5-18-94
L. HANSEN

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

010979

MAY 18 1994

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

**MEMORANDUM**

SUBJECT: TERBUTHYLAZINE. ID NO. 080814. Evaluation of Acute Toxicity and Mutagenicity Studies; Upgrades/ Supplements for Mutagenicity and Rabbit 28 Day Dermal Studies Submitted to Support Reregistration of Terbuthylazine.

Tox. Chem. No.: 125B
PC No.: 080814
Submission No.: S456669
Barcode No.: D203085
(subordinate to D198449)

FROM: William B. Greear, M.P.H.
Section IV, Tox. Branch I
Health Effects Division (7509C)
Linda T. Hansen (for) 5/10/94

TO: Bruce Sidwell, Manager, PM Team 53
Virginia Dietrich, Reviewer, PM Team 53
Special Review and Reregistration Division (7508W)

THRU: Marion P. Copley, D.V.M., D.A.B.T., Section Head
Section IV, Tox. Branch I
Health Effects Division (7509C)
Marion Copley 5/14/94

**CONCLUSIONS:**

The following acute and mutagenicity studies were reviewed and are summarized below:

**81-1 Acute Oral Toxicity**

MRID No. 416033-04 (DER attached): In an acute oral toxicity study, 5 male and 5 female RAIf(SPF) rats were administered 2000 mg/kg GS 13529 (terbuthylazine, technical) in distilled water by gavage and were observed for 14 days. Treatment-related clinical signs included piloerection, hunched posture, dyspnea and reduced locomotor activity. No clinical signs were observed after Day 6. One female rat died during the study and was reported to have an abnormal spotted thymus at sacrifice.

**$LD_{50}$: >2000 mg/kg for males and females**
**Tox. Category: III**
**Core-classification: Acceptable.** This study satisfies

1

the guideline requirement (81-1) for an acute oral toxicity study.

MRID Nos. 419077-02/418787-01 (DER attached): In an acute oral toxicity study, 5 male and 5 female OFA.SD rats per group were administered TK 12669/1 (terbuthylazine, technical) in distilled water by gavage at dose levels of 0, 1000, 1590 or 2510 mg/kg, and the animals observed for 14 days. Treatment-related signs of toxicity were observed for up to 2 days after dosing and included subdued behavior, diarrhea, piloerection and prostration. Body weight gain was adversely affected in males at all doses and in high-dose females. Mortality was observed at all doses. All animals that died on study had lung congestion. No gross lesions were observed in surviving rats.

**$LD_{50}$: between 1000 - 1590 (males); 1,503 mg/kg (females); 1,590 mg/kg (males and females combined, Bliss Method)**
**Tox. Category: III**
**Core-classification: Acceptable.** This study satisfies the guideline requirement (81-1) for an acute oral toxicity study.

**81-2 Acute Dermal Toxicity**

MRID No. 419077-03 (DER attached). In an acute dermal toxicity study, TK 12 669/1 (terbuthylazine, technical) was applied to the intact skin of OFA Sprague-Dawley rats (5/sex) at 2000 mg/kg (limit dose) for 24 hrs. No treatment-related signs of toxicity were noted with respect to mortality, body weight gain, clinical signs, cutaneous effects or gross lesions.

**$LD_{50}$: >2000 mg/kg, males and females**
**Tox. Category: III**
**Core-classification: Acceptable.** This study satisfies the guideline requirement (81-2) for an acute dermal toxicity study.

**81-3 Acute Inhalation Toxicity**

MRID No. 416033-05 (DER attached). In an acute inhalation limit test, 5 male and 5 female Tif:RAIf rats were exposed (nose only) to GS13429 (terbuthylazine, technical) for 4 hrs at concentrations of 0 or 5.324 mg/L. The MMAD, calculated assuming a log-normal distribution, was 2.1-2.4 $\mu$m (without using this assumption, the MMAD was approximately 9 $\mu$m). Following exposure, animals were observed for 14 days. Treatment-related signs of toxicity in exposed animals included piloerection, dyspnea, reduced locomotor activity (moderate to severe through day 1; slight, thereafter) and

hunched posture (slight to moderate through day 2). Recovery was complete by day 8. Decreased body weight gain was observed in males; females showed decreases at week 1 but recovered by the end of week 2.

**$LC_{50}$50: >5.3 mg/L, males and females**
**Tox. Category: IV**
**Core-classification: Acceptable.** This study satisfies the guideline requirement (81-3) for an acute toxicity study.

**81-4 Primary Ocular Irritation**

MRID No. 416033-06 (DER attached): In a primary eye irritation study, 49 mg (0.1 ml) GS 13529 (terbuthylazine, technical) was instilled into the conjunctival sac of 3 female New Zealand White rabbits. The test material was not washed from the eyes. One hour after administration, 2 animals exhibited iritis (severity score = 1) which resolved by 24 hrs.

**Minimal eye irritant**
**Tox. Category: IV**
**Core-classification: Acceptable.** This study satisfies the guideline requirement (81-4) for a primary eye irritation study in rabbits.

MRID No. 419077-04 (DER attached): In a primary eye irritation study, 52 mg (0.1 ml) TK 12669/1 (terbuthylazine technical) was instilled into the conjunctival sac of one eye of 6 male New Zealand White rabbits. Positive findings included iritis (grade 1) which resolved by 72 hrs and conjunctival redness (grade 2) which improved to grade 1 by 48 hrs.

**Moderate eye irritant**
**Tox. Category: III**
**Core-classification: Acceptable.** This study satisfies the guideline requirement (81-4) for a primary eye irritation study in rabbits.

**81-5 Primary Dermal Irritation**

MRID No. 419077-05 (DER attached): In a primary dermal irritation study, 6 male New Zealand White rabbits received an unknown amount of TK 12669/1 (terbuthylazine, technical) on the intact skin for 4 hrs. Two animals showed signs of slight erythema at 1 hr, but not 24 hrs. No other adverse dermal signs were reported. The Primary Irritation Index was calculated to be 0.0.

**Very slightly irritating**

**Tox. Category: IV**
**Core-classification: Supplementary (upgradable).** May be upgraded to acceptable with submission of additional information regarding the dose calculations so that the study author's dose calculation may be verified.

**81-6 Dermal Sensitization Potential**

MRID No. 419077-06 (DER attached): The dermal sensitization potential of TK 12669/1 (terbuthylazine, technical) was evaluated in Dunkin-Hartley guinea pigs (10/sex/group) in a maximization study. For induction, animals were injected twice with 0.1 ml each of 8.5 mg/L test material in water, a 50:50 mixture of 8.5 mg/L test material and 50% Freund's adjuvant and 50% Freund's adjuvant on study day 1 (each animal received a total of 6 injections). This was followed on day 8 by dermal application of 0.5 ml 10% sodium lauryl sulfate, and on day 9 by 0.5 ml of an aqueous 68% test material paste. This was followed 11 days later by application of 0.5 ml of 68% paste to a virgin site (challenge phase). No positive dermal sensitization response was observed in treated animals. A positive response was observed in the DNCB positive control group.

**Not a sensitizer under conditions of this study.**
**Core-classification: Acceptable.** This study satisfies the guideline requirement (81-6) for a dermal sensitization study in guinea pigs.

**84-2a Reverse Gene Mutation, Salmonella**

MRID No. 416340-01 (DER attached): In two independently performed Salmonella typhimurium/mammalian microsome plate incorporation assays, strains TA1535, TA1537, TA98 and TA100 were exposed to 20, 78, 313, 1250 or 5000 µg TK 12669/1 (terbuthylazine, technical)/plate with or without S9 activation. The S9 fraction was prepared from Arochlor 1254-induced rat livers and the S9 cofactor mix contained 30% liver homogenate. Dimethyl sulfoxide (DMSO) was used as solvent.

**No cytotoxicity or mutagenicity was observed in any strain at any dose tested in presence or absence of S9 activation. Compound insolubility was reported at ≥1250 µg/plate.** Results with positive controls confirmed the sensitivity of the test system to detect mutagenesis.

**Core-classification: Acceptable.** This study satisfies the guideline requirement (84-2) for a gene mutation study.

Supplements to the following studies are discussed below.

Studies were upgraded and/or Executive Summaries prepared:

**82-2 28-Day Dermal Toxicity, Rabbit**

MRID No. 420598-04 (supplement to MRID no. 405148-02, original study report): See HED Doc. nos. 006728 and 009678 for review and upgrade. An Executive Summary for this study is provided below:

Executive Summary: In a repeated dose dermal toxicity study in rabbits, technical terbuthylazine (97.1% ai) was applied daily to the intact skin of 5 males and 5 females for 29 consecutive days. Test material was moistened with distilled water and applications of 0 (distilled water), 0.05, 0.5 or 500 mg/kg/day were applied for 6 hrs/day under occlusive wrap.

At 500 mg/kg/day, reduced body weight gain was observed in males (-36% of controls at Day 28) and females (-39%). Food consumption was also decreased (-76% and -89% of controls during week 1 in males and females; between -11% to -54% of controls at other times). Reduced fecal output was observed sporadically among both sexes. Mortality occured in one female, preceded by cachexia, hypothermia and muscle wasting. **The LEL of 500 mg/kg/day is based on decreased body weight gain, hypothermia, weight loss, cachexia, food consumption and mortality of one female. The NOEL is 5.0 mg/kg/day.**

This study is **core-guideline** and satisfies the guideline requirement (82-2) for a subchronic dermal toxicity study in the rabbit.

**84-2b Mouse Bone Marrow Micronucleus Study**

MRID No. 420598-05 (supplement to MRID 414181-02, original study report): See HED Doc. Nos. 008077 and 009492 for review and study upgrade. An Executive Summary is provided below for this study:

Executive Summary: In a mouse bone marrow micronucleus assay, 8 Tif:MAGF, SPF (NMR-1 derived) mice/sex/dose were administered Belclene® 329 (terbuthylazine, technical) by gavage in 0.5% carboxymethylcellulose at single doses of 1250, 2500 or 5000 mg/kg (Part 1). Animals were sacrificed 24 hrs after dosing. In addition, a total of 24/sex were administered 5000 mg/kg and sacrificed at 16, 24 and 48 hrs after dosing (8 animals/sex at each time point; Part 2).

Deaths were observed at 5000 mg/kg in males (2 at 24 hr and 1 at 24 hr; Part 1) and in females (2 at 24 hr and 2 at 48 hr, Part 1; 1 at 2500 and 5000 mg/kg in Part 2). There was

no indication of a cytotoxic effect in the bone marrow (lowered PCE:NCE ratio) and no indication of other clinical signs of toxicity. However, the test material was tested up to the limit dose (5000 mg/kg) and mortality was stated by the study author to be treatment-related. **There was not a significant increase in the frequency of micronucleated polychromatic erythrocytes in bone marrow at any sampling time after treatment.**

This study is classified as an **Acceptable** study. It satisfies the guideline requirement (84-2) for an in vivo mouse micronucleus study.

**84-4 Unscheduled DNA Synthesis, Rat Hepatocytes (in vitro)**

MRID No. 420598-06 (See attached DER supplement; submission is supplement to MRID 413918-01, original study report; see HED Doc. nos. 8509 and 9680 for study review and upgrade).

Executive Summary: In a DNA repair study in rat hepatocytes obtained from male TifRAIf (SPF) rats, dose levels of 0.98, 3.9, 15.6, 62.5, 250 or 1000 µg/ml were used to test for DNA damage. A confirmatory test was conducted at concentrations of 0.250, 0.49, 0.98, 3.9, 15.6, 62.5, 250 or 1000 µg/ml. 2-AAF and 4-ABP were used as positive controls.

The test material did not produce a significant increase in mean gross or net nuclei grain counts when tested at levels up to 1000 µg/ml compared to vehicle controls, and a dose-response relationship was not observed. Additionally, the percental distribution of grain counts/nuclei were comparable for test material and vehicle control groups. **There was no evidence of increased DNA synthesis in the study due to treatment with terbuthylazine.**

**Core-classification: Acceptable.** This study satisfies the guideline requirement (84-4) for an unscheduled DNA synthesis study in rat hepatocytes.

**ACTION REQUESTED:**

Acute toxicity studies and a Salmonella gene mutation study on terbuthylazine were submitted by Ciba-Geigy to the Agency for review. In addition, supplemental information for 2 mutagenicity studies and a repeated-dose dermal toxicity study in rabbit were submitted. These data were required to support reregistration and for completion of the Reregistration Eligibility Document (RED) for terbuthylazine. A subordinate bean (D203085) was generated from the main package (D198449) by Tox. Branch I, HED, for submission of the data packages prior to completion of the RED. The RED will be completed following evaluation of terbuthylazine by the Cancer Peer Review Committee.

**FINAL**

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Acute Oral Toxicity

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

| | | |
|---|---|---|
| Principal Reviewer | Sara Lundgaard for<br>Donna Armstrong, Ph.D. | Date 4/8/94 |
| Independent Reviewer | <br>Carrie Rabe, Ph.D. | Date 4/8/94 |
| QA/QC Manager | William L McLellan<br>William McLellan, Ph.D. | Date 4/11/94 |

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 215
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: MCopley
Date: 4/19/94

**DATA EVALUATION REPORT**

**STUDY TYPE:** Acute Oral Toxicity / Rat **GUIDELINE:** §81-1

**PC CODE:** 080814 **MRID No.** 416033-04

**TEST MATERIAL:** GS 13529 technical

**SYNONYMS:** Terbuthylazine; Gardoprim; Primatol M.

**REGISTRANT:** Agricultural Division, Ciba-Geigy Limited, Basel, Switzerland

**TESTING LABORATORY:** Experimental Toxicology, CIBA-GEIGY Limited, Stein, Switzerland

**STUDY IDENTIFICATION:** 891215

**TITLE OF REPORT:** *"TERBUTHYLAZINE, ACUTE ORAL TOXICITY IN THE RAT, TEST NO. 891215"*

**AUTHORS:** H.R. Hartmann

**REPORT DATE:** August 31, 1989

EXECUTIVE SUMMARY: In an acute oral toxicity study, five male and five female rats (strain RAI f (SPF)) were administered 2000 mg/kg of GS 13529 technical in distilled water by gavage and were observed for 14 days. Treatment-related clinical signs included piloerection, hunched posture, dyspnea, and reduced locomotor activity. No clinical signs were observed after day 6. One female rat died during the study, and was reported to have an abnormal spotted thymus.

The results of this study determined that the acute oral $LD_{50}$ was greater than 2000 mg/kg for both sexes placing GS 13529 technical in TOXICITY CATEGORY: III.

CORE CLASSIFICATION: Acceptable; this study satisfies the guideline requirement [§ 81-1] for an acute oral toxicity study.

A. MATERIALS

Test Compound

| | |
|---|---|
| Test material: | GS 13529 technical |
| Description: | Solid |
| Batch number: | WKA 6925 |
| Purity: | 99% |
| Storage condition: | Room temperature |
| Stability: | Not reported; guaranteed by the sponsor |
| Vehicle: | Distilled water |
| Dosing volume: | 10 mL/kg |
| Dose Level: | 2,000 mg/kg |
| Controls: | None were used |

Test Animals

| | |
|---|---|
| Species: | Rat |
| Strain: | Tif: RAI f (SPF) |
| Source: | Animal production, CIBA-GEIGY Limited, Stein, Switzerland |
| Sex: | Males and females |
| Age: | 6-8 weeks |
| Initial Weight: | Males: 204 ± 7.5 on day of administration; Females: 182 ± 7.6 on day of administration; individual animal weights not reported |
| No. animals/dose: | 5/sex |
| Temperature: | 22±2°C |
| Humidity: | 55±10% |
| Photoperiod: | 12 hours |
| Air changes: | approximately 15/hour |
| Housing: | 5/cage; segregated by sex |
| Identification: | Fur staining and cage cards |
| Selection: | Random; approximately matched by weight |
| Feeding: | NAFAG 890 Tox rat chow, *ad libitum* |
| Water: | *Ad libitum* |
| Acclimation: | At least 5 days |

B. TEST PERFORMANCE

Method of administration: Oral gavage

Animals fasted: Overnight, prior to treatment

Dosing: Single administration

Observation period: 14 days

Observation frequency: 1, 3, 5 hours after intubation; thereafter, daily for clinical signs; mortality checks were made twice daily on weekdays and once daily on weekends and holidays

Body weight interval: Immediately before administration and days 7 and 14

Gross pathology: YES X ; NO ____

Histopathology: YES ____; NO X

C. RESULTS

Mortality: One death occurred 2 days after dosing. Results are summarized in Table 1 below.

Table 1. Incidence of Mortalities in Rats Given GS 13529 Technical[a]

| Dose (mg/kg) | (Number Dead/Number tested) | | |
|---|---|---|---|
| | Males | Females | Combined |
| 2,000 | 0/5 | 1/5 | 1/10 |

[a]Data extracted from Test No. 891215, p. 14.

Clinical Observations: Clinical signs, observed within 3 hours after administration, consisted of piloerection, hunched posture, dyspnea, and reduced locomotor activity. No major differences were noted between males and females. Piloerection persisted through day 6 in males and day 5 in females; hunched posture persisted through day 1 in both sexes; dyspnea persisted through day 3 in males and day 4 in females; and reduced locomotor activity persisted only for 3 hours in males and 5 hours in females. Clinical observations on individual rats were not submitted.

Body Weights: Mean body weight gain on day 14 was 35 g for females and 64 g for males. Body weight data from individual rats were not submitted.

Gross Necropsy: The female rat that died during the study had a spotted thymus. No deviations from normal morphology were observed in the remaining nine rats which were killed at the end of the study.

D. REVIEWER'S COMMENTS

The acute oral $LD_{50}$ in this study was determined to be greater than 2000 mg/kg for both sexes. This value corresponds to Toxicity Category III (Caution).

E. STUDY DEFICIENCIES

Only a single dose was tested. This dose was well below the limit dose (5,000 mg/kg). The low mortality observed indicated that higher doses should have been used to adequately assess mortality. In addition, individual animal data for clinical signs and weight gain were not submitted. However, this does not alter the conclusion that the Tox Category is III, therefore, a new study is not required.

F. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed September 4, 1989, was provided.

**FINAL**

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Acute Oral Toxicity

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

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| Independent Reviewer | Carrie Rabe<br>Carrie Rabe, Ph.D. | Date 4/8/94 |
| QA/QC Manager | William L. McLellan<br>William McLellan, Ph.D. | Date 4/11/94 |

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Numbers: 219, 224
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 4/19/99

DATA EVALUATION REPORT

STUDY TYPE: Acute Oral Toxicity

GUIDELINE: §81-1

PC CODE: 080814

MRID No. 419077-02
418787-01

TEST MATERIAL: TK 12669/1 (2-Chloro-4-tert. butylamino-6-ethylamino-s-triazine)

SYNONYMS: Terbuthylazine; Gardoprim; Primatol M; Bellacide 320.

REGISTRANT: Additives Division, Ciba-Geigy, Basel, Switzerland

TESTING LABORATORY: Hazleton France, L'Arbresle Cedex, France

STUDY IDENTIFICATION: Report number, 012333; project number, 904139

TITLE OF REPORT: *"TEST TO EVALUATE THE ACUTE TOXICITY FOLLOWING A SINGLE ORAL ADMINISTRATION (LD 50) IN THE RAT"*

AUTHOR: O. Mercier

REPORT DATE: February 8, 1991

EXECUTIVE SUMMARY: In an acute oral toxicity study, 5 male and 5 female rats (strain OFA.SD.) per group were administered TK 12669/1 in purified water once via gavage at dose levels of 0, 1,000, 1,590, or 2,510 mg/kg, and the animals were observed for 14 days. Treatment-related signs of toxicity were observed for up to 2 days after dosing and included subdued behavior, diarrhea, piloerection, and prostration. Body weight gain was adversely affected in males at all doses and in high-dose females. Mortality was observed at all doses. All animals that died on study had lung congestion. No gross lesions were observed in surviving rats.

The results of this study determined that the acute oral $LD_{50}$ was between 1,000 and 1,590 mg/kg for males, 1,503 mg/kg (1,030-2,194) for females, and 1,590 mg/kg (923-2,738) for both sexes combined (Bliss' method), placing TK 12669/1 in TOXICITY CATEGORY: III.

CORE CLASSIFICATION: Acceptable; this study satisfies the guideline requirement [§ 81-1] for an acute oral toxicity study.

A. MATERIALS

Test Compound

| | |
|---|---|
| Test material: | TK 12669/1 |
| Sample reference: | 01255 EO 003 |
| Description: | White powder |
| Batch number: | SG 6925 |
| Purity: | 96.4% |
| Storage condition: | Room temperature, in the dark |
| Stability: | Not reported; expiration date, 9/92 |
| Vehicle: | Purified water |
| Dose Levels: | 1,000, 1,590, or 2,510 mg/kg of test material prepared as a suspension in purified water |
| Dose volume: | 10 mL/kg |
| Controls: | Purified water |

Test Animals

| | |
|---|---|
| Species: | Rat |
| Strain: | OFA.SD (Sprague-Dawley) |
| Source: | Iffa-Credo, L'Arbresle Cedex, France |
| Sex: | Males and females |
| Age: | 5-7 weeks old |
| Weight: | Males 146-161 g on study day 1, females 121-150 g on study day 1 |
| No. animals/dose: | 5/sex |
| Temperature: | 21-25°C |
| Humidity: | 34-90% |
| Photoperiod: | 12 hours |
| Air changes: | At least 10/hour |
| Feeding: | U.A.R., formula A.04 (rat-mouse maintenance diet), *ad libitum* |
| Water: | *Ad libitum* |
| Identification: | Ear punch and cage cards |
| Selection: | Allocated to groups according to weight so that the mean weight of each group was comparable to others of the same sex |
| Housing: | 5/cage; segregated by sex |
| Acclimation: | At least 8 days |

B. TEST PERFORMANCE

Method of administration: Oral gavage

Animals fasted: At least 18 hrs prior to treatment

Dosing: Single administration

Observation period: 14 days after dosing

Observation frequency: 15 minutes after intubation; 1, 2, 4 hours; thereafter, daily

Body weight interval: Body weights were measured on study days -1, immediately before dosing on day 1, and on days 8 and 15, or at time of death.

Gross pathology: YES X ; NO ____

Histopathology: YES ___; NO X

C. RESULTS

Mortality: A dose-related increase in mortality was observed. Results are summarized below in Table 1.

Table 1. Incidence of Mortalities in Rats Given TK 12669/1[a]

| Dose (mg/kg) | (Number Dead/Number tested)[b] | | |
|---|---|---|---|
| | Males | Females | Combined |
| 0 | 0/5 | 0/5 | 0/10 |
| 1,000 | 2/5 | 1/5 | 3/10 |
| 1,590 | 3/5 | 3/5 | 6/10 |
| 2,510 | 2/5 | 4/5 | 6/10 |

[a]Data extracted from Report No. 012333, p. 21.

[b]All deaths occurred within 3 days of dosing.

Clinical Observations: Subdued behavior was observed in all treated animals 4 hours after dosing, and persisted until day 3 in two, three, and four animals, at 1,000, 1,590, and 2,510 mg/kg, respectively. Diarrhea was observed on day 3 in one rat at 1,000 mg/kg and in two rats at 1,590 mg/kg, but not in animals at 2,510 mg/kg. Piloerection was observed in five to six animals in each treatment group on day 2; prostration was observed in five to six animals at 1,590 and 2,510 mg/kg on day 2. All surviving rats had recovered by day 4. Data describing clinical signs in individual rats were not submitted.

Body Weights: Mean body weight gain of males at the low-, mid-, and high-doses were 67%, 49% and 60% of control, respectively. Insufficient numbers of animals survived at the mid-dose for statistical analysis, but the weight gains of low- and high-dose males were significantly lower than control. Mean body weight gains of females at the low-, mid-, and high-doses were 96%, 92%, and 39% of control, respectively. Weight gain of low-dose females was comparable to controls. Insufficient survival occurred at the mid- and high-doses for statistical analyses.

Gross Necropsy: All animals that died during the study were observed to have marked congestion of the lungs. One male at 1,590 mg/kg was observed with abdominal ascites (cloudy liquid). All animals that died prior to the terminal sacrifice, except one, had autolysis of the alimentary canal. No abnormalities were observed in animals killed at the end of the study.

D. REVIEWER'S COMMENTS

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The acute oral $LD_{50}$ in this study was calculated to be 1,590 mg/kg (923-2,738) for both sexes combined (Bliss's method). Body weight was adversely affected in males at all doses and in high-dose females. Mortality was observed at all doses. All animals that died on study had lung congestion.

E. STUDY DEFICIENCIES

Information on clinical observations was presented in summary tables with data from both sexes combined. No information was provided on clinical observations for control animals or individual treated rats.

F. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed February 12, 1991, was provided.

**FINAL**

010979

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Acute Dermal Toxicity

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

| | | |
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| Principal Reviewer | Sara Lundgaard for<br>Donna Armstrong, Ph.D. | Date 4/8/94 |
| Independent Reviewer | Carrie Rabe<br>Carrie Rabe, Ph.D. | Date 4/8/94 |
| QA/QC Manager | William L. McLellan<br>William McLellan, Ph.D. | Date 4/8/94 |

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 220
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 4/19/91

**DATA EVALUATION REPORT**

STUDY TYPE: Acute Dermal Toxicity - Rat GUIDELINE: §81-2

PC CODE: 080814 MRID No. 419077-03

TEST MATERIAL: TK 12669/1 (2-chloro-4-tert. butylamino-6-ethylamino-s-triazine

SYNONYMS: Terbuthylazine; Gadoprim; Primatol M.

REGISTRANT: Additives Division, Ciba-Geigy Limited, Basel, Switzerland

TESTING LABORATORY: Hazleton France, L'Arbresle Cedex, France

STUDY IDENTIFICATION: Project number, 904151; Report number, 012319

TITLE OF REPORT: *"TEST TO EVALUATE THE ACUTE TOXICITY FOLLOWING A SINGLE CUTANEOUS APPLICATION (LIMIT TEST) IN THE RAT"*

AUTHOR: O. Mercier

REPORT DATE: February 8, 1991

EXECUTIVE SUMMARY: In an acute dermal toxicity study, TK 12 669/1 (96.4% pure) was applied the the intact skin of OFA Sprague-Dawley rats (5/sex) at 2,000 mg/kg (limit dose) for 24 hours. No treatment-related signs of toxicity were noted with respect to mortality, body weight gain, clinical signs, cutaneous effects, or gross lesions.

**The results of this study determined that the acute dermal $LD_{50}$ was greater than 2,000 mg/kg, placing TK 12 669/1 in TOXICITY CATEGORY: III**

CORE CLASSIFICATION: **Acceptable**; this study satisfies the guideline requirement [§ 81-2] for an acute dermal toxicity study.

A. MATERIALS

Test Compound

| | |
|---|---|
| Test material: | TK 12669/01 |
| Physical description: | White powder |
| Batch number: | SG 6925 |
| Identification number: | 01255 E0 003 |
| Purity: | 96.4% |
| Storage condition: | Room temperature in the dark |
| Stability: | Not reported; expiration date, 9/92 |
| Vehicle: | Distilled water |
| Dose Level: | 2,000 mg/kg undiluted test material (limit test), prepared as a paste (at 76.46% w/v in purified water) and applied at a volume of 2.616 mL/kg |
| Controls: | No controls were used. |

Test Animals

| | |
|---|---|
| Species: | Rat |
| Strain: | OFA.SD. (IOPS Caw) |
| Source: | Iffa-Credo, 69592 L'Arbresle Cedex, France |
| Sex: | Males and females |
| Age: | 6-8 weeks |
| Weight: | Males 230-245 g on Day 1 (immediately before application)<br>Females 209-219 g on Day 1 (immediately before application) |
| No. animals/dose: | 5/sex |
| Temperature: | 21-25°C |
| Humidity: | 34-90% |
| Photoperiod: | 12 hours |
| Air changes: | 8/hour |
| Feeding: | U.A.R. formula A.04 (rat/mouse maintenance diet, pelleted), *ad libitum* |
| Water: | *Ad libitum* |
| Acclimation period: | 7 days |
| Housing: | Individual |
| Identification: | Ear punch and cage cards |
| Selection: | By weight, to obtain homogeneous weights by sex |

B. TEST PERFORMANCE

Skin preparation: The hair on the back and flanks of each animal was clipped (1/20th mm high) one day prior to testing.

Application: Test material, prepared as a paste, was applied to the test site and the area covered with a semi-occlusive dressing and an elastic

sleeve. The test site was approximately 10% of the animals body surface. After 24 hours, the dressing was removed and residual test material rinsed off with warm water.

Observation period: 14 days

Observation frequency: Observations for clinical signs and mortality were made 15 minutes, 1, 2, and 4 hours after administration and once daily thereafter. Cutaneous lesions were evaluated after removal of the test material and daily, thereafter.

Body weight intervals: Body weights were measured immediately before application (on study day 1) and at days 8 and 15.

Gross pathology: YES X ; NO ____

Histopathology: YES ____; NO X

C. RESULTS

Mortality: No mortality was observed. Animals were sacrificed on day 15.

Clinical Observations: No clinical signs of toxicity were observed in any animal.

Dermal Observations: Neither erythema nor edema was observed in any animal.

Body Weights: All animals gained weight. Mean body weights are summarized in Table 1 below.

Table 1. Mean Body Weights[a]

| Group | Day 1 | Day 8 | Day 15 |
|---|---|---|---|
| Males | 236 g | 257 g | 301 g |
| Females | 214 g | 231 g | 252 g |

[a]Data extracted from Report No. 012319, pp. 24-25.

Gross Necropsy: No compound-related gross findings were observed. (Individual gross pathology findings were not reported.)

$LD_{50}$ Determination: The estimated acute dermal $LD_{50}$ for males and females was > 2,000 mg/kg (limit test). This value corresponds to Toxicity Category III (Caution).

D. REVIEWER'S COMMENTS

No effects on survival or body weight were observed. Individual clinical signs and gross pathology data were not presented. However, no effects on these parameters were reported. TK 12669/1 was not irritating to the skin under the conditions of this study.

E. STUDY DEFICIENCIES

There were no deficiencies in the study.

E. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed February 12, 1991, was provided.

**FINAL**

010979

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Acute Inhalation Toxicity - Rat

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

Principal Reviewer Sara Lundgaard for Donna Armstrong, Ph.D. Date 4/12/94

Independent Reviewer Carrie Rabe, Ph.D. Date 4/12/94

QA/QC Manager William L. McLellan William McLellan, Ph.D. Date 4/12/94

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 216
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 4/19/90

**DATA EVALUATION REPORT**

**STUDY TYPE:** Acute Inhalation Toxicity **GUIDELINE:** §81-3

**PC CODE:** 080814 **MRID No.** 416033-05

**TEST MATERIAL:** GS 13529 technical

**SYNONYMS:** Terbuthylazine; Gardoprim; Primatol M

**REGISTRANT:** Agricultural Division, CIBA-GEIGY Limited, Basel, Switzerland

**TESTING LABORATORY:** Experimental Toxicology, CIBA-GEIGY Limited, Stein, Switzerland

**STUDY IDENTIFICATION:** 891219

**TITLE OF REPORT:** *"ACUTE INHALATION TOXICITY IN THE RAT"*

**AUTHORS:** H.R. Hartmann

**REPORT DATE:** December 18, 1989

EXECUTIVE SUMMARY: In an acute inhalation limit test, five male and five female rats (Tif: RAI f) were exposed (nose-only) to GS 13529 technical for 4 hours at concentrations of 0 or 5.324 mg/L. The MMAD, calculated assuming a log-normal distribution, was 2.1-2.4 μm (without using this assumption, the MMAD was approximately 9 μm). Following exposure, animals were observed for 14 days. Treatment-related signs of toxicity in exposed animals included piloerection, dyspnea, and reduced locomotor activity (moderate to severe through day 1; slight, thereafter) and hunched posture (slight to moderate through day 2). Recovery was complete by day 8. Decreased body weight gain was observed in males; females exhibited a decrease at 1 week, but recovered to control by the end of week 2.

**The results of this study determined that the acute inhalation $LC_{50}$ was greater that 5.300 mg/L for both males and females placing it in TOXICITY CATEGORY IV.**

CORE CLASSIFICATION: **Acceptable**; this study satisfies guideline requirement [81-3] for an acute inhalation toxicity study in rats.

A. MATERIALS

Test Compound

| | |
|---|---|
| Test material: | GS 13529 technical |
| Description: | Solid |
| Batch number: | WKA 6925 |
| Purity: | 99% |
| Density: | Not reported |
| Storage condition: | Room temperature |
| Stability: | Not reported |
| Exposure level: | 5.324 mg/L |
| Exposure period: | Single, 4-hour period |
| Control: | Filtered humidified air |

Test Animals

| | |
|---|---|
| Species: | Rat |
| Strain: | Tif: RAI f (SPF) |
| Source: | Animal production, CIBA-GEIGY Limited, Basel, Switzerland |
| Sex: | Males and females |
| Age: | 7-8 weeks at start of study |
| Weight: | 183-213 g at start of study (all animals combined; individual animal data not provided) |
| No. animals/dose: | 5/sex |
| Temperature: | 22± 2°C |
| Humidity: | 55±10% |
| Air changes: | 15/hour |
| Photoperiod: | 12 hours |
| Feeding: | NAFAG 890 Tox rat chow, *ad libitum* |
| Water: | *Ad libitum* |
| Identification: | Fur staining and cage cards |
| Selection: | Not reported |
| Housing: | 5/cage; sexes separated |
| Acclimation: | At least 5 days |

B. TEST PERFORMANCE

Inhalation Chamber: A cylindrical stainless steel exposure chamber fitted with nose-only exposure tubes was used. The chamber had an internal active volume of <1 L. Aerosol was delivered to each animal via individual delivery tubes (flow rate, 2 L/min) and exhaled air was removed via exhaust tubes.

Generation of Test Atmospheres: The test aerosol was generated using a brush-feed micronizing jet mill. The test material was micronized by impaction of particles against one another in two opposing air streams. A cyclone-type classifier was used to ensure that only particles of the desired size were allowed to leave the mill. The test aerosol was diluted with filtered humidified air to yield a total flow of 48 L/min. Controls were exposed to humidified air at an air flow rate of 32 L/min.

Analytical Determinations: Concentration of the test aerosol in the chamber was determined gravimetrically 5 times during the exposure period. Particle size analysis was conducted using an APS-33 Aerodynamic Particle Sizer. The number of determinations were not reported, but data were presented from two determinations that were reported to give the minimum and maximum values.

Table 1. Atmospheric Concentration and Particle Size Distribution of the Test Material, GS 13529 technical[a]

| Mean Gravimetric Exposure Conc.[b] (mg/L) | Nominal Conc. (mg/L) | MMAD[c] ($\mu$m) | GSD ($\mu$m) |
|---|---|---|---|
| 5.324 ± 0.149 | 5.443 | 2.1-2.4[d] | 1.6-1.7 |

[a]Data extracted from Test No. 891219, Table 1, p. 17.
[b]Mean ± standard deviation
[c]Mass median aerodynamic diameter
[d]Calculated assuming log-normal distribution; maximum and minimum provided

Chamber monitoring: Temperature, relative humidity, and oxygen content were monitored 5 times during exposure. Airflow was monitored an unspecified number of times during exposure. Results are presented in Table 2.

Table 2. Chamber Environmental Conditions[a,b]

| Exposure Level (mg/L) | Temperature (°C) | Relative Humidity (%) | Oxygen Content (%) | Flow Rate (L/min) |
|---|---|---|---|---|
| Control | 22.0 ± 0.3 | 36 ± 1 | 21.0 ± 0.0 | 32 |
| 5.324 | 23.6 ± 0.1 | 44 ± 0.5 | 21.0 ± 0.0 | 48 |

[a]Data extracted from Test No. 891219, Table 1, p. 17.
[b]Mean ± standard deviation

Exposure period: 4 hours

Observation Period: 14 days

Observation Frequency: During and immediately following exposure, then daily, thereafter

Body Weight Interval: Prior to exposure and on days 7 and 14

Gross Pathology: YES X ; NO ____

Histopathology: YES ____; NO X

C. RESULTS

Mortality: No mortalities were observed.

Clinical Observations: Individual animal data were not reported. Piloerection, hunched posture, dyspnea, and reduced locomotor activity were observed in treated males and females immediately after exposure. The number of animals was not reported. The severity of the symptoms (excluding hunched posture) gradually declined from moderate to severe or slight. Hunched posture was slight immediately after exposure, but became moderate on the day after exposure. These symptoms persisted for up to 7 days after exposure. All animals recovered completely by day 8. No clinical signs were observed in animals from the control group.

Body Weight: Individual animal data were not provided. Both male and female rats exposed to aerosols of GS 13529 technical had depressed weight gain 7 days after exposure. By the end of the study, female body weights recovered to control levels. Male body weights recovered somewhat but continued to lag behind controls. Total body weight gain in treated males was 70% of control. See Table 3 below.

Table 3. Mean Body Weight Data[a,b]

| Group/Sex | Day 0 | Day 7 | Day 14 |
|---|---|---|---|
| **Control** | | | |
| Males | 205 ± 2 | 255 ± 9 | 313 ± 13 |
| Females | 190 ± 13 | 205 ± 6 | 223 ± 11 |
| **5.324 mg/L** | | | |
| Males | 203 ± 9 | 212 ± 7* (83%) | 279 ± 9 (89%) |
| Females | 191 ± 5 | 190 ± 7* (93%) | 230 ± 14*(103%) |

[a]Data extracted from Test No. 891219, Table 3, p. 19.
[b]Mean ± S.D.; values in parentheses represent percent of control
*Significantly different from control, $p \leq 0.05$

Gross Necropsy: The study authors report that there were no treatment-related findings. However, individual gross pathology data were not submitted, and therefore, verification of these results was not possible.

$LC_{50}$ Determination: The study author report that the $LC_{50}$ was greater than 5.300 mg/L for both males an females.

D. REVIEWER'S COMMENTS

The results of this study determined that the acute inhalation $LC_{50}$ was greater that 5.300 mg/L for both males and females placing it in TOXICITY CATEGORY IV. Treatment-related signs of toxicity in exposed animals included piloerection, dyspnea, and reduced locomotor activity (moderate to severe through day 1; slight, thereafter) and hunched posture (slight to moderate through day 2). Recovery was complete by day 8. Decreased body weight gain was observed in males; females exhibited a decrease at 1 week, but recovered to control by the end of week 2.

E. STUDY DEFICIENCIES

Reporting deficiencies included a failure to report clinical signs, body weight, and gross pathology data for individual animals and the recording times for air flow measurements. However, these deficiencies would not alter the conclusions of the study.

E. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed December 22, 1989, was provided.

**FINAL**

010979

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Primary Eye Irritation

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

Principal Reviewer: Sara Lundgaard for Donna Armstrong, Ph.D. Date 4/8/94

Independent Reviewer: Carrie Rabe, Ph.D. Date 4/8/94

QA/QC Manager: William L. McLellan, Ph.D. Date 4/8/94

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 217
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 8/19/99

**DATA EVALUATION REPORT**

**STUDY TYPE:** Primary Eye Irritation **GUIDELINE: §81-4**

**PC CODE:** 080814 **MRID No.** 416033-06

**TEST MATERIAL:** GS 13529 technical

**SYNONYMS:** Terbuthylazine; Gardoprim; Primatol M.

**REGISTRANT:** Agricultural Division, CIBA-GEIGY Limited, Basel, Switzerland

**TESTING LABORATORY:** Experimental Toxicology, CIBA-GEIGY Limited, Stein, Switzerland

**STUDY IDENTIFICATION:** 891216

**TITLE OF REPORT:** *"ACUTE EYE IRRITATION/CORROSION STUDY IN THE RABBIT"*

**AUTHOR:** M. Schneider

**REPORT DATE:** August 9, 1989

EXECUTIVE SUMMARY: In a primary eye irritation study, 49 mg (volume equal to 0.1 mL) GS 13529 technical was instilled into the conjunctival sac of three female New Zealand White rabbits. The test material was not washed from the eyes. One hour after administration, two animals exhibited iritis (severity score = 1). The iritis was resolved by 24 hours.

**The results of this study determined that GS 13529 was a minimal eye irritant, placing it in TOXICITY CATEGORY: IV.**

CORE CLASSIFICATION: **Acceptable**; this study satisfies the guideline requirement [§ 81-4] for a primary eye irritation study in rabbits.

A. MATERIALS

010979

Test Compound

| | |
|---|---|
| Test material: | GS 13529 technical |
| Description: | Solid; the report did not state whether the test material was ground to a fine powder as recommended. |
| Batch number: | WKA 6925 |
| Purity: | 96.4% |
| Storage condition: | Room temperature |
| Stability: | Not reported |
| Vehicle: | None |
| Dose Level: | 49 mg undiluted test material; volume was equivalent to 0.1 mL |

Test Animals

| | |
|---|---|
| Species: | Rabbit |
| Strain: | New Zealand White (KFM-NZW) |
| Source: | Kleintierfarm Madoerin AG, Fuellinsdorf |
| No. of animals: | 3 |
| Sex: | Females |
| Age: | 12-14 weeks |
| Weight: | 2.27-2.97 kg at the start of the study |
| Temperature: | 20±3°C |
| Humidity: | 30-70% |
| Air changes: | Not reported |
| Photoperiod: | 12 hours |
| Feeding: | NAFAG No. 814 standard rabbit pellet, *ad libitum* |
| Water: | *Ad libitum* |
| Identification: | Ear tags |
| Selection: | Animals without preexisting eye defects were selected |
| Housing: | Individual |
| Acclimation: | At least 5 days |

B. TEST PERFORMANCE

Pretest Eye examination: Eyes were examined (method not reported) prior to administration of the test material.

Test Material Application: The test substance was placed in the lower conjunctival sac of the left eye. The eye lids were gently held together for several seconds. The untreated right eye of each animal served as the control. The study report did not indicate that the treated eyes were washed after treatment.

Observation period: Observations for ocular lesions were made at 1, 24, 48, and 72 hours and 7 days after administration of the test article using a slit lamp. Observations for mortality and clinical signs were made daily.

Scoring system: Eyes were examined and scored for ocular lesions using the scoring system of the OECD guidelines. This scoring system was identical to the Draize system.

C. RESULTS

A summary of ocular effects is presented below in Table 1.

Table 1. Summary of Incidences of Ocular Effects[a,b]

| | Observation Intervals | | | | |
|---|---|---|---|---|---|
| | Hour(s) | | | | Days |
| | 1 | 24 | 48 | 72 | 7 |
| Cornea | | | | | |
| Opacity | 0/3 | 0/3 | 0/3 | 0/3 | 0/3 |
| Iris | | | | | |
| Iritis | 2/3 (1) | 0/3 | 0/3 | 0/3 | 0/3 |
| Conjunctivae | | | | | |
| Redness | 3/3 (1) | 3/3 (1) | 1/3 (1) | 0/3 | 0/3 |
| Chemosis | 3/3 (1) | 0/3 | 0/3 | 0/3 | 0/3 |

[a]Data extracted from Test No. 891216, Table 1, pp. 10-11.
[b]Values shown in parenthesis indicate average severity score of affected animals.

The following grades for each tissue are considered positive:

| | |
|---|---|
| Opacity (Density) | - Grades 1, 2, 3, and 4 |
| Iris | - Grades 1 and 2 |
| Conjunctivae (Redness) | - Grades 2 and 3 |
| (Chemosis) | - Grades 2, 3, and 4 |

One hour after treatment, two animals exhibited iritis, each with a severity score of 1. Also, 1 hour after treatment, conjunctival redness and chemosis were observed in all three animals. Conjunctival redness persisted in all animals for 24 hours and in one animal for 48 hours; no score ever exceeded a grade of 1. The conjunctival effects were not considered to be positive responses.

D. REVIEWERS' COMMENTS

Only minimal eye irritation was observed in this study. One hour after administration, two animals exhibited iritis which was resolved by 24 hours.

E. STUDY DEFICIENCIES

According to EPA guideline requirements [81-4], at least six animals should be used in this type of study unless a justification for using fewer animals is provided. In this study only three animals were used,

and no explanation was provided by the study author. Also the report did not indicate that the test material was ground to find powder before instillation into the eye. However, this deficiency would not be expected to alter the conclusion of the study that the test compound is a Tox Category IV minimal irritant.

F. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed August 8, 1989, was provided.

**FINAL**

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Primary Eye Irritation

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

| | | |
|---|---|---|
| Principal Reviewer | Sara Lundgaard for Donna Armstrong, Ph.D. | Date 4/8/94 |
| Independent Reviewer | Carrie Rabe, Ph.D. | Date 4/8/94 |
| QA/QC Manager | William L. McLellan, Ph.D. | Date 4/8/94 |

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 221
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 4/19/91

C10979

DATA EVALUATION REPORT

STUDY TYPE: Primary Eye Irritation GUIDELINE: §81-4

PC CODE: 080814 MRID No. 419077-04

TEST MATERIAL: TK 12669/1 (2-chloro-4-tert. butylamino-6-ethylamino-s-triazine

SYNONYMS: Terbuthylazine, Gardoprim, Primatol M.

REGISTRANT: Additives Division, CIBA-GEIGY, Basel, Switzerland

TESTING LABORATORY: Hazleton France, L'Arbresle Cedex, France

STUDY IDENTIFICATION: Project number, 904140; Report number, 009324

TITLE OF REPORT: *"TERBUTHYLAZINE, TEST TO EVALUATE ACUTE OCULAR IRRITATION AND REVERSIBILITY IN THE RABBIT"*

AUTHORS: O. Mercier

REPORT DATE: December 11, 1990

EXECUTIVE SUMMARY: In a primary eye irritation study, 52 mg (equivalent to 0.1 mL) of TK 12669/1 (96.4% pure) was instilled into the conjunctival sac of one eye of New Zealand White rabbits (6 males). Positive findings included: iritis (grade = 1) which cleared within 72 hours and conjunctival redness (grade = 2) which decreased in severity to grade 1 by 48 hours post-treatment.

**The results of this study determined that TK 12669/1 was a moderate eye irritant, placing it in TOXICITY CATEGORY: III.**

CORE CLASSIFICATION: **Acceptable**; this study satisfies the guideline requirement [§ 81-4] for a primary eye irritation study in rabbits.

A. MATERIALS

Test Compound

| | |
|---|---|
| Test material: | TK 12669/1 |
| Batch number: | SG 6925 |
| Description: | White powder |
| Purity: | 96.4% |
| Storage condition: | Room temperature, in the dark |
| Stability: | Not reported; expiration date, 9/92 |
| Vehicle: | None |
| Dose Level: | 0.1 mL (52 mg) undiluted test material; it was not stated whether the test material was ground to a fine powder before instillation |

Test Animals

| | |
|---|---|
| Species: | Rabbit |
| Strain: | New Zealand White |
| Source: | Bancel, Saint Rambert d'Albon, France |
| No. animals: | 6 |
| Sex: | Males |
| Age: | Adult |
| Weight: | 2.35-2.50 kg at the start of the study |
| Temperature: | 21-30°C |
| Humidity: | 30-60% |
| Air changes: | Not reported |
| Photoperiod: | 12 hours |
| Feeding: | UAR formula 112 pelleted rabbit maintenance diet, approximately 150 g/day |
| Water: | *Ad libitum* |
| Identification; | Metal ear tag and cage cards |
| Selection: | Randomly "as they came to hand" and excluding animals with preexisting ocular lesions |
| Housing: | Individual |
| Acclimation: | 19 days |

B. TEST PERFORMANCE

Eye examination: Ocular examinations were performed with the aid of a Heine ophthalmoscope. Treated eyes were washed with fluorescein and examined after a 24-hour exposure period. All animals were reported to be free of ocular lesions prior to treatment. The time of the preliminary examination was not specified, however, it was assumed to be within 24 hours prior to test administration.

Test Material Application: The test substance was placed in the lower conjunctival sac of the right eye of each rabbit. Eye lids were gently held together for several seconds. The left eye served as the control. After 24 hours of exposure, the treated eyes were rinsed with fluorescein (concentration not specified) and then with water.

Observation period: Observations for ocular lesions were made at 1, 24, 48, and 72 hour(s) after administration of the test article. Observations for mortality and clinical signs were made twice daily.

Scoring system: Eyes were examined and scored for ocular lesions using the scoring system attached.

C. RESULTS: A summary of incidence and severity of ocular effects and mean scores of ocular effects are presented in Table 1 below.

Table 1. Incidence and Severity of Ocular Effects[a]

| | Observation Interval (Hours) | | | |
|---|---|---|---|---|
| | 1 | 24 | 48 | 72 |
| Conjunctivae | | | | |
| Chemosis | 6/6 (1)[b] | 4/6 (1) | 0/6 | 0/6 |
| Redness | 6/6 (1.8) | 6/6 (1.5) | 2/6 (1) | 1/6 (1) |
| Iris | | | | |
| Iritis | 6/6 (1) | 5/6 (1) | 3/6 (1) | 0/6 |
| Cornea | | | | |
| Opacity | 0/6 | 0/6 | 0/6 | 0/6 |

[a]Data extracted from Report No. 009324, pp. 22-23.
[b]The severity grade is listed in parenthesis.

The following grades for each tissue are considered positive:

| | |
|---|---|
| Conjunctivae (edema) | - grades 2, 3, and 4 |
| (redness) | - Grades 2 and 3 |
| Iritis | - Grades 1 and 2 |
| Cornea (opacity) | - Grades 1, 2, 3, and 4 |

Iritis (severity grade = 1) was observed in all rabbits 1 hour after the test material was administered. The iritis had resolved in all rabbits by 72 hours. Conjunctival chemosis, discharge, and redness were observed in all rabbits at the 1 hour observation period. Only the redness was sufficiently severe (severity grade of 2 in 5/6) to be considered a positive effect. The chemosis resolved by 48 hours and the discharge was no longer apparent at 24 hours. The conjunctival redness persisted in 1 rabbit (severity grade of 1) at 72 hours.

No mortality was observed during the study and the author reported no adverse behavior or clinical signs in any animal. Weight changes in animals during the study were not reported.

D. REVIEWERS' COMMENTS

All ocular effects considered to be positive (iritis, grade 1 and conjunctival redness, grade 2) had resolved by 48 hours. Minimal signs of conjunctival irritation (grade 1; not considered to be positive) had resolved in all rabbits but 1 by 72 hours. Based on these findings, TK 12669/1 is considered to be a moderate eye irritant and is classified as Toxicity Category III, Caution.

E. STUDY DEFICIENCIES

There were no deficiencies in the study.

F. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed December 7, 1990, was provided.

**FINAL**

010979

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Primary Dermal Irritation

Prepared for:

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
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Prepared by:

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Principal Reviewer Sara Lundgaard for Donna Armstrong, Ph.D. Date 4/8/94

Independent Reviewer Carrie Rabe Carrie Rabe, Ph.D. Date 4/8/94

QA/QC Manager William L. McLellan William McLellan, Ph.D. Date 4/11/94

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 218
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 9/19/90

010979

**DATA EVALUATION REPORT**

STUDY TYPE: Primary Dermal Irritation GUIDELINE: §81-5

PC CODE: 080814 MRID No. 416033-07

TEST MATERIAL: GS 13529 technical

SYNONYMS: Terbuthylazine; Gardoprim; Primatol M.

REGISTRANT: Agricultural Division, CIBA-GEIGY Limited, Basel, Switzerland

TESTING LABORATORY: Experimental Toxicology, CIBA-GEIGY Limited, Stein, Switzerland

STUDY IDENTIFICATION: 891217

TITLE OF REPORT: *"TERBUTHYLAZINE, ACUTE DERMAL IRRITATION/CORROSION STUDY IN THE RABBIT"*

AUTHORS: M. Schneider

REPORT DATE: June 19, 1989

EXECUTIVE SUMMARY: In a dermal irritation/corrosion study, a gauze patch with 0.5 g. of GS 13529 technical was moistened and applied for 4 hours to one flank each of three male New Zealand White rabbits. No signs of erythema, eschar, or edema were observed on any animal. The Primary Irritation Index was 0.0.

**The results of this study determined that GS 13529 was a non-irritant, placing it in TOXICITY CATEGORY: IV.**

CORE CLASSIFICATION: **Acceptable**; this study satisfies the guideline requirement [§ 81-5] for a primary dermal irritation study in rabbits.

A. MATERIALS

Test Compound

| | |
|---|---|
| Test material: | GS 13529 technical |
| Description: | Solid |
| Batch number: | WKA 6925 |
| Purity: | 99% |
| Storage condition: | Room temperature |
| Stability: | Not reported |
| Dose Level: | 0.5 g undiluted test material |

Test Animals

| | |
|---|---|
| Species: | Rabbit |
| Strain: | New Zealand White |
| Source: | Kleintierfarm Madoerin AG, Fuellinsdorf |
| Number animals: | 3 |
| Sex: | Males |
| Age: | 12-14 weeks old |
| Weight: | 2.30-2.46 kg |
| Temperature: | 20±3°C |
| Humidity: | 30-70% |
| Photoperiod: | 12 hours |
| Feeding: | Nafag No. 814 standard rabbit pellet, *ad libitum* |
| Water: | *Ad libitum* |
| Identification: | Ear tag |
| Selection: | Not reported |
| Acclimation: | At least 5 days |

B. TEST PERFORMANCE

Skin preparation: A 36 $cm^2$ area, on both flanks of each animal was clipped approximately 24 hours prior to testing.

Test Material Application: A 0.5 g aliquot of the test substance was placed on a gauze patch (12-16 $cm^2$), moistened with distilled water, and applied to one flank. The area was then covered loosely with aluminum foil that was held in place by adhesive tape. The contralateral flank was treated similarly except no test material was applied to the gauze patch. It was not reported whether the test material was removed by washing following removal of the patch.

Exposure time: 4 hours

Observation period: The degree of erythema and edema at the test site were determined 1, 24, 48, and 72 hour(s) after removal of the test material. Mortality and clinical signs were noted daily.

Scoring system: Skin reactions were scored using the OECD scoring system. The system is identical to the Draize system.

C. RESULTS

No signs of erythema, eschar, or edema were observed on any of the three animals after administration of the test substance. The Primary Irritation Index (average of the 24- and 72- hour scores) was 0.0, indicating that the compound was nonirritating.

D. REVIEWERS' COMMENTS

GS 13529 was a non-irritant to rabbit skin under the conditions of this study.

E. STUDY DEFICIENCIES

Only three animals were included in this study, rather than the six recommended in EPA guidelines. No justification for using fewer animals was provided by the study author. However, this deficiency is unlikely to alter the conclusions of the study.

F. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed June 21, 1989, was provided.

**FINAL**

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Primary Dermal Irritation

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

| | | |
|---|---|---|
| Principal Reviewer | Sara Lundgaard for<br>Donna Armstrong, Ph.D. | Date 4/8/94 |
| Independent Reviewer | Carrie Rabe<br>Carrie Rabe, Ph.D. | Date 4/8/94 |
| QA/QC Manager | William L. McLellan<br>William McLellan, Ph.D. | Date 4/8/94 |

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 222
Project Officer: Caroline Gordon

**EPA Reviewer and Section Head:**
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 4/20/94

## DATA EVALUATION REPORT

**STUDY TYPE:** Primary Dermal Irritation **GUIDELINE:** §81-5

**PC CODE:** 080814 **MRID No.** 419077-05

**TEST MATERIAL:** TK 12669/1 (2-Chloro-4-tert. butylamino-6-ethylamino-s-triazine)

**SYNONYMS:** Terbuthylazine; Gardoprim; Primatol M.

**REGISTRANT:** CIBA-GEIGY Limited, Additives Division, Basel, Switzerland

**TESTING LABORATORY:** Hazleton France, L'Arbresle Cedex, France

**STUDY IDENTIFICATION:** Report number: 008327; Project number: 904141

**TITLE OF REPORT:** *"TERBUTHYLAZINE; TEST TO EVALUATE ACUTE PRIMARY CUTANEOUS IRRITATION AND CORROSIVITY IN THE RABBITS"*

**AUTHOR:** O. Mercier

**REPORT DATE:** December 11, 1990

**EXECUTIVE SUMMARY:** In an primary dermal irritation study, six male New Zealand White rabbits received an unknown amount of TK 12669/1 on the intact skin for 4 hours. Two animals showed signs of slight erythema at 1 hour, but not at 24 hours; no other adverse dermal signs were reported. The Primary Irritation Index was calculated to be 0.0.

**The results of this study determined that TK 12669/1 was very slightly irritating.**
Toxicity Category IV

**CORE CLASSIFICATION: Supplementary**; this study does not satisfy the guideline requirement [§ 81-5] for a primary dermal irritation study because the author's dose calculation could not be verified. If the submitter provides addition information regarding the dose calculations, this study may be upgraded.

A. MATERIALS

Test Compound

| | |
|---|---|
| Test material: | TK 12669/1 |
| Description: | White powder |
| Batch number: | SG 6925 |
| Purity: | 96.4% |
| Storage condition: | Room temperature, in the dark |
| Stability: | Not reported; expiration date, 9/92 |
| Vehicle: | Purified water |
| Dose level: | 0.5 g undiluted test material; prepared by mixing 4 g of test material and 1.869 g of purified water and applying 0.6 mL of paste per animal<br>Note: Insufficient information was provided to verify that 0.6 mL of paste contained 0.5 g of test material |

Test Animals

| | |
|---|---|
| Species: | Rabbit |
| Strain: | New Zealand White |
| Source: | Bancel, France |
| Number animals: | 6 |
| Sex: | Males |
| Age: | Adults |
| Body Weight: | 2.45-2.55 kg |
| Temperature: | 21-27°C |
| Relative Humidity: | 18-54% |
| Photoperiod: | 12 hours |
| Housing: | Individual |
| Feeding: | U.A.R. formula 112 pelleted rabbit maintenance diet, 150 g/day |
| Water: | *Ad libitum* |
| Identification: | Metal ear tags and cage cards |
| Selection: | Randomly "as they came to hand"; excluding unhealthy animals, especially those with cutaneous lesions |
| Acclimation: | 12 days |

B. TEST PERFORMANCE

Skin preparation: Approximately 24 hours prior to testing, the flanks and back of each animal (about 14 x 14 cm) were clipped. Only animals with intact skin free from any irritation were used.

Test Material Application: The test material, prepared as a paste, was applied to 6 $cm^2$ of skin and covered with gauze and a semi-occlusive bandage. After 4 hours, the dressing was removed and surplus test material was removed using gauze moistened with purified water.

Observation Period: The degree of erythema and edema at the test site was determined 1, 24, 48, and 72 hours after removal of the test material. Mortality and clinical signs were noted twice each day.

Scoring system: Skin reactions were scored using a system described below.

Development of erythema and eschar formation

| | |
|---|---|
| No erythema | 0 |
| Very slight erythema (barely perceptible) | 1 |
| Well defined erythema | 2 |
| Moderate to severe erythema | 3 |
| Severe erythema (beetroot red) to slight eschar formation (deep lesion) | 4 |

Development of Edema

| | |
|---|---|
| No edema | |
| Very slight edema (barely perceptible) | 1 |
| Slight edema (edges of area well defined by definite raising) | 2 |
| Moderate edema (edges raised ≈1 mm) | 3 |
| Severe edema (raised >1 mm extending beyond the area of exposure) | 4 |

C. RESULTS

Table 1. Mean Skin Irritation Score[a]

| | Time (hours) | | | |
|---|---|---|---|---|
| | 1 | 24 | 48 | 72 |
| Erythema Mean Score | 0.33 | 0.0 | 0.0 | 0.0 |
| Edema Mean Score | 0.0 | 0.0 | 0.0 | 0.0 |

[a]Data extracted from Report No. 008327, p. 21.

Two animals showed signs of slight erythema 1 hour after administration of the test material, which did not persist to 24 hours. No edema or other dermal signs were observed. The Primary Irritation Index (average of the 1-, 24-, 48-, and 72- hour scores) was 0.08, indicating that the compound was very slightly irritating.

No changes in behavior or clinical signs were observed in any animal.

On this basis, TK 12269/1 was classified as Toxicity Category IV (Caution).

D. REVIEWER'S COMMENTS

Only very minimal irritation was observed in the rabbits. Two animals showed signs of slight erythema at 1 hour, but not at 24 hours. No other adverse dermal signs were reported.

E. STUDY DEFICIENCIES

The study author did not provide sufficient information to verify the dose calculation. Therefore, the study is classified as Supplementary. If the submitter provides information that allows verification that 0.6 mL of the paste contains 0.5 g of the test material, the study may be upgraded.

F. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed December 7, 1990, was provided.

**FINAL**

DATA EVALUATION REPORT

TERBUTHYLAZINE

Study Type: Dermal Sensitization

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031

Principal Reviewer Sara Lundgaard for Donna Armstrong, Ph.D. Date 4/8/94

Independent Reviewer Carrie Rabe Carrie Rabe, Ph.D. Date 4/8/94

QA/QC Manager William L. McLellan William McLellan, Ph.D. Date 4/11/94

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 223
Project Officer: Caroline Gordon

EPA Reviewer and Section Head:
Marion Copley, D.V.M.,
Section IV, Toxicology Branch I/HED (7509C)

Signature: Marion Copley
Date: 4/19/93

010979

# DATA EVALUATION REPORT

**STUDY TYPE:** Dermal Sensitization **GUIDELINE:** §81-6

**PC CODE:** 080814 **MRID No.** 419077-06

**TEST MATERIAL:** TK 12669/1 (2-chloro-4-tert. butylamino-6-ethylamino-S-triazine)

**SYNONYMS:** Terbutylazine; Gardoprim; Primatol M

**REGISTRANT:** Additives Division, CIBA-GEIGY Limited, Basel, Switzerland

**TESTING LABORATORY:** Hazleton France, L'Arbresle Cedex, France

**STUDY IDENTIFICATION:** Report number: 101322; Project number: 904152

**TITLE OF REPORT:** *"TERBUTHYLAZINE, TEST TO EVALUATE THE SENSITIZING POTENTIAL IN THE GUINEA PIG, GUINEA-PIG MAXIMIZATION TEST"*

**AUTHOR:** O. Mercier

**REPORT DATE:** April 22, 1991

EXECUTIVE SUMMARY: The dermal sensitization potential of TK 12669/1 was evaluated in Dunkin-Hartley guinea pigs (10/sex/group) in a maximization study. For induction, animals were injected twice with 0.1 mL each of 8.5 mg/L of TK 12669/1 in water, a 50:50 mixture of 8.5 mg/L TK 12669/1 and 50% Freund's adjuvant, and 50% Freund's adjuvant on study day 1. This was followed on day 8 by dermal application of 0.5 mL 10% sodium lauryl sulfate and on day 9 by 0.5 mL of test material prepared as a paste at 68% (w/w) in water. This was followed 11 days later by application of 0.5 mL of the 68% paste to a virgin site (challenge phase). No positive dermal sensitization response was observed in treated animals. A positive dermal response was observed in the positive control group (DNCB). Under the conditions of this study, TK 12669/1 was not shown to be a sensitizer in guinea pigs.

CORE CLASSIFICATION: **Acceptable**; this study satisfies guideline requirement [§ 81-6] for a dermal sensitization study in guinea pigs.



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A. MATERIALS

Test Compound

Test material: TK 12 669/1
Lot number: SG 6925
Purity: 96.4%
Physical description: White powder
Storage condition: Room temperature, in the dark
Stability: Not reported; expiration date, 9/92
Positive control material: 0.05% solution (w/w) of DNCB in 1,2 propylene glycol

Test Animals

Species: Guinea pig
Strain: Dunkin-Hartley albino
Source: Elevage Lebeau, Gambais, France
Sex: Males and females
Age: Young adult
Weight: Males 450-498 g (day 0)
Females 442-498 g (day 0)
Number and sex/group: 10 males and 10 females/induction and challenge
10 males and 10 females/positive control
10 males and 10 females/naive control irritation
6 males and 6 females/preliminary studies
Temperature: 19-23.5°C
Relative humidity: 42-69%
Photoperiod: 12 hours
Air changes: Not reported
Housing: 5-6/cage; sexes separate
Feeding: U.A.R. formula 106 complete pelleted guinea pig breeding diet, *ad libitum*
Water: *Ad libitum*
Identification: Ear tags and cage cards
Acclimation: 27 days for main study (14 days for preliminary study)
Selection: Random, "as they came to hand"; animals with cutaneous lesions excluded

B. TEST PERFORMANCE

Test procedure: The maximization test was used.

Test Site: Retroscapular and vertebral column areas (8 $cm^2$) were used for the induction phase and the right and left lateral abdominal areas (4 $cm^2$) were used for the challenge phase.

Skin Preparation: The test sites were prepared by clipping and shaving. The period of time prior to the application of the test material that the test sites were prepared was not reported.

Induction Phase

(a) Route of administration: On day 1, the test material and control solutions were administered by intradermal injection. On days 8 and 9, topical applications were made to the injection areas, in order to further induce a local irritation.

(b) Dosing: Each animal had a total of six injections; two injections each of the following solutions at a volume of 0.1 mL:

Dosed group
1) a 50:50 mixture (v:v) of 8.5 mg/L test material in 50% Freund's adjuvant (diluted with 0.9% NaCl)
2) 8.5 mg/L test material in water
3) 50% Freund's adjuvant (diluted with 0.9% NaCl)

Control group
1) 50% Freund's adjuvant (diluted with 0.9% NaCl)
2) water
3) a 50:50 mixture (v:v) of water and 50% Freund's adjuvant (diluted with 0.9% NaCl)

Positive control group
1) a 50:50 mixture (v:v) of 0.05% (w/w) solution of DNCB in 1,2-propylene glycol and 50% Freund's adjuvant (diluted with 0.9% NaCl)
2) 0.05% (w/w) solution of DNCB and 1,2 propylene glycol in water
3) 50% Freund's adjuvant (diluted with 0.9% NaCl)

(c) On day 8, injection areas of the dosed and positive control groups were painted with 0.5 mL of a 10% (w/w) suspension of sodium lauryl sulphate in Codex paraffin.

(d) On day 9, using a 8 $cm^2$ piece of filter paper held in contact with the skin by means of a 15 $cm^2$ impermeable and hypoallergenic bandage, the following topical applications were made to injection areas:

Dosed group
0.5 mL of test material prepared as a paste at 68% (w/w) in water

Control group
0.5 mL of water

Positive control group
0.5 mL of DNCB in a 0.05% (w/w) solution in 1,2 propylene glycol

(e) Length of exposure: 48 hours

(f) Rest period: 11 days, no treatment

Challenge Phase

(a) Route of administration: Test material and vehicle were applied topically to clipped and shaved left and right lateral abdominal regions (4 $cm^2$), respectively, and covered with filter paper held in place with a hypoallergenic patch. The patch was covered with gauze and an adhesive bandage.

(b) Length of exposure: 24 hours

(c) The following test applications were made to the left and right flanks:

Dosed group
left - 0.5 mL of test material prepared as a paste at 68% (w/w) in water

right - none

Control group
left - 0.5 mL of test material prepared as a paste at 68% (w/w) in water

right - none

Positive control group
left - 0.5 mL of DNCB in a 0.05% (w/w) solution in 1,2 propylene glycol

right - 0.5 mL of 1,2 propylene glycol

(d) Observation period: 24 and 48 hours

Evaluation System

A copy of the evaluation system is attached. Evaluation of macroscopic and histopathologic responses was performed blind according to the following definitions.

Positive reaction: an animal with a positive macroscopic cutaneous lesion or, histological examination demonstrated that a doubtful macroscopic reaction resulted from an allergic lesion

Negative reaction: an animal with no macroscopic abnormality or, histological examination failed to demonstrate that a doubtful macroscopic reaction resulted from an allergic lesion

Doubtful reaction: the origin of a doubtful macroscopic lesion could not be determined by histological examination

C. RESULTS

Body Weight: Mean body weight gains were comparable among the males in the treated and naive control groups, but body weight gain among the positive control males was approximately 50% lower. A slight loss in body weight was observed among females in the positive control group and no body weight gain was seen among females in the treatment group. Mean body weights (g) are summarized in Table 1.

Table 1. Mean Body Weight Data[a]

| Treatment Group | Prestudy (grams) | Terminal (grams) |
|---|---|---|
| Induction and challenge | | |
| Males | 483 | 556 |
| Females | 472 | 472 |
| Challenge control | | |
| Males | 485 | 571 |
| Females | 474 | 510 |
| Positive Control | | |
| Males | 478 | 527 |
| Females | 476 | 468 |

[a]Data extracted from Report No. 101322, pp. 56-59.

Skin Reaction: No skin reactions were observed at challenge for the test group or naive controls (scores for erythema and edema were 0). All animals in the positive control group displayed signs of irritation at the 24- and 48-hour observation periods. At 24 hours, 7/10 male and 9/10 female animals were reported to have moderate symptoms (score, 3) and the remaining animals were reported to have mild symptoms (score, 2). At 48 hours, 6/10 male and 6/10 female animals were reported to have moderate symptoms and the remaining animals were reported to have mild symptoms. The scoring system used to evaluate erythema and edema was not described.

D. REVIEWERS' COMMENTS

No positive dermal sensitization response in guinea pigs, following exposure to the test material, was observed. A positive dermal response was observed in the positive control group.

E. STUDY DEFICIENCIES

This study satisfies minimal data requirements according to Guideline [81-6]. Categories used to report and score results of macroscopic examinations were not clearly described. However, this study is minimally acceptable for regulatory purposes since no evidence of any dermal response was observed in animals exposed to the test substance.

E. QUALITY ASSURANCE

The test was performed under Good Laboratory Practice Standards. A Quality Assurance Statement, signed April 25, 1991, was provided.

TERBUTHYLAZINE TOX R 010979

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Pages 52 through 53 are not included in this copy.

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_____ Identity of product impurities.

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_____ 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.

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_____ The document is not responsive to the request.

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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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**FINAL**

DATA EVALUATION REPORT

TERBUTHYLAZIN

Study Type: Mutagenicity: Salmonella typhimurium/Mammalian Microsome Mutagenicity Assay

Prepared for:

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

Prepared by:

Clement International Corporation
9300 Lee Highway
Fairfax, VA 22031-1207

| | | |
|---|---|---|
| Principal Reviewer | Marisa B. Brett<br>Marisa B. Brett, B.A. | Date 3/18/94 |
| Independent Reviewer | Nancy E. McCarroll<br>Nancy E. McCarroll, B.S. | Date 3/18/94 |
| QA Manager | William L. McLellan<br>William L. McLellan, Ph.D. | Date 3/18/94 |

Contract Number: 68D10075
Work Assignment Number: 3-51
Clement Number: 225
Project Officer: Caroline Gordon

MUTAGENICITY STUDIES

EPA Reviewer: Irving Mauer, Ph.D.
Immediate Office (7509C) TB1
EPA Section Head: Marion Copley, DVM, DABT
Review Section IV,
Toxicology Branch I (7509C)

Signature: M Copley for I Mauer 3/30/94
Date:

Signature: M Copley
Date: 3/30/94

DATA EVALUATION REPORT

STUDY TYPE: Salmonella typhimurium/mammalian microsome gene mutation assay

TEST MATERIAL: TK 12669/1 (Terbuthylazin)

TOX. CHEM. NUMBER: 125B

PC CODE: 080814

MRID Number: 416340-01

SYNONYM(S): None provided

SPONSOR: Ciba-Geigy Ltd., Plastics and Additives Division, Basle, Switzerland

TESTING FACILITY: Ciba-Geigy Ltd., Basle, Switzerland

TITLE OF REPORT: Salmonella/Mammalian-microsome Mutagenicity Test

AUTHOR: Ogorek, B.

STUDY NUMBER: Test No. 874192

REPORT ISSUED: August 20, 1987

EXECUTIVE SUMMARY:

In two independently performed Salmonella typhimurium/mammalian microsome plate incorporation assays, strains TA1535, TA1537, TA98 and TA100 were exposed to 20, 78, 313, 1250, or 5000 µg/plate TK 12669/1 (terbuthylazin) with or without S9 activation. The S9 fraction was prepared from Aroclor 1254-induced rat livers and the S9 cofactor mix contained 30% liver homogenate. Dimethyl sulfoxide (DMSO) was used as the solvent to deliver TK12669/1 to the test system.

**No cytotoxicity or mutagenicity was observed in any strain at any dose either in the presence or absence of S9 activation; compound insolubility was reported at ≥1250 µg/plate.** Results with the positive controls confirmed the sensitivity of the test system to detect mutagenesis.

This study is classified as **Acceptable** and satisfies the guideline requirement for a gene mutation study (84-2). For future submissions, however, it is recommended that the study author provide justification for the use of a high S9 level; otherwise, assays should be performed using the screening concentration of S9 (4-10%) recommended for this test system.[1]

A. MATERIALS

1. Test Material: TK 12669/1 (Terbuthylazin)

Description: Not specified
Identification number: Batch number SG 6015
Purity: 99.2%
Receipt date: Not reported
Stability: Reported to be stable; expiration date ensured by sponsor
Contaminants: None listed
Solvent used: Dimethyl sulfoxide (DMSO)
Other provided information: Neither the test material storage conditions nor the frequency of dosing solution preparation were reported. No analytical determinations were reported.

2. Control Materials:

Solvent/final concentration: DMSO/0.1 mL/plate

Positive:

Nonactivation:

| | | |
|---|---|---|
| Sodium azide | 2.5, 5.0 | µg/plate TA1535 |
| 9(5)-Aminoacridine | 50, 100 | µg/plate TA1537 |
| Daunorubicin | 5, 10 | µg/plate TA98 |
| 4-Nitroquinoline-N-oxide (4-NQO) | 0.125, 0.25 | µg/plate TA100 |

Activation:

| | | |
|---|---|---|
| Cyclophosphamide (CP) | 250 | µg/plate TA1535 |
| 2-Aminoanthracene (2-AA) | 5 | µg/plate TA1537, TA98, TA100 |

3. Activation: S9 derived from Tif:RAIf(SPF)

| | | | |
|---|---|---|---|
| x Aroclor 1254 | x induced | x rat | x liver |
| ___ phenobarbital | ___ noninduced | ___ mouse | ___ lung |
| ___ none | | ___ hamster | ___ other |
| ___ other | | ___ other | |

The rat liver S9 homogenate was obtained from Analabs., Inc., North Haven, CT. The S9 activation mixture contained 0.3 mL S9 fraction/mL and 0.7 mL cofactor solution/mL. The use of 30% S9 was not justified.

[1]Maron, D.M., Ames, B.N. (1983). Revised methods for the Salmonella mutagenicity test. Mutat. Res. 113: 173-215.

4. Test Organism Used: S. typhimurium strains

| | | | | | |
|---|---|---|---|---|---|
| _____ TA97 | __x__ TA98 | __x__ TA100 | _____ TA102 | _____ TA104 | |
| __x__ TA1535 | __x__ TA1537 | _____ TA1538 | | | |

list any others:

Test organisms were properly maintained? Not reported.
Checked for appropriate genetic markers (rfa mutation, R factor)? Not reported.

5. Test Compound Concentrations Used:

(a) Preliminary cytotoxicity assay: Nine concentrations (0.08, 0.31, 1.2, 4.9, 19.5, 78.1, 312.5, 1250.0, and 5000.0 µg/plate) were evaluated without S9 activation with S. typhimurium strain TA100. Duplicate plates were used per dose, per strain, and solvent (DMSO) and positive (4-NQO) controls were included.

(b) Mutation assay:

- Initial trial: Five concentrations (20, 78, 313, 1250, and 5000 µg/plate +/- S9) were evaluated with all S. typhimurium tester strains.
- Confirmatory trial: As above for the initial trial.

Note: Triplicate plates per dose, per strain, per condition were prepared for both the initial and confirmatory mutation assays.

B. TEST PERFORMANCE

1. Type of Salmonella Assay:
- __x__ Standard plate test
- ___ Pre-incubation (_____) minutes
- ___ "Prival" modification
- ___ Spot test
- ___ Other (describe)

2. Preliminary Cytotoxicity/Mutation Assays: Similar procedures were used for the preliminary cytotoxicity and the mutation assays. Aliquots of 0.1 mL of the appropriate tester strain culture were prepared in nutrient broth and mixed with 2 mL of top agar and 0.1 mL of the appropriate test material dose, solvent or positive control; 0.5 mL of the S9 cofactor mix was added for the S9-activated trials. The contents of each tube were mixed and poured over 20 mL of minimum agar containing Vogel-Bonner Medium E salts and glucose and incubated at 37±1.5°C for 48 hours. Following incubation, plates were scored for revertant colonies and the means and standard deviations were determined per strain, per dose, per condition for the mutation assay.

3. Evaluation Criteria:

(a) Valid assay: The assay was considered valid if (1) the mean colony counts for the solvent controls were within the provided historical ranges of the performing laboratory and (2) the results with the positive controls satisfied the criteria for a positive response.

(b) Positive response: The test material was considered positive if it caused a reproducible dose-related ≥2-fold increase in mutant colonies of S. typhimurium strains TA1535, TA1537 or TA98, or a reproducible dose-related ≥1.5-fold increase in revertant colonies of strain TA100.

C. REPORTED RESULTS

1. Test material solubility: The report stated that the test material precipitated at concentrations of 1250 and 5000 µg/plate. No further information was provided. It was assumed by our reviewers that this precipitation occurred in the preliminary cytotoxicity assay and in both trials of the nonactivated and S9-activated mutagenicity assays.

1. Preliminary Cytotoxicity Assay: S. typhimurium strain TA100 was evaluated in the absence of S9 activation. No clear evidence of cytotoxicity was observed at any dose. Based on these findings, levels ranging from 20 to 5000 µg/plate +/- S9 were selected for the initial and confirmatory mutation assays.

2. Mutation Assays: Representative data from the initial and confirmatory assays are presented in Tables 1 and 2, respectively. The results from both trials were in good agreement and indicated that the selected doses of the test material were not cytotoxic and did not exert a mutagenic effect in any strain either in the presence or absence of S9 activation. All strains responded to the mutagenic action of the appropriate nonactivated or S9-activated positive control. However, our reviewers noted that the response of strains TA98 and TA100 to the S9-activated positive control (5 µg/plate 2-AA) was lower than expected in both trials. Based on our reviewers experience with this test system, the lack of an optimum response was attributed to the use of a high concentration of S9 (30%) in the S9 cofactor mix.

Based on the overall results, the study author concluded that TK 12669/1 (terbuthylazin) was not mutagenic.

D. REVIEWERS' DISCUSSION/CONCLUSIONS: We assess that the test material was assayed to an appropriately high level with and without S9 activation and was neither cytotoxic nor mutagenic in any tester strain. As stated previously, all strains responded to the mutagenic action of the nonactivated and S9-activated positive controls, and the lower than expected reversion to histidine prototrophy of strains TA98 and TA100 following exposure to 5 µg/plate 2-AA probably resulted from the use of 30% S9 in the S9-cofactor

Table 1. Representative Results of the Initial Salmonella typhimurium/Mammalian Microsome Mutation Assay with TK 12669/1 (Terbuthylazin)

| Substance | Dose per Plate | S9 Activation | Revertants per Plate of Bacterial Tester Strain[a] S. typhimurium TA1535 | TA1537 | TA98 | TA100 |
|---|---|---|---|---|---|---|
| Solvent Control | | | | | | |
| Dimethyl sulfoxide | 0.1 mL | - | 12±4 | 6±2 | 19±5 | 166±7 |
| | | + | 11±8 | 8±2 | 27±4 | 145±8 |
| Positive Controls[b] | | | | | | |
| Sodium azide | 2.5 µg | - | 696±46 | -- | -- | -- |
| 9(5)-Aminoacridine | 50 µg | - | -- | 31±7 | -- | -- |
| Daunorubicin | 5 µg | - | -- | -- | 610±62 | -- |
| 4-Nitroquinoline -N-oxide | 0.125 µg | - | -- | -- | -- | 719±53 |
| Cyclophosphamide | 250 µg | + | 702±6 | -- | -- | -- |
| 2-Aminoanthracene | 5 µg | + | -- | 153±34 | 1527±157 | 1511±93 |
| Test Material | | | | | | |
| TK 12669/1 | 313 µg[c] | - | 8±1 | 6±1 | 29±4 | 139±20 |
| | 1250 µg[d] | - | 9±3 | 5±3 | 18±8 | 136±25 |
| | 5000 µg[d] | - | 8±1 | 4±1 | 13±1 | 156±13 |
| | 313 µg[c] | + | 14±3 | 9±3 | 20±5 | 166±9 |
| | 1250 µg[d] | + | 12±4 | 9±3 | 32±11 | 160±4 |
| | 5000 µg[d] | + | 9±3 | 8±1 | 26±7 | 156±10 |

[a]Means and standard deviations from the counts of three plates.
[b]Two levels of the nonactivated positive control were assayed; data from the lower doses were selected as representative.
[c]Highest soluble dose; results for lower levels (20 or 78 µg/plate +/-S9) did not suggest a mutagenic effect.
[d]Test material precipitation reported at this level.

Note: Data were extracted from the study report, pp. 14-17.





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Table 2. Representative Results of the Confirmatory Salmonella typhimurium/Mammalian Microsome Mutation Assay with TK 12669/1 (Terbuthylazin)

| Substance | Dose per Plate | S9 Activation | Revertants per Plate of Bacterial Tester Strain[a] S. typhimurium TA1535 | TA1537 | TA98 | TA100 |
|---|---|---|---|---|---|---|
| Solvent Control | | | | | | |
| Dimethyl sulfoxide | 0.1 mL | - | 15±2 | 6±1 | 21±4 | 157±20 |
| | | + | 13±5 | 16±1 | 33±4 | 180±14 |
| Positive Controls[b] | | | | | | |
| Sodium azide | 2.5 µg | - | 676±9 | -- | -- | -- |
| 9(5)-Aminoacridine | 50 µg | - | -- | 28±10 | -- | -- |
| Daunorubicin | 5 µg | - | -- | -- | 433±96 | -- |
| 4-Nitroquinoline -N-oxide | 0.125 µg | - | -- | -- | -- | 781±38 |
| Cyclophosphamide | 250 µg | + | 575±32 | -- | -- | -- |
| 2-Aminoanthracene | 5 µg | + | -- | 130±75 | 1341±244 | 1207±337 |
| Test Material | | | | | | |
| TK 12669/1 | 313 µg[c] | - | 12±4 | 5±2 | 23±6 | 143±4 |
| | 1250 µg[d] | - | 10±3 | 8±5 | 17±6 | 156±3 |
| | 5000 µg[d] | - | 8±4 | 8±4 | 14±3 | 141±8 |
| | 313 µg[c] | + | 8±1 | 20±4 | 32±5 | 176±12 |
| | 1250 µg[d] | + | 13±0 | 12±5 | 31±4 | 171±2 |
| | 5000 µg[d] | + | 13±5 | 9±3 | 22±6 | 165±9 |

[a]Means and standard deviations from the counts of three plates.
[b]Two levels of the nonactivated positive control were assayed; data from the lower doses were selected as representative.
[c]Highest soluble dose; results for lower levels (20 or 78 µg/plate +/-S9) did not suggest a mutagenic effect.
[d]Test material precipitation reported at this level.

Note: Data were extracted from the study report, pp. 18-21.

mix. There was, however, no compelling evidence that the use of an excessive concentration of S9 adversely affected the outcome of the study. We conclude, therefore, that the assay is acceptable, but for future submissions, justification for the use of 30% S9 in the S9-cofactor mix should be provided; otherwise, assays should be performed using the screening concentration (4-10%) recommended by Maron and Ames (1983).[2]

E. QUALITY ASSURANCE MEASURES: Was the test performed under GLP? Yes. (A Quality Assurance statement was signed and dated August 19, 1987.)

[2]Maron, D.M. and Ames, B.N. (1983). Mutat. Res. 113:173-215.

Reviewed by: William B. Greear, M.P.H. W.B. Greear 4/20/94
Review Section IV, Toxicology Branch I (7509C)
Secondary Reviewer: Marion P. Copley, D.V.M. Marion Copley 5/3/94
Review Section IV, Toxicology Branch I (7509C)

SUPPLEMENTAL DATA EVALUATION REPORT
(Original MRID #41391801, Report #831174; New MRID #420598-06, Report #881606)

Study Type: Guideline Series: DNA Repair-rat

Tox. Chem. No.: 125B
PC No.: 080814
MRID No.: 420598-06

Test Material: GS13529 TECH., Belclene 329

Sponsor: Ciba-Geigy Corporation, Hawthorne, NY 10532-2188

Testing Facility: Toxicology II, Ciba-Geigy, Ltd., Basle, Switzerland

Title of Report: Autoradiographic DNA Repair Test on Rat Hepatocytes (OECD Conform).

Author: Original (Thomas Hertner); Supplement E. Puri

Study Number: 881606

Report Issued: Original (June 19, 1989); Supplement (July 26, 1991)

Executive Summary: In a DNA repair study using rat hepatocytes obtained from male Tif.RAIF (SPF) rats dose levels of 0.98, 3.9, 15.6, 62.5, 250 or 1000 ug/ml w.ere used to test for DNA damage. A confirmatory test was conducted at concentrations of 0.250, 0.49, 0.98, 3.9, 15.6, 62.5, 250 or 1000 ug/ml. 2-AAF and 4-ABP were used as positive controls. The test material did not produce a significant increase in the mean gross or net nuclei grain counts when tested at levels up to 1000 ug/ml when compared to vehicle controls and a dose-response relationship was not observed. Additionally, the percental distribution of grain counts per nuclei among the test and vehicle control groups were comparable(see Tables attached). There was no evidence of increased DNA synthesis in the study.

[In the supplement, it was explained that the highest dose tested was done to the solubility limit of 100 mg/ml in DMSO. Also, precipitation in the nuclei occurred at dose levels from 3.9 to 1000 ug/ml. This explanation was also provided in the initial evaluation of the study. At that time, the study was considered to be acceptable. The explanation provided in this supplement is adequate. Further, it was decided that if this study was acceptable then the previously submitted DNA repair

test on human fibroblast (Report # 831175; 6/18/84, Acc # 259814); Refer to Tox Doc #008509, William B. Greear, 8/12/91).] would also be acceptable.

TERBUTHYLAZINE TOX R 6/0979

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Pages 64 through 76 are not included in this copy.

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_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

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X FIFRA registration data.

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