3-21-91 FILE COPY : 121011

DATA EVALUATION RECORD

1.) CHEMICAL: clethodim (Select 2EC) herbicide

2.) TEST MATERIAL: clethodim technical (RE 45601) 83.3%.

3.) STUDY TYPE: 123-2, Tier II, Aquatic Plant Growth. (Non-target Area Phytotoxicity)

4.) CITATION: Forbis, A. D. and J.W. Blasberg. 1990. Acute Toxicity of RE-45601 Technical to the Freshwater Algae (Selenastrum capricornutum) Printz. 09/21/90.

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ABC Final Report #38373

FOR: Valent U.S.A. Corp.
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EPA MRID No.: 416851-06

5.) REVIEWED BY:

Richard C. Petrie
Agronomist
EEB/EFED

Signature: Richard C. Petrie

Date: 3/21/91

6.) APPROVED BY:

Charles Lewis
Acting Head, Section 3
EEB/EFED

Signature: Charles Lewis

Date: 3/22/91

7.) CONCLUSIONS:

This study satisfies the Guideline requirement for a 5 day acute non-target Selenastrum capricornutum plant phytotoxicity study (123-2) using the technical material. No adverse growth reductions greater than 50% occurred for Selenastrum capricornutum at any concentration tested up to 11.4 mg ai/L measured

concentration. The maximum nominal concentration tested was 8.0 mg ai/L. The maximum concentration tested was far greater than 3 times the estimated environmental concentration value of 550 ppb.

8.) RECOMMENDATIONS:

No further testing of Selenastrum capricornutum is required.

9.) BACKGROUND:

This is one of 5 non-target aquatic phytotoxicity studies requested when herbicides are applies aerially.

10.) DISCUSSION OF INDIVIDUAL TESTS:

11.) MATERIALS AND METHODS:

The alga Selenastrum capricornutum was used in this static 120 hour EC50 study. The parent stock of this culture was obtained from the Department of Botany, University of Texas at Austin. The test procedure for this static algal assay was submitted with the study (Chevron Protocol #89-404). This study was conducted in 250 ml Erlenmeyer flasks containing 100 ml synthetic algal culture medium. The micronutrients used in the stock solutions contained Na2EDTA at 300 mg. Algal cell counts were taken using a hemacytomer with an Olympus Model CHA microscope.

A 120 hour range finding study was conducted using 0.001, 0.01, 0.10, and 1.0 mg/L test material. The cell counts at the end of the 5 day range-finding study were: 90%, 99%, 81%, and 94% of the control populations, respectively.

For the definitive study, the following concentrations were used:

ai ppm nominal	mean ppm ai measured	% of target conc.
0.50	DAY 0 - 0.44	DAY 0 - 87
	DAY 5 - 0.28	DAY 5 - 56*
1.00	DAY 0 - 0.96	DAY 0 - 96
	DAY 5 - 1.04	DAY 5 - 104
2.00	DAY 0 - 2.31	DAY 0 - 116
	DAY 5 - 2.15	DAY 5 - 108
4.00	DAY 0 - 5.00	DAY 0 - 125
	DAY 5 - 5.12	DAY 5 - 128
8.00	DAY 0 - 11.60	DAY 0 - 145
	DAY 5 - 11.40	DAY 5 - 143

* Sample bottle cracked during thawing.

The test material was RE-45601 technical clethodim containing 83.3% active ingredient. The test material was unstable at extreme pH,

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temperature, and upon exposure to sunlight. The solubility was found to be <11 mg/L. For the 120 hour period, the temperature ranged from 24 to 25 degrees C and the pH ranged from 7.1 to 8.2. Test vessels were constantly rotary agitated at approximately 100 rpm. Light intensity was maintained at 4300 lux under continuous fluorescent lighting.

12.) REPORTED RESULTS:
The following results were reported:

MEAN* MEASURED ai CONC.	%DIFFERENCE FROM THE SOLVENT CONTROL				
	24 HR.	48 HR.	72 HR.	96 HR.	120 HR.
0.28	-24	0	0	-25	-4
1.04	-9	0	-8	-23	-19
2.15	-15	-17	0	-14	0
5.12	-35	-12	-8	0	-4
11.40	-35	-46	-26	-30	-27

* 5 DAY VALUES

13.) STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:

A valid EC50 could not be determined because the cell growth at the maximum 11.40 mg ai/L (8.0 mg ai/L nominal) concentration was not significantly different from the solvent and non-solvent controls after 120 hours. The no-effect concentration for this study is 8.0 mg ai/L nominal.

Test results were subjected to analysis of variance and treatment means were compared using a multiple means test (Dunnett's).

This study was conducted following the intent of Good Laboratory Practice Regulations and the final report was reviewed by the ABC Labs Quality Assurance Unit.

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

A. TEST PROCEDURES:

Test protocols were included in the submission and properly referenced. Deviations from protocol were: 1.) the nutrient chelate EDTA was added to the cultures at 300 ug/L. EPA Subdivision J guidelines specifically prohibit the addition of EDTA to algal cultures. The EEB has recently determined that the

addition of EDTA to algal cultures is an acceptable practice after discussions with algae experts at our Corvallis Laboratory. Experimental data demonstrates that the addition of up to 400 ug/L is an acceptable practice that will not result in over-stimulation of algal growth. 2.) the Subdivision J guidelines calls for 4000 lux light intensity. A light intensity of 4300 lux was used.

These deviations from protocol are not expected to significantly affect test results.

B. STATISTICAL ANALYSIS:

Statistical analyses was performed using the Stephens program. Runs on 120 and 96 hour test results were made. Calculation of an EC50 was not possible for either data set due to a lack of 50% mortality at any treatment level. The no-effect level was determined to be 8.0 mg ai/L (NOMINAL), 11.40 mg ai/L MEASURED.

C . Discussion/Results:

The number of cells in the 8.0 mg ai/L (NOMINAL), 11.40 mg ai/L (MEASURED) concentration were not significantly different from the number of cells in the solvent control after 120 hours. An EC50 value could not be calculated due to a lack of greater than 50% mortality at the highest concentration tested. Based on the maximum proposed label rate of 0.25# ai/Acre, if applied directly to a water body 6 inches deep, the resulting concentration of clethodim would be 184 ppb. Three times this concentration is approximately 550 ppb. The maximum concentration used in this study was well above this 3X concentration. Additional testing to establish an EC50 is not necessary.

D. Adequacy Of The Study:

(1) Classification: Core

(2) Rationale: N/A

(3) Repairability: N/A

Clethodim (Select 2EC) Selenastrum Capricornutum

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
8	100	27	27	0
4	100	4	4	0
2	100	0	0	0
1	100	19	19	0
.5	100	4	4	0

120 hr

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 0

THE MOVING AVERAGE METHOD CANNOT BE USED WITH THIS DATA SET BECAUSE NO SPAN WHICH PRODUCES MOVING AVERAGE ANGLES THAT BRACKET 45 DEGREES ALSO USES TWO PERCENT DEAD BETWEEN 0 AND 100 PERCENT.

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
4	16.86172	14.88823	0

A PROBABILITY OF 0 MEANS THAT IT IS LESS THAN 0.001.

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE = .5473921
95 PERCENT CONFIDENCE LIMITS =-1.700365 AND 2.795149

LC50 = 418.4684
95 PERCENT CONFIDENCE LIMITS = 4.387072 AND +INFINITY

LC10 = 2.002493
95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

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R. PETRIE CLETHODIM SELENASTRUM CAPRICORNUTUM 03/19/91

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CONC. PPM	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
8	100	30	30	0
4	100	0	0	0
2	100	14	14	0
1	100	23	23	0
.5	100	25	25	0

96hr

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 0

THE MOVING AVERAGE METHOD CANNOT BE USED WITH THIS DATA SET BECAUSE NO SPAN WHICH PRODUCES MOVING AVERAGE ANGLES THAT BRACKET 45 DEGREES ALSO USES TWO PERCENT DEAD BETWEEN 0 AND 100 PERCENT.

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
4	126.9724	12.77267	0

A PROBABILITY OF 0 MEANS THAT IT IS LESS THAN 0.001.

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE = -.15494

95 PERCENT CONFIDENCE LIMITS =-1.900835 AND 1.590955

LC50 = 3.010745E-06

95 PERCENT CONFIDENCE LIMITS = 0 AND 1.077946

LC10 = 473.5823

95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

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CONC. PPM	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
8	100	26	26	0
4	100	8	8	0
2	100	0	0	0
1	100	8	8	0
.5	100	0	0	0

72h.

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 0

THE MOVING AVERAGE METHOD CANNOT BE USED WITH THIS DATA SET BECAUSE NO SPAN WHICH PRODUCES MOVING AVERAGE ANGLES THAT BRACKET 45 DEGREES ALSO USES TWO PERCENT DEAD BETWEEN 0 AND 100 PERCENT.

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
4	2.701332	7.691292	0

A PROBABILITY OF 0 MEANS THAT IT IS LESS THAN 0.001.

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE = 1.316597
95 PERCENT CONFIDENCE LIMITS =-.847326 AND 3.480519

LC50 = 31.97454
95 PERCENT CONFIDENCE LIMITS = 6.284626 AND +INFINITY

LC10 = 3.468934
95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

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