatments were repeated daily for a total of 14 treatments. Between the treatments, the skin was washed with soap and water, dried and left for 30 minutes (MRID No.: 43496003; Report No.: CTL/R/308).

Data were not presented to substantiate the conclusions that repeated dermal application of pirimicarb did not result in clinical toxicity. In addition the effect of ethylene glycol on the bioavailability of pirimicarb was not demonstrated.

The study is classified as **core-Supplementary, not upgradable** and **does not satisfy** the requirements, for a § 82-2 repeated dose dermal toxicity (21-day) in rabbits.

MATERIALS:

1. **Test Compound:** Pirimicarb (PP 062), Purity : 94% (w/w), Lot # was not given.

2. **Test Animals:** Species: Albino Rabbits, Strain: not furnished, Weight: males and females - 2 - 3 kg. Source: not furnished. Housing, environmental controls, feed and watering information were not provided.

METHODS:

Pirimicarb was applied to the clipped backs (≈ area 40 sq. cm.) of four male and female albino rabbits at a dose of 500 mg/kg pirimicarb in a dosage volume of 250 mg/ml in ethylene glycol for 24 hours. No occlusive bandage was used; however, the animals were restrained using a plastic neck collar. The treatments were repeated daily for a total of 14 treatments. Between the treatments, the skin was washed with soap and water, dried and left for 30 minutes.

QUALITY ASSURANCE:

A statement of Confidentiality of Data and a GLP statement indicating the study did not meet the regulations were attached.

RESULTS:

The author stated that no treatment-related clinical signs were observed. Rabbits gained weights during the study.

DISCUSSION

The study as conducted totally inadequate to satisfy the requirements of subacute dermal toxicity in rabbits. No data are presented on animals/source, housing, food consumption, body weight gains, hematology, clinical chemistry and gross and histopathology.

The study is classified as **core-Supplementary, not upgradable** and **does not satisfy** the requirements, for a § 82-2 repeated dose dermal toxicity (21-day) in rabbits.

[82-2. Pirimicarb. Repeated dose dermal toxicity - rabbit/1971]

GReddy\pirimor\82-2\4-26-95

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Reviewed by: Guruva B. Reddy, D.V.M., Ph.D.
Section IV, Tox. Branch I (7509C)
Secondary Reviewer: John Doherty, Ph.D., Acting Section Head
Section IV, Tox. Branch I (7509C)

5/24/95

5/24/95

DATA EVALUATION REPORT

STUDY TYPE: Acute Inhalation Toxicity/Rat/81-3

TOX. CHEM. NO.: 359C

P. C. NO.: 106101

MRID NO.: 43496001

TEST MATERIAL: Pirimicarb Technical

SYNONYMS: 2-(dimethylamino)-5,6-dimethyl-4-pyrimidinyl dimethylcarbamate

STUDY/PROJECT NUMBERS: Report No.: CTL/P/4522
Study No.: HR2247

SPONSOR: Zeneca Inc.
Wilmington, DE 19897

TESTING FACILITY: Zeneca Central Toxicology Laboratory
Alderley Park, Macclesfield
Cheshire, UK

TITLE OF REPORT: Pirimicarb: 4-Hour Acute Inhalation Toxicity Study in the Rat

AUTHOR: R J Parr-Dobrzanski

STUDY COMPLETION DATE: October 28, 1994

EXECUTIVE SUMMARY: In a acute inhalation toxicity study, groups (5/sex/dose) of Wistar-derived male and female rats were exposed nose-only to gravimetric concentrations of 410, 746 or 1205 µg/l pirimicarb with MMAD of 3.02 ± 2.03 to 3.46 ± 1.89 for 4 hours (MRID No.: 43496001; Study No.: HR2247).

One high-dose male and three females died during the exposure. Six high-dose and one mid-dose animals were sacrificed in moribund condition. Clinical signs included salivation, slow deep respirations and auditory hypoesthesia. Necropsy of the dead and terminal sacrifice were unremarkable. The calculated LC_{50}, based on the mortality was 948 µg/l for males (95% C.I. 746 to ~~1047~~ 1204) and 858 µg/l for females (95% C.I. 703 to 1047).

The study is classified as **Core-Acceptable**, assigned a **Toxicity Category III** and **satisfies the requirements**, for a Series 81-3 acute inhalation toxicity study in rats.

MATERIALS:

1. **Test Compound:** Technical grade Pirimicarb, CTL Ref. # Y00032/047, Purity 97.4% and described as milled white solid, was used in this study.

   Sample preparation: Sponsor provided milled samples. No further milling was done by the testing facility.

2. **Test Animals:** Species: rats, Strain: Alpk:APfSD (wistar-derived), Age: not given but were described as young adults, Weight: Males - 280 to 333, and Females - 227 to 249 g, Source: Alderley Park, Cheshire, UK. The animals were housed five to a cage (sexes separately) during the acclimatization (5 days) and post-exposure periods; and maintained at a temperature of 20-24°C, relative humidity of 40-60% and 12 hours of light/dark cycles. The animals had access to CTI Diet (Special Diet Services Limited, Witham, Essex, UK) and water ad libitum.

   Three groups of five males and five females were exposed to analytical/gravimetric concentrations of 414/410, 747/746 or 1065/1205 µg/l, for 4 hours, respectively. Preliminary trials were run to determine the target atmospheric pirimicarb concentrations. The study design, the analytical and gravimetric concentrations and mortality are presented in Table 1.

TABLE 1. SUMMARY OF CHAMBER CONCENTRATION AND MORTALITY

GROUP	CHAMBER CONCENTRATION (µg/L)			MORTALITY		
	TARGET	ANALYTICAL	GRAVIMETRIC	♂	♀	TOTAL
I	400	414 ± 47	410 ± 45	0/5	0/5	0/10
II	800	747 ± 100	746 ± 101	0/5	1/5	1/10
III	1200	1065 ± 207	1205 ± 193	5/5	5/5	10/10

METHODS:

LC_{50} Study: Exposure by nose-only for 4 hours. Animals were restrained using tubes supplied by Battlles, Geneva, Switzerland, which were inserted into PERSPEX exposure chamber (9.2 L; Appendix A), once the target concentration

had been achieved and stable over 30 minutes. The attached figure is a combination of 2 units which has a total volume of 27.6 liters. Temperature and relative humidity were measured at frequent intervals. Oxygen concentration was not measured during the study. Dust was generated using a Wright's dust-feed mechanism and carried to the exposure chamber. Clean, undiluted dry air was passed through the dust feed mechanism at flow rates of 10 - 12, 8 - 15 or 9 - 11 liters/min to generate gravimetric concentrations of 410, 746 or 1205 µg/l, respectively. Air flow rates were measured and were altered at frequent intervals to maintain the desired concentration.

The nominal concentration of the test substance in the exposure chamber was not calculated since the investigators felt that these values were not meaningful.

The particulate/gravimetric concentration of the pirimicarb in the breathing zone was determined, at approximately 30 - 60 intervals during the exposure period by drawing the test atmosphere, at a known flow rate for a known time, through a 25 mm diameter Vinyl Metricel (Gelman Sciences Limited, Northampton, UK) filter housed in a Delrin open-faced filter holder. The particulate or gravimetric concentration was calculated by wt. gain of filter by volume of air.

The aerodynamic particle size range in the test atmosphere was determined using a Marple Cascade Impactor. The mean amount of aerosol, by weight, in each size range, was then used to calculate the aerodynamic particle size distribution of the aerosol. The mass median aerodynamic diameter (MMAD) and geometric standard deviation (σ_g) were calculated from regression line derived from the cumulative data based on the amount of material collected on the impactor stages.

The animals were observed immediately prior to exposure, frequently during the exposure, at the end of exposure, and once daily for the remainder of the study for clinical signs and mortality.

Rats were weighed Day-1, shortly before exposure and on days 2, 3, 8 and 15.

Detailed gross necropsy was performed on all animals which died or sacrificed at the end of the study.

QUALITY ASSURANCE:

A statement of Data Confidentiality, GLP compliance and Quality Assurance are attached.

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RESULTS:

The mean analytical/gravimetric (particulate) concentration of the test substance in the I, II and III group was 414/410, 747/746 and 1065/1205 μg/l, respectively (see Table 1). There was no variation between the mean analytical and gravimetric concentrations for group I and II. For group III, discrepancy existed (3/6 samples collected) between the analyzed pirimicarb concentration expressed as a percent of the total particulate and the purity of the test substance. The reason for this discrepancy is probably due to 3/6 samples collected were analyzed 25% less than the target concentration.

Table 2 summarizes the analytical concentration, MMAD, σ_g and percent cumulative particle sizes. Percent of MMAD particles with $\leq$ 3.5 μm exceeded 35.5% of total particle distribution, in all three groups and meets the interim guidelines for acute inhalation toxicity. The test groups were exposed to mean aerosol particle concentrations of 410, 746, and 1205 μg/l, with MMAD of 3.05, 3.46 and 3.02 μm and geometric standard deviations of 2.04, 1.89 and 2.03, respectively.

TABLE 2. SUMMARY OF PARTICLE SIZE DISTRIBUTION

GROUP	GRAVIMETRIC CONC. (μg/l)	PARTICLE SIZE DISTRIBUTION*						
		MMAD (μm)	σ_g	CUMULATIVE % OF PARTICLES				
				$\leq$ 1.5μm	$\leq$ 3.5μm	$\leq$ 6.0μm	$\leq$ 9.8μm	$\geq$ 9.8μm
I	410	3.05	2.04	7.5	40.4	35.6	12.9	3.4
II	746	3.46	1.89	5.7	35.5	38.8	17.2	2.7
III	1205	3.02	2.03	11.2	48.3	32.1	6.7	1.8

* = MMAD
σ_g = Geometric standard deviation

The calculated LC_{50}, based on the mortality data was 948 μg/l for males (95% C.I. 746 to 1204) and 858 μg/l for females (95% C.I. 703 to 1047).

Clinical signs in males and females are consistent with moderate toxicity and respiratory tract irritation. One high dose male and three females died during exposure. One female (group II) and four males and two females in the high-dose deteriorated during exposure; became moribund following exposure, therefore, humanely killed. Clinical signs included salivation, slow deep breathing, auditory hypoesthesia. Clinical conditions of surviving animals

improved by day 2/3 of the study and were normal by terminal day.

All surviving males/females gained weight by study termination.

Gross pathology of the dead or sacrificed were unremarkable.

DISCUSSION:

The data reporting was thorough and the summary means were supported by individual animal data. The experimental animals were exposed nose-only for 4 hours to the mean analytical/gravimetric (particulate) concentration of the test substance in the I, II and III group was 414/410, 747/746 and 1065/1205 µg/l, respectively. The Percent of MMAD particles with ≤ 3.5 µm exceeded 35.5% of total particle distribution, in all three groups and meets the interim guidelines for acute inhalation toxicity. The test groups were exposed to mean aerosol particle concentrations of 410, 746, and 1205 µg/l, with MMAD of 3.05, 3.46 and 3.02 µm and geometric standard deviations of 2.04, 1.89 and 2.03, respectively. Clinical signs indicative of moderately irritating to the respiratory tract. Based on the data presented the acute inhalation toxicity of pirimicarb is as follows:

LC_{50} (95% C.I.) = 948 (746 to 1204) µg/l, males
LC_{50} (95% C.I.) = 858 (703 to 1047) µg/l, females

The study is classified as **Acceptable** and **satisfies** the requirement, of a Series § 81-3 acute inhalation toxicity study in rats. Pirimicarb is assigned to **Toxicity Category III.**

PIRIMICARB TOXR 011605

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Reviewed by: Irving Mauer, Ph.D., Geneticist
Toxicology Branch I, HED (7509C)
Secondary Reviewer: Karl P. Baetcke, Ph.D., Chief
Toxicology Branch I, HED (7509C)

5-11-95

5/22/95

DATA EVALUATION REPORT

MRID No.: 434960-04
PC No.: 106101
RD Record No.: S479870
EPA ID No.: 010182-GAO-Pirimicarb Tech
Tox Chem No.: 359C
Project No.: D210945

I. SUMMARY

STUDY TYPE: (84-2a) Mutagenicity -- Reverse gene mutation in Salmonella typhimurium (Ames)

CHEMICAL: Pirimicarb

SPONSOR: Zeneca, Wilmington, DE

TESTING FACILITY: (ICI, now Zeneca) Central Toxicology Laboratory (CTL), Alderley Park (UK)

TITLE OF REPORT: PIRIMICARB: Short-term predictive tests for carcinogenicity results from the Salmonella microsome reverse mutation test

AUTHOR(S): E. Longstaff

STUDY NUMBER: CTL/P/428

DATE ISSUED: September 22, 1978

EXECUTIVE SUMMARY: Cultures of Salmonella typhimurium (TA-strains 1535, 1538, 98 and 100) were exposed to graded concentrations of test article for 48 hours in the presence of S-9 activation. No increased frequencies of revertents (reverse gene mutation from his-to his+) were evident in any strain treated up to the HDT, 2500 ug/plate.

TB-I Evaluation: ACCEPTABLE

II. DETAILED REVIEW:

A. Test Material: Pirimicarb

Description: (not provided)
Batches (Lots): 189
Purity (%): 97.7
Solvent/carrier/diluent: Dimethylsulfoxide (DMSO)

B. Test Organism: Bacterial cultures

Species: Salmonella typhimurium LT2
Strain: TA1535, TA1538, TA98, TA100 (all his-)
Source: (not provided)

C. Study Design (Protocol): This study was designed to assess the mutagenic potential of the test article when administered in vitro to cultures of Salmonella typhimurium TA strains, and assaying for reverse mutation at the histidine locus (his-to-his+), according to established (published) procedures and FIFRA/OECD Test guidelines.

A Statement of Quality Assurance measures (inspections/audits) was not provided.[1]

A Statement of adherence to Good Laboratory Practice (GLP) was not provided.[1]

D. Procedures/Methods of Analysis: Replicate cultures of the four TA tester strains of Salmonella typhimurium were exposed to DMSO solvent or 5 concentrations of test article (4, 20, 100, 500 and 2500 ug/plate) in the presence of a mammalian metabolite activation system.[2] The carcinogens, 1,3 -propane sultone (PS) and N-2-flurenyl-acetamide (AAF) served as positive controls, at concentrations of 40, 1000, and 2500 ug/ml.

After 48 hours incubation, culture plates were examined for revertent (his+) colonies.

E. Results: As detailed in the (single) summary tabulation provided in this Report (attached to this DER), none of the pirimicarb doses, up to the HDT 2500 ug/plate,

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[1] This assay preceded the promulgation of these FIFRA requirements.

[2] Microsomal fractions (S9) of liver homogenates prepared from young adult male Sprague Dawley rats pre-treated with the PCB, Aroclor 1254, plus NADP(H)-generating co-factors.

increased the incidence over control of revertents at the histidine locus. By contrast, both reference chemicals (PS, AAF) caused significantly positive results in their strain-specific target strains, some 10 to 50-fold background.

The author concluded that pirimicarb was not mutagenic under activation conditions.

F. TB-I Evaluation: Although many of the essential elements currently required by FIFRA Test Guidelines when conducting such mutagenic assays were not included in this report (such as GLP/QA; identification of test organism source; methods to assure identity of test system; criteria for determining a positive; individual replicate data, etc.), the negative results for this singular trial of the test article may be accepted in view of the positive controls validating the response of the bacterial test system.

ATTACHMENT (Summary Data Table)

PIRIMICARB TOXR 011605

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Reviewed by: Irving Mauer, Ph.D., Geneticist
Toxicology Branch I, HED (7509C)
Secondary Reviewer: Karl P. Baetcke, Ph.D., Chief
Toxicology Branch I, HED (7509C)

05-11-95
5/22/95

011605

DATA EVALUATION REPORT

MRID No.: 434960-05
PC No.: 106101
RD Record No.: S479870
EPA ID No.: 010182-GAO-Pirimicarb Tech
Tox Chem No.: 359C
Project No.: D210945

I. SUMMARY

STUDY TYPE: (84-2a) Mutagenicity -- Reverse gene mutation in Salmonella typhimurium (Ames)

CHEMICAL: Pirimicarb

SPONSOR: Zeneca, Wilmington, DE

TESTING FACILITY: (ICI, now Zeneca) Central Toxicology Laboratory (CTL), Alderley Park (UK)

TITLE OF REPORT: An Examination of Pirimicarbi for Potential Mutagenicity Using the Salmonella/Microsome Reverse Mutation Assay

AUTHOR(S): R. W. Trueman

STUDY NUMBER: CTL/P/540 (CTL Ref. Y00032/001/002)

DATE ISSUED: February 21, 1980

EXECUTIVE SUMMARY: Replicate cultures of the Ames battery of Salmonella typhimurium mutant strains were exposed in repeat experiments to test article for 48 hours. No increase in reverse mutation was found at doses up to 2500 ug/plate, with/without S9 activation.

TB-I Evaluation: ACCEPTABLE

II. DETAILED REVIEW:

A. Test Material: Pirimicarb (technical)

Description: (not provided)
Batches (Lots): (not provided)
Purity (%): 98
Solvent/carrier/diluent: Dimethylsulfoxide (DMSO)

B. Test Organism: Bacterial cultures

Species: Salmonella typhimurium LT2
Strain: TA1535, TA1537, TA1538, TA98, TA100 (all his-)
Source: (not provided)

C. Study Design (Protocol): This study was designed to assess the mutagenic potential of the test article when administered in vitro to cultures of Salmonella typhimurium, and determining frequency of revertent colonies (his+), resulting from reverse gene mutation, according to established (published) procedures and FIFRA/OECD Test guidelines.

A Statement of Quality Assurance measures (inspections/audits) was not provided.[1]

A Statement of adherence to Good Laboratory Practice (GLP) was not provided.[1]

D. Procedures/Methods of Analysis: Replicate cultures (5 plates per dose) were exposed to DMSO solvent, or a graded series of five test article doses, both in the absence and presence of a mammalian metabolic activation system.[2] Strain-specific mutagens[3] served as positive controls for non-activated and activated test series.

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[1] This assay preceded the promulgation of these FIFRA requirements.

[2] Post-mitochondrial fraction of hepatic homogenates derived from adult male Sprague Dawley rats pre-treated with Aroclor 1254, plus NADP(H)-generating co-factors.

[3] Without activation (-S9): TA1535/TA100: 1, 3 propane sulfone.
TA1537: 9-Aminoacridine.
TA1538/TA98: 2-Acetylaminofluorene

With activation (+S9): All strains: 2-Aminofluorene, or Aflatoxin B1.

Two complete (independent) assays were run. Revertent colonies were counted after 48 hours exposure, and mean colony counts of pirimicarb and positive-control plates were compared statistically to solvent controls using software provided for the Data General M600 Computer.

E. Results: In contrast to the strongly significant positive response induced by the mutagens (up to 200-fold increase in revertent colonies, Report Tables 1 and 3) none of the test article series in either trial showed increases over DMSO solvent controls in colony counts up to the HDT, 2500 ug/plate. (Report Tables 2 and 4, attached here).

Hence the author concluded that pirimicarb was "unequivocally negative" in this Ames test.

F. TB-I Evaluation: ACCEPTABLE, as demonstrating negative response to reverse gene mutation at the histidine locus in the battery of Salmonella (Ames) strains.

ATTACHMENT (Summary Data Tables)

PIRIMICARB TOXR 011605

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Reviewed by: Irving Mauer, Ph.D., Geneticist
Toxicology Branch I, HED (7509C)
Secondary Reviewer: Karl P. Baetcke, Ph.D., Chief
Toxicology Branch I, HED (7509C)

05-12-95

5/22/95

DATA EVALUATION REPORT

MRID No.: 434960-06
PC No.: 106101
RD Record No.: S479870
EPA ID No.: 010182-GAO-Pirimicarb Tech
Tox Chem No.: 359C
Project No.: D210945

I. SUMMARY

STUDY TYPE: (84-2b) Mutagenicity - in vitro Chromosome damage in human lymphocytes (HLC)

CHEMICAL: Pirimicarb

SPONSOR: Zeneca, Wilmington, DE

TESTING FACILITY: (ICI, now Zeneca) Central Toxicology Laboratory (CTL), Alderley Park (UK)

TITLE OF REPORT: Pirimicarb: A cytogenetic study in human lymphocytes in vitro

AUTHOR(S): J. Wildgoose, C.A. Howard, C.R. Richardson and V. Randall

STUDY NUMBER: CTL/P/1655 (CTL Ref. Y00032/001/002/Study No. SV0227)

DATE ISSUED: March 6, 1987

EXECUTIVE SUMMARY: Primary lymphocyte cultures from two volunteer blood donors were exposed for 3 hours to test article, with and without S-9 activation.

No chromosome aberrations were found at any dose up to the limit of solubility, 500 ug/ml.

TB-I Evaluation: ACCEPTABLE

II. DETAILED REVIEW:

A. Test Material: Pirimicarb

Description: Pale (fawn) solid
Batches (Lots): WED/G/9680 RS 306/C
Purity (%): 98.2
Solvent/carrier/diluent: Dimethylsulfoxide (DMSO)

B. Test Organism: Primary cultures of lymphocytes

Species: Human
Source: Two healthy adult donors: one male, on female

C. Study Design (Protocol): This study was designed to assess the clastogenic potential of the test article when administered in vitro to human lymphocyte cultures, according to established (published) procedures and FIFRA/OECD Test guidelines.

A Statement of Quality Assurance measures (inspections/audits) was provided.

A Statement of adherence to Good Laboratory Practice (GLP) was provided.

D. Procedures/Methods of Analysis: Approximately 44 hours after culture initiation, duplicate cultures of lymphocytes from blood collected in phytohemmaglutinin-supplemented tissue culture medium were exposed for 3 hours to a graded series (six) of test article concentrations (ranging from 10 to 500 ug/ml, the latter the limit of solubility), in the absence and presence of a mammalian metabolic activation system.[1] Separate cultures of lymphocytes were exposed to the reference clastogens, mitomycin C (MMC, 0.5 ug/ml) or cyclophosphamide (CP, 100 ug/ml), to serve as positive controls for, respectively, the non-activated (-S9) and activated (+S9) test series.

After the 3 hour exposure, all cultures were washed free of treatment media. Approximately a day later (70 hours after culture initiation), all cultures were treated with the mitotic-arresting c-metaphase-inducing alkaloid, colchicine, and processed by conventional cytological techniques onto standard glass microscope slides, fixed

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[1] Post-mitochondrial supernatant (S9) from hepatic homogenates of adult male Sprague Dawley rats pre-treated with Aroclor 1254, plus NAPD(H)-generating co-factors.

in Carnoy's fluid (acetic acid:methanol), air-dried, stained with buffered Giemsa, and finally mounted under DPX resin. Four slides were prepared for each culture analyzed.

Under oil-immersion microscopy, mitotic indices (percent cells in c-metaphase) were determined for each coded culture to be analyzed, as an indirect measure of cytotoxicity. As well, 100 metaphase from each slide were scored for the conventional array of structural chromosome aberrations.

Statistical analysis of the resulting chromosome data was considered by the authors "not [to be] necessary"; they claimed to rely solely on visual inspection for interpretation.

E. Results: Doses chosen for cytogenetic analysis were 50, 250 and 500 ug/ml for lymphocyte cultures from both donors, the HDT being at the limit of solubility.

From both donors, a dose-related decrease in MI was evident (Report Tables 1, 2, attached to this DER), but no significant increases in structural chromosomal damage in any test cultures treated with pirimicarb.

By contrast, the appropriate (positive) responses were elicited in cultures treated with the reference compounds.

Hence, the investigators concluded that pirimicarb was not clastogenic in human lymphocyte cultures.

F. TB-I Evaluation: ACCEPTABLE, in demonstrating pirimicarb was not clastogenic in lymphocyte cultures from two blood donors.

ATTACHMENT (Summary Data Tables)

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PIRIMICARB TOXR 011605

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Reviewed by: Irving Mauer, Ph.D., Geneticist 05-12-95
Toxicology Branch I, HED (7509C)
Secondary Reviewer: Karl P. Baetcke, Ph.D., Chief 5/22/95
Toxicology Branch I, HED (7509C)

DATA EVALUATION REPORT

MRID No.: 434960-07
PC No.: 106101
RD Record No.: S479870
EPA ID No.: 010182-GAO-Pirimicarb Tech
Tox Chem No.: 359C
Project No.: D210945

I. SUMMARY

STUDY TYPE: (84-4) Mutagenicity - Other genotoxicity: DNA damage/repair in vivo (HPC/UDS)

CHEMICAL: Pirimicarb

SPONSOR: Zeneca, Wilmington, DE

TESTING FACILITY: (ICI, now Zeneca) Central Toxicology Laboratory (CTL), Alderley Park (UK)

TITLE OF REPORT: Pirimicarb: Assessment for the induction of unscheduled DNA synthesis in rat hepatocytes in vivo

AUTHOR(S): J.C. Kennelly

STUDY NUMBER: CTL/P/2824 (Study No. SR0367)

DATE ISSUED: March 15, 1990

EXECUTIVE SUMMARY: Male Sprague Dawley rats were gavaged once at doses of 50, 100 and 250 mg/kg, and hepatocyte (HPC) prepared in culture to sample repair unscheduled DNA synthesis (UDS), measured as increased net nuclear (silver) grain counts (NNGC), at two sampling times (4 and 12-hours post-dose).

There was no induced increase in NNGC at any dose up to clinically toxic MTDs (200-250 mg/kg).

TB-I Evaluation: ACCEPTABLE

II. DETAILED REVIEW:

A. Test Material: Pirimicarb

Description: Cream-colored powder
Batches (Lots): [not provided]
Purity (%): 98.3
Solvent/carrier/diluent: Corn oil

B. Test Organism: Rodent

Species: Rat
Strain: Alpk:APf Sprague Dawley
Weights: Males (only): 204-273 g.
Source: ICI Barriered Animal Breeding Unit (currently Zeneca), Alderley Park (UK)

C. Study Design (Protocol): This study was designed to assess the genotoxic potential of the test article when administered in vivo to male Sprague-Dawley rats, and sampling primary hepatocyte cultures for unscheduled DNA synthesis, according to established (published) procedures, as modified below (Section D).

A Statement of Quality Assurance measures (inspections/audits) was provided.

A Statement of adherence to Good Laboratory Practice (GLP) was provided.

D. Procedures/Methods of Analysis: Following preliminary toxicity testing,[1] groups of male rats (5/dose group) were gavaged once at three dose levels (50, 100, 200 mg/kg), and sacrificed at two post-dose times (4 hours and 24 hours) in order to sample target tissue cells (hepatocytes, see below). In addition to corn oil solvent controls, separate groups of rats were dosed with the genotoxic carcinogen, 6-p-dimethylaminophenylazo-benzthiazole (6BT), and also sacrificed at the same sampling times.

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[1] Two studies mentioned:

(1) An older assay, contained in ICI Report No. HO-1H/P/67, which recorded an oral LD_{50} value for pirimicarb as "168-221 mg/kg" in "females" (!sic!) of the same strain.

(2) Concurrently with this submission (See: E. RESULTS, below), in which groups of four male Alpk:APf Sprague Dawley rats were dosed at 150, 200 and 250 mg/kg, and observed over a four-day period.

Two independent experiments were conducted at each time point.

At each sampling time, hepatocytes were established in culture on sterile coverslips, following perfusion and excision of the liver from each animal. After a settling-down period (up to 2 hours), each culture was exposed to tissue culture medium containing tritiated thymidine (3HTdR) and returned to the incubator for 4 hours, at which time cultures were washed with medium containing a surfeit of unlabelled thymidine (a "cold chase," meant to remove excess radiolabel from cultures). Cultures were then re -incubated overnight. At least 2 to 3 slides were prepared per treatment.

Next day, coverslip cultures were fixed with Carnoy's (glacial acetic acid: methanol), air-dried, then mounted cell side out on standard glass microscope slides, and dipped under darkroom safe-lighting in Ilford K-2 photographic emulsion. After removal and "gelling" of the emulsion, treated slides were placed in microscope boxes, sealed and stored in a refrigerator.

After 14 days exposure at 4°C, slides were developed and fixed by conventional photographic procedures, stained with haemalum, counterstained with eosin-Y phloxine, dehydrated and covered with DPX mounting resin.

At least 25 (normally 50) morphologically-unaltered cells per coded slide (accumulating at least 100 cells per animal) were examined for nuclear grain counts (number of silver grains over each nucleus), for grain counts over adjacent nuclear-sized cytoplasmic areas, and mean net nuclear grain count determined (NNGC, calculated as nuclear count less cytoplasmic count); as well, percent of cells in repair was recorded for each animal, both determinations representing unscheduled DNA synthesis (UDS), i.e., an attempt to repair damage to DNA damage, separate from the normal DNA replicative (cell division) cycle.

A positive (UDS) response at any individual dose level is recorded when the mean NNGC is 5 or greater; but this response must be reproducible between independent trials.

Silver grain data were accumulated by a microscope-mounted image analyzer linked to a computer. Strict criteria were adhered to for: (a) Assay acceptance, by considering parameters for background counts among solvent slides, (no more than 30-40 cytoplasmic counts; cytoplasmic counts of 40 or more are unacceptable), and normally responsive positive control preparations (NNGC

should be 5+, with at least 20% of cells in repair); as well as for: (b) Definition of a genotoxic response in test series.

E. Results: In concurrent preliminary oral toxicity testing, one of 4 males given 250 mg/kg died the day after dosing. Hence, the next lower dose, 200 mg/kg, (which was non-lethal) was considered the MTD, and selected as the highest dose to be tested (HDT) in the main assay; two lower doses, 50 and 100 mg/kg, completed the test article dose schedule.

In the main assay, 4/10 animals died at the HDT, 200 mg/kg (Report APPENDIX B, attached), and signs of acute toxicity were evident (salivation, trembling, piloerection) in mid dose (100 mg/kg) animals, but no deaths. Thus, in the longer sampling trial (12 hours), 100 mg/kg was selected as the HDT, supplemented by the one lower dose level, 50 mg/kg. Hepatocytes cultured from low (50 mg/kg) and mid-dose (100 mg/kg) pirimicarb-treated rats were apparently morphologically normal, and showed no increased net nuclear grain counts greater than 0, nor any increased percentage of cells in repair (Report Tables 1 and 2, attached here). By contrast, the positive control (6BT) responded appropriately, with mean NNGC 10-12 times background, and 60-70% cells in repair.

Hence, the author concluded that pirimicarb did not induce repair UDS in hepatocytes of rats orally dosed up to the MTD, 200 mg/kg.

F. TB-I Evaluation: ACCEPTABLE, at demonstrating negative results (no increased UDS) in primary hepatocytes from male Sprague-Dawley rats treated acutely with pirimicarb doses up to clinically toxic doses, 200-250 mg/kg.

ATTACHMENTS: Data Tables

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PIRIMICARB TOXR 011605

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PIRIMICARB

Mutagenicity---Chromosome damage in vivo (Mouse MT) (84-2b)

EPA Reviewer: Irving Mauer, PH.D. Date: 05-18-95

Immediate Office, Toxicology Branch-I (7509C)
Secondary Reviewer: Karl P. Baetcke, Ph.D. Date: 5/22/95
Immediate Office, Toxicology Branch-I (7509C)

DATA EVALUATION RECORD

MRID No.: 434960-08
PC No.: 106101
RD Record No.: S479870
EPA ID No.: 010182-GAO-Pirimicarb Tech.
Tox Chem. No.: 359C
Project No.: D210945

I. SUMMARY

STUDY TYPE: (84-2b) Mutagenicity---Chromosome damage in vivo (Mouse MT)

CHEMICAL: Pirimicarb

SPONSOR: Zeneca, Wilmington, DE

TESTING FACILITY: (ICI; successor, Zeneca) Central Toxicology Laboratory (CTL), Alderley Park (UK)

TITLE OF REPORT: PIRIMICARB (TECHNICAL): AN EVALUATION IN THE MOUSE MICRONUCLEUS TEST

AUTHOR(S): K. Jones and C. A. Howard

STUDY NUMBER: CTL/P/2641 (SMO310)

DATE ISSUED: 29 Aug 1989

EXECUTIVE SUMMARY: Male and female mice were orally intubated once at two doses (50% and 80% of the MLD), and bone marrow polychromatic erythrocytes scored for the presence of micronuclei M-PCE, representing the results of chromosome aberrations.

No increased M-PCE were induced at dose levels causing cytotoxicity, 43.3 and 69.3 mg/kg.

TB-I EVALUATION: ACCEPTABLE

II. DETAILED REVIEW

A. TEST MATERIAL: Pirimicarb Technical

Description:	Cream-colored powder
Batches (Lots):	[Not provided]
Purity (%):	97.3%
Solvent/carrier/diluent:	Corn oil

B. TEST ORGANISM: Rodent

Species:	Mouse
Strain:	C57Bl/CJfCD-1/Alpk
Age:	9-11 weeks
Weights - males:	24.2-28.6 g
females:	18.6-21.2 g
Source:	Barriered Animal Breeding Unit, Alderley Park (UK)

C. STUDY DESIGN (PROTOCOL): This study was designed to assess the clastogenic potential of the test article when administered in vivo to mice, and the incidence of micronuclei in polychromatic erythrocytes determined, according to established (published) procedures and FIFRA Test Guidelines.

A Statement of Quality Assurance measures (inspections/audits) was provided.

A Statement of adherence to Good Laboratory Practice (GLP) was provided.

D. PROCEDURES/METHODS OF ANALYSIS: Following preliminary toxicity testing (at single doses of 50, 100, 125, 150 mg/kg), groups of mice (5/sex/dose) were intubated at single doses of 43.3 or 69.3 mg/kg, and sacrificed 24, 48 and 72 hours later (in order to include at least one complete cell cycle). In addition to corn oil solvent controls, two final groups (five males, five females) were treated with the clastogen, cyclophosphamide (CP, 65 mg/kg), and sacrificed 24 hours later.

At scheduled sacrifice, bone marrow cells smears were smeared onto glass microscope slides by conventional cytological technique, and stained with polychrome methylene blue and eosin. Coded slides were examined (1000 polychromatic erythrocytes per slide) for the presence of micronuclei (M-PCE). In addition, 1000 cells were counted to determine the ratio of PCE to total erythrocytes (as an indirect measure of cytotoxicity).

50

PCE data were analyzed by ANOVA for trend, and group differences by one-sided Student's t-test.

E. RESULTS: In preliminary toxicity testing a median lethal dose (MLD) was estimated by linear log = dose interpolations as 86.6 mg/kg for both sexes. The doses selected for the main assay were 43.3 and 69.3 mg/kg, representing 50% and 80%, respectively, of the MLD.

In the main assay, no statistically significant increases over solvent background in M-PCE were found in pirimicarb-treated male or female mice at either of the two dose levels, or at any sampling time (Report Tables 1 to 3, attached). Dose-dependent reductions in percent of PCE were evident in test animals at 24 hours (Report Table 4) attributable mainly to treated females (Report Table 6), indicating the test article was cytotoxic at the doses tested. Finally, the positive control responded appropriately, with significantly increased in M-PCE 10 fold background.

F. TOX EVALUATION: ACCEPTABLE

ATTACHMENT: Data Tables

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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.

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Chemical:	**Pirimicarb**
PC Code:	**106101**
HED File Code	**13000 Tox Reviews**
Memo Date:	***07/13/95***
File ID:	**TX011605**
Accession Number:	***412-02-0281***

HED Records Reference Center
04/23/2002