DATA EVALUATION RECORD
ALGAE OR DIATOM EC_{50} TEST
GUIDELINE 123-2 (TIER II)

1. **CHEMICAL:** Metolachlor PC Code No.: 108801

2. **TEST MATERIAL:** CGA-51202 Purity: Not reported

3. **CITATION:** Author: A. Vial
   Title: Report on the Growth Inhibition Test of CGA-51202 to Green Algae (*Scenedesmus subspicatus*)
   Study Completion Date: August 12, 1991
   Laboratory: Ciba-Geigy Ltd., Crop Protection Division, Basle, Switzerland
   Sponsor: Novartis Crop Protection, Inc., Greensboro, NC
   Laboratory Project ID: 918152
   DP Barcode: D260010
   MRID No.: 449295-15

4. **REVIEWED BY:** Karl Bullock, M.S., Environmental Scientist,
   Golder Associates Inc.
   Signature: **Date:** 11/10/99

   APPROVED BY: Pim Kosalwat, Ph.D., Senior Scientist,
   Golder Associates Inc.
   Signature: **Date:**

5. **APPROVED BY:** Brian Montague, Fisheries Biologist
   Environmental Fate and Effects Division, OPP
   Signature: **Date:** April 2000

6. **STUDY PARAMETERS:**
   Definitive Test Duration: 72 hours
   Type of Concentrations: Initial measured

7. **CONCLUSIONS:** This study is scientifically sound but does not fulfill the guideline requirements for a Tier-II algal toxicity test. The percent purity of the test substance was not reported, and the test was conducted for only 3 days.
   Results Synopsis
   72-hour EC_{50} = 57.1 (29.3 to inf.) **NOEC** 29.3 ppm **Slope**= N/A

8. **ADEQUACY OF THE STUDY:**
   A. Classification: Supplemental.

B. Rationale: The percent purity of the test substance was not reported and the test was conducted for only 3 days. Species is not generally accepted by the Agency.

C. Repairability: No.

9. GUIDELINE DEVIATIONS:

1. The percent purity of the test substance was not reported.
2. The study was conducted for 3 days rather than the required 4 days.
3. The maximum labelled rate was not reported.
4. The nutrients used in the test were not reported.

10. SUBMISSION PURPOSE:

11. MATERIALS AND METHODS:

A. Test Organisms

Guideline Criteria	Reported Information
Species *Skeletonema costatum* *Anabaena flos-aquae* *Selenastrum capricornutum* *Navicula pelliculosa*	*Scenedesmus subspicatus*
Initial Number of Cells 3,000 - 10,000 cells/mL	9,400 cells/mL
Nutrients Standard formula, e.g. 20XAAP	Not reported

B. Test System

Guideline Criteria	Reported Information
Solvent	None
Temperature Skeletonema: 20°C Others: 24-25°C	23 ±1°C
Light Intensity Anabaena: 2.0 KLux (±15%) Others: 4.0-5.0 KLux (±15%)	10.8 KLux
Photoperiod Skeletonema: 14 h light, 10 h dark or 16 h light, 8 h dark Others: Continuous	Continuous
pH Skeletonema: approx. 8.0 Others: approx. 7.5	Initial: 6.7-7.9 Final: 7.6-7.9

C. Test Design

Guideline Criteria	Reported Information
Dose range 2X or 3X progression	3X
Doses at least 5	1.23, 3.7, 11, 33, and 100 mg/L, not corrected for percent active ingredient
Controls negative and/or solvent	Negative control group
Replicates per dose 3 or more	6 replicates in the control and 3 replicates per treatment
Duration of test 120 hours	72 hours
Daily observations were made?	Yes

Guideline Criteria	Reported Information
Method of Observations	"TOA" cell counter
Maximum Labeled Rate	Not reported

12. REPORTED RESULTS:

Guideline Criteria	Reported Information
Initial and 72 h cell densities were measured?	Yes
Control cell count at 72 hr ≥2X initial count?	Yes
Initial chemical concentrations measured? (Optional) 1. Percent of nominal 2. Detection limit 3. Method validation	 1. 89 - 95% of nominal 2. <1.0 mg/L 3. 108.1% of nominal
Raw data included?	Yes

Analytical Results

Toxicant Concentration (mg/L)				
Nominal	Hour of Study		Mean Measured (SD)	Percent of Nominal
	0	96		
Control	<1.0	<1.0	-	-
1.23	1.1	1.0	1.1 (0.07)	85
3.7	3.5	3.5	3.5 (0)	95

11	10.4	10.3	10.4 (0.07)	94
33.0	29.3	32.2	30.8 (2.05)	93
100.0	92.2	81.7	87.0 (7.42)	87

Dose Response

Initial Measured Concentration (ppm)	72-hour Avg. Cell Density (x 10^4 cells/mL)	Inhibition* (%)	Final pH
Control	146.4	-	7.9
1.1	155.3	-6.1	7.9
3.5	154.7	-5.7	7.9
10.4	168.0	-14.8	7.9
29.3	174.0	-18.9	7.9
92.2	8.8	94.0	7.6

*Negative inhibition indicates stimulation

Other Significant Results: None.

Statistical Results for Cell Density:

Statistical Methods: The EC_{50} values were calculated according to Berkson (1953); NOEC calculated according to the modified Dunnett's test. Results are based on nominal concentrations and areas under the growth curves.

EC_{50}: 77.6 mg/L 95% C.I.: 73.1-80.8 mg/L

Probit Slope: N/A NOEC: 33 mg/L

13. **VERIFICATION OF STATISTICAL RESULTS:**

Statistical Methods: Analyses were based on initial measured concentrations and cell density. The probit method could not be used since it requires at least two concentrations at which the percent inhibition is between 0 and 100. Non-linear regression could not be used to estimate the EC_{50} value because it relies upon estimates generated from the probit analysis. Therefore, the EC_{50} was estimated using the binomial method, although 95% confidence intervals could not be estimated. Williams' test was used to determine the NOEC.

EC_{50}: 57.1 ppm 95% C.I.: N/A

Probit Slope: N/A NOEC: 29.3 ppm

14. **REVIEWER'S COMMENTS:** This study is scientifically sound but does not fulfill the guideline requirements for an algal toxicity test. The percent purity of the test material was not reported and the test was only conducted for 3 days rather than the required 4 days. Based on initial measured concentrations, the 72-hour EC_{50} and NOEC for *S. subspicatus* were 57.1 and 29.3 ppm, respectively. This study can be categorized as **Supplemental**.

CGA-51202: Acute Green Algae Test
File: 44929515 Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Control	6	146.417	146.417	157.472
2	1.1	3	155.333	155.333	157.472
3	3.5	3	154.667	154.667	157.472
4	10.4	3	168.000	168.000	157.472
5	29.3	3	174.000	174.000	157.472
6	92.2	3	8.833	8.833	8.833

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WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE WILLIAMS P=.05	DEGREES OF FREEDOM WILLIAMS
Control	157.472				
1.1	157.472	0.574		1.75	k= 1, v=15
3.5	157.472	0.574		1.84	k= 2, v=15
10.4	157.472	0.574		1.87	k= 3, v=15
29.3	157.472	0.574		1.88	k= 4, v=15
92.2	8.833	7.149	*	1.89	k= 5, v=15

s = 27.219

Note: df used for table values are approximate when v > 20.