File

DP Barcode : D178840,
D178841, D178881, ~~D169296~~,
~~D169589, D170681~~
PC Code No : 077101
EEB Out : 7/21/92

To: Linda Deluise
Product Manager 52
Special Review and Reregistration Division (H7508W)

From: Douglas J. Urban, Acting Chief
Ecological Effects Branch/EFED (H7507C)

Attached, please find the EEB review of...

Reg./File # : 077101-001913
Chemical Name : Trichloromelamine
Type Product : algaecide/bacteriocide/microbiocide
Product Name :
Company Name : The Drackett Co., D.B.K. Inc., U.S. Army Natick RD&E Center
Purpose : Phase IV Review

Action Code : 604 Date Due : 8/11/92
Reviewer : Tracy Perry

EEB Guideline/MRID Summary Table: The review in this package contains an evaluation of the following:

GDLN NO	MRID NO	CAT	GDLN NO	MRID NO	CAT	GDLN NO	MRID NO	CAT
71-1(A)	42250801	P Y	72-2(A)			72-7(A)		
71-1(B)			72-2(B)			72-7(B)		
71-2(A)	42280801	P Y	72-3(A)			122-1(A)		
71-2(B)	42247401	P Y	72-3(B)			122-1(B)		
71-3			72-3(C)			122-2		
71-4(A)			72-3(D)			123-1(A)		
71-4(B)			72-3(E)			123-1(B)		
71-5(A)			72-3(F)			123-2		
71-5(B)			72-4(A)			124-1		
72-1(A)	41934901	P Y	72-4(B)			124-2		
72-1(B)			72-5			141-1		
72-1(C)			72-6			141-2		
72-1(D)						141-5		

Y=Acceptable (Study satisfied Guideline)/Concur
P=Partial (Study partially fulfilled Guideline but additional information is needed

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

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

JUL 21 1992

MEMORANDUM

SUBJECT: Trichloromelamine: Phase IV Review.

FROM: Douglas Urban, Acting Branch Chief
Ecological Effects Branch
Environmental Fate and Effects Division (H7507C) 7/21/92

TO: Linda Deluise, PM 52
Reregistration Branch
Special Review and Reregistration Division (H7508W)

EEB has completed the Phase IV Review for the List C chemical Trichloromelamine. According to the Phase 2 response of The Drackett Company (attached), only the indoor food and indoor non-food use groups are being retained. Therefore, four acute toxicity studies are required: 71-1(a) Acute Avian Oral - Quail; 71-2(a) Acute Avian Dietary - Quail; 72-1(a) Acute Fish Toxicity - Rainbow Trout; 72-2(a) Acute Aquatic Invertebrate Toxicity - Daphnia.

The following studies were included in this submission:

Fletcher, D.W. and C.A. Pedersen. 1988. Trichloromelamine: 21-Day Acute Dietary LD_{50} Study in Bobwhite Quail. Performed by Bio-Life Associates. Ltd., Neillsville, WI. Submitted by the U.S. Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. MRID No. 422508-01.

Fletcher, D.W. and C.A. Pedersen. 1988. Trichloromelamine: 8-Day Acute Dietary LC_{50} Study in Mallard ducklings. Performed by Bio-Life Associates. Ltd., Neillsville, WI. Submitted by the U.S. Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. MRID No. 4223474-01.

Fletcher, D.W. and C.A. Pedersen. 1988. Trichloromelamine: 8-Day Acute Dietary LC_{50} Study in Bobwhite Quail. Performed by Bio-Life Associates. Ltd., Neillsville, WI. Submitted by the U.S. Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. MRID No. 422808-01.

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Bowman, J. 1986. Acute Toxicity of Trichloromelamine Bluegill Sunfish (*Lepomis macrochirus*). Performed by Analytical Bio-Chemistry Laboratories, Inc., Columbia MO. Submitted by the U.S. Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. MRID No. 419349-01.

EEB has reviewed these studies and found them to be supplemental as information was missing on the percent active ingredient of the test material. These studies may be upgraded to core and will fulfill guideline requirements upon the submission of this information.

All applicable data requirements for tricloromelamine and their statuses can be found in the attached table. If you have any questions, please contact Tracy Perry at 305-6451 or Henry Craven at 305-5320.

Date: 07/20/92
Case No: 813180
Chemical No: 077101

PHASE IV
DATA REQUIREMENTS FOR
ECOLOGICAL EFFECTS BRANCH

Data Requirements	Composition[1]	Use Pattern[2]	Does EPA Have Data To Satisfy This Requirement? (Yes, No)	Bibliographic Citation	Must Additional Data Be Submitted under FIFRA3(c)(2)(B)?
6 Basic Studies in Bold					
71-1(a) Acute Avian Oral, Quail/Duck	(TGAI)	L,M	YES	42250801	~~YES~~[3] NO TLP
71-1(b) Acute Avian Oral, Quail/Duck	(TEP)	-	-	-	-
71-2(a) Acute Avian Diet, Quail	(TGAI)	L,M	YES	42280801	~~YES~~[3] NO 2/23/94
71-2(b) Acute Avian Diet, Duck	(TGAI)	L,M	YES	42247401	~~YES~~[3] NO TLP
71-3 Wild Mammal Toxicity	(TGAI)	-	-	-	-
71-4(a) Avian Reproduction Quail	(TGAI)	-	-	-	-
71-4(b) Avian Reproduction Duck	(TGAI)	-	-	-	-
71-5(a) Simulated Terrestrial Field Study	(TEP)	-	-	-	-
71-5(b) Actual Terrestrial Field Study	(TEP)	-	-	-	-
72-1(a) Acute Fish Toxicity Bluegill	(TGAI)	L,M	YES	41934901	~~YES~~[3] NO TLP
72-1(b) Acute Fish Toxicity Bluegill	(TEP)	-	-	-	-
72-1(c) Acute Fish Toxicity Rainbow Trout	(TGAI)	L,M	YES	42010601	~~YES~~[3] NO TLP
72-1(d) Acute Fish Toxicity Rainbow Trout	(TEP)	-	-	-	-
72-2(a) Acute Aquatic Invertebrate Toxicity	(TGAI)	L,M	YES	42020801	~~YES~~[3] NO TLP
72-2(b) Acute Aquatic Invertebrate Toxicity	(TEP)	-	-	-	-
72-3(a) Acute Estu/Mari Tox Fish	(TGAI)	-	-	-	-
72-3(b) Acute Estu/Mari Tox Mollusk	(TGAI)	-	-	-	-
72-3(c) Acute Estu.Mari Tox Shrimp	(TGAI)	-	-	-	-

* In Bibliographic Citation column indicates study may be upgradeable

1.Composition: TGAI = Technical grade of the active ingredient; PAIRA = Pure active ingredient, radiolabeled; TEP = Typical end-use product

2.Use Patterns: A = Terrestrial Food Crop; B = Terrestrial Feed Crop; C = Terrestrial Non-Food Crop; D = Aquatic Food Crop; E = Aquatic Non-Food Outdoor; F = Aquatic Non-Food Industrial; G = Aquatic Non-Food Residential; H = Greenhouse Food Crop; I = Greenhouse Non-Food Crop; J = Forestry; K = Outdoor Residential; L = Indoor Food; M = Indoor Non-Food; N = Indoor Medical; O = Indoor Residential; Z = Use Group for Site 00000

3. This study may be upgraded to core upon submission of percent active ingredient of the test material.

4. This study is not required to support indoor food/non-food use patterns.

DATA EVALUATION RECORD

1. **CHEMICAL:** Trichloromelamine.
Shaughnessey No. 077101.

93.8% purity (TLP) 5/13/94

2. **TEST MATERIAL:** Trichloromelamine; Lot No. 2342; purity not reported; a white powder.

3. **STUDY TYPE:** Avian Dietary LC_{50} Test. Species Tested: Bobwhite quail (*Colinus virginianus*).

4. **CITATION:** Fletcher, D.W. and C.A. Pedersen. 1988. Trichloromelamine: 8-Day Acute Dietary LC_{50} Study in Bobwhite Quail. Laboratory Study ID - BLAL No. 88 QC 110. Study performed by Bio-Life Associates, Ltd., Neillsville, WI. Submitted by U.S. Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. EPA MRID No. 422808-01.

5. **REVIEWED BY:**

Carolyn F. Poppell, Sc.M.
Senior Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: for Carolyn Poppell
Date: 7/10/92

6. **APPROVED BY:**

Mark Mossler, M.S.
Associate Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature:
Date: 7/10/92

Henry T. Craven, M.S.
Supervisor, EEB/EFED
USEPA

Signature: Henry T. Craven
Date: Tracy L. Perry 7/20/92

7. **CONCLUSIONS:** This study is scientifically sound, but does not fulfill the requirements for an avian dietary LC_{50} test since the purity of the test substance was not reported. With an LC_{50} estimated to be greater than 5000 ppm (based on nominal concentrations), trichloromelamine is classified as practically non-toxic to bobwhite quail. The NOEC was 5000 ppm.

8. **RECOMMENDATIONS:** N/A.

upgraded to Core

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9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. **MATERIALS AND METHODS:**

A. **Test Animals:** The birds used in the study were 10-day old bobwhite quail (*Colinus virginianus*). Birds were hatched at Bio-Life Associates, Ltd., from eggs purchased from Oak Ridge Game Farm, Gravette, AR. The birds could not be differentiated by sex. All birds were phenotypically indistinguishable from wild birds and from the same hatch. Birds were acclimated to the facilities for 10 days prior to initiation of the study. Forty-eight deaths were recorded during the acclimation period. Thirty-two of these deaths occurred during the first 72 hours of the acclimation period. All other birds were normal and active during the quarantine period. Prior to initiation of the project, all birds were evaluated for suitability for testing.

B. **Test System:** Birds were housed indoors in wire pens. Pen dimensions were 45.7 cm x 61.0 cm x 45.7 cm. Fluorescent light was provided 24 hours per day. The minimum and maximum temperature and humidity of the room were recorded once daily throughout the study. Room temperatures ranged from 94°F to 104°F with relative humidity between 35% and 58% during the 8-day project.

C. **Dosage:** Eight-day dietary LC_{50} test. Nominal dietary concentrations selected for the study were 312, 625, 1250, 2500, and 5000 parts per million (ppm). Dietary concentrations were not adjusted for purity of the test substance. Therefore, all dietary concentrations and the LC_{50} value are reported as parts per million of the test substance as received.

D. **Design:** Arbitrary selections were made from the entire population of birds. Groups of ten birds were then assigned to each of five control groups and five treatment groups. All birds were fed Purina® Game Bird Startena during the quarantine and study periods. Food and water were supplied *ad libitum* during the study period.

The highest concentration test diet was prepared by mixing a weighed amount of test compound (65 g) with untreated diet (12.935 kg) three days before test initiation. Each successively lower test level diet was

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prepared by mixing equal amounts of stock diet with the next higher test level diet. The birds were fed the appropriate dietary concentrations for five days, and then given untreated food for three days.

Samples of the diet were collected immediately after preparation. Stability samples were left in the test room during the five day exposure period. The samples were frozen after collection and sent to the sponsor for confirmation of dietary levels.

Observations were made daily for mortalities, signs of toxicity, abundance of feed and water, and feed spillage. Birds were group-weighed at the beginning of the treatment period (Day 1), and at the conclusion of the study on Day 8. Group food consumption was determined for the five-day exposure period (Days 0-5), and at the end of the three-day recovery period (Days 6-8).

At the termination of the study, gross pathological examinations were conducted on four surviving birds from each of the control and treatment groups.

E. **Statistics:** An estimation of an LC_{50} was made by a visual inspection of the data.

12. **REPORTED RESULTS:** There were no mortalities in any of the control or treatment groups during the study. All birds were normal in appearance and behavior throughout the study.

Body weights and food consumption values in the test groups were comparable to those of the control groups during the test and recovery periods (Table 1).

Gross pathological examinations of four arbitrarily selected birds from each control and treatment group revealed no abnormal pathological findings.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** No deaths were recorded, and no abnormal behavioral reactions or systemic signs of toxicity were noted during the study. The 8-day acute dietary LC_{50} of trichloromelamine was determined to be greater than 5000 ppm. The no-observed-effect concentration (NOEC) was determined to be 5000 ppm.

The report stated that the study was examined for conformance with EPA Good Laboratory Practice regulations,

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and was signed by the study director from Bio-Life Associates, Ltd., the sponsor, and the submitter. A Quality Assurance statement was included in the report.

14. **REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:**

A. **Test Procedure:** The test procedures were in accordance with Subdivision E, ASTM, and SEP guidelines except for the following deviations:

Birds were arbitrarily assigned to control and test groups. The guidelines specify that birds must be randomly assigned to pens.

Body weights were measured by group. Individual body weights should have been measured.

The pen dimensions (45.7 cm x 61 cm = 2788 cm^2) were smaller than the recommended dimensions (35 cm x 100 cm = 3500 cm^2) for bobwhite quail chicks.

Brooder temperatures are not specified. Maximum room temperatures recorded during the study (ranging from 99°F to 104°F) exceed the recommended maximum of 81°F (27°C).

The concentration of the test substance in the diet was not confirmed by chemical analysis. These analyses are recommended, but not required, by the guidelines.

The body weight gain data was not presented separately for the exposure and observation periods.

B. **Statistical Analysis:** Due to the absence of mortality, the LC_{50} could not be calculated. The LC_{50} is estimated based on visual observation of the data.

C. **Discussion/Results:** Although there was 12% mortality in the test birds during acclimation, no control birds died during the study. This indicated that the test birds were suitable for testing by initiation.

This study is scientifically sound, but does not meet the requirements for an avian dietary LC_{50} test since the purity of the test material was not reported.

The dietary LC_{50} of trichloromelamine in bobwhite quail was estimated to be greater than 5000 ppm (based on nominal concentrations). This value classifies

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trichloromelamine as practically non-toxic to bobwhite quail. The NOEC was 5000 ppm (based on nominal concentrations).

D. **Adequacy of the Study:**

(1) **Classification:** Supplemental.

(2) **Rationale:** The purity of the test substance was not reported.

(3) **Repairability:** Yes, this study can be upgraded to "core" pending the submission and evaluation of the purity data.

15. **COMPLETION OF ONE-LINER:** Yes; July 8, 1992.

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RIN 1281-98

Page 11 is not included in this copy.

Pages _____ through _____ are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

DATA EVALUATION RECORD

1. **CHEMICAL:** Trichloromelamine.
Shaughnessey No. 077101

93-8% purity
TLP 2/23/94

2. **TEST MATERIAL:** Trichloromelamine; Lot No. 2342; purity not reported; a white powder.

3. **STUDY TYPE:** Avian Single Dose Oral LD_{50} Test. Species Tested: Bobwhite quail (*Colinus virginianus*).

4. **CITATION:** Fletcher, D.W. and C.A. Pedersen. 1988. Trichloromelamine: 21-Day Acute Dietary LD_{50} Study in Bobwhite Quail. Laboratory Study ID - BLAL No. 88 QD 109. Study performed by Bio-Life Associates, Ltd., Neillsville, WI. Submitted by U.S. Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. EPA MRID No. 422508-01.

5. **REVIEWED BY:**

Carolyn F. Poppell, Sc.M.
Senior Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: Mark Mossler for Carolyn Poppell
Date: 7/10/92

6. **APPROVED BY:**

Mark Mossler, M.S.
Associate Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: Mark Mossler
Date: 7/10/92

Henry T. Craven, M.S.
Supervisor, EEB/EFED
USEPA

Signature: Henry T. Craven
Date: Tracy L. Perry 7/20/92

7. **CONCLUSIONS:** This study is scientifically sound, but does not fulfill the requirements for an avian oral LD_{50} test since the purity of the test material was not reported. With an LD_{50} of greater than 2150 mg/kg (based on nominal concentrations), trichloromelamine is considered to be practically non-toxic to bobwhite quail. The NOEL for trichloromelamine could not be determined due to signs of toxicity at the lowest treatment level.

8. **RECOMMENDATIONS:** N/A.

upgraded to core
2/23/94

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9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. **MATERIALS AND METHODS:**

A. **Test Animals:** The birds used in the study were adult bobwhite quail (*Colinus virginianus*) approximately 23 weeks of age. The birds were obtained from Oak Ridge Game Farm, Gravette, Arkansas at approximately 16 weeks of age. All birds were phenotypically indistinguishable from wild birds. Birds were acclimated to test facilities for 48 days prior to test initiation. Four of the 269 birds died during the quarantine period. All remaining birds were normal and active throughout the quarantine period.

B. **Test System:** All birds were housed indoors in steel wire mesh pens maintained over galvanized steel pans. Pen dimensions were 53.3 cm x 45.7 cm x 38.1 cm. Fluorescent lights provided 8 hours of light per day. The temperature and the relative humidity of the animal room were recorded once daily. The temperature during the study ranged from 67°F to 87°F with relative humidity between 56% and 86%.

C. **Dosage:** Twenty-one-day single dose oral LD_{50} test. Based upon an initial range-finding study, nominal dosages selected for the study were 1470 and 2150 mg/kg of body weight. The dietary concentrations were not adjusted for purity of the test substance. Therefore, the dietary concentrations and the LD_{50} are reported for the test substance as received.

D. **Design:** Groups of ten birds (five males and five females) were randomly allocated to each of two treatment groups and one control group. Water was available at all times and food was offered *ad libitum* with the exception of a fasting period of approximately 19 hours prior to dosing. The birds were fed Purina® Duck Grower throughout the quarantine and study periods. The test substance was ground with a mortar and pestle and administered to each bird via one gelatin capsule. Control birds received an empty gelatin capsule.

Each bird was individually weighed at test initiation (Day 1) and on test days 3, 7, 14, and 21. Group feed consumption values were recorded on test days 3, 7, 14, and 21. Daily inspections were made for mortalities,

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clinical signs of toxicity, abundance of feed and water, and feed spillage.

All birds that died during the study were subjected to gross pathological examinations. At the conclusion of the study, gross pathological examinations were also performed on four arbitrarily selected birds (two male and two female) from each of the control and test groups.

E. **Statistics:** Due to the pattern of mortality in the study, the LD_{50} was not calculated.

12. **REPORTED RESULTS:** There were no mortalities in the control group or in the 1470 mg/kg dosage group. Three mortalities occurred in the 2150 mg/kg dosage group (Table 2, attached).

No clinical signs of toxicity were noted in the control group. The first two deaths in the 2150 mg/kg test group occurred on Day 1 of the study. Diarrhea was noted in both test groups at the end of Day 1. Droppings were normal in appearance by the end of Day 3 in the 1470 mg/kg group and by the end of Day 7 in the 2150 mg/kg test group. The third death in the 2150 mg/kg dosage group occurred on Day 10 of the study. No other signs of toxicity were noted throughout the study.

A statistically significant depression in body weights was noted on Day 3 in the 2150 mg/kg dosage group (Table 3, attached). Severe food avoidance was noted in this group during the first three test days. A significant increase in body weights in the 1470 mg/kg group was noted on Day 21. No other statistically significant differences were noted with regard to body weights.

Food avoidance was noted in both test groups during the first three test days (Table 3, attached). Thereafter, food consumption values were comparable to the control group.

Gross pathological examinations revealed no abnormal findings in the two birds that died on Day 1 of the study. The bird in the higher dosage group which died on Day 10 had a friable liver. Gross pathological examinations of 12 arbitrarily selected survivors at study termination revealed no abnormal pathological findings.

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13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** The acute oral LD_{50} of trichloromelamine was determined to be greater than 2150 mg/kg of body weight. The no-observed-effect level (NOEL) was determined to be less than 1470 mg/kg of body weight.

The report stated that the study was conducted in conformance with EPA Good Laboratory Practice regulations. The statement was signed by the study director at Bio-Life Associates, Ltd., the sponsor, and the submitter. A Quality Assurance statement was included in the report.

14. **REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:**

A. **Test Procedure:** The test procedures were in accordance with Subdivision E and SEP guidelines with the exception that the purity of the test substance was not reported.

B. **Statistical Analysis:** The acute oral LD_{50} could not be calculated and is assumed to be greater than 2150 mg/kg of body weight.

C. **Discussion/Results:** With an LD_{50} of greater than 2150 mg/kg (based on nominal concentrations), the test material is considered to be practically non-toxic to bobwhite quail. The NOEL was not established due to signs of toxicity (reduced feed consumption and diarrhea) at the lowest treatment level.

This study is scientifically sound, but does not meet the requirements for an avian oral LD_{50} test since the purity of the test material was not reported.

D. **Adequacy of the Study:**

(1) **Classification:** ~~Supplemental~~ Core.

(2) **Rationale:** The purity of the test material was not reported.

(3) **Repairability:** Yes, this study can be upgraded to "core" pending the submission and evaluation of the purity data.

15. **COMPLETION OF ONE-LINER:** Yes; July 7, 1992.

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RIN 1281-98

Page _____ is not included in this copy.

Pages 16 through 17 are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

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_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

DATA EVALUATION RECORD

1. **CHEMICAL:** Trichloromelamine.
Shaughnessey No. 077101.

93.8% purity
TLP 2/23/94

2. **TEST MATERIAL:** Trichloromelamine; Lot No. 2342; purity not reported; a white powder.

3. **STUDY TYPE:** Avian Dietary LC_{50} Test. Species Tested: Mallard duck (*Anas platyrhynchos*).

4. **CITATION:** Fletcher, D.W. and C.A. Pedersen. 1988. Trichloromelamine: 8-Day Acute Dietary LC_{50} Study in Mallard Ducklings. Laboratory Study ID - BLAL No. 88 DC 109. Study performed by Bio-Life Associates, Ltd., Neillsville, WI. Submitted by U.S. Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. EPA MRID No. 422474-01.

5. **REVIEWED BY:**

Carolyn F. Poppell, Sc.M.
Senior Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: Mark Mossler for Carolyn Poppell
Date: 7/9/92

6. **APPROVED BY:**

Mark Mossler, M.S.
Associate Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: Mark Mossler
Date: 7/9/92

Henry T. Craven, M.S.
Supervisor, EEB/EFED
USEPA

Signature: Henry T. Craven
Date: Tracy L. Perry 7/20/92

7. **CONCLUSIONS:** This study is scientifically sound but does not fulfill the requirements for an avian dietary LC_{50} test since the purity of the test substance was not reported. With an LC_{50} estimated to be greater than 5000 ppm (nominal concentration), trichloromelamine is classified as practically non-toxic to mallard ducklings. The NOEC was 2500 ppm.

8. **RECOMMENDATIONS:** N/A.

upgraded to core
TLP 2/23/94

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9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. **MATERIALS AND METHODS:**

A. **Test Animals:** The birds used in the study were 5-day old mallard ducklings (*Anas platyrhynchos*), obtained at two days of age from Whistling Wings, Inc., Hanover, IL. The birds could not be differentiated by sex. All birds were phenotypically indistinguishable from wild birds. Birds were acclimated to the facilities for 3 days prior to initiation of the study. Seven out of 437 birds died during the quarantine period. All other birds were normal and active during this period. Prior to initiation of the project, all birds were evaluated to determine suitability for testing.

B. **Test System:** Birds were housed indoors in steel wire pens maintained over concrete. Pen dimensions were 45.7 cm x 61 cm x 45.7 cm. Fluorescent light was provided 24 hours per day. The minimum and maximum temperature and humidity of the room were recorded once daily throughout the study. The room temperature ranged from 74°F to 82°F during the study. Relative humidity ranged between 67% and 91%.

C. **Dosage:** Eight-day dietary LC_{50} test. Nominal dietary concentrations selected for the study were 312, 625, 1250, 2500, and 5000 parts per million (ppm), based on nominal concentrations. The dietary concentrations were not adjusted for purity of the test substance. Therefore, the dietary concentrations and the LC_{50} are reported for the test substance as received.

D. **Design:** Birds were not formally randomized, but rather arbitrary selections were made from the entire population of birds. Ten birds were assigned to each of five control groups and five treatment groups. All birds were fed Purina® Game Bird Startena during the quarantine and study periods. Food and water were supplied *ad libitum* during the study period.

The highest concentration test diet was prepared by mixing a weighed amount of test compound (65 g) with untreated diet (12.935 kg) three days before test initiation. Lower concentration diets were prepared by serial dilution of the feed. The birds were fed the

appropriate dietary concentrations for five days, and then given untreated food for three days.

Samples of the diet were collected immediately after preparation. Stability samples were left in the test room during the five day exposure period. The samples were frozen after collection.

Observations were made daily for mortalities, signs of toxicity, abundance of feed and water, and feed spillage. Birds were group-weighed at the beginning of the treatment period (Day 1), and at the conclusion of the study on Day 8. Group food consumption was determined for the five-day exposure period (Days 0-5), and at the end of the three-day observation period (Days 6-8).

At the conclusion of the study, gross pathological examinations were conducted on the two birds that died during the investigation and on four arbitrarily selected surviving birds from each control and treatment group.

E. **Statistics:** An estimation of the LC_{50} was made by visual inspection of the data.

12. **REPORTED RESULTS:** There were no mortalities in any of the control groups, or in the 625, 1250, or 2500 ppm test groups. One mortality was reported in each of the 312 and 5000 ppm groups on days 6 and 2, respectively (Table 1, attached).

All birds in the control groups were normal in appearance and behavior throughout the study. The mortality in the 5000 ppm treatment group occurred at the end of Day 2. Near the end of Day 4, one bird at this treatment level was weak in appearance, and smaller than its penmates. On Day 5, one bird in the 312 ppm group also appeared weak. The single mortality in this treatment group occurred on Day 6 of the study. The study director concluded that the mortality which occurred in the lowest treatment level was not treatment-related. No other signs of toxicity or behavioral changes were observed during the study.

Body weights and feed consumption in test groups were comparable to those in the control groups (Table 2, attached).

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Gross pathological examinations of the two test birds that died during the study and of birds examined at termination revealed no abnormal pathological findings.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** The 8-day acute dietary LC_{50} of trichloromelamine in mallard ducklings was determined to be in excess of 5000 ppm, based on nominal concentrations. The no-observed-effect concentration (NOEC) was determined to be 2500 ppm.

The report stated that the study was examined for conformance with EPA Good Laboratory Practice regulations, and was signed by the study director from Bio-Life Associates, Ltd., the sponsor, and the submitter. A Quality Assurance statement was included in the report.

14. **REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:**

A. **Test Procedure:** The test procedures were in accordance with Subdivision E, ASTM, and SEP guidelines except for the following deviation:

Birds were assigned to test groups by arbitrary selection rather than statistically random assignment.

The pen dimensions (45.7 cm x 61 cm = 2788 cm^2) were smaller than the recommended dimensions (70 cm x 100 cm = 7000 cm^2)

The purity of the test substance was not reported.

Body weights were measured by group. Individual body weights should have been measured.

The maximum relative humidity reported in the study area (91%) exceeds the recommended maximum of 80%.

The concentration of the test substance in the diet was not confirmed by chemical analyses. These analyses are recommended but not required.

The body weight gain data was not presented separately for the exposure and observation periods.

B. **Statistical Analysis:** Due to the pattern of mortality, the LC_{50} could not be calculated. The LC_{50} is estimated based on visual observation of the data.

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C. **Discussion/Results:** The study is scientifically sound, but has several deviations from the guidelines (listed in Section 14-A). The most serious deviation is that the purity of the test material was not reported.

The dietary LC_{50} of trichloromelamine in mallard ducklings was estimated to be greater than 5000 ppm (based on nominal concentrations). This value classifies trichloromelamine as practically non-toxic to mallard ducklings. The NOEC was 2500 ppm.

The author's conclusion of no apparent effects on body weight change or food consumption is accepted after a visual inspection of the data in Table 2.

The authors also concluded that the single mortality which occurred at the 312 ppm dosage level was not treatment-related. The reviewer concurs with this conclusion, based on the pattern of mortality at higher dosage levels. The only other mortality during the study occurred at the 5000 ppm concentration, the highest dosage tested.

D. **Adequacy of the Study:**

(1) **Classification:** ~~Supplemental~~. Core (TLP) 2/23/94

(2) **Rationale:** The purity of the test substance was not reported.

(3) **Repairability:** Yes, this study can be upgraded to "core" pending the submission and evaluation of the purity data.

15. **COMPLETION OF ONE-LINER:** Yes; July 7, 1992.

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RIN 1281-98

Page _____ is not included in this copy.

Pages 23 through 24 are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

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DATA EVALUATION RECORD

1. **CHEMICAL:** Trichloromelamine.
Shaughnessey No. 077101. 95.9% purity

2. **TEST MATERIAL:** Trichloromelamine; Lot No. 1933; a yellow powder.

3. **STUDY TYPE:** Freshwater Fish Static Acute Toxicity Test. Species Tested: Bluegill Sunfish (*Lepomis macrochirus*)

4. **CITATION:** Bowman, J. 1986. Acute Toxicity of Trichloromelamine to Bluegill Sunfish (*Lepomis macrochirus*). Laboratory Project ID No. 34811. Prepared by Analytical Bio-Chemistry Laboratories, Inc., Columbia, MO. Submitted by US Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD. EPA MRID No. 419349-01.

5. **REVIEWED BY:**

Mark A. Mossler, M.S.
Associate Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: Mark A. Mossler
Date: 7/6/92

6. **APPROVED BY:**

Louis M. Rifici, M.S.
Associate Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: Louis M. Rifici
Date: 7/7/92

Henry T. Craven, M.S.
Supervisor, EEB/EFED
USEPA

Signature: Henry T. Craven 7/21/92
Date: Stacy L. Perry 7/20/92

7. **CONCLUSIONS:** This study is scientifically sound but does not satisfy the guideline requirements for a static acute toxicity test. The purity of the test material was not reported. The 96-hour LC_{50} of 4.5 mg/l (based on nominal concentrations of total product) classifies trichloromelamine as moderately toxic to bluegill sunfish. The NOEC was 1.0 mg/l.

8. **RECOMMENDATIONS:** Submit data concerning the test material purity.

upgraded to core
TLP 2/23/94

9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. **MATERIALS AND METHODS:**

A. **Test Animals:** Bluegill Sunfish (*Lepomis macrochirus*) were obtained from Osage Catfisheries in Osage Beach, MO. The fish were maintained in culture tanks on a 16-hour light photoperiod for at least 2 weeks prior to testing. The fish were fed a commercially-available fish food daily with occasional supplements of newly hatched brine shrimp. The condition of the fish was monitored daily and records of disease treatments were kept. The fish were acclimated to the dilution water and test temperature and held without food for 48-96 hours prior to or during testing.

Mean weight and standard length of the control fish were 0.53 (±0.10) g and 28 (±1.7) mm. Biomass loading rate in the control was 0.36 g/l.

B. **Test System:** Vessels used in the test were 5-gallon glass containers filled with 15 l of soft reconstituted water (control) or test solution. The reconstituted water was prepared to yield a total hardness of 40-45 mg/l as $CaCO_3$, a total alkalinity of 30-35 mg/l as $CaCO_3$, and an initial pH of 7.2-7.6. The vessels were kept in a water bath set to maintain 22 ±1.0°C. The test concentrations were prepared by adding appropriate volumes of a test material stock solution [prepared in dimethylformamide (DMF)] directly to the test chambers.

C. **Dosage:** Ninety-six-hour static test. Based on preliminary tests, five nominal concentrations (1.0, 1.8, 3.2, 5.6, and 10 mg/l), a solvent (0.1 ml DMF/l) and a dilution water control were used. The concentrations made were based on total product.

D. **Design:** Ten fish were randomly added to each test chamber within 30 minutes of test solution preparation. All chambers were observed once every 24 hours for mortality and sublethal effects. Dead fish were removed from the chambers at each observation period.

Temperature, pH, and dissolved oxygen concentration (DO) were measured in the controls and selected test concentrations at 0, 48, and 96 hours.

E. **Statistics:** The 96-hour median lethal concentration (LC_{50}) and associated 95% confidence interval (C.I.) were calculated using binomial probability.

12. **REPORTED RESULTS:** The mortality of the bluegill sunfish are given in Table 3 (attached). The 96-hour LC_{50}, based on nominal concentrations, was 4.5 mg/l (95% C.I. = 3.2-5.6 mg/l). Sublethal effects (dark discoloration and/or rapid respiration) were observed in all concentrations greater than 1.0 mg/l; therefore, the no-observed-effect concentration (NOEC) was 1.0 mg/l.

At test initiation, the DO of the controls, low, medium, and high concentrations were 8.9 to 9.1 mg/l or 101-103% of saturation. After 96 hours, oxygen levels in the control, solvent control, 1.0, 3.2, and 5.6 mg/l test chambers ranged from 61 to 85% of saturation. The pH values ranged from 7.0 to 7.6 (Table 3). The temperature was 22-23°C throughout the test.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** The author presented no conclusions.

Quality Assurance and Good Laboratory Compliance Statements were included in the report, indicating that the study was conducted in accordance with EPA Good Laboratory Practice standards (40 CFR Part 160).

14. **REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:**

A. **Test Procedure:** The test procedures were generally in accordance with the SEP, except for the following:

The purity of the test material was not reported.

The acclimation period of the sunfish to the test conditions was 2 to 4 days. The SEP recommends that the acclimation period to the test conditions be at least two weeks.

The test temperature was not monitored every six hours as recommended.

Thirty-minute dawn and dusk simulation periods are recommended in the SEP. Transition periods were not used in the study.

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No length range of the fish was given. The SEP states that the longest fish should not be more than twice the length of the shortest fish.

Each selected nominal concentration was between 55% and 57% of the next highest concentration. The SEP recommends that each concentration be 60% of the next highest concentration.

The DO at test initiation ranged from 104 to 106% of saturation at 22°C. The SEP recommends that the DO be between 60 and 100% of saturation during the first 48 hours of the test.

B. **Statistical Analysis:** The reviewer used EPA's Toxanal program to calculate the LC_{50} value and obtained the same results (see attached printout).

C. **Discussion/Results:** This study is scientifically sound but does not satisfy the guideline requirements for a static acute toxicity test. The 96-hour LC_{50} of 4.5 mg/l (based on nominal concentrations of total product) classifies the test material as moderately toxic to bluegill sunfish. The NOEC can be estimated as 1.0 mg/l (based on nominal concentration of total product).

D. **Adequacy of the Study:**

(1) **Classification:** Supplemental. Core TLP 2/23/94

(2) **Rationale:** The purity of the test material was not reported.

(3) **Repairability:** Yes, this study can be upgraded to "core" pending the submission of the purity data.

15. **COMPLETION OF ONE-LINER FOR STUDY:** Yes, 7-1-92.

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MOSSLER TRICHLOROMELAMINE LEPOMIS MACROCHIRUS 7-1-92

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB.(PERCENT)
10	10	10	100	9.765625E-02
5.6	10	9	90	1.074219
3.2	10	0	0	9.765625E-02
1.8	10	0	0	9.765625E-02
1	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT 3.2 AND 5.6 CAN BE USED AS STATISTICALLY SOUND CONSERVATIVE 95 PERCENT CONFIDENCE LIMITS, BECAUSE THE ACTUAL CONFIDENCE LEVEL ASSOCIATED WITH THESE LIMITS IS GREATER THAN 95 PERCENT.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS 4.490551

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

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