US EPA ARCHIVE DOCUMENT

13544

# 030886

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|---|---|
| **Chemical:** | **Diuron** |
| **PC Code:** | **035505** |
| **HED File Code** | **13000 Tox Reviews** |
| **Memo Date:** | **07/29/93** |
| **File ID:** | **TX010441** |
| **Accession Number:** | **412-02-0012** |

**HED Records Reference Center**
**03/08/2002**

410

CASWELL FILE

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

010441

OPP OFFICIAL RECORD
HEALTH EFFECTS DIVISION
SCIENTIFIC DATA REVIEWS
EPA SERIES 361

JUL 29 1993

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

**MEMORANDUM**

**SUBJECT:** Diuron: 21-day dermal toxicity study in rabbits

**TO:** Peg Perreault/Linda Propst PM 73
Special Review and Reregistration Division

**FROM:** K. Clark Swentzel K. Clark Swentzel 7/27/93
Section Head, Section 2
Toxicology Branch II
HED

**THROUGH:** Marcia van Gemert, Ph.D. Marcia van Gemert 7/27/93
Branch Chief
Toxicology Branch II
HED

| | |
|---|---|
| **CASE** | 818790 |
| **BARCODE:** | D191321 |
| **MRID** | 427183-01 |
| **SUBMISSION:** | S440777 |
| **PC NO.** | 035505 |
| **CASWELL NO.** | 410 |
| **REGISTRANT:** | DuPont Agricultural Products |

Requested Action

Review study.

Submission

Diuron was administered dermally to New Zealand white rabbits daily (6 hours/day) for a total of 21 (males) or 22 (females) consecutive applications at doses of 0, 50, 500 and 1200 mg/kg/day. There was no evidence of systemic or dermal toxicity based on the investigated parameters indicated in the attached DER prepared by Clement.

The Clement reviewer stated that no data on the stability of the test material were provided. This reviewer called Ian Wellings of DuPont who referred me to a hydrolysis study (MRID 41418804) with diuron which the Agency has accepted (letter sent by fax from Dr.

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Wellings is attached). This data is relevant since the test material was made up in water. Hydrolysis studies at pH's 5, 7 and 9 at 25 ± 1 and in darkness show that the 1/2 life in water is greater than 500 days. After 30 days, no significant degradation of diuron had taken place in any of the studies.

**Dermal toxicity NOEL = 1200 mg/kg/day**
**Dermal toxicity LOEL = Not determined**

**Systemic toxicity NOEL = 1200 mg/kg/day**
**Systemic toxicity LOEL = Not determined**

**Classification:** Core-minimum. This study meets the requirement set forth under EPA Guideline Series 82-2 for a dermal toxicity study in rabbits.

attachment

**FINAL**

# DATA EVALUATION REPORT

Diuron

Study Type:
21-Day Repeated Dermal Toxicity Study

Prepared for:

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

Prepared by:

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

| | | |
|---|---|---|
| Principal Reviewer | Patricia B. Bittner<br>Patricia Bittner, M.S. | Date 7/1/93 |
| Independent Reviewer | John Liccione<br>John Liccione, Ph.D. | Date 7/1/93 |
| QA/QC Manager | Sharon A. Segal<br>Sharon Segal, Ph.D. | Date 7/1/93 |

Contract Number: 68D10075
Work Assignment Number: 2-120
Clement Number: 385
Project Officer: Caroline Gordon

EPA Reviewer, Section Head: K. Clark Swentzel
Review Section II, Toxicology Branch II,
Health Effects Division

Signature: K. Clark Swentzel
Date: 7/27/93

DATA EVALUATION REPORT

STUDY TYPE: 21-Day repeated dermal toxicity study

TEST MATERIAL: Diuron

MRID NUMBER: 427183-01

CAS REGISTRY NUMBER: 330-54-1

TOX CHEM NUMBER: 410

SYNONYMS: DPX-14740-166; dichlorofenidim; Cekiuron® (Cequisa); Dailon®; Daiter®; Farmco®; Toterbane 50F®; Vonduron®; IN-14740-166; IN 14740; 14740-166 Milled Diuron technical

STUDY REPORT NUMBER: HLR 484-92

MEDICAL RESEARCH NUMBER: 9122-001

SPONSOR: DuPont Agricultural Products
E.I. du Pont de Nemours and Company
Wilmington, DE

TESTING FACILITY: E.I. du Pont de Nemours and Company
Haskell Laboratory for Toxicology and Industrial Medicine
Newark, DE

TITLE OF REPORT: Repeated Dose Dermal Toxicity: 21-Day Study with DPX-14740-166 (Diuron) in Rabbits

AUTHOR: S. MacKenzie

REPORT ISSUED: September 21, 1992

QUALITY ASSURANCE STATEMENT: A signed Good Laboratory Compliance Statement (dated September 16 and October 21, 1992) and a signed Quality Assurance Document with a list of inspection dates (dated September 21, 1992) were included.

CONCLUSIONS: Diuron was administered dermally to New Zealand White rabbits daily (6 hours/day) for a total of 21 (males) or 22 (females) consecutive applications at doses of 0, 50, 500, and 1200 mg/kg/day.

NOEL for systemic toxicity = 1200 mg/kg/day.
LEL for systemic toxicity was not determined.

NOEL for dermal toxicity = 1200 mg/kg/day.
LEL for dermal toxicity was not determined.

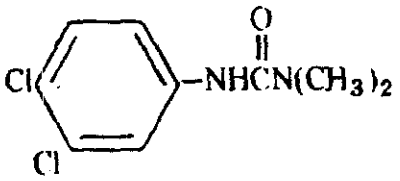
CORE CLASSIFICATION: Core Minimum. This study satisfies the requirements of Guideline Series 82-2 for a 21-day dermal toxicity study in rabbits; data on the stability of the test material were not provided. This study meets the requirements of the limit test.

A. MATERIALS, METHODS, AND RESULTS

1. Test Article Description

Name: *N'*(3,4-dichlorophenyl)-*N*,*N*-dimethyl-urea (Diuron)

Structure:




Diuron

Lot number: 2507

CAS number: 330-54-1

Purity: 96.8%

Physical property: White powder

Stability: Not reported

Vehicle: Deionized water

2. Test Article Analyses for Purity and Stability

Purity and stability analyses of the test material were not performed as part of this study. Purity (as provided by the Sponsor) was reported to be 96.8%. No data on the stability of the test material were provided.

3. Animals

Male and female New Zealand White rabbits (approximately 16 weeks old), were received from Hazleton Research Products, Denver, PA. Animals were housed individually in suspended, stainless steel cages and acclimated for approximately 2 weeks prior to treatment. Water was provided *ad libitum*; approximately 125 g of food (Purina® High Fiber Rabbit Chow # 5325) was offered to each animal daily. Animal

rooms were environmentally controlled at targeted temperatures of 20±2°C, relative humidity of 50±10%, and a 12-hour photoperiod; small differences observed in these parameters were not believed to affect the study outcome.

Rabbits were assigned to test groups (5/sex/dose) as shown below using a computer-generated randomization procedure based on body weights. Animals were identified with ear tags. Dose levels were adjusted based on each animal's most recent body weight determination. At the initiation of treatment, rabbit body weight ranges were 1899-2518 g for males and 1814-2934 g for females. Deionized water was applied to control animals.

The group assignments and dose levels for rabbits exposed to diuron were as follows:

| Dose Group | Dose Level (mg/kg/day) |
|---|---|
| Group 1 (Control) | 0 |
| Group 2 (Low-) | 50 |
| Group 3 (Mid-) | 500 |
| Group 4 (High-) | 1200 |

Rationale for dose selection: Doses were selected on the basis of a range-finding study in which rabbits dermally exposed 6 hours/day for 5 days to doses up to 1500 mg/kg/day showed no clinical signs of toxicity; mild/slight erythema was noted at 1500 mg/kg/day at days 1 and 2. In addition, in an acute dermal toxicity study (HLR 773-79) in which rabbits received a single dermal dose of 2000 mg/kg, no clinical signs of toxicity were noted. Based on these results, 1200 mg/kg/day was selected as the high dose.

4. Test Procedure

Hair was clipped from the back of each rabbit approximately 1 day prior to treatment. Individually specified amounts of test material were weighed and mixed in a weighing boat with approximately 5 mL of de-ionized water to form a paste or slurry. The suspension was further washed from the boat with deionized water, and the mixture was applied to a gauze pad; this was placed on the clipped, intact skin of the rabbit to cover at least 10% of the total body surface area. Control animals received the vehicle (5 mL/animal/day) as a treatment applied to their gauze pads. The gauze was wrapped with layers of plastic film, stretch gauze bandage, and elastic adhesive bandage, successively. Animals were exposed for 6 hours/day for a total of 21 (males) or 22 (females) consecutive days. Following each exposure, the wrappings were removed and the test sites were washed

with lukewarm tap water or bottled spring water; the skin was patted dry.

5. Statistical Methods

-- Body weight, body weight gain, and organ weight data were analyzed using one-way analysis of variance. If significant ($p < 0.05$) differences were noted in body weight or weight gain data, Dunnett's test was applied; organ weight data were also examined using Dunnett's test.

-- Clinical observations were analyzed using Fisher's Exact test with a Bonferroni correction.

-- Clinical laboratory data were analyzed using one-way analysis of variance and Bartlett's test. If significant differences among group means were noted, Dunnett's test was applied. If the results of Bartlett's test were significant ($p < 0.005$), the Kruskal-Wallis and Mann-Whitney U tests were used.

6. General Observations

(a) Mortality/moribundity/survival

Observations for mortality and moribundity were made daily.

No animals died prior to termination of the study and no moribundity was observed.

(b) Clinical observations/dermal reactions

Daily observations were made for clinical signs of toxicity. The dermal application site was evaluated daily for signs of irritation, approximately 1 hour after bandage removal. Erythema and edema were scored using the Draize scoring system. Other dermal changes were noted separately, if present.

No treatment-related clinical effects were reported. Occurrences of mucus in stool were noted in 1 male from each treatment group and diarrhea in 1 male at 1200 mg/kg/day; these effects were considered incidental.

In most treated and control groups, slight erythema was observed by days 10-11 progressing to mild erythema by days 10-14. Similar incidences were noted across all groups. Moderate erythema was noted at 1200 mg/kg/day in 2 females. Slight (1 male) or mild (1 female) transient edema was noted at 1200 mg/kg/day. The dermal effects observed are considered to be the result of mechanical injury to the skin since slight to mild erythema was noted in control animals and other effects (scabs, scar tissue, red spots, and cuts) were also observed sporadically in all dose groups.

(c) Body weight

Animals were weighed twice weekly (3-4-day intervals) before and during the treatment period.

No treatment-related effects on body weights or weight gains were noted for any treatment group relative to controls.

(d) Food consumption

Group food consumption was measured weekly, with mean daily food consumption (g/rabbit) being estimated from this data.

No treatment-related effect on food consumption or food efficiency was reported.

7. Clinical Pathology

Blood samples were taken from the auricular artery of each rabbit 8 days prior to initiation of treatment and approximately 1 day after the last treatment (just prior to euthanasia). It was not specified whether the animals were fasted prior to sampling. Pretest data were used to identify and eliminate outliers from the study; these data were not used for statistical comparison with posttest data. The parameters checked (X) below were examined:

(a) Hematology

X Hematocrit (HCT)*
X Hemoglobin (HGB)*
X Leukocyte count (WBC)*
X Erythrocyte count (RBC)*
X Platelet count*
Reticulocyte count (RETIC)
Red cell morphology
X Leukocyte differential count*
Corrected leukocyte count (COR WBC)
X Mean corpuscular HGB (MCH)
X Mean corpuscular HGB concentration (MCHC)
X Mean corpuscular volume (MCV)
Thrombotest
Leukoctye and platelet morphology

* = Recommended by Subdivision F(November 1984) Guidelines

Hematocrit, hemoglobin, and cell count analyses were performed using a Serono Baker 9000® hematology analyzer. Leukocyte differential counts were determined manually.

No treatment-related changes in the hematologic parameters examined were reported.

(b) Clinical chemistry:

Electrolytes

X Calcium*
X Chloride*
Magnesium
X Phosphorus*
X Potassium*
X Sodium*

Enzymes

X Alkaline phosphatase (ALP)
Cholinesterase
Lactic acid dehydrogenase
X Serum alanine aminotransferase (SGPT)*
X Serum aspartate aminotransferase (SGOT)*
Gamma glutamyltransferase (GGT)
Creatinine phosphokinase

Other

X Albumin*
Albumin/globulin ratio
X Blood creatinine*
X Blood urea nitrogen*
X Cholesterol
X Globulin (calculated)
X Glucose*
X Total bilirubin*
Direct bilirubin
X Total protein*
Triglycerides

* = Recommended by Subdivision F (November 1984) Guidelines

Blood samples were analyzed for the above parameters using a Coulter DACOS® clinical chemistry analyzer.

No treatment-related changes in clinical chemistry parameters were observed. A significant increase (57%, $p < 0.05$) in mean cholesterol levels was noted in females at 1200 mg/kg/day when compared to controls. The study author indicated that the cholesterol levels in the high-dose females were within the normal range; however, historical control data for this parameter were not provided.

8. Sacrifice and Pathology

Animals were anesthetized and exsanguinated approximately 1 day after the last treatment. Gross necropsy was performed on all animals. Animals in the control and high-dose groups had histological examinations conducted for those organs checked (X) below. Treated and untreated skin, liver, gall bladder, kidneys, and lesions from animals in the low- and mid-dose groups were also examined. Double-checked (XX) organs from animals in all dose groups were also weighed.

Digestive System

Tongue
X Salivary glands
X Esophagus
X Stomach
X Duodenum
X Jejunum
X Ileum
X Cecum
X Colon
X Rectum
XX Liver*
X Gall bladder
X Pancreas

Respiratory

X Trachea
X Lung

Cardiovascular/Hematologic

X Aorta
X Heart
X Bone marrow
X Lymph nodes
X Spleen
X Thymus

Urogenital

XX Kidneys*
X Urinary bladder
XX Testes
X Epididymides
X Prostate
Seminal vesicle
X Ovaries
X Uterus
X Vagina

Neurologic

X Brain
X Peripheral nerve (sciatic nerve)
Spinal cord (three levels)
X Pituitary
X Eyes

Glandular

XX Adrenals
Lacrimal gland
Mammary gland
X Thyroids
X Parathyroids
Harderian glands

Other

X Bone (sternum and femur)
X Skeletal muscle
X Skin (treated and untreated)*
X All gross lesions and masses*

* = Recommended by Subdivision F (November 1984) Guidelines

(a) Macroscopic

No treatment-related lesions were noted in animals in any group. Sporadic observations occurring as single events included skin nodules in females (50, 500, and 1200 mg/kg/day), and small testes, skin ulceration/erosion, and lung discoloration in males (1200 mg/kg/day).

(b) Organ weights and body weight ratios

No treatment-related effects were observed in absolute or relative organ weights in any group.

(c) Microscopic

An increased incidence of cardiomyopathy (minimal or mild) was observed in females at 1200 mg/kg/day (2/5) compared to controls (0/5). The cardiomyopathy may have been due to the aortic degeneration/mineralization (3/5, minimal) that was also observed in control animals (2/5, mild), although severity was slightly increased among the high-dose females. An increased incidence of retinal folds (minimal) was observed in males at 1200 mg/kg/day (2/5) compared to controls (0/5), but the biological significance of this finding is unknown.

B. DISCUSSION

The conduct of the study was adequate. No treatment-related changes in clinical observations, body weight, body weight gain, food consumption, hematology, or histopathology parameters were noted. Based on these results, the NOEL for systemic toxicity was 1200 mg/kg/day; the LEL was not determined. However, the highest dose tested (i.e., 1200 mg/kg/day) was a limit test for a repeated dermal toxicity study. The NOEL for dermal toxicity was 1200 mg/kg/day.

STUDY/REPORTING DEFICIENCIES

Deficiencies in the reporting of data were as follows:

- No data on the stability of the test material were reported. The study report (CBI p. 13) states that the compound was assumed to be stable "in the absence of visible evidence to the contrary."
- Glucose levels were not determined in fasted animals as per Guidelines.

These deficiencies were not sufficient to affect the outcome of the study.

Core Classification: Minimum, due to lack of data on the stability of the test material.

DU PONT

010441

AGRICULTURAL PRODUCTS
Walker's Mill, Barley Mill Plaza
P.O. Box 80038
Wilmington, DE 19880-0038
Registration & Regulatory Affairs
Fax: 302-992-6470

July 12, 1993

**SENT BY FAX**

Clark Swentzel,
Section Chief, Toxicity Branch
U.S. Environmental Protection Agency
Document Processing Desk (H7509C)
Room 820E, Crystal Mall 2
1921 Jefferson Davis Highway
Arlington, VA 22202

7-12-93 ;10:35AM ; DU PONT→ ;# 2/ 2

Dear Mr. Swentzel:

In our recent telephone conversation, you mentioned that you would like to have some information on the stability of the diuron that is applied as a slurry/solution in water in the 21-day study in rabbits (MRID No. 42718301). As you are aware, the diuron was applied to the skin of the rabbits and occluded for approximately 6 hours daily. It was then washed off. This procedure was repeated for 21 to 22 consecutive days.

The test material was made up in water and applied within 1 to 4 hours. Hydrolysis studies at pH's 5, 7, and 9, at 25 ± 1°C and in darkness show that the 1/2 - life in water is greater than 500 days. After 30 days, no significant degradation of the diuron had taken place in any of the studies. The report containing these studies has been accepted by the Agency (MRID No. ~~41488804~~).

0 1 KCS 41418804

These data suggest that the diuron should be stable throughout the preparation and application periods involved in this repeated dose dermal toxicity study in rabbits. Please let me know if there are any further questions related to this study.

Sincerely,

Ian Wellings

Ian Wellings, Ph.D.
Product Registration Manager

IW/msd

DRAFT

# DATA EVALUATION REPORT

Diuron

Study Type:
21-Day Repeated Dermal Toxicity Study

Prepared for:

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

Prepared by:

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

| | | |
|---|---|---|
| Principal Reviewer | Patricia Bittner, M.S. | Date 6/10/93 |
| Independent Reviewer | John Liccione, Ph.D. | Date 6/10/93 |
| QA/QC Manager | Sharon Segal, Ph.D. | Date 6/10/93 |

Contract Number: 68D10075
Work Assignment Number: 2-120
Clement Number: 385
Project Officer: Caroline Gordon

EPA Reviewer, Section Head: K. Clark Swentzel Signature: _____
Review Section II, Toxicology Branch II,
Health Effects Division Date: _____

DATA EVALUATION REPORT

STUDY TYPE: 21-Day repeated dermal toxicity study

TEST MATERIAL: Diuron

MRID NUMBER: 427183-01

CAS REGISTRY NUMBER: 330-54-1

TOX CHEM NUMBER:

SYNONYMS: DPX-14740-166; dichlorofenidim; Cekiuron® (Cequisa); Dailon®; Daiter®; Farmco®; Toterbane 50F®; Vonduron®; IN-14740-166; IN 14740; 14740-166 Milled Diuron technical

STUDY REPORT NUMBER: HLR 484-92

MEDICAL RESEARCH NUMBER: 9122-001

SPONSOR: DuPont Agricultural Products
E.I. du Pont de Nemours and Company
Wilmington, DE

TESTING FACILITY: E.I. du Pont de Nemours and Company
Haskell Laboratory for Toxicology and Industrial Medicine
Newark, DE

TITLE OF REPORT: Repeated Dose Dermal Toxicity: 21-Day Study with DPX-14740-166 (Diuron) in Rabbits

AUTHOR: S. MacKenzie

REPORT ISSUED: September 21, 1992

QUALITY ASSURANCE STATEMENT: A signed Good Laboratory Compliance Statement (dated September 16 and October 21, 1992) and a signed Quality Assurance Document with a list of inspection dates (dated September 21, 1992) were included.

CONCLUSIONS: Diuron was administered dermally to New Zealand White rabbits daily (6 hours/day) for a total of 21 (males) or 22 (females) consecutive applications at doses of 0, 50, 500, and 1200 mg/kg/day.

NOEL for systemic toxicity = 1200 mg/kg/day.
LEL for systemic toxicity was not determined.

NOEL for dermal toxicity = 1200 mg/kg/day.
LEL for dermal toxicity was not determined.

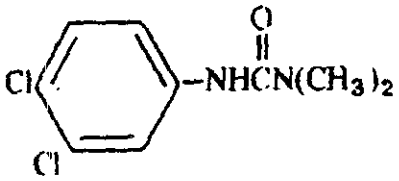
CORE CLASSIFICATION: Core Minimum. This study satisfies the requirements of Guideline Series 82-2 for a 21-day dermal toxicity study in rabbits; data on the stability of the test material were not provided. This study meets the requirements of the limit test.

A. MATERIALS, METHODS, AND RESULTS

1. Test Article Description

Name: *N'*(3,4-dichlorophenyl)-*N*,*N*-dimethyl-urea (Diuron)

Structure:




Diuron

Lot number: 2507

CAS number: 330-54-1

Purity: 96.8%

Physical property: White powder

Stability: Not reported

Vehicle: Deionized water

2. Test Article Analyses for Purity and Stability

Purity and stability analyses of the test material were not performed as part of this study. Purity (as provided by the Sponsor) was reported to be 96.8%. No data on the stability of the test material were provided.

3. Animals

Male and female New Zealand White rabbits (approximately 16 weeks old), were received from Hazleton Research Products, Denver, PA. Animals were housed individually in suspended, stainless steel cages and acclimated for approximately 2 weeks prior to treatment. Water was provided *ad libitum*; approximately 125 g of food (Purina® High Fiber Rabbit Chow # 5325) was offered to each animal daily. Animal

rooms were environmentally controlled at targeted temperatures of 20±2°C, relative humidity of 50±10%, and a 12-hour photoperiod; small differences observed in these parameters were not believed to affect the study outcome.

Rabbits were assigned to test groups (5/sex/dose) as shown below using a computer-generated randomization procedure based on body weights. Animals were identified with ear tags. Dose levels were adjusted based on each animal's most recent body weight determination. At the initiation of treatment, rabbit body weight ranges were 1899-2518 g for males and 1814-2934 g for females. Deionized water was applied to control animals.

The group assignments and dose levels for rabbits exposed to diuron were as follows:

| Dose Group | Dose Level (mg/kg/day) |
|---|---|
| Group 1 (Control) | 0 |
| Group 2 (Low-) | 50 |
| Group 3 (Mid-) | 500 |
| Group 4 (High-) | 1200 |

Rationale for dose selection: Doses were selected on the basis of a range-finding study in which rabbits dermally exposed 6 hours/day for 5 days to doses up to 1500 mg/kg/day showed no clinical signs of toxicity; mild/slight erythema was noted at 1500 mg/kg/day at days 1 and 2. In addition, in an acute dermal toxicity study (HLR 773-79) in which rabbits received a single dermal dose of 2000 mg/kg, no clinical signs of toxicity were noted. Based on these results, 1200 mg/kg/day was selected as the high dose.

4. Test Procedure

Hair was clipped from the back of each rabbit approximately 1 day prior to treatment. Individually specified amounts of test material were weighed and mixed in a weighing boat with approximately 5 mL of de-ionized water to form a paste or slurry. The suspension was further washed from the boat with deionized water, and the mixture was applied to a gauze pad; this was placed on the clipped, intact skin of the rabbit to cover at least 10% of the total body surface area. Control animals received the vehicle (5 mL/animal/day) as a treatment applied to their gauze pads. The gauze was wrapped with layers of plastic film, stretch gauze bandage, and elastic adhesive bandage, successively. Animals were exposed for 6 hours/day for a total of 21 (males) or 22 (females) consecutive days. Following each exposure, the wrappings were removed and the test sites were washed

with lukewarm tap water or bottled spring water; the skin was patted dry.

5. Statistical Methods

-- Body weight, body weight gain, and organ weight data were analyzed using one-way analysis of variance. If significant ($p < 0.05$) differences were noted in body weight or weight gain data, Dunnett's test was applied; organ weight data were also examined using Dunnett's test.

-- Clinical observations were analyzed using Fisher's Exact test with a Bonferroni correction.

-- Clinical laboratory data were analyzed using one-way analysis of variance and Bartlett's test. If significant differences among group means were noted, Dunnett's test was applied. If the results of Bartlett's test were significant ($p < 0.005$), the Kruskal-Wallis and Mann-Whitney U tests were used.

6. General Observations

(a) Mortality/moribundity/survival

Observations for mortality and moribundity were made daily.

No animals died prior to termination of the study and no moribundity was observed.

(b) Clinical observations/dermal reactions

Daily observations were made for clinical signs of toxicity. The dermal application site was evaluated daily for signs of irritation, approximately 1 hour after bandage removal. Erythema and edema were scored using the Draize scoring system. Other dermal changes were noted separately, if present.

No treatment-related clinical effects were reported. Occurrences of mucus in stool were noted in 1 male from each treatment group and diarrhea in 1 male at 1200 mg/kg/day; these effects were considered incidental.

In most treated and control groups, slight erythema was observed by days 10-11 progressing to mild erythema by days 10-14. Similar incidences were noted across all groups. Moderate erythema was noted at 1200 mg/kg/day in 2 females. Slight (1 male) or mild (1 female) transient edema was noted at 1200 mg/kg/day. The dermal effects observed are considered to be the result of mechanical injury to the skin since slight to mild erythema was noted in control animals and other effects (scabs, scar tissue, red spots, and cuts) were also observed sporadically in all dose groups.

(c) Body weight

Animals were weighed twice weekly (3-4-day intervals) before and during the treatment period.

No treatment-related effects on body weights or weight gains were noted for any treatment group relative to controls.

(d) Food consumption

Group food consumption was measured weekly, with mean daily food consumption (g/rabbit) being estimated from this data.

No treatment-related effect on food consumption or food efficiency was reported.

7. Clinical Pathology

Blood samples were taken from the auricular artery of each rabbit 8 days prior to initiation of treatment and approximately 1 day after the last treatment (just prior to euthanasia). It was not specified whether the animals were fasted prior to sampling. Pretest data were used to identify and eliminate outliers from the study; these data were not used for statistical comparison with posttest data. The parameters checked (X) below were examined:

(a) Hematology

X Hematocrit (HCT)*
X Hemoglobin (HGB)*
X Leukocyte count (WBC)*
X Erythrocyte count (RBC)*
X Platelet count*
Reticulocyte count (RETIC)
Red cell morphology
X Leukocyte differential count*
Corrected leukocyte count (COR WBC)
X Mean corpuscular HGB (MCH)
X Mean corpuscular HGB concentration (MCHC)
X Mean corpuscular volume (MCV)
Thrombotest
Leukoctye and platelet morphology

* = Recommended by Subdivision F(November 1984) Guidelines

Hematocrit, hemoglobin, and cell count analyses were performed using a Serono Baker 9000® hematology analyzer. Leukocyte differential counts were determined manually.

No treatment-related changes in the hematologic parameters examined were reported.

(b) Clinical chemistry:

Electrolytes

X Calcium*
X Chloride*
Magnesium
X Phosphorus*
X Potassium*
X Sodium*

Enzymes

X Alkaline phosphatase (ALP)
Cholinesterase
Lactic acid dehydrogenase
X Serum alanine aminotransferase (SGPT)*
X Serum aspartate aminotransferase (SGOT)*
Gamma glutamyltransferase (GGT)
Creatinine phosphokinase

Other

X Albumin*
Albumin/globulin ratio
X Blood creatinine*
X Blood urea nitrogen*
X Cholesterol
X Globulin (calculated)
X Glucose*
X Total bilirubin*
Direct bilirubin
X Total protein*
Triglycerides

* = Recommended by Subdivision F (November 1984) Guidelines

Blood samples were analyzed for the above parameters using a Coulter DACOS® clinical chemistry analyzer.

No treatment-related changes in clinical chemistry parameters were observed. A significant increase (57%, $p < 0.05$) in mean cholesterol levels was noted in females at 1200 mg/kg/day when compared to controls. The study author indicated that the cholesterol levels in the high-dose females were within the normal range; however, historical control data for this parameter were not provided.

8. Sacrifice and Pathology

Animals were anesthetized and exsanguinated approximately 1 day after the last treatment. Gross necropsy was performed on all animals. Animals in the control and high-dose groups had histological examinations conducted for those organs checked (X) below. Treated and untreated skin, liver, gall bladder, kidneys, and lesions from animals in the low- and mid-dose groups were also examined. Double-checked (XX) organs from animals in all dose groups were also weighed.

Digestive System

Tongue
X Salivary glands
X Esophagus
X Stomach
X Duodenum
X Jejunum
X Ileum
X Cecum
X Colon
X Rectum
XX Liver*
X Gall bladder
X Pancreas

Respiratory

X Trachea
X Lung

Cardiovascular/Hematologic

X Aorta
X Heart
X Bone marrow
X Lymph nodes
X Spleen
X Thymus

Urogenital

XX Kidneys*
X Urinary bladder
XX Testes
X Epididymides
X Prostate
Seminal vesicle
X Ovaries
X Uterus
X Vagina

Neurologic

X Brain
X Peripheral nerve (sciatic nerve)
Spinal cord (three levels)
X Pituitary
X Eyes

Glandular

XX Adrenals
Lacrimal gland
Mammary gland
X Thyroids
X Parathyroids
Harderian glands

Other

X Bone (sternum and femur)
X Skeletal muscle
X Skin (treated and untreated)*
X All gross lesions and masses*

* = Recommended by Subdivision F (November 1984) Guidelines

(a) Macroscopic

No treatment-related lesions were noted in animals in any group. Sporadic observations occurring as single events included skin nodules in females (50, 500, and 1200 mg/kg/day), and small testes, skin ulceration/erosion, and lung discoloration in males (1200 mg/kg/day).

(b) Organ weights and body weight ratios

No treatment-related effects were observed in absolute or relative organ weights in any group.

(c) Microscopic

An increased incidence of cardiomyopathy (minimal or mild) was observed in females at 1200 mg/kg/day (2/5) compared to controls (0/5). The cardiomyopathy may have been due to the aortic degeneration/mineralization (3/5, minimal) that was also observed in control animals (2/5, mild), although severity was slightly increased among the high-dose females. An increased incidence of retinal folds (minimal) was observed in males at 1200 mg/kg/day (2/5) compared to controls (0/5), but the biological significance of this finding is unknown.

B. DISCUSSION

The conduct of the study was adequate. No treatment-related changes in clinical observations, body weight, body weight gain, food consumption, hematology, or histopathology parameters were noted. Based on these results, the NOEL for systemic toxicity was 1200 mg/kg/day; the LEL was not determined. However, the highest dose tested (i.e., 1200 mg/kg/day) was a limit test for a repeated dermal toxicity study. The NOEL for dermal toxicity was 1200 mg/kg/day.

STUDY/REPORTING DEFICIENCIES

Deficiencies in the reporting of data were as follows:

- No data on the stability of the test material were reported. The study report (CBI p. 13) states that the compound was assumed to be stable "in the absence of visible evidence to the contrary."
- Glucose levels were not determined in fasted animals as per Guidelines.

These deficiencies were not sufficient to affect the outcome of the study.

Core Classification: Minimum, due to lack of data on the stability of the test material.