

254841
RECORD NO.

1/17/90

031301
SHAUGHNESSEY NO.

REVIEW NO.

EEB REVIEW

DATE: IN 11-20-89 OUT

FILE OR REG. NO. 45639-128

PETITION OR EXP. NO.

DATE OF SUBMISSION

DATE RECEIVED BY EFED 11-13-89

RD REQUESTED COMPLETION DATE 01-13-90

EEB ESTIMATED COMPLETION DATE 01-13-90

RD ACTION CODE/TYPE OF REVIEW 660

TYPE PRODUCT(S) Fungicide

DATA ACCESSION NOS. 405831-02

PRODUCT MANAGER NO. E. Feris (21)

PRODUCT NAME(S) DCNA (Dichloran) Technical

COMPANY NAME NOR-AM Chemical Company

SUBMISSION PURPOSE Submitted in response to RS

SHAUGHNESSEY NO.	CHEMICAL AND FORMULATION	% AI
<u>031301</u>	<u>Dichoran</u>	<u>97.0</u>
<u> </u>	<u> </u>	<u> </u>
<u> </u>	<u> </u>	<u> </u>



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

MEMORANDUM

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

Subject: Review of Freshwater Invertebrate Acute Toxicity Test
(Daphnia magna) for DCNA (Dichloran Technical)

From: James W. Akerman, Chief
Ecological Effects Branch
Environmental Fate and Effects Division (H7507C)

To: Eric Feris (PM-21)
Reregistration Branch
Special Review/Reregistration Division (H7508C)

Attached is the Ecological Effects Branch review of a freshwater invertebrate acute toxicity study using Daphnia for the reregistration of DCNA.

The study is scientifically sound with a few minor deviations from standard methodologies. This study fulfills the basic requirements of current guidelines and would be acceptable for use in a risk assessment. The 48-hour EC_{50} with 95% confidence limits for the toxicity of Dichloran Technical to Daphnia magna was calculated to be 2.07 mg/l. This value places Dichloran Technical in a category of moderately toxic to freshwater invertebrates.

DATA EVALUATION RECORD

1. **CHEMICAL:** DCNA
2. **TEST MATERIAL:** Dichloran Technical (97% a.i.),
a yellow powder
3. **STUDY TYPE:** Freshwater Invertebrate Acute Toxicity Test
Species Tested: Daphnia magna
4. **STUDY IDENTIFICATION:**

Author: Hill, R.W., Moffat, A.M., and M.H.I.
Comber
Laboratory: The Brixham Laboratory
Chester Park Research Station
Saffron Walden, Essex England
Study No.: ENVIR/88/19
Study Date: 03/11/88
Submitted By: NOR-AM Chemical Company
Accession No: 40583102
5. **REVIEWED BY:**

Art Roybal
Wildlife Biologist
Ecological Effects Branch, EFED
Signature: *Art Roybal*
Date: 1-12-90
6. **APPROVED BY:**

Norman Cook
Supervisory Biologist
Ecological Effects Branch, EFED
Signature: *Norman J. Cook*
Date: 1-17-90
7. **CONCLUSIONS:** The study is scientifically sound with a few minor deviations from standard methodologies. This study fulfills the basic requirements of current guidelines and would be acceptable for use in a risk assessment. The 48-hour EC₅₀ with 95% confidence limits for the toxicity of Dichloran Technical to Daphnia magna was calculated to be 2.07 mg/l. This value places Dichloran Technical in a category of moderately toxic to freshwater invertebrates.
8. **RECOMMENDATIONS:** N/A

9. BACKGROUND:

10. DISCUSSION OF INDIVIDUAL TESTS: N/A

11. MATERIALS AND METHODS:

A. Test Animals: The test organism was the freshwater crustacean, Daphnia magna, obtained from continuous laboratory cultures. The stock cultures of Daphnia were maintained in a reconstituted water medium, identical to the test dilution water, at a temperature of $20 \pm 2^{\circ}\text{C}$, and a photoperiod of 16 hours light: 8 hours dark. The cultures were fed a defined diet of algae and yeast. Daphnia less than 24 hours old were used for testing. (From brood stock 17 days old. No symptoms of disease had been observed in this culture).

B. Preparation of Test Solution: A 10 ml stock solution of the test substance was prepared in acetone (11,200 mg/l). The test concentrations were prepared by the addition of aliquots of this stock solution to the dilution medium using microlitre syringes. The test solutions were stirred for one hour prior to use. The following nominal exposure concentrations were employed: control, solvent control, 0.32, 0.56, 1.0, 1.8, 3.2, and 5.6 mg/l dichloran technical. Apart from the control, each exposure concentration contained 500 ul/l of acetone.

C. Test Water: The water was aerated for greater than 2 hours before use. The pH of the dilution water was 8.42.

D. Test Procedure: Borosilicate glass beakers (250 ml) were used as test vessels, with four replicates per exposure concentration, each containing 200 ml of test solution. When the test solutions were at test temperature, five Daphnia were randomly added to each test vessel in <2.0 ml of dilution water, giving a total of 20 Daphnia per concentration. A random number table was used for this purpose. The temperature was maintained at $20 \pm 1^{\circ}\text{C}$ with a photoperiod of 16 hours light: 8 hours dark. The test vessels were not aerated during the course of the test.

E. Observation Of Effects: Assessments of the response of the Daphnia were made at 24 and 48 hours after commencement of the test. Each Daphnia was viewed by eye, and was defined as affected if showing no whole body movement relative to the water within a period of 15 seconds, even if movement of individual appendages was visible. Daphnia so affected were termed immobile.

F. Physical and Chemical Analysis: The dissolved oxygen concentration of the control (dilution water used to prepare the test solution) was measured prior to the start of the test. The initial pH of each test solution was determined

using the excess remaining after filling the test vessels. At the end of the test the pH and dissolved oxygen concentration of two replicates of each control and treatment were measured. The temperature of the water in an additional test vessel containing no Daphnia was measured by thermometer at 0, 24 and 48 hours, and at hourly intervals by an automatic recording system (platinum resistance sensor). The concentration of the test solution was measured at the start (excess solution) and end of the test by the method described in Appendix 2. At the end of the test one replicate of each test concentration was sampled.

F. Statistics: Statistical analysis of the concentration vs. effect data was obtained by using Stephan et al. (1978), a computerized LC₅₀ program. This program calculated the LC₅₀ and its 95% confidence limits by using the binomial, moving average and probit tests. All calculations were based on the nominal concentrations of Dichloran Technical.

12. REPORTED TEST RESULTS:

Biological Data: The number of Daphnia immobilized at each concentration, after 24 and 48 hours, are given in Table 1. The authors state, "No symptoms of toxicity were observed in this study." The numbers immobilized in the replicates were summed for each treatment and control during each time period. Where the data permitted, the 24 and 48 hour median effective concentrations and their 95% confidence limits were calculated by probit analysis using a computer program. A graph of the concentration:percentage response after 48 hours, showing the regression line fitted by the analysis, is given in Figure 1. The median effective concentration (EC₅₀) was defined as the concentration, calculated from the data obtained, resulting in 50% immobilization of the Daphnia in the time period specified. The results obtained were:

	EC ₅₀	Confidence Limits
24 hours (slope of probit = 4.2)	4.43	3.62 - 6.03 mg/l
48 hours (slope of probit = 8.6)	2.07	1.80 - 2.39 mg/l

Chemical and Physical Analysis: It was noted in this study that all test exposure concentrations appeared to be clear and no precipitation of the test material was observed. The levels of Dichloran in the exposure vessels were determined on all exposure concentrations and the Daphnia media control and solvent control by high performance liquid chromatography at 0 and 48 hours. Dissolved oxygen levels ranged from 9.0 to 9.3 mg/l and the pH values ranged from 8.28 to 8.42. The temperature recorded by thermometer at 0,

24 and 48 hours were 20.3, 20.5, and 20.3°C respectively (see Table 3). The range of temperatures recorded automatically at hourly intervals was 20.0 to 20.5°C. The concentrations of the test substance determined in the test solutions at the start and finish of the test are given in Table 2. The measured concentrations at the start of the test ranged from 91 to 110% of the nominal values. The measured concentrations at the end of the test ranged from 96 to 110% of the nominal value, therefore, the nominal values were used in this report.

13. STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:
"The 24 hour and 48 hour EC₅₀ values were determined to Daphnia magna using dicloran technical. The 48-hour EC₅₀ value was 2.07 mg/l dicloran technical based on nominal concentration. According to the Standard Evaluation Procedure this formulation would be classified as moderately toxic."(This is a direct quote including spelling, etc.)

The authors state the study was conducted following the intent of the Good Laboratory Practice Regulations.

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

- A. Test Procedure: The study is in general accordance with EPA Standard Evaluation Procedures and ASTM procedures.
- B. Statistical Analysis: The EPA Toxanal computer program was used to verify EC₅₀ values at the 24 and 48 hour intervals. EC₅₀ values obtained with the EPA Toxanal program coincide with the values calculated in this study. See attached documentation.
- C. Discussion/Results: A 48-hour EC₅₀ value of 2.07 mg/l would place Dichloran Technical in the classification of moderately toxic to Daphnia magna.
- D. Adequacy of the Study:
- (1) Classification: Core
 - (2) Rationale: Minor inconsistencies with standard methodologies, however, the deviations do not detract from the study's soundness or intent
 - (3) Repairability: N/A

15. COMPLETION OF ONE-LINER FOR STUDY: Yes, 01/12/90
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Roybal dichloran technical Daphnia 01-10-90

24-Hour

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
5.6	20	13	65	13.1588
3.2	20	6	30	5.765915
1.8	20	1	5	2.002716E-03
1	20	0	0	9.536742E-05
.56	20	0	0	9.536742E-05
.32	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 0 AND +INFINITY 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 4.412896

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
1	.8069815	4.412896	2.870203	9.605119

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
6	.2013353	1	.9975969

SLOPE = 4.183476
95 PERCENT CONFIDENCE LIMITS = 2.306334 AND 6.060619

LC50 = 4.428219
95 PERCENT CONFIDENCE LIMITS = 3.615435 AND 6.025872

LC10 = 2.201171
95 PERCENT CONFIDENCE LIMITS = 1.361506 AND 2.779418

Art dichloran technical Daphnia 01-10-90

48-Hour

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
5.6	20	20	100	9.536742E-05
3.2	20	19	95	2.002716E-03
1.8	20	6	30	5.765915
1	20	0	0	9.536742E-05
.56	20	0	0	9.536742E-05
.32	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 1 AND 3.2 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 2.107342

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
4	5.135013E-02		2.076829	1.747534

2.508648

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
7	.1975888	1	.9995357

SLOPE = 8.92993
95 PERCENT CONFIDENCE LIMITS = 4.96049 AND 12.89937

LC50 = 2.074524
95 PERCENT CONFIDENCE LIMITS = 1.804893 AND 2.390401

LC10 = 1.495203
95 PERCENT CONFIDENCE LIMITS = 1.100391 AND 1.733395

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Study/Species/Lab/ Access # _____ Chemical # a.i. _____ Results _____ Reviewer/ Date _____ Validation Status _____

14-Day Single Dose Oral LD50, LD50 = mg/kg (95% C.L.) Contr. Mort.(%) =
 Species: Slope= # Animals/Level= Age(Days)=
 Lab.: Sex =
 Acc. #: 14-Day Dose Level mg/kg/(% Mortality)
 () , () , () , () , () , ()
 Comments:

14-Day Single Dose Oral LD50, LD50 = mg/kg (95% C.L.) Contr. Mort.(%) =
 Species: Slope= # Animals/Level= Age(Days)=
 Lab.: Sex =
 Acc. #: 14-Day Dose Level mg/kg/(% Mortality)
 () , () , () , () , () , ()
 Comments:

8-Day Dietary LC50, LC50 = ppm (95% C.L.) Contr. Mort.(%) =
 Species: Slope= # Animals/Level= Age(Days)=
 Lab.: Sex =
 Acc. #: 8-Day Dose Level ppm/(% Mortality)
 () , () , () , () , () , ()
 Comments:

8-Day Dietary LC50, LC50 = ppm (95% C.L.) Contr. Mort.(%) =
 Species: Slope= # Animals/Level= Age(Days)=
 Lab.: Sex =
 Acc. #: 8-Day Dose Level ppm/(% Mortality)
 () , () , () , () , () , ()
 Comments:

96-hour LC50, LC50 = pp (95% C.L.) Contr. Mort.(%) =
 Species: Slope= # Animals/Level= Sol. Contr. Mort.(%) =
 Lab.: Temperature =
 Acc. #: 96-Hour Dose Level pp/(% Mortality)
 () , () , () , () , () , ()
 Comments:

96-hour LC50, LC50 = PP (95% C.L.) Con. Mor(%) =
 Species: Slope= # Animals/Level= Sol. Con. Mor.(%) =
 Lab.: Temp. =
 Acc. #: 96-Hour Dose Level pp/(% Mortality)
 () , () , () , () , () , ()
 Comments:

48-hour Invertebrate, LC50 = 2.07 ppm (95% C.I.) Con. Mort.(%) =
 Species: Daphnia Slope= 8.93 # Animals/Level= 20 Sol. Con. Mort.(%) =
 Lab.: Brixham Lab 77023.i Temp. = 20.3
 Acc. #: H0583102 96-Hour Dose Level pp/(% Mortality)
 .32 (0%), .56 (5%), 1.0 (9%), 1.6 (17%), 2.2 (45%), 5.6 (100%)
 Comments:

Study/Species/Lab/ Accession #	Chemical & Active	Results				Reviewer/ Date	Validation Status
		Group	Dose(ppm)	Effectd/Parameters	Mort.(%)		
Avian Reproduction, Species: _____	_____	Control	_____	_____	_____	_____	_____
Lab: _____	_____	Treatment I	_____	_____	_____	_____	_____
Acc. #: _____	_____	Treatment II	_____	_____	_____	_____	_____
		Treatment III	_____	_____	_____	_____	_____
		Study Duration:	_____				
		Comments:	_____				

Field Study(Simulated/Actual)	Species:	Group	Rate(ai/a)	Treatment Interval	Total # Treatments	Mor.(%)	Validation Status
Lab: _____	_____	Control	_____	_____	_____	_____	_____
Acc. #: _____	_____	Treatment I	_____	_____	_____	_____	_____
		Treatment II	_____	_____	_____	_____	_____
		Treatment III	_____	_____	_____	_____	_____
		Crop/Site:	_____				
		Study Duration:	_____				
		Comments:	_____				

Chronic fish,

Species _____

Lab: _____

Acc. #: _____

Concentrations Tested (pp_) = _____

MAIC = > _____ < _____ PP _____

Effectd Parameter = _____

Contr. Mort.(%) = _____ Sol. Contr. Mort.(%) = _____

Comments: _____

Chronic invertebrate,

Species _____

Lab: _____

Acc. #: _____

Concentrations Tested (pp_) = _____

MAIC => _____ < _____ PP _____

Effectd Parameter(s) = _____

Contr. Mort.(%) = _____ Sol. Contr. Mort.(%) = _____

Comments: _____

Use this form for individual studies & to submit pesticide applications.



United States Environmental Protection Agency
Office of Pesticide Programs
Washington, DC 20460

Data Review Record

Confidential Business Information - Does not contain
National Security Information (E.O. 12065)

031301

Pack Number

49820

EFED

Date Received

11-13-89

1. Product Name

Chemical Name

DCNA

2. Identifying Number	3. Record Number	4. Action Code	5. MRID/ Accession Number	6. Study Guideline or Narrative
45639-128	254841	660	40583102	72-2 A

7. Reference No.	8. Date Rec'd (EPA)	9. Prod/Review Mgr/DCI	10. PM/RM Team No.	11. Date to HED/EFED/RD/BEAD	12. Proj Return Date	13. Date Returned to RD/SRRD
	4-12-88	E.FERIS	21	11-13-89	1-13-90	

Instructions

Please review data

031301-S/0113

This Section Applies to Review of Studies Only

<p>14. Check Applicable Box</p> <p><input type="checkbox"/> Adverse 6(a)(2) Data (405)</p> <p><input type="checkbox"/> Special Review Data (870)</p> <p><input checked="" type="checkbox"/> Generic Data (Reregistration)(660)</p> <p><input type="checkbox"/> Product Specific Data (Reregistration)(655)</p>	<p>15. No. of Individual Studies Submitted</p> <p>1</p>
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<p>16. Have any of the above studies (in whole or in part) been previously submitted for review?</p> <p><input type="checkbox"/> Yes (Please identify the study(ies))</p> <p><input type="checkbox"/> No</p>	<p>17. Related Actions</p>
--	----------------------------

18.	To	Type of Review	19. Reviews Also Sent to	20. Data Review Criteria
HED		Science Analysis & Coordination	<input type="checkbox"/> SAC	<p>A. Policy Note No. 31</p> <p><input type="checkbox"/> 1 = data which meet 6(a)(2) or meet 3(c)(2)(B) flagging criteria</p> <p><input checked="" type="checkbox"/> 2 = data of particular concern from registration standard</p> <p><input type="checkbox"/> 3 = data necessary to determine tiered testing requirements</p> <p>B. Section 18</p> <p><input type="checkbox"/> 1 = data in support of section 3 in lieu of section 18</p> <p>C. Inert Ingredients</p> <p><input type="checkbox"/> 1 = data in support of continued use of List 1 inert</p>
		Toxicology/HFA	<input type="checkbox"/> TOX/HFA	
		Toxicology/IR	<input type="checkbox"/> TOX/IR	
		Dietary Exposure	<input type="checkbox"/> DEB	
		Nondietary Exposure	<input type="checkbox"/> NDE	
EFED	<input checked="" type="checkbox"/>	Ecological Effects	<input type="checkbox"/> EEB	
		Environmental Fate & Groundwater	<input type="checkbox"/> EFGWB	
SRRD		Special Review	<input type="checkbox"/> SR	
		Reregistration	<input type="checkbox"/> RER	
		Generic Chemical Support	<input type="checkbox"/> GSC	
RD		Insecticide-Rodenticide	<input type="checkbox"/> IR	
		Fungicide-Herbicide	<input type="checkbox"/> FH	
		Antimicrobial	<input type="checkbox"/> AM	
		Product Chemistry		
BEAD		Precautionary Labeling		
		Economic Analysis		
		Analytical Chemistry		
		Biological Analysis		

<input type="checkbox"/> Confidential Statement of Formula (EPA Form 8570-4) Attached (Trade Secrets)	<input type="checkbox"/> Label Attached
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11

Use this form for individual studies & to submit pesticide applications.



United States Environmental Protection Agency
Office of Pesticide Programs
Washington, DC 20460
Data Review Record
Confidential Business Information - Does not contain
National Security Information (E.O. 12065)

Pack Number

Date Received

49820

EFED

11-13-89

1. Product Name

Chemical Name

DCNA

2. Identifying Number	3. Record Number	4. Action Code	5. MRID/ Accession Number	6. Study Guideline or Narrative
45639-128	254841	660	40583102	72-2A

7. Reference No.	8. Date Rec'd (EPA)	9. Prod/Review Mgr/DCI	10. PM/RM Team No.	11. Date to HED/EFED/RD/BEAD	12. Proj Return Date	13. Date Returned to RD/SRRD
	4-12-88	E.FERIS	21	11-13-89	1-13-90	

Instructions

Please review data

031301-5/0113

This Section Applies to Review of Studies Only

14. Check Applicable Box	15. No. of Individual Studies Submitted
<input type="checkbox"/> Adverse 6(a)(2) Data (405) <input type="checkbox"/> Special Review Data (870) <input checked="" type="checkbox"/> Generic Data (Reregistration)(660) <input type="checkbox"/> Product Specific Data (Reregistration)(655)	1

16. Have any of the above studies (in whole or in part) been previously submitted for review?	17. Related Actions
<input type="checkbox"/> Yes (Please identify the study(ies)) <input type="checkbox"/> No	

18.	To	Type of Review	19. Reviews Also Sent to	20. Data Review Criteria
HED		Science Analysis & Coordination	<input type="checkbox"/> SAC <input type="checkbox"/> PC	A. Policy Note No. 31 <input type="checkbox"/> 1 = data which meet 6(a)(2) or meet 3(c)(2)(B) flagging criteria <input checked="" type="checkbox"/> 2 = data of particular concern from registration standard <input type="checkbox"/> 3 = data necessary to determine tiered testing requirements B. Section 18 <input type="checkbox"/> 1 = data in support of section 3 in lieu of section 18 C. Inert Ingredients <input type="checkbox"/> 1 = data in support of continued use of List 1 inert
		Toxicology/HFA	<input type="checkbox"/> TOX/HFA <input type="checkbox"/> PL	
		Toxicology/IR	<input type="checkbox"/> TOX/IR	
		Dietary Exposure	<input type="checkbox"/> DEB <input type="checkbox"/> EA	
EFED	<input checked="" type="checkbox"/>	Ecological Effects	<input type="checkbox"/> NDE <input type="checkbox"/> AC	
		Environmental Fate & Groundwater	<input type="checkbox"/> EEB <input type="checkbox"/> BA	
SRRD		Special Review	<input type="checkbox"/> EFGWB	
		Reregistration		
		Generic Chemical Support		
RD		Insecticide-Rodenticide	<input type="checkbox"/> SR	
		Fungicide-Herbicide	<input type="checkbox"/> RER	
		Antimicrobial	<input type="checkbox"/> GSC	
		Product Chemistry	<input type="checkbox"/> IR	
BEAD		Precautionary Labeling	<input type="checkbox"/> FH	
		Economic Analysis	<input type="checkbox"/> AM	
		Analytical Chemistry		
		Biological Analysis		

Confidential Statement of Formula (EPA Form 8570-4) Attached (Trade Secrets)

Label Attached

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