

⑤ 2-EEB-306
12-20-93
12116193
MRID No. 429490-01
IV-a-DAP

Reviewer



2001064

DATA EVALUATION RECORD

- 1. **CHEMICAL:** Proxel Press Paste (Benzisothiazolin).
Shaughnessey No. 098901.
- 2. **TEST MATERIAL:** Proxel press paste; 1,2-benzisothiazol-3(2H)-one; CAS No. 2634-33-5; purity of 76.1% w/w; a moist brown powder
- 3. **STUDY TYPE:** 72-2. Freshwater Invertebrate Acute Static Test. Species Tested: *Daphnia magna*.
- 4. **CITATION:** Kent, S.J., G.C. Roberts, and A.J. Grinell. 1993. Proxel Press Paste: Acute Toxicity to *Daphnia magna*. Project ID No. X060/B. Prepared by Brixham Environmental Laboratory, ZENECA Limited, Brixham, Devon, UK. Submitted by ZENECA Inc., Wilmington, DE. EPA MRID No. 429490-01.

5. **REVIEWED BY:**

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

Signature: *Mark A. Mossler*
Date: 12/10/93

6. **APPROVED BY:**

Rosemary Graham Mora, M.S.
Associate Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: *Rosemary Graham Mora*
Date: 12/10/93

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

Signature: *Henry T. Craven*
Date: 12/10/93

7. **CONCLUSIONS:** This study is scientifically sound and meets the guideline requirements for an acute toxicity test using *Daphnia magna* with *Daphnia magna*. Based on mean measured concentrations, the 48-hour EC₅₀ was 4.4 mg proxel press paste/l. Therefore, proxel press paste is classified as moderately toxic to *Daphnia magna*. The NOEC was 1.9 mg proxel press paste/l. *formulation*

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

9. **BACKGROUND:**

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

11. MATERIALS AND METHODS:

- A. **Test Animals:** *Daphnia magna* (<24 hours old) used in the test were taken from in-house cultures (21 ±1 days old) maintained at 20 ±1°C under a 16-hour light photoperiod. The cultures were fed a diet of *Chlorella vulgaris* and a commercially-available microencapsulated food.
- B. **Test System:** The test was conducted under static conditions. The test vessels were covered, 250-ml glass beakers containing 200 ml of solution. The test solution depth was 60 mm. The beakers were randomly arranged within the test area. The test temperature (20 ±1°C) was maintained by control of the room temperature. A 16-hour light photoperiod with 15-minute transition periods were provided.

The test dilution water was reconstituted water prepared by dissolving NaHCO₃ (192 mg/l), CaSO₄·2H₂O (120 mg/l), MgSO₄·7H₂O (245 mg/l), and KCl (8 mg/l) in glass-distilled water.

A primary stock solution was prepared in dilution water. The treatment solutions were prepared by adding appropriate amounts of the stock to dilution water.

- C. **Dosage:** Forty-eight-hour, static test. Six nominal concentrations (0.56, 1.0, 1.8, 3.2, 5.6, and 10 mg proxel press paste/l) and a dilution water control were selected for testing.
- D. **Design:** Five daphnids were added to each of four beakers per treatment or control. The number of immobilized daphnids was determined after 24 and 48 hours. Overt signs of toxicity were also noted. Daphnids were not fed during the study and solutions were not aerated.

The dissolved oxygen concentration (DO) of the dilution water was measured at test initiation and in two replicates of each treatment and control group at test termination. The pH of each test solution was measured at test initiation and in two replicates of each treatment and control group at test termination. Treatments demonstrating 100% mortality at the 24-hour period were immediately measured for pH and DO. The temperature of the water in a vessel containing no daphnids was recorded manually daily and hourly.

Samples collected at 0 and 48 hours were analyzed for 1,2-benzisothiazol-3(2H)-one using high pressure liquid chromatography.

E. Statistics: The 48-hour median effective concentration (EC_{50}) and confidence interval (C.I.) were calculated using the moving average angle method.

12. REPORTED RESULTS: The mean measured concentrations were 0.58, 1.1, 1.9, 3.3, 5.8, and 10 mg proxel press paste/l (Table 1, attached). These values represent 100-110% of nominal concentrations.

There was no mortality in the control or three lowest-concentration treatment groups (Table 2, attached). The 48-hour EC_{50} value was 4.3 mg proxel press paste/l (95% C.I.= 3.7-5.3 mg proxel press paste/l). The no-observed-effect concentration (NOEC) was 1.9 mg proxel press paste/l.

During the test, the DO was 9.0-9.3 mg/l. The pH was 8.31-8.50 and the temperature ranged from 19.3 to 20.5°C.

13. STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:
No conclusions were presented.

Good Laboratory Practice and Quality Assurance statements were included in the report stating compliance with OECD principles and, indirectly, with 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 report did not state whether the daphnids were randomly placed in the test beakers. Test daphnids must be randomly distributed to the test vessels.

B. Statistical Analysis: The reviewer assumes that corrections were made to the analytical results so that results are in terms of the total product (proxel press paste). Using mean measured concentration of proxel press paste data, the reviewer used EPA's Toxanal program to determine the 48-hour EC_{50} and obtained similar results as the authors (see attached printout). However, since the probit model supplied a slope and narrower confidence interval, the results from this model will be reported. The 48-hour EC_{50} value was 4.4 mg proxel press paste/l (95% C.I.= 3.9-4.9 mg proxel

press paste/l). The slope of the probit curve was 13.4.

- C. Discussion/Results: This study is scientifically sound and meets the guideline requirements for an acute toxicity test using *Daphnia magna*. Based on mean measured concentrations, the 48-hour EC₅₀ was 4.4 mg proxel press paste/l. Therefore, proxel press paste is classified as moderately toxic to *Daphnia magna*. The NOEC was 1.9 mg proxel press paste/l.
This test may not reflect technical level active ingredient toxicity however.
- D. Adequacy of the Study:
- (1) Classification: Core. *for paste formulation*
 - (2) Rationale: N/A.
 - (3) Repairability: N/A.

15. COMPLETION OF ONE-LINER FOR STUDY: Yes, 12-7-93.

TABLE 1

PROXEL PRESS PASTE: ACUTE TOXICITY TO *Daphnia magna*CONCENTRATIONS OF PROXEL PRESS PASTE IN TEST VESSELS
DETERMINED BY QUANTIFICATION OF 1,2-BENZISOTHAZOL-3-(2H)-ONE (BIT)

Sponsor	ZENECA Biocides
Test substance:	Proxel press paste
Test organism:	<i>Daphnia magna</i>
Test water:	Reconstituted water medium (Appendix 1)

Nominal concn. of Proxel press paste (mg/l)	Measured concn. of Proxel press paste (mg/l)		Mean measured concn. of Proxel press paste	
	0 h (22.2.93)	48 h (24.2.93)	(mg/l)	% of nominal
10	10	10	10	100
5.6	5.8	5.8	5.8	104
3.2	3.4	3.2	3.3	103
1.8	1.9	1.8	1.9	106
1.0	1.1*	1.1*	1.1	110
0.56	0.59	0.56	0.58	104
Dilution water control	<0.0068	<0.0065	<0.0068	-

* Mean of triplicate injections:

0 hour: 1.1, 1.1, 1.1 mg/l

48 hour: 1.1, 1.1, 1.1 mg/l

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TABLE 2

PROXEL PRESS PASTE: ACUTE TOXICITY TO *Daphnia magna**Daphnia* RESPONSE

Sponsor	ZENECA Biocides
Test substance:	Proxel press paste
Test organism:	<i>Daphnia magna</i>
Test water:	Reconstituted water medium (Appendix 1)

Time (hours)	Nominal concn. of Proxel press paste (mg/l)	Mean measured concn. of Proxel press paste (mg/l)	Number immobilised per replicate				Total number tested	Total number immobilised	% immobilised
			A	B	C	D			
24	10	10	5	5	5	5	20	20	100
	5.6	5.8	5	5	3	5	20	18	90
	3.2	3.3	0	0	0	0	20	0	0
	1.8	1.9	0	0	0	0	20	0	0
	1.0	1.1	0	0	0	0	20	0	0
	0.56	0.58	0	0	0	0	20	0	0
	Dilution water control	<0.0068	0	0	0	0	20	0	0
48	10	10	5	5	5	5	20	20	100
	5.6	5.8	5	5	4	5	20	19	95
	3.2	3.3	0	1	0	0	20	1	5
	1.8	1.9	0	0	0	0	20	0	0
	1.0	1.1	0	0	0	0	20	0	0
	0.56	0.58	0	0	0	0	20	0	0
	Dilution water control	<0.0068	0	0	0	0	20	0	0

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MOSSLER PROXEL PRESS PASTE DAPHNIA MAGNA 12-7-93

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
10	20	20	100	9.536742E-05
5.8	20	19	95	2.002716E-03
3.3	20	1	5	2.002716E-03
1.9	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 3.3 AND 5.8 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 4.374929

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
3	5.135012E-02	4.369579	3.783579	5.046337

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
5	.1584156	1	.9999389

SLOPE = 13.43237
95 PERCENT CONFIDENCE LIMITS = 8.086093 AND 18.77865

LC50 = 4.374928
95 PERCENT CONFIDENCE LIMITS = 3.871028 AND 4.944411

LC10 = 3.519031
95 PERCENT CONFIDENCE LIMITS = 2.878537 AND 3.963503

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Ecological Effects Branch One-Liner Data Entry Form

Chemical Paraquat (Bamithalozol) Shaughnessy No. 088901 Pesticide Use Unknown

INVERTEBRATE ACUTE TOXICITY	% AI	EC ₅₀ (95%CL) SLOPE	HRS/TYPE	NOEC	STUDY/REVIEW DATES	MRID/CATEGORY	LAB	RC
1. <i>Daphnia magna</i>	76.1	44 mg/l* (3.9-4.9) 134-PROGIC	48 hr Static	1.9 mg/l*	1993/1993	429490-01 Core	ZEN**	MM
2.								
3.								
4.								
5.								
6.								
7.								
CHRONIC TOX.	% AI	MATC LC ₅₀	DAYS	AFFECTED PARA.	STUDY/REVIEW DATES	MRID/CATEGORY	LAB	RC
1.								
2.								
3.								

COMMENTS:

* - Based on mean measured concentrations of paraquat in PARC, ** ZEN = ZENECA Lab Inc.

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