

(4)

DATA EVALUATION RECORD
BLUE-GREEN ALGAE EC₅₀ TEST
GUIDELINE 123-2 (TIER II)

1. CHEMICAL: Metolachlor
Shaughnessey No.: 108801

2. TEST MATERIAL: Metolachlor technical
Purity: 97.3%

3. CITATION:

Authors: Hoberg, J.R.

Title: Metolachlor technical - 5-day toxicity to
the freshwater green alga, *Anabaena*
flos-aquae

Date: 1994

Laboratory: Springborn Laboratories, Inc., Wareham,
MA

Sponsor: Ciba Crop Protection, Greensboro, NC

Lab. Report No.: 94-7-5382

MRID No.: 434871-04

4. REVIEWED BY:

William Erickson
Biologist
EEB/EFED

Signature: *W. Erickson*

Date: *1/26/95*

5. APPROVED BY:

Harry Craven
Section Head 4
EEB/EFED

Signature: *Harry T. Craven*

Date: *2/15/95*

6. CONCLUSION: The study is scientifically sound and fulfills the guideline requirement for a Tier 2 aquatic plant growth study with a blue-green algae. The 120-h EC₅₀ value is 1.2 mg ai/l, which classifies technical metolachlor as moderately toxic to *Anabaena flos-aquae*.

7. ADEQUACY OF THE STUDY: Core.

8. MAJOR GUIDELINE DEVIATIONS: None.

9. MATERIALS AND METHODS:

A. Test Organisms:

Guideline Criteria	Reported Information
Species: <i>Skeletonema costatum</i> <i>Anabaena flos-aquae</i> <i>Selenastrum capricornutum</i> <i>Navicula pelliculosa</i>	<i>Anabaena flos-aquae</i>
Number of Cells: 3,000 - 10,000	10,000
Nutrients:	AAP medium

B. Test System:

Guideline Criteria	Reported Information
Solvent used:	None
Site of test: Growth Chamber	Environmental chamber
Temperature: 20° C for <i>Skeletonema</i> sp.; others are 24-25°C	24 °C
Light Intensity: 2.0 Lux for <i>Anabaena</i> sp.; others are 4-5.0	1.1-3.2
Photoperiod:	continuous illumination
pH: 7.5-8.0	7.5

C. Test Design:

Guideline Criteria	Reported Information
Dose range: 2X or 3X	3X
Doses: at least 5	6
Controls: negative and/or solvent	Negative
Three or more replicates per dose:	3
Duration of test: 120 hours	120 h
Daily Observations?	Yes
Method of Observations:	Compound microscope
Maximum Labeled Rate:	Not reported

10. REPORTED RESULTS:

Guideline Criteria	Reported Information
Initial and 120 hours Cell Measurements?	Yes
Control cell count at 120 hr from initial increase $\geq 2X$?	Yes
Measured Initial Chemical Concentrations? Optional	Yes
Raw data included? (Y/N)	Excerpted
Quality Assurance Measures?	Yes

Dose Response:

doses→ mg ai/l	control	0.019	0.063	0.19	0.72	2.1	6.8
inhibit %		1	5	16	44	56	82
120 hr pH	9.7	9.5	8.7	8.2	7.9	7.7	7.7

Observations: At test termination, cell fragments were observed in the 0.72, 2.1, and 6.8 mg ai/l concentrations. Bloated cells were observed in the 6.8 mg ai/l concentration. The cells exposed to the remaining concentrations and the control were normal.

Statistical Results:

Method: Linear Regression (EC_{50}) and Williams' Test (NOEC)

EC_{50} : 1.1 mg ai/l 95% C.L.: 0.31-3.9 mg ai/l

NOEC: 0.063 mg ai/l

11. REVIEWER'S DISCUSSION/INTERPRETATION:

Test Procedures: Test procedures met guideline criteria.

Statistical Analysis:

Method: EPA Toxanol program (EC_{50}); Williams' Test (NOEC)

EC_{50} : 1.2 mg ai/l 95% C.L.: 0.9-1.6 mg ai/l

Slope: 1.2 NOEC: 0.063 mg ai/l

Discussion: The study is scientifically sound and fulfills the guideline requirement for a Tier 2 aquatic plant growth study with a blue-green algae.

12. COMPLETION OF STUDY ONE-LINER: 1/26/95.

WILLIAMS TEST (Isotonic regression model)

TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
0	87.333				
0.019	86.000	0.323		1.76	k= 1, v=14
0.063	82.333	1.210		1.85	k= 2, v=14
0.19	73.333	3.388	*	1.88	k= 3, v=14
0.72	49.000	9.276	*	1.89	k= 4, v=14
2.1	36.667	12.260	*	1.90	k= 5, v=14
6.8	16.000	17.261	*	1.91	k= 6, v=14

s = 5.062

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

W. ERICKSON METOLACHLOR ANABAENA TOXICITY

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
6.8	100	82	82	0
2.1	100	56	56	0
.72	100	44	44	0
.19	100	16	16	0
.063	100	5	5	0
.019	100	1	1	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 1.229634

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS
3	3.905641E-02	1.227328	.9407113
1.60024			

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H
3	2.118531E-02	1
.6560646		

SLOPE = 1.219745
95 PERCENT CONFIDENCE LIMITS = 1.042209 AND 1.397281

LC50 = 1.251928
95 PERCENT CONFIDENCE LIMITS = .9875165 AND 1.619197

LC10 = .1138619
95 PERCENT CONFIDENCE LIMITS = 7.518421E-02 AND .1585785

ANABAENA CELL DENSITIES

File: A:ANABAENA.DAT

Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 1 of 2

GRP	IDENTIFICATION	N	MIN	MAX	MEAN
1	0	3	83.000	93.000	87.333
2	0.019	3	81.000	89.000	86.000
3	0.063	3	77.000	88.000	82.333
4	0.19	3	65.000	80.000	73.333
5	0.72	3	45.000	55.000	49.000
6	2.1	3	33.000	39.000	36.667
7	6.8	3	14.000	19.000	16.000

ANABAENA CELL DENSITIES

File: A:ANABAENA.DAT

Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 2 of 2

GRP	IDENTIFICATION	VARIANCE	SD	SEM
1	0	26.333	5.132	2.963
2	0.019	19.000	4.359	2.517
3	0.063	30.333	5.508	3.180
4	0.19	58.333	7.638	4.410
5	0.72	28.000	5.292	3.055
6	2.1	10.333	3.215	1.856
7	6.8	7.000	2.646	1.528

ANABAENA CELL DENSITIES

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WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	0	3	87.333	87.333	87.333
2	0.019	3	86.000	86.000	86.000
3	0.063	3	82.333	82.333	82.333
4	0.19	3	73.333	73.333	73.333
5	0.72	3	49.000	49.000	49.000
6	2.1	3	36.667	36.667	36.667
7	6.8	3	16.000	16.000	16.000

ANABAENA CELL DENSITIES