(8-17-92) 121011
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MRID No. 420297-06

DATA EVALUATION RECORD

1. **CHEMICAL:** Clethodim.
Shaughnessey No. 121011.

2. **TEST MATERIAL:** Select 2.0 EC; SX-1839; 10894-05; 25.6% active ingredient.

3. **STUDY TYPE:** Growth and Reproduction of Aquatic Plants -- Tier 2. Species Tested: *Navicula pelliculosa*.

4. **CITATION:** Thompson, S.G., C.M. Holmes, and G.T. Peters. 1991. Select 2.0 EC: A 5-Day Toxicity Test with the Freshwater Diatom (*Navicula pelliculosa*). Laboratory Project ID 162A-118. Conducted by Wildlife International Ltd., Easton, MD. Submitted by Valent U.S.A. Corporation, Walnut Creek, CA. EPA MRID No. 420297-06.

5. **REVIEWED BY:**

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Date: 11/22/91

6. **APPROVED BY:**

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Date: 12/2/91

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Signature: Henry T. Craven 8/7/92 Richard C. Peters 7/28/92
Date: Daniel Rieder 8-17-92

7. **CONCLUSIONS:** This study is scientifically sound and meets the guideline requirements for a Tier 2 aquatic plant growth and reproduction test. The 5-day EC_{50} and NOEC for clethodim were 11 and 3.1 mg ai/l, respectively. These values translate to an EC_{50} and NOEC for Select 2.0 EC of 42 and 12 mg/l of formulated product, respectively.

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

9. **BACKGROUND:**

Core Study

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

11. **MATERIALS AND METHODS:**

A. **Test Species:** *Navicula pelliculosa* cultures used in the test came from laboratory stock cultures originally obtained from University of Texas, Austin, Texas. Cultures that were exponentially growing were used for test inoculum.

B. **Test System:** Test vessels used were 250-ml flasks. The test medium (Table 2, attached) was the same as that used for culturing with the pH adjusted to 7.5 ±0.1 and filter sterilized (0.2 µm).

One-hundred milliliters of the appropriate test or control solution were placed into each flask which were kept at 24 ±2°C in an environmental chamber under continuous illumination (3,477-4,815 lux). The test vessels were shaken at 100 rpm.

C. **Dosage:** Five-day growth and reproduction test. Based on the results of a preliminary test, seven nominal concentrations of 0.10, 0.26, 0.64, 1.6, 4.0, 10, and 25 mg ai/l, a solvent control (0.125 ml acetone/l), a formulation control, and a medium control were selected for the definitive test. The test material was dissolved in acetone to prepare an initial stock solution. The test concentrations were prepared by diluting appropriate volumes of the stock solution in 1000 ml of medium.

D. **Test Design:** An inoculum of *Navicula pelliculosa* designed to provide 10,000 cells/ml was added to each flask (3 containers per treatment). Diatom growth was monitored daily by conducting cell counts on a microscope using a hemocytometer.

The pH was measured at the beginning and end of the study. Temperature within the growth chamber was monitored continuously.

At the beginning and end of the test, samples were removed from exposure and control solutions, frozen, and sent to Chevron Chemical Company, Richmond, CA, for analysis by reverse phase high-pressure liquid chromatography (HPLC).

E. **Statistics:** All calculations were made using mean measured concentrations. The growth rate was computed

from cell density data. The 5-day EC values and associated 95% confidence intervals were computed using the binomial method on growth rate versus mean measured concentration data. The no-observed-effects concentration (NOEC) was estimated using a Kruskal-Wallis test.

12. **REPORTED RESULTS:** The mean measured concentrations were 0.05, 0.16, 0.43, 1.16, 2.99, 8.0, and 20 mg ai/l (Table 1, attached).

Cell counts for each concentration after five days are given in Table 4 (attached). Percent inhibition increased with increasing toxicant concentration for the two highest test concentrations. Mean cell density in the formulation blank and solvent control was significantly lower than that of the negative control.

The 5-day EC_{50} was calculated to be 10.9 mg ai/l with a 95% confidence limit of 8.0-20 mg ai/l based on growth rate inhibition. The NOEC was 2.99 mg ai/l.

The pH ranged from 6.8 to 7.2 in all test solutions and the controls at test initiation and from 6.8 to 7.4 at test termination. The temperature ranged from 22.4 to 25.4°C.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** The authors concluded that if Select 2.0 EC were applied at 0.25 lb ai/A to a water depth of 6 inches, the estimated environmental concentration of clethodim would be 0.184 mg/l, or 6.2% of the NOEC.

Good Laboratory Practice and Quality Assurance statements were included in the report indicating compliance with EPA Good Laboratory Practice Standards, 40 CFR Part 160, under the Federal Insecticide, Fungicide, and Rodenticide Act.

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

A. **Test Procedure:** The test procedure and the report were generally in accordance with the SEP and Subdivision J guidelines, except for the following deviations:

The cell inoculum (10,000 cells/ml) was greater than recommended (3,000 cells/ml).

The conductivity of the test solutions was not measured.

The light intensity during the test was occasionally higher or lower than recommended.

The concentrations were not of a 2x progression.

B. **Statistical Analysis:** The reviewer used EPA's Toxanal program and ANOVA (coupled with Dunnett's test) analyses on the 5-day inhibition and cell count data to determine the EC and NOEC values, respectively. The values obtained were less conservative than those of the authors. The EC_{50} and NOEC for Select 2.0 EC are 42 and 12 mg/l of formulated product, respectively, based on inhibition of growth rate.

C. **Discussion/Results:** Since the title of this study indicates that the formulated product Select 2.0 EC is the subject of the test, the reviewer reported the EC and NOEC values in terms of mg/l of formulated product.

Based on mean measured concentrations transformed to mg/l formulated product, the 5-day EC_{50} was calculated to be 42 mg/l with a 95% confidence limit of 31-77 mg/l. The NOEC was 12 mg/l. These values are equivalent to EC_{50} and NOEC values of 11 and 3.1 mg ai/l, respectively, for clethodim.

This study is scientifically sound and meets the guideline requirements for a Tier 2 toxicity study using non-target aquatic plants.

D. **Adequacy of the Study:**

(1) **Classification:** Core for Select 2.0 EC only.

(2) **Rationale:** N/A.

(3) **Repairability:** N/A.

15. **COMPLETION OF ONE-LINER:** Yes, 11-15-91.

CLETHODIM

Page _____ is not included in this copy.

Pages 5 through 8 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.

X 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.

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
77	100	88	88	0
30.8	100	5	5	0
11.5	100	0	0	0
4.5	100	0	0	0
1.7	100	0	0	0
.6	100	0	0	0
.2	100	0	0	0

BECAUSE THE NUMBER OF ORGANISMS USED WAS SO LARGE, THE 95 PERCENT CONFIDENCE INTERVALS CALCULATED FROM THE BINOMIAL PROBABILITY ARE UNRELIABLE. USE THE INTERVALS CALCULATED BY THE OTHER TESTS.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS 51.62083

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
1	2.006091E-02	51.62083	48.37718	55.2135

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
9	3.429906E-02	1	1

SLOPE = 7.086282
95 PERCENT CONFIDENCE LIMITS = 5.773902 AND 8.398662

LC50 = 52.56262
95 PERCENT CONFIDENCE LIMITS = 48.26435 AND 57.07569

LC10 = 34.79023
95 PERCENT CONFIDENCE LIMITS = 30.34381 AND 38.62223

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Based on inhibition of cell density compared to solvent control.
Raw data from Table 4 (attached).

Navicula cell density

Summary Statistics and ANOVA

Transformation = None

Group	concentration (mg/l)*	n	Mean	s.d.	cv%
1 = control		3	92333.3333	45610.6713	49.4
2	0.2	3	142666.6667	71472.6055	50.1
3	0.6	3	383666.6667	173508.8855	45.2
4	1.7	3	294333.3333	146377.3662	49.7
5	4.5	3	100333.3333	26083.2002	26.0
6	11.5	3	283333.3333	48013.8869	16.9
7	30.8	3	87333.3333	42453.8966	48.6
8	77.0	3	11000.0000	1732.0508	15.7

i = solvent control

Raw data from Table 4 (attached).

*) the mean for this group is significantly less than the control mean at alpha = 0.05 (1-sided) by Dunnett's test

NOEC = 77 mg/l of Select 2.0 EC.

* - based on measured concentrations of ai converted to mg/l of formulated product

Minumum detectable difference for Dunnett's test = -186240.855286
This difference corresponds to -201.70 percent of control

Between groups sum of squares =********** with 7 degrees of freedom.

Error mean square = ********** with 16 degrees of freedom.

Bartlett's test p-value for equality of variances = .003

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*                                              *
* Warning - the test for equality of variances *
* is significant (p less than 0.01).  The      *
* results of this analysis should be inter-    *
* preted with caution.                         *
*                                              *
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Shaughnessey # 121011 Chemical Name Clethodim Chemical Class _____ Page 1 of 1

Study/Species/Lab/ MRID #	Chemical % a.i.	Results	Reviewer/ Date	Validation Status
14-Day EC_{50}	_____	EC_{50} = . pp (95% C.L.) # plants/vessel = Slope = Species: Temperature = Lab: 14-Day Dose Level pp /(% Effect) ( ), ( ), ( ), ( ), ( ) MRID # Comments:	_____	_____
5-Day EC_{50}	25.6	EC_{50} = 42 mg/l* (95% C.L. 31 - 77) - for select 2.0 EC - Gnamd probability EC_{50} = 11. mg ai/l (7.9 - 20) - for clethodim Slope = N/A # Cells/ml = 10,000 Species: Navicula pelliculosa Temperature = 24°C Lab: Wildlife Intern. 5-Day Dose Level mg/l* pp /(% Effect) 0.2 (0), 0.6 (0), 1.7 (0), 4.5 (0), 11.5 (0) 30.8 (5), 77 (88) MRID # 420297-06 Comments: NOEC = 12 mg/l* of formulated product, for select 2.0 EC NOEC = 3.1 mg ai/l for clethodim * - Based on mean measured conc. of ai converted to mg/l of formulated product	M. Hassler 11/15/91	Core.