

8-16-84



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

003991

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT : Application for registration of SN 584 in utility poles
Accession No. 251810 Caswell No. 573

TO : Henry Jacoby, PM 21
Registration Division (TS-767)

FROM : Quang Q. Bui, Ph.D. *Quang Q. Bui 8/16/84*
Section V, Toxicology Branch
Hazard Evaluation Division (TS-769)

THRU : Laurence D. Chitlik, DABT *LOC 8/17/84*
Section Head, Section V
Toxicology Branch/HED (TS-769)

THRU : William Burnam, Chief
Toxicology Branch
Hazard Evaluation Division (TS-769)

Action Requested: Review of acute and sub-chronic studies

Registrant: Nor Am Agricultural Products, Inc.,
Naperville, Illinois 60566

Recommendation:

From the available toxicology data on methylisothiocyanate it can be concluded that this chemical should be classified as Tox. Cat. I based upon its eye-irritating properties.

Since SN 584 may only be applied by the use of capsules inserted in utility poles (see Background Section below) by qualified persons employed or contracted by utility companies, the applicator exposure would be expected to be low if appropriate protective measures are taken. However, Toxicology Branch does not find that the registration of SN-584 for use in utility poles to be toxicologically supported at this time. Nearly all of the newly submitted studies (acute and subchronic dermal) have reporting deficiencies and are classified as Core Supplementary Data (see individual study review). If these studies can be upgraded, no additional toxicology studies will be required (see page 3 for the list of studies that may potentially be upgraded if additional data are submitted).

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Background:

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SN-584 is an encapsulated product containing 90% technical methyl isothiocyanate (MITC) and 10% inert ingredients. Each capsule weighs [REDACTED] and the active ingredient MITC (19 grams).

In addition to this application for registration of SN-584, MITC is also a component (20%) of the fumigant Vorlex.

Available Toxicity Data:

Previously submitted data are summarized as follows:

A. Methyl isothiocyanate

1. W. Dykstra's memo of 01/31/78 (EPA registration # 2139-55)

- a) Acute inhalation, rat : LC50 = 1.9 mg/L/1 hour, Tox. II, Minimum Data, Huntingdon Res. Cen. 7/4/77.
- b) Eye irritation, rabbit : Tox. I, Minimum Data (a) Huntingdon Res. Center. 12/23/76

2. G. Burin's memos of 12/21/82 and 04/15/83

- a) Eye irritation - antidotal study, rabbit: Supplementary Data; Sodium bicarbonate and Cortisone enanthate are effective.
- b) (a) Eye irritation, rabbit: Huntingdon Res. Cen. 12/23/76 reclassified as Supplementary Data. However, no new study is requested and the chemical is classified as Tox. Cat. I
- c) Dermal irritation, rabbits: Unspecified testing lab., study report, and final date. Invalid Data.

B. Vorlex

1. W. Dystra's memo of 01/31/78 (EPA registration # 2139-55)

- a) Acute inhalation, rats: LC50 = 11 mg/L/1 hour, Tox. III Huntingdon Res. Cen. 08/22/77, Minimum Data.
- b) Eye irritation, rabbits: Tox. Cat. I, Huntingdon Res. Center 12/23/76, Minimum Data (b)

2. G. Burin's memos of 12/21/82 and 04/15/83

- a) Acute oral, rats: LD 50 = 538 mg/kg, Tox. Cat. III Schering AG, 04/12/79, Minimum Data.
- b) Acute dermal, rabbits: LD 50 = 470 mg/kg, Tox. Cat. II Schering AG, 05/11/79, Minimum Data.

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- c) Acute dermal, rats: LD50 = 961 mg/kg, Tox. Cat. II
Schering AG., 04/11/79; Minimum Data.
- d) Acute intraperitoneal, rats: LD50 = 259 mg/kg
Schering AG., 05/26/79; Minimum Data.
- e) (b) Eye irritation, rabbits: Huntingdon Res. Cen.
12/23/76. Reclassified as Supplementary Dat. However,
no new study is requested and the compound is classified
as Tox. Cat. I
- f) Dermal irritation, rabbits: Invalid Data
- g) 13-week inhalation, rats: Schering AG., 04/10/79
Supplementary Data.

Summary of newly submitted studies conducted with MITC

- 1) Acute oral LD50, rats: LD50 = 95 mg/kg (males and females)
Tox. Cat. II, Minimum Data; Schering AG., 7/6/79.
- 2) Acute oral LD50, rats: LD50 = 175 mg/kg (males)
Supplementary Data*; Tokyo Dental College, 1970.
- 3) Acute oral LD50, mice: LD50 = 90 mg/kg (males)
Supplementary Data*; Tokyo Dental College, 1970.
- 4) Acute oral LD50, mice: LD50 = 104 mg/kg (females)
Minimum Data; Matsumoto Dental School, 1974.
- 5) Acute oral LD50, rabbits: LD50 undetermined
Supplementary Data, Hazleton Lab., 4/28/76.
- 6) Acute oral LD50, dogs: LD50 undetermined
Supplementary Data, Hazleton Lab., 4/28/76.
- 7) Acute oral LD50, monkeys: LD50 undetermined
Supplementary Data, Hazleton Lab., 4/28/76.
- 8) 30-day dermal, rats: Systemic NOEL < 120 mg/kg/day
Supplementary Data; Nara Medical College.
- 9) 31-day dermal, rats: Systemic NOEL undetermined
Supplementary Data*; Schering AG., 11/26/75.
- 10) Acute subcutaneous LD50, rats: LD50 = 60 mg/kg (males)
and 59 mg/kg (females). Supplementary Data*;
Matsumoto Dental College, 1974.
- 1) Acute subcutaneous LD50, mice: LD50 = 75 mg/kg (males)
and 89 mg/kg (females). Supplementary Data*.
Matsumoto Dental College, 1974.

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- 12) Acute intraperitoneal LD50, rats: LD50 = 54 mg/kg (males) and 56 mg/kg (females); Supplementary Data*; Matsumoto Dental College.
- 13) Acute intraperitoneal LD50, mice: LD50 = 82 mg/kg (males) and 89 mg/kg (females); Supplementary Data*; Matsumoto Dental College.

Supplementary Data* : study that may be upgraded if additional requested data are submitted (see individual reviewed study).

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DATA REVIEWED

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Study I

Study Title : Systemic tolerance study (LD50) of ZK 3.318 of rats (M+F) after a single P.O. administration.
Testing Facility : Schering AG
Report No. : 142/79
Final Report Date: July 6, 1979

Groups of 5 Wistar SPF male and female rats each were exposed to single oral administration of 0, 60, 80, 100, 120, 140, or 160 mg/kg of methylisothiocyanate technical (95.7%) dissolved in sesame oil (1% solution). The control groups received only sesame oil (12 ml/kg). All animals were weighed prior to dosing and at the end of the experiment (21 days). Clinical observations were recorded daily and necropsy was performed on all animals that died and/or survived throughout the entire experiment. All animals were housed individually under climate-controlled conditions with food (pelleted Altromin®) and water ad libitum.

Results:

Administration of methylisothiocyanate resulted in a dose-related increased incidence of apathy and atactic walk. Convulsions were noted at dosage levels above 120 mg/kg. However, all clinical symptoms disappeared by day 4 in those animals that survived.

The mortality rate was recorded as follows : 0, 10, 70, 90, 100, and 100% for the groups receiving 60, 80, 100, 120, 140, and 160 mg/kg, respectively. Death usually occurred within 48 hours after chemical treatment. Necropsy of dead animals did not reveal any compound related findings.

Discussion and Conclusions:

The LD50 was calculated to be 95 mg/kg (86 - 104 mg/kg) for males and females.

Clinical observations were made up to 21 days after treatment instead of the required 14 days. The volumes of sesame oil used for the group receiving 120, 140, and 160 mg/kg were respectively 12, 14, and 16 ml/kg which were relatively high for rats.

Core Classification: Minimum Data

Toxicity Category: II

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Study II

Study Title : Acute oral toxicity test in rats with Methyl Isothiocyanate
Testing Facility : Tokyo Dental College, Tokyo, Japan
Final Report No. : unspecified
Final Report Dated : 1970

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Groups of 10 male rats (Donryu strain) each weighing from 100 to 120 grams were given methyl isothiocyanate by gavage at 88, 133, 167, 200, and 300 mg/kg. Crystal methyl isothiocyanate was dissolved in olive oil and the volume used was 5 ml/kg of body weight. Clinical observations were made for 1 week after chemical treatment. The LD50 was determined by the Litchfield-Wilcoxon method.

Results:

Immediately after administration, increased motor activity was noted among the test animals. Increased lacrimation and nasal discharge were also recorded. Convulsions and ocular hemorrhage were observed in animals of the higher dosage groups.

The mortality rate was recorded as follows : 0, 30, 40, 70, and 100% for the groups receiving 88, 133, 167, 200, and 300 mg/kg, respectively. All deaths occurred within 48 hours after treatment.

Discussion and Conclusions:

The LD50 was calculated to be 175 mg/kg (147 - 205) for male rats. Observations should be made for a period of at least 14 days after chemical treatment although all deaths apparently occurred within 2 days. Terminal body weight and necropsy data for all animals investigated were not available. The purity of the test chemical was not stated and female rats should have been investigated.

Core Classification: Supplementary Data

This study may be upgraded to Core Minimum Data if the purity of the test chemical is submitted.

Toxicity Category: Not assigned (probably II)

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Study III

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Study title : Acute oral toxicity test in mouse with methyl
isothiocyanate
Testing Facility : Tokyo Dental College, Tokyo, Japan.
Report No. : Unspecified
Final Report Date : 1970

Male mice (dd strain) weighing approximately 20 grams were divided into 5 groups of 10 each. Methyl isothiocyanate (39, 59, 88, 133, and 200 mg/kg) was given by gavage in a constant volume (0.2 ml/20 grams of body weight) of olive oil. All animals were observed for a period of 7 days. The Litchfield Wilcoxon method was used to determine the LD50.

Results:

Increased motor activity was noted in all tested animals immediately after chemical treatment. Lacrimation, nasal discharge, convulsion, and ocular hemorrhage were the clinical signs observed.

Mortality recorded in the groups receiving 59, 88, 133, and 200 mg/kg were respectively 20, 50, 70, and 100%. All deaths occurred within 48 hours after chemical administration.

Discussions and Conclusions:

The calculated LD50 was 90 mg/kg (70 - 123) for male mice.

Terminal body weight and purity of the chemical tested were not addressed in the report. Animals were not observed for at least 14 days after treatment and female mice should have been investigated.

Core classification : Supplementary Data

This study may be upgraded to Core Minimum Data if the purity of the test compound and body weight data are submitted.

Toxicity category: Not assigned (probably II)

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Study IV

Study title : Acute toxicity test of methyl isothiocyanate
Testing Facility : Matsumoto Dental School, Japan
Report No. : Unspecified
Report Final Date : 1974

Methyl isothiocyanate (90%) was dissolved in olive oil and given to female mice (dd strain) at a constant volume of 0.1 ml/20 grams of body weight. Six groups of 10 female mice each were used and received a single dose of 70, 83, 100, 120, 140, and 170 mg/kg by gavage. All animals were observed for 7 days.

Litchfield Wilcoxon and probit analysis methods were used to determine the LD50.

Results:

Decreased motor activity were the only clinical signs observed. Death occurred within 48 hours after treatment and was recorded as follows: 0, 30, 40, 50, 90, 100% for the groups receiving 70, 83, 100, 120, 140, and 170 mg/kg.

Discussion and Conclusions:

The LD50 was calculated to be 104 mg/kg (88 - 123) for female mice. However the observation period should cover 14 days post-treatment and terminal body weight reported. Male mice (dd strains) should have also been investigated.

Core Classification: Minimum Data

Toxicity Category: II

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Study V

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Study title : Acute oral toxicity study in rabbits
with methyl isothiocyanate
Testing Facility : Hazleton Laboratories, Vienna, Virginia
Report No. : 947-100
Final Report Date: 04/28/76

Methyl isothiocyanate (96.1% purity, Lot No. 1098-118) was administered at 10.0 and 21.5 mg/kg as a single oral dose in corn oil to two groups of 2 rabbits each. The animals were housed individually with food and water ad libitum. All dosages were based on the weight of the animals (3.05 - 3.34 kg) at study initiation.

Clinical observations were recorded for 14 days. Individual body weight were recorded on day 14.

Results:

No deaths were produced at the dosage levels used. All animals appeared normal throughout the entire study except for one animal of the 10 mg/kg that exhibited slight anorexia on day 8 post-treatment.

Discussion and Conclusions:

The LD50 apparently was greater than 21.5 mg/kg.

The minimum requirements for a dermal LD50 study are 4 animals per sex per dose level and at least 3 dose levels should be used. In addition, the sex of the test animals was not mentioned.

Core Classification: Supplementary Data

The LD50 could not be adequately assessed since the highest dose used is not the limit dose (2,000 mg/kg) for a dermal study. Furthermore 4 animals per sex should be used for each dose level.

Toxicity Category: Not assigned.

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Study VI

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Study title : Acute oral toxicity study in dogs with methyl isothiocyanate
Testing Facility : Hazleton Laboratories, Vienna, Virginia
Final Report No. : 947-100
Final Report Date: 04/28/76

Methyl isothiocyanate (Lot No. 1098-118, 96.1% purity) was administered either in corn oil at 0.1, 0.5, 1.0, 4.64, 10, and 21.5 mg/kg or "as received" in gelatin capsules at 10, 21.5, 46.6, and 100 mg/kg to male beagle dogs. A total of eight animals were used with 2 animals treated twice on the same day due to emesis (see Discussion and Conclusions Section below).

Individual body weights were recorded initially and weekly thereafter. After two weeks of observations, the animals were sacrificed and necropsy performed.

Results:

Death was recorded for two animals that received 21.5 and 100 mg/kg in gelatin capsules. Necropsy observations of these animals revealed congested livers, focal hemorrhagies on the stomach, congested kidneys, and hemolyzed blood-filled intestines with hemorrhagic areas evident on the mucosa.

No signs of toxicity were found in animals receiving 0.1 and 0.5 mg/kg. Emesis, sanguineous emesis, decreased motor activity, generalized weakness, and dark colored stools were observed in dogs receiving higher dosage levels. Necropsy findings in the surviving dogs did not reveal any compound related effects.

Discussion and Conclusions:

Since only one animal was used for each dosage level, the acute oral LD50 in dogs could not be determined from the submitted data. This approach precludes the determination of an acute oral LD50.

Emesis was observed in animals that received dose levels of 1 mg/kg and above, and in several instances, different dosage levels were given to the same animal:

Dog # 17589 : 21.5*, 21.5*, and 0.0 mg/kg
Dog # 17605 : 10.0* and 1.0 mg/kg
Dog # 17588 : 10.0*, 10.0*, and 4.64 mg/kg
Dog # 17587 : 46.4* and 10.0 mg/kg
(*) emesis

Methyl isothiocyanate at a dosage level of 1 mg/kg and above induced toxicity characterized by generalized weakness, decreased motor activity, and dark colored stool without any compound related gross pathological changes.

Core Classification: Supplementary Data

An oral LD50 could not be calculated since only one animal was treated per dose level.

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Study VII

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Study title : Acute oral toxicity study in monkeys with methyl isothiocyanate
Testing Facility : Hazleton Laboratories, Vienna, Virginia.
Final Report No. : 947-100
Final Report Date: 04/28/76

Methyl isothiocyanate (Lot No. 1098-118, 96.1% purity) was dissolved in corn oil and given as a single administration to 4 groups of one male rhesus monkey each at levels of 10, 21.5, 46.4, and 100 mg/kg.

The animals were housed in individual metal cages with food and water ad libitum under a climate-controlled environment. Daily observations were made for one week after treatment. Necropsies were performed on all animals dying during the study or surviving until study termination (one week).

Results:

The animal receiving 100 mg/kg was found dead on the second day after treatment. Necropsy of this animal revealed mottled liver, hemorrhaging stomach mucosa, and bloody stomach. All other animals survived the entire experiment and necropsy of these animals did not reveal any compound changes at the 10 through 46.4 mg/kg dosage levels. Diarrhea (green colored) was observed in the lowest dose group for three consecutive days (days 4-7). Emesis, mydriasis, and diarrhea were the clinical signs associated with the 46.4 mg/kg group.

Discussion and Conclusions:

There were no individual data to confirm the clinical observations and necropsy examinations reported. Furthermore, the use of one animal per group precludes a determination of a LD50.

Core Classification: Supplementary Data

The LD50 could not be calculated since only one animal was used per group.

Study VIII

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Study title : One month toxicological study of MITC in rats by dermal application.
Testing Facility : Nara Medical College, Japan.
Final Report No. : Unspecified
Final Report Date: Unspecified

Procedures:

Groups of 10 Sprague Dawley rats of each sex per dose level were exposed dermally to 0, 120, 240, and 480 mg/kg of MITC for one month. The test compound was dissolved in benzol and applied to a hair-clipped area (4x4 cm) of the skin. Control animals were treated with benzol only.

Body weights were recorded three times a week whereas food and water consumption were measured weekly. Clinical observations were made daily and necropsy was performed on all animals that died during the study or survived until study termination. At necropsy, the following organs were removed and weighed: heart, liver, spleen, lung, kidney, adrenal, thymus, testis, ovaries, prostate, uterus, brain, and pituitary. Histopathologic examinations were performed on 23 organs.

At study termination, hematology and urinalysis were conducted on all animals. Clinical chemistry determinations included calcium, chloride, urea nitrogen, glucose total protein, hepatic enzymes, and cholinesterase enzymes.

Results:

1) Mortality

Two animals died during the experiment: one each in the control and low dose groups. A large abscess in the lung was attributed as the cause of death in each case.

2) Body weight

Body weight data of the treated and control groups are presented as follows:

Body weight (in grams)	Control		Low		Mid		High	
	M	F	M	F	M	F	M	F
Initial	236.0	181.2	238.6	179.8	233.2	182.4	237.8	182.6
Final	336.6	232.8	269.2	220.0	274.0	222.6	279.5	223.2
BW gain	100.6	51.6	30.6	40.2	40.8	40.2	41.7	40.6
% increase(a)†	42.6	28.5	12.8	22.4	17.5	22.0	17.5	22.2
% increase(b)	100.0	100.0	30.4	77.9	40.6	77.9	41.5	78.7

Data extracted from Table 1.

(a) as compared to the respective initial body weight

(b) as compared to control body weight gain

† calculated by this reviewer

Dermal application of methyl isothiocyanate (MITC) to male rats resulted in body weight gain depression observed at all dosage levels tested. The low, mid, and high dose males gained 69.6, 59.4, and 58.5% less than their concurrent controls, respectively. The body weight gain in the treated females was slightly lower but comparable to that of control females.

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3. Food and Water Consumption

	Control		Low		Mid		High	
	M	F	M	F	M	F	M	F
Food cons. (grams/day/ animal)	18.4	16.1	15.9	14.3	16.4	14.4	15.7	14.3
Food (% of control) †	100.0	100.0	86.4	88.8	89.1	89.4	85.3	88.8
Water cons. (ml/day/ animal)	32.3	29.4	33.9	27.7	30.5	31.3	34.9	31.2
Food effic.	0.177	0.078	0.059	0.062	0.065	0.080	0.063	0.082
Food effic. (% control)	100.0	100.0	33.3	79.5	36.7	102.6	35.6	105.1

Data extracted from Tables 2, 3, and 4.

The food efficiency index (ratio of body weight increase/food consumed) of the treated females was comparable or greater to that of the respective control animals. This index, however, was greatly reduced in the males with the values being 33.3, 36.7, and 35.6% of the controls for the low, mid, and high dose groups, respectively. This finding suggests that the decreased body weight gain observed in the males of the treated groups was compound induced.

4. Urinalysis

No compound related findings were noted in urinalysis of the treated groups in comparison to control animals.

5. Hematology

In the males, a slight decrease in RBC, WBC, and hemoglobin was noted in the highest dosage group but not dose related.

In the females, these values in the treated groups were also lower than control findings but were not considered as biologically significant by this reviewer. Differential leucocyte count, was not performed or not reported.

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6. Clinical chemistry *

The most relevant findings are tabulated as follows :

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	<u>Control</u>	<u>Low</u>	<u>Mid</u>	<u>High</u>
Total Serum protein				
male	6.7	6.5	6.1	6.2
female	6.9	6.3	6.1	6.2
Albumin				
male	51.6	45.3	46.9	45.0
female	50.3	47.4	50.6	47.5
SGOT				
male	177	220	213	205
female	211	219	236	211
LDH				
male	2930	3878	3110	3462
female	3477	3357	3744	3448
Glucose				
male	143	137	127	139
female	125	117	121	115

Data extracted from Table 6

* Units were not indicated in the report

In both males and females of the treated groups, the blood levels of total serum protein were lower than those of the controls. Similar findings were noted in albumin levels except for females of the mid-dose group. Slight to moderate increased in SGOT values were recorded in males at all dosage levels and females in the low and mid-dose groups. The male LDH levels in all treated groups were also higher than those of controls.

The blood glucose levels of all males and females of the treated groups were lower than those of the control group. However, no dose-response effects were found and, consequently, these findings were not biologically meaningful.

All other parameters investigated were comparable to control values.

Clinical chemistry determinations should also include : Na⁺, K⁺, creatinine, and total bilirubin.

7. Organ weights

The absolute weight of several organs (lung, liver, thymus, and kidneys in both males and females and genital organs in males) in all treated groups including the lowest dose used were less than control values. The significance of these findings, however, could not be properly assessed due to the absence of relative organ weight data (ratio of organ weight/terminal body weight) and statistical evaluations.

8. Gross dermal pathology

Erosion changing to ulcer was observed in the skin of treated animals two weeks after application. This observation intensified proportionally to the doses used. Thickening, severe cornification, necrosis of the epithelium, and formation of crust were noted in treated animals.

No dermal damage was recorded from the controls that were treated with the vehicle alone (benzol).

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9. Necropsy data

Most relevant histopathological findings of importance are summarized in the following table.

	<u>Control</u>	<u>Low</u>	<u>Mid</u>	<u>High</u>
<u>Lung</u>				
Bronchitis	6M,3F	10M,7F	9M,8F	7M,6F
Pneumonia	1M,1F	7M,7F	7M,5F	2M,2F
<u>Liver</u>				
Infilt.focus	2M,3F	1M,1F	2M,3F	1M,1F
Fatty degene.	0M,0F	3M,2F	0M,2F	0M,1F
Congestion	1M,0F	0M,1F	1M,0F	1M,0F
<u>Urinary bladder</u>				
Inflammation	0M,0F	2M,1F	3M,0F	3M,2F
Hemorrhage	2M,2F	0M,0F	4M,0F	0M,0F
<u>Skin</u>				
Subc.gran.	0M,2F	8M, 9F	10M, 9F	9M,10F
Ulcerat.	0M,0F	9M, 8F	10M,10F	8M,10F
Hyperpl.	2M,6F	9M,10F	10M,10F	9M,10F
Hyperker.	1M,2F	9M,10F	9M,10F	8M,10F
<u>Spleen</u>				
Inc. hemosid.	1M,2F	2M, 6F	2M, 6F	2M, 4F

Data extracted from Table 8

Pneumonitis and bronchitis occurred at a high frequency among all animals including the controls. The report indicated that pleurisy was not found. However, due to the high incidence of lung diseases observed in all groups, the health status of the animals investigated is questionable.

Skin alterations characterized by ulceration were observed in all treated groups but not the control. This finding clearly indicated that the dermal changes were compound-related. However, ulceration was not accompanied by either splenectasis or enlargement of the lymph nodes.

No specific compound related findings were observed in all other organs investigated.

Discussion and Conclusions:

Ulceration, formation of crust, infiltration of neutrocytes were observed and limited to the applied area of all treated animals. Splenomegaly and enlargement of the lymph nodes were not associated with the dermal changes. Consequently, MITC, based upon the submitted data, did not appear to affect the hematopoietic system.

However, dermal application of MITC suppressed the body weight gain of male animals at all dosage levels including the lowest dose tested (120 mg/kg). The decrease in body weight gain was associated with a slight decrease in food intake resulting in significant reduction in the food efficiency index.

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Core Classification : Supplementary Data

A systemic NOEL could not be demonstrated from the dosage levels selected (decreased body weight gain and food consumption at the lowest dose used, 120 mg/kg). Moreover, the following comments are noted by this reviewer:

- Statistical evaluations were not identified and/or performed.
- Hematologic, clinical chemistry determinations, and urinalysis should also be performed at study initiation.
- Individual male blood and urine determinations are not available.
- Hematologic determinations should also include differential leucocyte count.

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Study title : Subacute (31-day) dermal toxicity to the rat
Testing Facility : Schering AG Berlin, Germany
Final Report No. : 4411
Final Report Date : 11/26/75

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Procedures:

A copy of the procedures used is appended. In general, the methods described are unremarkably different from those commonly employed for a subchronic dermal study and therefore are acceptable.

Results:

Evaluation of this study could not be performed at the present time due to the following reasons:

- a) Only findings with either suspected compound related or statistical significance were mentioned in the report or summarized in tables.
- b) There were no individual data to verify the reported findings.

Conclusions:

The registrant is requested to submit a complete study with all data representing all parameters investigated. Individual data should also be included to validate the reported findings.

Core Classification Data: Supplementary Data

This study, however, may be upgraded if all pertaining data are submitted for complete evaluation.

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Investigational Outline

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Test Animals and Method of Application

Groups of 10 male and 10 female rats each (weight: 140-243 g, Wistar strain, SPF quality, breeder: Zentralinstitut für Versuchstierkunde, Hannover) fed a standard diet (Altromin R) and water ad libitum, housed under conventional conditions in Makrolon cages type II with perforated bottom (one rat per cage) in air-conditioned (ambient temperature 21 - 25 °C, relative humidity: 40-65 %) rooms with controlled lighting (12 hours' day-night rhythm) were treated daily, once per day, during 31 days in total with either sesame oil alone or 1, 10 and 100 mg of methyl isothiocyanate (dissolved, suspended) in 5 ml/kg applied onto a dorsal skin surface of 20-30 cm².

The test material was distributed over the application size of each animal using a sprayer tarped with a mull bandage and allowed to contact the skin for 5 hours. At the end of the contact period all residual material was removed from the skin with a soft pulp ("Kleenex"). Because of the high volatility of MITC gas masks were used throughout the study when handling the material.

The following parameters were investigated:

Clinical studies

General observations every day;
food consumption on 24 days (determined by weighing the portion left every 2 to 3 days);
water consumption on 17 days (determined by weighing every 3 to 4 days);
body weight: once a week

Hematologic studies

Hemoglobin concentration, hematocrit value, erythrocyte count, leucocyte count, differential blood count, thrombocyte count in weeks -2, 0, 4.

Bone marrow examinations

Count of nucleated cells per mg of bone marrow and establishment of myelogram in week 5.

Urine analyses

pH-value, specific weight, albumin, glucose, acetone, blood, urobilinogen and sediment during weeks -2, -1, 2, 4.

LDH determined in the 24 hours urine during weeks -1, 2, 4, 5 in 5 males and 5 females of each group.

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Clinical chemistry in blood, serum and brain

in the blood: Fasting glucose determination
during weeks -2, 2, 4

in the serum: GPT (Glutamate pyruvate transaminase
activity), AP (alkaline phosphatase
activity)
weeks -2, 2, 4
COH weeks -1, 2, 4, 5

blood urea nitrogen concentration
total cholesterol level
weeks -2, 2, 4

total albumin and serum albumin
electrophoresis
weeks -2, 2, 4

in the plasma and
erythrocytes: cholinesterase activity during weeks
-1, 2, 3, 4

in the brain: determination of cholinesterase activity
during week 5 in 5 males and females
each per group

autopsy and organ
weighing autopsy was made of all decedents and
of all animals sacrificed after test
closure.

The following organs were weighed:
liver, kidney, heart, ovary, adrenals,
thyroid gland, testicles, brain

Histologic examinations were made on the following organs:

liver fixing: liquid according to Carnoy,
buffered formalin following Lillie
staining: PAS / H.E., oil-red-O

kidney, adrenals, heart fixing: buffered formalin following Lillie
staining: H.E., oil-red-O

pancreas fixing: Bouin's mixture
staining: aldehyde rubine according to
Scott

stomach, duodenum, jejunum,
Ileum, caecum, colon,
thyroid gland, thymus gland, lungs,
spleen, lymphatic node, cerebrum,
cerebellum, medulla obl.
spinal marrow, peripheric nerve

fixing: buffered formalin according to Lillie

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STUDY X

Study Title : Acute toxicity tests of methylisothiocyanate
Testing Facility : Matsumoto Dental College, Japan
Final Report No. : Unspecified
Final Report Date : 1974

003991

Methyl isothiocyanate (96% purity) was dissolved in olive oil and administered subcutaneously to groups of 10 rats (Donryu strain) per sex each at a constant volume of 0.1 ml/100 grams of body weight. All animals were observed daily for mortality and clinical observations for one week. The dosage levels used were 40, 48, 58, 69, and 83 mg/kg.

Results:

Decreased motor activity was observed in all rats immediately after chemical injection. Tonic convulsion and salivation followed by death were noted in the rats of the higher groups.

Mortality occurred within 24 hours after chemical treatment and all surviving animals were free of clinical symptoms by three days.

<u>Dose levels</u>	<u>Mortality</u>	
	<u>Male</u>	<u>Female</u>
40 mg/kg	0/10	0/10
48 mg/kg	2/10	1/10
58 mg/kg	4/10	4/10
69 mg/kg	8/10	9/10
83 mg/kg	10/10	10/10

Discussion and Conclusions:

The subcutaneous LD50 was reported as 60 mg/kg and 59 mg/kg for males and females respectively.

The observation period should cover at least 14 days post-treatment and initial and final body weight must be presented.

Core Classification Data: Supplementary Data

This study may be upgraded to Core Minimum Data if body weight data are submitted for evaluation.

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STUDY XI

Study Title : Acute toxicity test with methyl isothiocyanate
Testing Facility : Matsumoto Dental College, Japan
Final Report No. : Unspecified
Final Report Date : 1974

003991

Groups of 10 mice (dd strains) per sex each received single subcutaneous injection of methyl isothiocyanate dissolved in olive oil at 58, 70, 83, 100, and 120 mg/kg. The animals were observed daily for clinical signs and mortality for 1 week after treatment. The LD50 was determined by the Litchfield-Wilcoxon with probit analysis methods.

Results:

Decreased motor activity was noted in all treated animals. Tonic convulsion, salivation, and lacrimation followed by death were observed in the higher dosage groups. Death occurred within 3 days post-treatment. For the groups receiving 58, 70, 83, 100, and 120 mg/kg, the mortality rate was respectively 0, 40, 60, 80, and 100% for males and 0, 20, 30, 80, and 100% for females.

Discussion and Conclusions:

The reported subcutaneous LD50s are 75 mg/kg and 89 mg/kg for male and female mice, respectively.

Although all reported deaths occurred within 3 days, observation of the animals should be conducted for at least 14 days post-treatment. Initial and final body weight must be submitted.

Core Classification: Supplementary Data

This study can be upgraded to Core Minimum Data if body weight data are submitted for evaluation.

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STUDY XII

Study title : Acute toxicity test of methylisothiocyanate
Intraperitoneal toxicity

003991

Testing Facility : Matsumoto Dental College
Final Report No. : Unspecified
Final Report Date : Unspecified

Methyl isothiocyanate (96% purity) dissolved in olive oil was injected intraperitoneally to 10 groups of 10 rats (Donryu strains) per sex each. The dosage levels administered were 40, 48, 58, 69, and 83 mg/kg in a constant volume of 0.1 ml/100 grams of body weight. Clinical observations and mortality were recorded for one week. The LD50s were determined by the Litchfield-Wilcoxon with probit analysis methods.

Results:

Decreased motor activity, tonic convulsion, and salivation followed by death were the reported toxic signs. All deaths occurred within 24 hours and all animals that survived were cleared of clinical signs by 3 days post-treatment.

The mortality rate for the groups receiving 40, 48, 58, 69, and 83 mg/kg was 0, 20, 70, 90, and 100% for males and 0, 30, 50, 90, and 100% for females.

Discussion and Conclusions :

The calculated intraperitoneal LD50s were reported as 54 mg/kg for males and 56 mg/kg for females.

The observation period should extend to 14 days post-treatment. Body weight data were not available.

Core Classification: Supplementary Data

This study may be upgraded to Core Minimum Data if body weight data are submitted.

003991

STUDY XIII

Study title : Acute toxicity test with methylisothiocyanate
Intraperitoneal toxicity
Testing Facility : Matsumoto Dental College
Final Report No. : Unspecified
Final Report Date : 1974

The acute intraperitoneal LD50 of methylisothiocyanate in olive oil was investigated in 10 groups of 10 mice (dd strains) per sex each. Animals received either 58, 70, 83, 100, or 120 mg/kg in a constant volume of 0.1 ml/20 grams of body weight. Determinations of the LD50 were made by using the Litchfield Wilcoxon with probit analysis methods.

Results:

Salivation, lacrimation, decreased motor activity, and tonic convulsion followed by death were noted in the treated animals. Deaths occurred within 48 hours post-treatment.

The mortality rate for the groups receiving 58, 70, 83, 100, and 120 mg/kg' were respectively 0, 40, 60, 80, and 100% for males and 0, 20, 30, 80, and 100% for females.

Discussion and Conclusions:

The intraperitoneal LD50 for male and female mice were respectively 82 and 89 mg/kg.

Observation for 14 days post-treatment is required for an acute study and body weight data are requested to verify the dosage levels used.

Core Classification: Supplementary Data

This study may be upgraded to Core Minimum Data if body weight data are available.

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