

(3)

DP Barcode : D210584
PC Code No : 108801
EEB Out : 13/06/95

To: Walter Waldrop/Jane Mitchell
Product Manager 71
Special Review and Reregistration Division (7508W)

From: Anthony F. Maciorowski, Chief
Ecological Effects Branch/EFED (7507C)

Attached, please find the EEB review of...

Reg./File # : 108801
Chemical Name : Metolachlor
Type Product : Fungicide
Product Name :
Company Name : Ciba Crop Protection, Greensboro, NC
Purpose : Review guideline studies 72-3a,b,c; 123-1a,b;
123-2
Action Code : 627 Date Due : 04/24/95
Reviewer : William Erickson Date In : 01/11/95

EEB Guideline/MRID Summary Table: The review in this package contains an evaluation of the following:

Gdln No.	MRID No.	Cat.	Gdln No.	MRID No.	Cat.	Gdln No.	MRID No.	Cat.
71-1(a)			72-2(a)			72-7(a)		
71-1(b)			72-2(b)			72-7(b)		
71-2(a)			72-3(a)	434871-01	Y	122-1(a)		
71-2(b)			72-3(b)	434871-02	Y	122-1(b)		
71-3			72-3(c)	434871-03	Y	122-2		
71-4(a)			72-3(d)			123-1(a)	434871-07	Y
71-4(b)			72-3(e)			123-1(b)	434871-08	Y
71-5(a)			72-3(f)			123-2	434871-04 434871-05 434871-06	Y Y Y
71-5(b)			72-4(a)			124-1		
72-1(a)			72-4(b)			124-2		
72-1(b)			72-5			141-1		
72-1(c)			72-6			141-2		
72-1(d)						141-5		

Y=Acceptable (Study satisfied Guideline)/Concur

P=Partial (Study partially fulfilled Guideline but
additional information is needed)

S=Supplemental (Study provided useful information but Guideline was
not satisfied)

(1)



UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Metolachlor: review of toxicity studies

FROM: Anthony F. Maciorowski, Chief
Ecological Effects Branch
Environmental Fate and Effects Division (7507C) 2/16/95

TO: Walter Waldrop/Jane Mitchell
Product Manager 71
Special Review and Reregistration Division (7508W)

Ciba Crop Protection, Greensboro, NC submitted the following studies to support reregistration of metolachlor:

Machado, M.W. 1994. Metolachlor technical - acute toxicity to sheepshead minnow (*Cyprinodon variegatus*) under flow-through conditions. Conducted by Springborn Laboratories, Inc., Wareham, MA. Lab. Report No. 94-7-5378. MRID No. 434871-01.

Dionne, E. 1994. Metolachlor technical - acute toxicity to Eastern oyster (*Crassostrea virginica*) under flow-through conditions. Conducted by Springborn Laboratories, Inc., Wareham, MA. Lab. Report No. 94-7-5365. MRID No. 434871-02.

Machado, M.W. 1994. Metolachlor technical - acute toxicity to mysid shrimp (*Mysidopsis bahia*) under flow-through conditions. Conducted by Springborn Laboratories, Inc., Wareham, MA. Lab. Report No. 94-7-5402. MRID No. 434871-03.

Hoberg, J.R. 1994. Metolachlor technical - 5-day toxicity to the freshwater green alga, *Anabaena flos-aquae*. Conducted by Springborn Laboratories, Inc., Wareham, MA. Lab. Report No. 94-7-5382. MRID No. 434871-04.

Hoberg, J.R. 1994. Metolachlor technical - toxicity to duckweed (*Lemna gibba*). Conducted by Springborn Laboratories, Inc., Wareham, MA. Lab. Report No. 94-8-5404. MRID No. 434871-05.



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Hoberg, J.R. 1994. Metolachlor technical - toxicity to the marine diatom, *Skeletonema costatum*. Conducted by Springborn Laboratories, Inc., Wareham, MA. Lab. Report No. 94-7-5382. MRID No. 434871-06.

Chetram, R.S. 1994. Tier 2 seedling emergence nontarget phytotoxicity study using metolachlor. Conducted by Pan-Agricultural Labs, Inc, Madera, CA. Lab. Report # 94349. MRID No. 434871-07.

Chetram, R.S. 1994. Tier 2 vegetative vigor nontarget phytotoxicity study using metolachlor. Conducted by Pan-Agricultural Labs, Inc, Madera, CA. Lab. Report # 94350. MRID No. 434871-08.

Chetram, R.S. 1994. Tier 2 seed germination nontarget phytotoxicity study using metolachlor. Conducted by Pan-Agricultural Labs, Inc, Madera, CA. Lab. Report # 94348. MRID No. 434973-01.

The DERs are attached. The Agency no longer requires seed germination studies; therefore, EEB has not reviewed that study (MRID No. 434973-01). The remainder of the studies submitted are acceptable and fulfill the guideline requirements for 72-3a,b,c and 123-1a,b. Guideline 123-2 (Aquatic Plant Growth) has not been fully satisfied, because the registrant did not submit required phytotoxicity studies for *Selenastrum capricornutum* and for a freshwater diatom.

If you have any questions, please contact Bill Erickson at 305-6212 or Harry Craven at 305-5320.

(4)

DATA EVALUATION RECORD
ACUTE LC₅₀ TEST WITH AN ESTUARINE/MARINE FISH
§ 72-3 (A)

1. CHEMICAL: Metolachlor
Shaughnessey No.: 108801

2. TEST MATERIAL: Metolachlor technical
Purity: 97.3%

3. CITATION

Authors: Machado, M.W.

Title: Metolachlor technical - acute toxicity to
sheepshead minnow (*Cyprinodon variegatus*)
under flow-through conditions

Date: 1994

Laboratory: Springborn Laboratories, Inc., Wareham,
MA

Sponsor: Ciba Crop Protection, Greensboro, NC

Lab. Report No.: 94-7-5378

MRID No.: 434871-01

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 Craven*

Date: *2/15/95*

6. CONCLUSIONS: The study is scientifically sound and satisfies the guideline requirement for an acute marine/estuarine fish toxicity test. The LC₅₀ value of 9.8 mg ai/l classifies technical metolachlor as moderately toxic to the sheepshead minnow. The NOEC is 3.6 mg ai/l.

7. ADEQUACY OF THE STUDY: Core.

8. MAJOR GUIDELINE DEVIATIONS: None.

9. MATERIALS AND METHODS:

A. Test Organisms

Guideline Criteria	Reported Information
<u>Species</u> Preferred species are the sheepshead minnow (<i>Cyprinodon variegatus</i>) or the Silverside (<i>Menidia sp.</i>)	Sheepshead minnow
<u>Mean Wet Weight</u> 0.5 - 5 g	Mean: 0.22 g Range: 0.11-0.35 g
<u>Mean Standard Length</u> Longest not > 2x shortest	Mean: 23 mm Range: 15-27 mm
<u>Supplier</u>	Aquatic Research Organisms, Hampton, NH
All fish from same source?	Yes
All fish from the same year class?	Not reported

B. Source/Acclimation

Guideline Criteria	Reported Information
<u>Acclimation Period</u> minimum 14 days	14 days
Wild caught organisms were quarantined for 7 days?	N/A
Were there signs of disease or injury?	Not reported
If treated for disease, was there no sign of the disease remaining during the 48 hours prior to testing?	Not reported
<u>Feeding</u> No feeding during the study	Last fed 48 h prior to test
<u>Pretest Mortality</u> <3% mortality 48 hours prior to testing	No mortality within 48 h of testing

C. Test System

Guideline Criteria	Reported Information
<u>Source of dilution water</u> Soft reconstituted water or water from a natural source, not dechlorinated tap water	Filtered seawater
<u>Does water support test animals without observable signs of stress?</u>	Yes
<u>Salinity</u> 30-34 ‰ salinity, weekly range < 6 ‰	31 ‰
<u>Water Temperature</u> 22 ± 1 °C	21-22 °C
<u>pH</u> 8.0-8.3 for marine-stenohaline fishes, 7.7-8.0 for estuarine-euryhaline fishes, monthly range < 0.8	8.0
<u>Dissolved Oxygen</u> Static: ≥ 60% during 1 st 48 hrs and ≥ 40% during 2 nd 48 hrs, flow-through: ≥ 60%	Lowest % DO was 73% at 96 h
<u>Test Aquaria</u> 1. <u>Material</u> : Glass or stainless steel 2. <u>Size</u> : Volume of 19 L (5 gal) or 30 x 60 x 30 cm 3. <u>Fill volume</u> : 15-30 L of solution	<u>Material</u> : not described <u>Size</u> : 39 x 20 x 25 cm <u>Fill vol.</u> : 15 l solution
<u>Type of Dilution System</u> Must provide reproducible supply of toxicant	Mount and Brungs intermittent-flow proportional diluter, temp.-controlled water bath, and set of 14 aquaria
<u>Flow Rate</u> Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period	6.5 vol/24 hours

Guideline Criteria	Reported Information
<u>Biomass Loading Rate</u> Static: ≤ 0.8 g/L at $\leq 17^{\circ}\text{C}$, ≤ 0.5 g/L at $> 17^{\circ}\text{C}$; flow-through: ≤ 1 g/L/day	0.023 g/L
<u>Photoperiod</u> 16 hours light, 8 hours dark	16 h light
<u>Solvents</u> Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests	Solvent: acetone Maximum conc.: 0.092 mL/L

D. Test Design

Guideline Criteria	Reported Information
<u>Range Finding Test</u> If $\text{LC}_{50} > 100$ mg/L with 30 fish, then no definitive test is required.	100% mortality at 20 mg ai/L
<u>Nominal Concentrations of Definitive Test</u> Control & 5 treatment levels; each conc. should be 60% of the next highest conc.; concentrations should be in a geometric series	2.6, 4.3, 7.2, 12, and 20 mg ai/L.
<u>Number of Test Organisms</u> Minimum 10/level, may be divided among containers	20/level 10/rep.
<u>Test organisms randomly or impartially assigned to test vessels?</u>	Yes
<u>Biological observations made every 24 hours?</u>	Yes

Guideline Criteria	Reported Information
Water Parameter Measurements 1. <u>Temperature</u> Measured constantly or, if water baths are used, every 6 hrs, may not vary > 1°C 2. <u>DO and pH</u> Measured at beginning of test and every 48 h in the high, medium, and low doses and in the control	Temp. continuously monitored in one rep; measured once daily in both reps of each level DO, pH, and salinity measured once daily in both reps of each level
Chemical Analysis needed if solutions were aerated, if chemical was volatile, insoluble, or known to absorb, if precipitate formed, if containers were not steel or glass, or if flow-through system was used	Yes

10. REPORTED RESULTS

General Results:

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
<u>Recovery of Chemical</u> % of nominal	83-110%
<u>Control Mortality</u> Not more than 10% of control organisms may die or show abnormal behavior	2.5% (1 of 40)
Raw data included?	Yes
Signs of toxicity (if any) were described?	Yes

Mortality:

Concentration (ppm)		Number of Fish	Cumulative Number Dead			
Nominal	Mean Measured		Hour of Study			
			24	48	72	96
Control		20	0	1	1	1
Solvent Control		20	0	0	0	0
2.6	2.8	20	1	1	1	1
4.3	3.6	20	0	0	0	0
7.2	6.2	20	0	0	1	1
12	11	20	0	0	2	12
20	19	20	1	7	20	20

Other Findings: Sublethal effects, including lethargy and loss of equilibrium, were observed among all surviving fish exposed to the 11 mg ai/l treatment level and to one fish in the 6.2 mg ai/l treatment level.

Statistical Results

Method: Moving Average 96-hr LC_{50} : 9.8 mg ai/l

95% C.I.: 8.5-11 mg ai/l NOEC: 3.6 mg ai/l

11. REVIEWER'S VERIFICATION OF STATISTICAL RESULTS: (attached)

Method: Moving Average 96-hr LC_{50} : 9.8 mg ai/l

95% C.I.: 8.5-11.4 mg ai/l NOEC: 3.6 mg ai/l

12. REVIEWER'S COMMENTS: Deviations from guideline procedures include the following:

The size of the test fish (0.11-0.35 g) was smaller than recommended (0.5-5 g).

The report did not mention if any fish were diseased and if any treatment was made.

The study is scientifically sound and satisfies the guideline requirement for an acute marine/estuarine fish toxicity test. The EC_{50} value of 9.8 mg ai/l classifies technical metolachlor as moderately toxic to the sheepshead minnow.

THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE
 E LOWER CONCENTRATIONS HAD ZERO MORTALITY.
 FORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

KSON METOLACHLOR SHEEPSHEAD MINNOW ACUTE TEST

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
19	20	20	100	9.536742E-05
11	20	12	60.00001	25.17223
6.2	20	1	5	2.002716E-03
3.6	20	0	0	9.536742E-05
2.8	20	1	5	2.002716E-03

THE BINOMIAL TEST SHOWS THAT 6.2 AND 19 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 10.05811

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS
3	5.026151E-02		9.802751 8.525102
11.47825			

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H
5	2.539791	8.462025

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 = 5.375903
 95 PERCENT CONFIDENCE LIMITS = -3.191524 AND 13.94333

LC50 = 9.560683
 95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

LC10 = 5.549446
 95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

(4)

DATA EVALUATION RECORD
ACUTE LC₅₀ TEST WITH AN ESTUARINE/MARINE MOLLUSK
§ 72-3(B)

1. **CHEMICAL:** Metolachlor
Shaughnessey No.: 108801
2. **TEST MATERIAL:** Metolachlor technical
Purity: 97.3%
3. **STUDY TYPE:** 72-3b - Acute toxicity study to marine/estuarine mollusk.

4. **CITATION:**

Author: Dionne, E.
Title: Metolachlor technical - acute toxicity to Eastern Oyster (*Crassostrea virginica*) under flow-through conditions.
Date: 1994
Lab. Report #: 94-7-5365
Laboratory: Springborn Laboratories, Inc., Wareham, MA
Sponsor: Ciba Crop Protection, Greensboro, NC
MRID No.: 434871-02

5. **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*

7. **CONCLUSIONS:** This study is scientifically sound and fulfills the guideline requirement for an acute toxicity test with a marine/estuarine mollusk. The 96-hour EC₅₀ is 1.7 mg ai/l, which classifies technical metolachlor as moderately toxic to the Eastern oyster. The NOEC is 0.71 mg ai/l.

8. **CLASSIFICATION:** Core.

9. **MAJOR GUIDELINE DEVIATIONS:** None.

10. **MATERIALS AND METHODS:**

A. Test Organism: Eastern Oyster

Guideline Criteria	Reported Information
Species (Scientific Name)	<i>Crassostrea virginica</i>
Mean valve height (25 - 50 mm -- the long axis)	39 mm
Supplier	P. Cummins Oyster Co., Pasadena, MD
All oysters from same source (y/n)	Yes
All oysters from the same year class (y/n)	Yes

B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period (minimum 10 days)	17 days
Wild caught 7 day quarantine (y/n)	Not reported
Check for signs of disease or injury (y/n; describe)	Yes; checked for mortality and after the shell growth was grounded off, checked for stress.
If diseased it can be treated in 48-hr pretest no sign of the disease remains (Report hours prior to test in which no sign of disease or N/A)	Examined for boring sponges and/or mudworms; several opened to confirm no parasites present
Was peripheral shell growth removed prior to testing? If so how much.	Yes; 3-5 mm
Feeding during the acclimation	Fed a supplemental algal diet of <i>Isochrysis galbana</i> Parke, clone T-Iso
<3% mortality 48 hours prior to testing (% mortality, if any)	No mortality within 7 days of testing

C. Test System:

Guideline Criteria	Reported Information
Describe source of dilution water (natural unfiltered seawater)	Natural unfiltered seawater from Cape Cod Canal, Bourne, MA
Does water support test animals without observable signs of stress?	Yes
Salinity of Test water:	31‰ ppt
Water Temperature (between 15°C and 30°C -- but must be consistent)	20 ± 1°C
pH	7.9
Dissolved Oxygen (Static 1 st 48 hrs 40%; 2 nd 48 hrs 60%; Flow-through 60%) (% of lowest conc. & hour)	Measured daily; 62-88% saturation at 96 h in definitive test
Total Organic Carbon	1.3 mg/l
Test Aquaria 1. Material (glass or stainless steel) 2. a. Static volume (18.9 L (5 gal or 19000 cc) with 15 L solution) b. Static or flow-through volume (300x600x300 = 54000 cc.)	Glass aquaria 18 l 49.5 X 25.5 X 29 cm
Type of Dilution System (Reproducible supply of toxicant)	Harvard Apparatus peristaltic pump was calibrated to deliver 0.1875 ml/min of the 11 mg ai/ml stock solution.
Flow rate Consistent flow rate-meter systems calibrated before study and checked 2*24 hours - 5 to 10 vol/24 hours	6 solution volume replacements per 24 hours
Biomass Loading Rate (all oysters should be able to sit on the bottom with water flowing freely around them)	Yes
Photoperiod (16 L & 8 D)	16 hours light and 8 hours dark

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Solvents (Do not exceed 0.5 ml/L for flow-through)	0.5 ml/L acetone
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D. Test Design:

Guideline Criteria	Reported Information
<u>Range Finding Test</u> (LC ₅₀ >100 mg/L with 30 shrimp, no definitive test required.)	Tested concentrations of 0.039 to 6.3 mg ai/l. After 96-hours, growth reduction of 0-97% was observed.
<u>Definitive Test</u>	
Nominal Concentrations (control+5 treatment levels; dosage should be 60% of the next highest concentration; concentrations should be geometric series)	0.71, 1.2, 2.0, 3.3, and 5.5 mg ai/l; also negative and solvent controls
Controls (Minimum control mortality; static 10%; flow-through 5%)	No mortality reported
Number of Test Organisms; (Minimum 10/level can be divided among containers)	20 oysters in each test aquarium (40 per treatment level)
All organisms must be randomly assigned to test vessels (y/n)	Impartially assigned
Biological Observations (y/n)	Yes
Water Parameter Measurements 1. Temperature - record every 6 hrs; >1°C. 2. D.O. beginning, 48 hrs, end for control high, medium, and low dose. 3. pH beginning, 48 hrs, end for control, high, medium, and low dose.	21-22°C Yes; lowest value was 62% at 96 hours Yes; ranged was 7.5-7.9
Chemical Analysis (needed if aeration, volatile, insoluble, precipitate, not steel or glass, known to adsorb, and flow-through) (yes or no)	Yes

11. REPORTED RESULTS:

Guideline Criteria	Reported Information														
Mean Measured Concentrations	0.71, 1.1, 1.7, 2.9, 4.5														
Recovery of Chemical (% recovery)	83-100% of nominal														
Mortality & Observations (Describe observations & attach mortality tables)	No mortality at any treatment level														
Mean shell growth (mm) of control and treatment groups after 96 h exposure	<table><tr><td>water control</td><td>2.2 mm</td></tr><tr><td>solvent contr</td><td>2.4</td></tr><tr><td>0.71 mg ai/l</td><td>2.6</td></tr><tr><td>1.1</td><td>1.9</td></tr><tr><td>1.7</td><td>1.1</td></tr><tr><td>2.9</td><td>0.36</td></tr><tr><td>4.5</td><td>0.0</td></tr></table>	water control	2.2 mm	solvent contr	2.4	0.71 mg ai/l	2.6	1.1	1.9	1.7	1.1	2.9	0.36	4.5	0.0
water control	2.2 mm														
solvent contr	2.4														
0.71 mg ai/l	2.6														
1.1	1.9														
1.7	1.1														
2.9	0.36														
4.5	0.0														
Ratio of mean shell growth of treated to untreated oysters after 96 h exposure	<table><tr><td>0.71 mg ai/l</td><td>+12%</td></tr><tr><td>1.1</td><td>-18%</td></tr><tr><td>1.7</td><td>-52%</td></tr><tr><td>2.9</td><td>-84%</td></tr><tr><td>4.5</td><td>-100%</td></tr></table> note: control groups pooled	0.71 mg ai/l	+12%	1.1	-18%	1.7	-52%	2.9	-84%	4.5	-100%				
0.71 mg ai/l	+12%														
1.1	-18%														
1.7	-52%														
2.9	-84%														
4.5	-100%														
Statistical Results: EC ₅₀ : 95% CL: NOEC:	Regression analysis 1.8 mg ai/l 1.2-2.8 mg ai/l 0.71 mg ai/l														

Other findings: Reduced feeding and reduced fecal and pseudofecal production were observed at the 24-, 48-, 72-, and 96-hour intervals at the highest treatment level (4.5 mg ai/l). These effects were not observed at the four lowest treatment levels.

12. QUALITY ASSURANCE MEASURES: Yes.

13. REVIEWER'S DISCUSSION/INTERPRETATION:

Test Procedures: Test procedures followed guideline criteria.

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Statistical Analysis:

Method: EPA's Nuthatch program EC_{50} : 1.6 mg ai/l

95% CL: 1.4-1.9 mg ai/l NOEC: 0.71 mg ai/l

Slope: 4.97

Discussion/Results: The study is scientifically sound and satisfies the guideline requirement for an acute toxicity test with a marine/estuarine mollusk. The EC_{50} value of 1.6 mg ai/l classifies metolachlor technical as moderately toxic to the Eastern oyster.

14. COMPLETION DATE OF ONE-LINER FOR STUDY: 1/26/95.

!!!Williams: not implemented for 234 degrees of freedom.

Parameter	Estimate	95% Bounds Lower Upper	Std.Err.	Lower Bound /Estimate
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Goodness of fit: $\chi^2_p = 0.084$ based on $df = 3.0$ 2.3e+02
 OLS Est. var: 0.5184 small deviation

Dose	Flaps.	Obs. Mean	Pred. Mean	Obs. - Pred.	Pred. Control	% Change
0.00	40.0	39	38	-0.0871	100	0.000000
0.75	40.0	37.58	37.39	-0.1913	99.5	0.000000
1.00	40.0	37.18	36.99	-0.1919	99.5	0.000000
1.50	40.0	36.10	35.91	-0.1919	99.5	0.000000
2.00	40.0	35.10	34.91	-0.1919	99.5	0.000000
2.50	40.0	34.00	33.81	-0.1919	99.5	0.000000

Analysis of Weighted Variance

Residual	131.34	237.00	0.5516
Means Lack-of-Fit	3.667	3.000	1.2218
Within Groups	127.67	234.00	0.54569

Parameter	Estimate	Std. Err.	Correlations
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	Control Mean	=	2.4795	0.1505	1.0000	-0.71602	0.51894
Control P-50	=	0.21246	0.025414	-0.71602	1.0000	-0.71602	-0.71602
Control Sucrose	=	0.20740	0.02162	0.31894	-0.31894	0.31894	1.0000

Starting Values	2.4875	0.20020	0.23583
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File: A:\WATCH.OUT Page 4

File: A:\BUTTERFLY.001 Page 3

⑤

Program: MUTATCH
Path: C:\MUTATCH
Date: 1/22/95

Toxicity measurement for continuous endpoints, using weighted nonlinear regression, weighting proportional to predicted means.

Reference

R.D. Bruce and D.J. Versteeg. 1992. A statistical procedure for modeling continuous toxicity data. Env. For. and Chem. 11:1485-1494.

OYSTER.DAT : OYSTER SHELL DEPOSITION

Raw data:

OYSTER SHELL DEPOSITION

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5 40
6 40
7 40
8 40
9 40
10 40
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DATA EVALUATION RECORD
ACUTE LC₅₀ TEST WITH AN ESTUARINE/MARINE SHRIMP
§ 72-3(C)

1. CHEMICAL: Metolachlor
Shaughnessey No.: 108801

2. TEST MATERIAL: Metolachlor technical
Purity: 97.3%

3. CITATION

Author: Machado, M.W.
Title: Metolachlor technical - acute toxicity
to mysid shrimp (*Mysidopsis bahia*) under
flow-through conditions.
Date: 1994
Lab. Report #: 94-7-5402
Laboratory: Springborn Laboratories, Inc., Wareham,
MA
Sponsor: Ciba Crop Protection, Greensboro, NC
MRID No.: 434871-03

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 Craven

Date: 2/15/95

6. CONCLUSIONS: The 96-h LC₅₀ of 4.9 mg ai/l classifies
technical metolachlor as moderately toxic to mysid shrimp.
The NOEC is 2.3 mg ai/l.

7. ADEQUACY OF THE STUDY: Core.

9. MAJOR GUIDELINE DEVIATIONS: None.

10. MATERIALS AND METHODS:

A. Test Organisms

Guideline Criteria	Reported Information
Species Preferred species are <i>Mysidopsis bahia</i> , <i>Penaeus setiferus</i> , <i>P. duorarum</i> , <i>P. aztecus</i> and <i>Palaemonetes</i> sp.	<i>Mysidopsis bahia</i>
Age Juvenile, mysids should be ≤ 24 hours old	≤ 24 h old
Supplier	Springborn Laboratories culture facility
All shrimp are from same source?	Yes
All shrimp are from the same year class?	Yes

B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period minimum 10 days	14 days
Wild caught organisms were quarantined for 7 days?	N/A
Were there signs of disease or injury?	Not reported
If treated for disease, was there no sign of the disease remaining during the 48 hours prior to testing?	N/A
Feeding No feeding during the study and no feeding for 24 hour before the beginning of the test if organisms are over 0.5 g each.	Time of last feeding was not reported
Pretest Mortality $<3\%$ mortality 48 hours prior to testing	% mortality prior to testing was not reported

C. Test System

Guideline Criteria	Reported Information
<u>Source of dilution water</u> Soft reconstituted water or water from a natural source, not dechlorinated tap water	Filtered seawater from Cape Cod Canal, Bourne, MA
Does water support test animals without observable signs of stress?	Yes
<u>Salinity</u> 30-34 ‰ for marine (stenohaline) shrimp and 10-17 ‰ for estuarine (euryhaline) shrimp, weekly range < 6 ‰	31-32 ‰
<u>Water Temperature</u> 22 ± 1 °C	25 ± 1 °C
<u>Ph</u> 8.0-8.3 for marine (stenohaline) shrimp, 7.7-8.0 for estuarine (euryhaline) shrimp, monthly range < 0.8	7.9
<u>Dissolved Oxygen</u> Static: ≥ 60% during 1 st 48 hrs and ≥ 40% during 2 nd 48 hrs, Flow-through: ≥ 60%	80-81% at 72 h at highest dosage
<u>Total Organic Carbon</u>	1.4 mg/L
<u>Test Aquaria</u> 1. <u>Material</u> : Glass or stainless steel 2. <u>Size</u> : 19.6 L is acceptable for organisms ≥ 0.5 g (e.g. pink shrimp, white shrimp, and brown shrimp), 3.9 L is acceptable for smaller organisms (e.g. mysids and grass shrimp). 3. <u>Fill volume</u> : 15 L is acceptable for organisms ≥ 0.5 g, 2-3 L is acceptable for smaller organisms.	Glass 39 x 20 x 25 cm 7.0-11 l

Guideline Criteria	Reported Information
<u>Type of Dilution System</u> Must provide reproducible supply of toxicant	Continuous-flow serial diluter calibrated to deliver 50 ml/min of exposure solution to each replicate aquarium
<u>Flow Rate</u> Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period	6.5 vol/24 hours
<u>Biomass Loading Rate</u> Static: ≤ 0.8 g/L at $\leq 17^{\circ}\text{C}$, ≤ 0.5 g/L at $> 17^{\circ}\text{C}$; flow-through: ≤ 1 g/L/day	0.00014 g/L
<u>Photoperiod</u> 16 hours light, 8 hours dark	16 h light, 8 h dark.
<u>Solvents</u> Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests	Solvent: acetone Maximum conc.: 0.1 ml/L.

D. Test Design

Guideline Criteria	Reported Information
<u>Range Finding Test</u> If $\text{LC}_{50} > 100$ mg/L with 30 shrimp, then no definitive test is required.	3 preliminary tests - 100% mortality at 8.0 mg ai/l
<u>Nominal Concentrations of Definitive Test</u> Control & 5 treatment levels; a geometric series in which each concentration is at least 60% of the next higher one.	0.50, 1.0, 2.0, 4.0, and 8.0 mg ai/l
<u>Number of Test Organisms</u> Minimum 20/level, may be divided among containers	20
Test organisms randomly or impartially assigned to test vessels?	Yes
Biological observations made every 24 hours?	Yes

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Guideline Criteria	Reported Information
Water Parameter Measurements 1. Temperature Measured constantly or, if water baths are used, every 6 hrs, may not vary > 1°C 2. DO and pH Measured at beginning of test and ever 48 h in the high, medium, and low doses and in the control	Temp. continuously monitored in one rep. of control and once daily in all others DO, pH, and salinity measured once daily in both reps of each treatment and control
Chemical Analysis needed if solutions were aerated, if chemical was volatile, insoluble, or known to absorb, if precipitate formed, if containers were not steel or glass, or if flow-through system was used	Samples from each replicate analyzed for metolachlor at 0 h and 96 h (except for 2.0 mg ai/l dosage, which was sampled at 24 h and 96 h due to suspected sampling error at 0 h)

11. REPORTED RESULTS

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
<u>Recovery of Chemical</u>	89-120% of nominal
<u>Control Mortality</u> Not more than 10% of control organisms may die or show abnormal behavior.	None
Raw data included?	Yes
Signs of toxicity (if any) were described?	Yes

Mortality:

Concentration (ppm)		Number of Shrimp	Cumulative Number Dead			
Nominal	Mean Measured		Hour of Study			
			24	48	72	96
Control		20	0	0	0	0
Solvent Control		20	0	0	0	0
0.5	0.61	20	0	0	0	0
1.0	1.0	20	0	0	0	0
2.0	2.3	20	0	0	0	0
4.0	4.0	20	0	1	4	7
8.0	7.1	20	0	1	13	16

Other Findings: At the two highest dosages, surviving mysids exhibited dark pigmentation and were lethargic by the end of the study.

B. Statistical Results:

Method: Probit Analysis 96-h LC_{50} : 4.9 mg ai/l

95% C.I.: 4.2-5.9 mg ai/l NOEC: 2.3 MG AI/l

Probit Slope: (not reported)

12. VERIFICATION OF STATISTICAL RESULTS:

Method: Probit Analysis 96-h LC_{50} : 4.9 mg ai/l

95% CL: 4.2-5.9 mg ai/l NOEC: 2.3 mg ai/l

- 13. REVIEWER'S COMMENTS:** The study is scientifically sound and fulfills the guideline requirement for an acute toxicity test with mysid shrimp. Technical metolachlor is classified as moderately toxic to marine/estuarine mysid shrimp.

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W. ERICKSON METOLACHLOR MYSID SHRIMP ACUTE TEST

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
7.1	20	16	80	.5908966
4	20	7	35	13.1588
2.3	20	0	0	9.536742E-05
1	20	0	0	9.536742E-05
.61	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 2.3 AND 7.1 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 4.813171

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS
2	9.753802E-02	4.861711	4.172503

5.902251

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H
7	.1756815	1

GOODNESS OF FIT PROBABILITY
.8140603

SLOPE = 6.06238
95 PERCENT CONFIDENCE LIMITS = 3.521371 AND 8.603389

LC50 = 4.951274
95 PERCENT CONFIDENCE LIMITS = 4.20845 AND 5.944308

LC10/ = 3.056527
95 PERCENT CONFIDENCE LIMITS = 2.106609 AND 3.684414

(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

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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 $\rightarrow$ 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

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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

GOODNESS OF FIT PROBABILITY

.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

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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

File: A:ANABAENA.DAT

Transform: NO TRANSFORM

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

DATA EVALUATION RECORD
LEMNA GIBBA EC₅₀ TEST
GUIDELINE 123-2 (TIER II)

1. CHEMICAL: Metolachlor
Shaughnessey #: 108801

2. TEST MATERIAL: Metolachlor technical
Purity: 97.3%

3. CITATION:

Authors: Hoberg, J.R.

Title: Metolachlor technical - toxicity to
duckweed (*Lemna gibba*)

Date: 1994

Laboratory: Springborn Laboratories, Inc., Wareham,
MA

Sponsor: Ciba Crop Protection, Greensboro, NC

Lab. Report No.: 94-8-5404

MRID No.: 434871-05

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 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 duckweed. The EC₅₀ value is 0.048 mg ai/l, which classifies technical metolachlor as very highly toxic to duckweed. The NOEC is 0.0084 mg ai/l.

7. ADEQUACY OF THE STUDY: Core.

8. MAJOR GUIDELINE DEVIATIONS: None.

9. MATERIALS AND METHODS:

A. Test Organism:

Guideline Criteria	Reported Information
Species: <i>Lemna gibba</i>	<i>Lemna gibba</i> G3
Number of Plants: 5	5
Number of fronds per plant: 3	3
Nutrients:	Hoagland's 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	24-25 °C
Light Intensity: 2.0 Lux for <i>Anabaena</i> sp.; others are 4-5.0	3.8-4.8
Photoperiod:	Continuous illumination
pH: 5.0	5.0
Static or Renewal System	Static

C. Test Design:

Guideline Criteria	Reported Information
Dose range: 2X or 3X	2X
Doses: at least 5	7
Controls: negative and/or solvent	Negative
Three or more replicates per dose:	3
Duration of test: 14 days	14 days
Daily Observations?	No; 3-day intervals
Method of Observations:	Not specified
Maximum Labeled Rate:	Not reported

10. REPORTED RESULTS:

Guideline Criteria	Reported Information
Initial and 14 day frond count?	Yes
control frond count at 14 days increase $\geq 2X$?	Yes
Measured Initial Chemical Concentrations? Optional	Yes
Raw data included? (Y/N)	Excerpted
Quality Assurance Measures?	Yes

Dose Response

doses $\rightarrow$ mg ai/l	control	0.0011	0.0021	0.0042	0.0084	0.015	0.030	0.058
inhibit %	-	2	11	6	2	18	20	61
pH: 0 /14 day	5.0 6.4	5.0 6.4	5.0 6.4	4.9 6.4	4.9 6.4	4.9 6.2	4.9 6.2	4.9 6.0

Observations: Fronds at the 0.058 mg ai/l level had little root formation and were smaller and chlorotic in comparison to the controls. Fronds at 0.0084-0.030 mg ai/l were slightly chlorotic. Several fronds in control and at dosages of 0.0011-0.0042 mg ai/l were observed to be chlorotic.

Statistical Results:

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

Frond density:

EC_{50} : 0.051 mg ai/l 95% C.L.: 0.037-0.066 mg ai/l

NOEC: 0.0084 mg ai/l

Frond biomass:

EC_{50} : 0.043 mg ai/l 95% C.L.: 0.018-0.12 mg ai/l

NOEC: 0.015 mg ai/l

11. REVIEWER'S DISCUSSION/INTERPRETATION:

Test Procedures: Test procedures met guideline criteria.

Statistical Analysis: Frond density

Method: EPA Toxanol program; Williams' Test

EC₅₀: 0.048 mg ai/l 95% C.L.: 0.043-0.056 mg ai/l

NOEC: 0.0084 mg ai/l

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

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

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 4.897321E-02

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS
1	.1043968	4.897321E-02	4.396628E-02

5.662954E-02

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H
4	.7148472	9.371648

GOODNESS OF FIT PROBABILITY

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 = 1.169028
95 PERCENT CONFIDENCE LIMITS = .1806308 AND 2.157426

LC50 = 7.721045E-02
95 PERCENT CONFIDENCE LIMITS = 2.679345E-02 AND 704.6784

LC10 = 6.328546E-03
95 PERCENT CONFIDENCE LIMITS = 2.764061E-05 AND 1.640182E-02

LEMNA GIBBA FROND PRODUCTION

File: A:LEMNA.DAT

Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 1 of 2

GRP	IDENTIFICATION	N	MIN	MAX	MEAN
1	0	3	417.000	481.000	442.667
2	0.0011	3	413.000	474.000	435.667
3	0.0021	3	376.000	404.000	393.333
4	0.0042	3	380.000	478.000	417.333
5	0.0084	3	415.000	444.000	434.000
6	0.015	2	357.000	371.000	364.000
7	0.030	3	323.000	375.000	353.333
8	0.058	3	169.000	180.000	173.667

LEMNA GIBBA FROND PRODUCTION

File: A:LEMNA.DAT

Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 2 of 2

GRP	IDENTIFICATION	VARIANCE	SD	SEM
1	0	1144.333	33.828	19.531
2	0.0011	1114.333	33.382	19.273
3	0.0021	229.333	15.144	8.743
4	0.0042	2809.333	53.003	30.601
5	0.0084	271.000	16.462	9.504
6	0.015	98.000	9.899	7.000
7	0.030	732.333	27.062	15.624
8	0.058	32.333	5.686	3.283

LEMNA GIBBA FROND PRODUCTION

File: A:LEMNA.DAT

Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	0	3	442.667	442.667	442.667
2	0.0011	3	435.667	435.667	435.667
3	0.0021	3	393.333	393.333	414.889
4	0.0042	3	417.333	417.333	414.889
5	0.0084	3	434.000	434.000	414.889
6	0.015	2	364.000	364.000	364.000
7	0.030	3	353.333	353.333	353.333
8	0.058	3	173.667	173.667	173.667

WILLIAMS TEST (Isotonic regression model)

TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
0	442.667				
0.0011	435.667	0.294		1.75	k= 1, v=15
0.0021	414.889	1.166		1.84	k= 2, v=15
0.0042	414.889	1.166		1.87	k= 3, v=15
0.0084	414.889	1.166		1.88	k= 4, v=15
0.015	364.000	2.954	*	1.89	k= 5, v=15
0.030	353.333	3.751	*	1.90	k= 6, v=15
0.058	173.667	11.294	*	1.90	k= 7, v=15

s = 29.171

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



DATA EVALUATION RECORD
MARINE DIATOM 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 - toxicity to
the marine diatom, *Skeletonema costatum*
Date: 1994
Laboratory: Springborn Laboratories, Inc., Wareham,
MA
Sponsor: Ciba Crop Protection, Greensboro, NC
Lab. Report No.: 94-7-5382
MRID No.: 434871-06

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 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 marine diatom. The 120-h EC₅₀ value is 0.061 mg ai/l, which classifies technical metolachlor as very highly toxic to *Skeletonema costatum*.

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>Skeletonema costatum</i>
Number of Cells: 3,000 - 10,000	10,000
Nutrients:	AES

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	19-20 °C
Light Intensity: 2.0 Lux for <i>Anabaena</i> sp.; others are 4-5.0	3.2-4.8
Photoperiod:	continuous illumination
pH: 7.5-8.0	8.1

C. Test Design:

Guideline Criteria	Reported Information
Dose range: 2X or 3X	3X
Doses: at least 5	7
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

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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	0.0017	0.0048	0.014	0.043	0.15	0.56	1.7
inhibit %		-2	20	26	55	56	83	92
120 hr pH	8.5	8.5	8.4	8.4	8.0	8.3	8.0	7.8

Observations: Cell fragments and bloated cells were observed at the three highest concentrations. Thin cell walls also were observed at the two highest test concentrations.

Statistical Results:

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

EC_{50} : 0.0093 mg ai/l 95% C.L.: 0.0028-0.030 mg ai/l

NOEC: 0.0017 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} : 0.061 mg ai/l 95% C.L.: 0.049-0.076 mg ai/l

NOEC: 0.0017 mg ai/l

Discussion: The study is scientifically sound and fulfills the guideline requirement for a Tier 2 aquatic plant growth study with a marine diatom.

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

W. ERICKSON METOLACHLOR SKELETONEMA TOXICITY

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
1.7	100	92	92	0
.56	100	83	83	0
.15	100	56	56	0
.043	100	55	55	0
.014	100	26	26	0
.0048	100	20	20	0
.0017	100	0	0	0

THE BINOMIAL TEST SHOWS THAT .014 AND .043 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 3.565843E-02

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS
6	1.001462E-02		6.123163E-02
4.915391E-02		7.666621E-02	

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H
4	.1003167	3.651154
2.642334E-03		

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

SLOPE = .9827709
95 PERCENT CONFIDENCE LIMITS = .6714999 AND 1.294042

LC50 = 6.097744E-02
95 PERCENT CONFIDENCE LIMITS = 3.142891E-02 AND .1222098

LC10 = 3.111032E-03
95 PERCENT CONFIDENCE LIMITS = 6.803341E-04 AND 7.568381E-03

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SKELETONEMA CELL DENSITIES

File: A:SKELETON.DAT

Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 1 of 2

GRP	IDENTIFICATION	N	MIN	MAX	MEAN
1	0	3	91.000	104.000	99.000
2	0.0017	3	95.000	107.000	101.000
3	0.0048	3	76.000	81.000	79.000
4	0.014	3	65.000	78.000	73.333
5	0.043	3	41.000	48.000	44.333
6	0.15	3	37.000	49.000	43.667
7	0.56	3	9.000	21.000	16.667
8	1.7	3	7.000	9.000	8.000

SKELETONEMA CELL DENSITIES

File: A:SKELETON.DAT

Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 2 of 2

GRP	IDENTIFICATION	VARIANCE	SD	SEM
1	0	49.000	7.000	4.041
2	0.0017	36.000	6.000	3.464
3	0.0048	7.000	2.646	1.528
4	0.014	52.333	7.234	4.177
5	0.043	12.333	3.512	2.028
6	0.15	37.333	6.110	3.528
7	0.56	44.333	6.658	3.844
8	1.7	1.000	1.000	0.577

SKELETONEMA CELL DENSITIES

File: A:SKELETON.DAT

Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	0	3	99.000	99.000	100.000
2	0.0017	3	101.000	101.000	100.000
3	0.0048	3	79.000	79.000	79.000
4	0.014	3	73.333	73.333	73.333
5	0.043	3	44.333	44.333	44.333
6	0.15	3	43.667	43.667	43.667
7	0.56	3	16.667	16.667	16.667
8	1.7	3	8.000	8.000	8.000

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SKELETONEMA CELL DENSITIES

File: A:SKELETON.DAT

Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model)

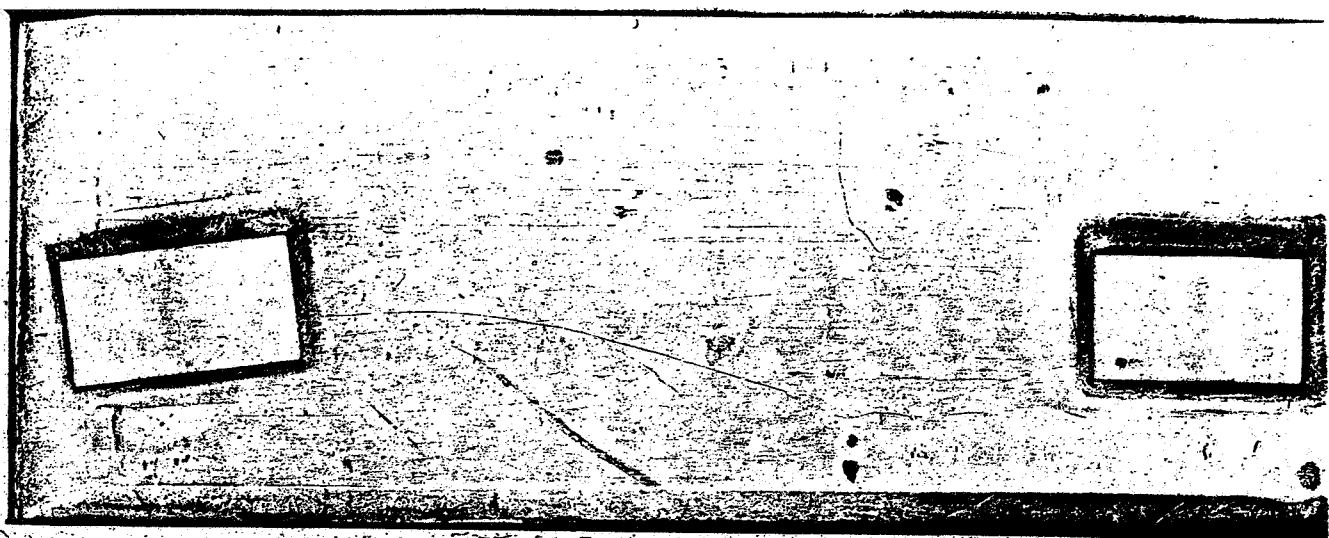
TABLE 2 OF 2

IDENTIFICATION		ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
	0	100.000				
	0.0017	100.000	0.224		1.75	k= 1,
v=16						
	0.0048	79.000	4.478	*	1.83	k= 2,
v=16						
	0.014	73.333	5.747	*	1.86	k= 3,
v=16						
	0.043	44.333	12.241	*	1.87	k= 4,
v=16						
	0.15	43.667	12.390	*	1.88	k= 5,
v=16						
	0.56	16.667	18.436	*	1.89	k= 6,
v=16						
	1.7	8.000	20.377	*	1.89	k= 7,
v=16						

s = 5.470

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

4/6



DATA EVALUATION RECORD
SEEDLING EMERGENCE EC₂₅ TEST
GUIDELINE 123-1a (TIER II)

(4)

1. CHEMICAL: Metolachlor
Shaughnessey No.: 108801

2. TEST MATERIAL: Metolachlor technical
Purity: 97.3%

3. CITATION:

Author: Chetram, R.S.
Date: 1994
Title: Tier 2 seedling emergence nontarget
phytotoxicity study using metolachlor
Sponsor: Ciba Crop Protection, Greensboro, NC
Laboratory: Pan-Agricultural Labs, Inc., Madera, CA
Lab. Report #: 94349
MRID No.: 434871-07

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: *H.T. Craven*

Date: *2/15/95*

6. CONCLUSION: The study is scientifically sound and fulfills the guideline requirement for a Tier 2 plant emergence study. Ryegrass, with an EC₂₅ of 0.02 lb ai/acre for plant dry weight and height, is the most sensitive of the four monocot species. Lettuce and cucumber, with EC₂₅ values of 0.09 lb ai/acre for dry weight, were the most sensitive of the six dicot species.

7. ADEQUACY OF THE STUDY: Core.

8. MAJOR GUIDELINE DEVIATIONS: None.

9. MATERIALS AND METHODS:

A. Test Organisms:

Guideline Criteria	Reported Information
Species: 6 dicots in 4 families including soybean and rootcrop; 4 monocots in 2 families including corn	Monocots: corn, ryegrass, oat, onion Dicots: soybean, lettuce, radish, tomato, cucumber, cabbage
Number of seeds/rep: 10 Number of reps/dose: 3	10 seeds/pot 4 reps/dose
Source & Germination % of Seed:	Yes

B. Test System:

Guideline Criteria	Reported Information
Solvent used:	50% acetone
Site of test:	Greenhouse
Soil type:	Pasteurized sandy loam soil (1.1% organic matter) with perlite added to facilitate drainage
Fertilizer:	17:9:13
Pot size:	7.5 x 7.5 x 6 cm
No. species/pot	1
Method of pesticide application:	Overhead flat-fan nozzle (TeeJet 8001E)
Watering method:	Hand watered immediately after treatment; thereafter, a HydroRain controlled irrigation system was used
Test duration:	21 days
Growth stage at application: (seed)	Seed

C. Test Design:

Guideline Criteria	Reported Information
Dose range: 2X or 3X	2X
Doses: at least 5	5
Controls: negative and solvent	Yes
Parameters observed and measured	% emergence, % survival, phytotoxicity, plant height, plant dry weight
Maximum Labeled Rate:	Not stated; highest dose rate was 4.0 lb ai/acre

10. REPORTED RESULTS:

Guideline Criteria	Reported Information
NOEC Observed?	Yes
Measured Initial Chemical Concentrations? (optional)	Yes
Raw data included? (Y/N)	Yes
Quality Assurance Measures?	Yes

Emergence, plant dry weight, plant height:

The parameter with the most sensitive EC_{25} value is tabulated below for each of the 10 species.

Species	Parameter	EC_{25}	NOEC
corn	dry weight, height	>0.025	0.025
ryegrass	dry weight, height	0.017 0.018	0.012 0.0061
oat	dry weight	0.11	0.049
onion	height	0.091	0.049
soybean	dry weight	>0.15	0.15
radish	dry weight	1.3	0.44
lettuce	dry weight, height	>0.025	0.025

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cabbage	dry weight	0.50	0.049
cucumber	dry weight	0.14	0.049
tomato	dry weight	0.33	0.025

Phytotoxicity:

Phytotoxic symptoms included stunting, leaf chlorosis, leaf curling, necrosis leaf desiccation, and plant death. NOEC values (lb ai/acre) are tabulated below.

Species	NOEC (lb ai/acre)
corn	0.025
ryegrass	0.0061
oat	0.15
onion	0.049
soybean	4.0
radish	0.44
lettuce	0.025
cabbage	0.15
cucumber	0.049
tomato	0.44

Mortality:

No mortality occurred for soybean, radish, cucumber, or cabbage. The following mortality was reported for other species:

Lettuce: 100% mortality at 0.44, 1.3, and 4.0 lb ai/acre; 65% at 0.15 lb ai/acre; 8% at 0.049 lb ai/acre.

Tomato: 26% mortality at 4.0 lb ai/acre.

Oat: 14% and 6% mortality at 4.0 and 1.3 lb ai/acre, respectively.

Ryegrass: 47% mortality at 0.44 lb ai/acre.

Corn: 49% mortality at 4.0 lb ai/acre.

Onion: 63% mortality at 4.0 lb ai/acre; 16% and 5% at 1.3 and 0.44 lb ai/acre, respectively.

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Statistical Results:

Method: Regression Analysis

Most sensitive dicot: lettuce Parameter: dry wt, ht

Most sensitive monocot: ryegrass Parameter: dry wt, ht

11. REVIEWER'S DISCUSSION/INTERPRETATION OF STUDY RESULTS:

Test Procedures: Test procedures met guideline criteria

Statistical Analysis:

Methods: EPA's Toxanol, EC₂₅, and Nuthatch programs

Most sensitive monocot: ryegrass Parameter: dry wt, ht

EC₂₅: 0.02 lb ai/acre 95% C.L.: 0.017-0.033

Slope: 6.23 NOEC: 0.0031

Most sensitive dicot: lettuce, cucumber Parameter: dry wt

EC₂₅: 0.09 lb ai/acre 95% C.L.: 0.012-0.62 (lettuce)
0.043-0.19 (cucumber)

Slope: 0.52 (lettuce) NOEC: 0.012 (lettuce)
1.01 (cucumber) 0.049 (cucumber)

Discussion: The study is scientifically sound and fulfills the guideline requirement for a Tier 2 plant emergence study.

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

Program: NUTHATCH
Path : C:\NUTHATCH
Date : 1/25/95

Toxicity measurement for continuous endpoints, using weighted nonlinear regression, weighting proportional to predicted means.

Reference

R.D. Bruce and D.J. Versteeg. 1992. A statistical procedure for modeling continuous toxicity data. Env. Tox. and Chem. 11:1485-1494.

EMMT-RYE.DAT : EMERGENCE - RYEGRASS WT

Raw data:

EMERGENCE - RYEGRASS WT

6
4
4
4
4
4
4
0
0.235
0.146
0.192
0.249
0.0031
0.156
0.161
0.189
0.186
0.0061
0.112
0.122
0.134
0.147
0.012
0.136
0.097
0.131
0.136
0.025
0.18
0.155
0.077
0.055
0.049
0.009
0.024
0.019
0.016

EMMT-RYE.DAT : EMERGENCE - RYEGRASS WT

Williams Test

[One-Sided Test for Decrease]

Dose Isotone T-bar P-value Significance
Means

0 0.205
0.0031 1.377 M.S.
0.0061 3.251 <0.005 *
0.012 3.41 <0.005 *
0.025 3.759 <0.005 *
0.049 7.984 <0.005 *

***Significant (p < 0.05) ***M.S. = Not Significant.

Estimates of EC₅₀

Parameter	Estimate	95% Bounds	Lower	Upper	Std.Err.	Lower Bound	Upper Bound	Estimate
EC1	0.013	0.0071	0.024	0.13	0.54			
EC5	0.017	0.010	0.028	0.10	0.61			
EC25	0.024	0.018	0.033	0.067	0.73			
EC50	0.031	0.025	0.039	0.046	0.80			

Slope = 6.23 Std.Err. = 1.56

!!Poor fit: p = 0.021 based on DF= 3.0 18.

EMMT-RYE.DAT : EMERGENCE - RYEGRASS WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. - Pred.	%Change
0.00	4.00	0.205	0.159	0.046	100
0.00310	4.00	0.173	0.159	0.014	100
0.00610	4.00	0.129	0.159	-0.030	100
0.0120	4.00	0.125	0.158	-0.0329	99.5
0.0250	4.00	0.117	0.115	0.00219	72.2
0.0490	4.00	0.0170	0.0173	-0.000311	10.9

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	0.26945	21.000	0.012831
Means Lack-of-Fit	0.11055	3.0000	0.036852
Within Groups	0.15890	18.000	0.0088275

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	0.15873	0.011457	1.0000
log10 EC50	-1.5076	0.045897	-0.44285
Bruce & V. SIGMA	0.16062	0.040280	0.28427
Starting Values	0.20550	-1.8365	0.62015

EMMT-ONI.DAT : EMERGENCE - ONION HT

Raw data:

EMERGENCE - ONION HT

6
38
37
37
24
6

0
122
111
172
87
118
130
109
75
124
120
109
115
112
141
87
57
112
38
118
131
146
40
77
123
141
168
110
109
51
166
153
153
147
167
118
153
34
145
0.049
102
76
105
71
57
123
55
64
127
123
139
88
150
99
125
110
88
105
58
155
145
172
153
77
103
34
143
123
117

127
126
154
140
131
153
143
164
0.15
24
138
78
23
67
61
110
51
38
84
127
50
57
12
24
40
67
30
35
25
32
133
55
27
67
112
137
72
50
58
39
64
82
30
70
41
116
0.44
26
42
30
20
36
42
29
6
36
3
20
20
10
35
20
20
52
51
45
125
64
77
80

!!!Poor fit: $p < 0.001$ based on DF= 173.
 EMHT-ONI.DAT : EMERGENCE - ONION HT

Observed vs. Predicted Treatment Group Means						
Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. Pred.	Pred. %Control	%Change
0.00	38.0	116.0	122.0	-6.91	100.0	0.00
0.0490	37.0	114.0	94.0	19.7	76.8	23.2
0.150	37.0	62.9	71.3	-8.45	58.3	41.7
0.440	37.0	41.6	47.1	-5.49	38.4	61.6
1.30	24.0	22.4	25.9	-3.57	21.2	78.8
4.00	6.00	33.7	11.3	22.3	9.27	90.7

!!!Warning: EC1 not bracketed by doses evaluated.

!!!Warning: EC5 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	3003.5	176.00	17.066
Means Lack-of-Fit	503.85	3.0000	167.95
Within Groups	2499.7	173.00	14.449

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	122.41	7.2771	1.0000
log10 EC50	-0.62997	0.098948	-0.79631
Bruce&V. SIGMA	0.93037	0.11632	0.45805

Starting Values 115.50 -0.74602 1.2086

EMHT-ONI.DAT : EMERGENCE - ONION HT

Raw data:

EMERGENCE - ONION HT

6	0.061
4	0.048
4	0.068
4	0.085
4	0.049
4	0.048
4	0.062
4	0.082
4	0.073
4	0.15
4	0.042
4	0.061
4	0.051
4	0.055

EMHT-ONI.DAT : EMERGENCE - ONION HT

Williams Test

!!!Williams: not implemented for 173 degrees of freedom.

Parameter	Estimate	95% Bounds		Std.Err.	Estimates of EC%	
		Lower	Upper		Estimate	Lower Bound
EC1	0.0016	0.00035	0.0074	0.34	0.22	
EC5	0.0069	0.0021	0.023	0.26	0.31	
EC25	0.055	0.027	0.11	0.16	0.49	
EC50	0.23	0.15	0.37	0.099	0.64	

Slope = 1.07 Std.Err. = 0.134

56

0.44
0.018
0.017
0.039
0.039
1.3
0.008
0.009
0.004
0.010
4.0
0
0
0.002
0

Residual = 21.000 0.0023095
Means Lack-of-Fit = 3.0000 0.00021087
Within Groups = 0.047866 18.000 0.0026592

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	= 0.066322	0.0047078	1.0000 -0.69306 0.49121
Log10 EC50	= -0.43894	0.078336	-0.69306 1.0000 -0.71298
Bruce&V. SIGMA	= 0.44998	0.060774	0.49121 -0.71298 1.0000
Starting Values	0.065875	-0.43638	0.42507

EMHT-RYE.DAT : EMERGENCE - RYEGRASS HT

Raw data:

EMERGENCE - RYEGRASS HT

6
39
39
33
37
40
33
0
192
191
175
129
161
166
170
169
158
142
155
65
115
160
144
132
147
139
163
140
135
165
137
215
164
175
131
197
159
99
136
134
173
132
173
99
153
154
136

EMHT-ONI.DAT : EMERGENCE - ONION WT

Williams Test

(One-Sided Test for Decrease)

Dose Isotone T-bar P-value Significance

Dose	Isotone Means	T-bar	P-value	Significance
0	0.0659	-0.04988	N.S.	
0.049	0.0559	1.762	N.S.	
0.15	0.0522	4.954	<0.005	*
0.44	0.0283	7.631	<0.005	*
1.3	0.00775	8.845	<0.005	*
4	0.0005			

***=Significant (p < 0.05) "N.S." = Not Significant.

Estimates of EC₅₀

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err.	Lower Bound /Estimate
EC1	0.033	0.012	0.087	0.20	0.38
EC5	0.066	0.030	0.15	0.17	0.45
EC25	0.18	0.11	0.31	0.11	0.59
EC50	0.36	0.25	0.53	0.078	0.69

Slope = 2.22 Std.Err. = 0.300

Goodness of fit: p = 0.97 based on Df = 3.0 18.

EMHT-ONI.DAT : EMERGENCE - ONION WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	%Pred. -Obs.	%Change
0.00	4.00	0.0655	0.0663	-0.000822	100.0
0.0490	4.00	0.0663	0.0646	0.00168	97.4
0.150	4.00	0.0522	0.0533	-0.00106	80.4
0.440	4.00	0.0283	0.0283	-0.00000	42.7
1.30	4.00	0.00775	0.00727	0.000481	11.0
4.00	4.00	0.000500	0.000686	-0.000186	1.03

Warning: EC1 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source SS DF MS

0.0031
85
151
165
147
78
130
62
104
116
150
185
169
132
146
156
178
139
93
177
172
138
144
140
187
161
145
123
135
190
187
137
176
141
178
147
128
156
137
130
0.0061
82
147
163
172
95
153
193
136
145
53
145
151
154
111
143
192
162
91
136
138
92
109
168
127
128
65
158
161

185
108
147
138
52
0.012
88
67
139
98
160
167
154
131
173
44
69
173
96
174
143
56
62
104
172
93
125
147
97
146
93
109
139
87
86
145
126
133
75
155
125
140
134
0.025
175
166
175
112
69
171
147
72
139
62
176
157
66
155
162
50
140
115
67
168
45
93
66
79
55

57

Slope = 2.9% Std.Err. = 0.385

!!!Poor fit: p = 0.028 based on DF= 3.0 2.2e+02
 ENHT-RYE.DAT : EMERGENCE - RYEGRASS HT

Observed vs. Predicted Treatment Group Means						
Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. Pred.	Pred. %Control	%Change
0.00	39.0	151.	142.	8.71	100.	0.00
0.00310	39.0	144.	142.	2.04	99.9	0.0869
0.00610	33.0	133.	140.	-7.04	98.8	1.19
0.0120	33.0	119.	130.	-11.1	91.8	8.19
0.0250	40.0	103.	95.6	7.63	67.3	32.7
0.0490	33.0	45.6	48.1	-2.49	33.9	66.1

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	2357.8	218.00	10.815
Means Lack-of-Fit	97.151	3.0000	32.384
Within Groups	2260.6	215.00	10.514

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	142.05	3.8586	1.0000
Log10 EC50	-1.4503	0.024791	-0.58905
Bruce&V. SIGMA	0.33794	0.043938	0.46596

Starting Values 150.77 -1.5026 0.45930

ENHT-LET.DAT : EMERGENCE - LETTUCE DRY WT

Raw data:

EMERGENCE - LETTUCE DRY WT

6	0.419
4	0.386
4	0.453
4	0.361
4	0.0031
4	0.348
4	0.417
4	0.334
4	0.308
4	0.0061
4	0.391
4	0.348
4	0.379
4	0.34
4	0.012
4	0.365

ENHT-RYE.DAT : EMERGENCE - RYEGRASS HT

Williams Test

!!!Williams: not implemented for 215 degrees of freedom.

Estimates of ECx

Parameter	Estimate	95% Bounds Lower Upper	Std.Err.	Lower Bound /Estimate
EC1	0.0058	0.0035 0.0097	0.11	0.60
EC5	0.0099	0.0068 0.014	0.083	0.68
EC25	0.021	0.017 0.026	0.044	0.82
EC50	0.035	0.032 0.040	0.025	0.89

38

0.354
0.417
0.368
0.025
0.314
0.3
0.397
0.294
0.049
0.371
0.301
0.319
0.264

EMWT-LET.DAT : EMERGENCE - LETTUCE DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	Significance
0	0.405			
0.0031	0.364	1.451	N.S.	
0.0061	0.364	1.451	N.S.	
0.012	0.364	1.451	N.S.	
0.025	0.326	2.802	0.007	*
0.049	0.314	3.248	<0.005	*

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC₅₀

Parameter	Estimate	95% Bounds Lower Upper	Std.Err.	Lower Bound /Estimate
EC1	6.2E-05	2.3E-08 0.16	1.6	0.00038
EC5	0.0012	1.3E-05 0.12	0.95	0.011
EC25	0.088	0.012 0.62	0.41	0.14
EC50	1.7	0.016 1.8E+02	0.97	0.0096

Slope = 0.524 Std.Err. = 0.296

Goodness of fit: p = 0.42 based on DF= 3.0 18.

EMWT-LET.DAT : EMERGENCE - LETTUCE DRY WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	0.405	0.402	0.00280	100.	0.00
0.00310	4.00	0.352	0.372	-0.0198	92.4	7.55
0.00610	4.00	0.364	0.362	0.00274	90.0	10.0
0.0120	4.00	0.376	0.350	0.0262	87.0	15.0
0.0250	4.00	0.326	0.334	-0.00801	83.2	16.8
0.0490	4.00	0.314	0.318	-0.00389	79.0	21.0

!!!Warning: EC1 not bracketed by doses evaluated.

!!!Warning: EC5 not bracketed by doses evaluated.

!!!Warning: EC25 not bracketed by doses evaluated.

!!!Warning: EC50 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	0.093592	21.000	0.0044568
Means Lack-of-Fit	0.013206	3.0000	0.0044020
Within Groups	0.080387	18.000	0.0044659

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean =	0.40195	0.021047	1.0000
log10 EC50 =	0.23008	0.97128	0.18364
Bruce&V. SIGMA =	1.9075	1.0758	0.50540
Starting Values	0.40475	-0.019712	1.7037

EMWT-TOM.DAT : EMERGENCE - TOMATO DRY WT

Raw data:

EMERGENCE - TOMATO DRY WT

6
4
4
4
4
4
4
0
0.483
0.691
0.666
0.671
0.049
0.488
0.558
0.54
0.469
0.15
0.519
0.567
0.675
0.477
0.44
0.485
0.489
0.522
0.484
1.3
0.365
0.36
0.306
0.309
4.0
0.092
0.032
0.302
0.1

EMWT-TOM.DAT : EMERGENCE - TOMATO DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	significance
0	0.628	1.72	N.S.	
0.049	0.537	1.72	N.S.	
0.15	0.537	2.506	0.013	*
0.44	0.495	5.527	<0.005	*
1.3	0.335	5.527	<0.005	*
4	0.132	5.569	<0.005	*

EMUT-CRN.DAT : EMERGENCE - CORN DRY WT

One-Sided Test for Decrease

Dose	Isotone Means	T-bar	P-value	Significance
0	1.34	1.784	0.046	*
0.049	0.964	1.784	N.S.	
0.15	0.964	1.784	N.S.	
0.44	0.964	1.784	N.S.	
1.13	0.57	3.651	<0.005	*
4	0.0998	5.882	<0.005	*

Goodness of fit: $p = 0.21$ based on DF= 3.0 18.

EMWT-CUC.DAT : EMERGENCE - CUCUMBER DRY WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. - Pred.	%Control	%Change
0.00	4.00	1.34	1.08	0.261	100.	0.00
0.0490	4.00	0.823	1.08	-0.255	100.	0.00156
0.150	4.00	1.07	1.08	-0.00872	99.7	0.281
0.440	4.00	1.00	0.996	0.00470	92.3	7.69
1.30	4.00	0.570	0.572	-0.00154	53.0	47.0
4.00	4.00	0.0998	0.0995	0.000286	9.22	90.8

Analysis of Weighted Variance

Source	SS	DF	MS
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Residual 2.2838 21.000 0.10875
 Means Lack-of-Fit 0.49453 3.0000 0.16484
 Within Groups 1.7893 18.000 0.099406

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
-----------	----------	----------	--------------

Control Mean = 1.0788 0.095368 1.0000 -0.53815 0.36235
 Log10 EC50 = 0.13998 0.10160 -0.53815 1.0000 -0.64332
 Bruce&V. SIGMA = 0.34813 0.095615 0.36235 -0.64332 1.0000

Starting Values 1.3395 -0.30092 0.98516

EMWT-CUC.DAT : EMERGENCE - CUCUMBER DRY WT

Raw data:

EMERGENCE - CUCUMBER DRY WT

6
4
4
4
4
4
4
0
2.999
2.861
4.123
3.942
0.049
3.235
3.019
3.536
3.257
0.15
1.748
2.087
2.412
2.520
0.44
1.578
2.059
1.697
1.948
1.3

0.945
1.088
1.026
1.044
4.0
0.653
0.644
0.522
0.785

EMWT-CUC.DAT : EMERGENCE - CUCUMBER DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar P-value	Significance
0	3.48	0.7279	N.S.
0.049	3.31	5.538	<0.005 *
0.15	2.19	7.154	<0.005 *
0.44	1.82	10.55	<0.005 *
1.3	1.03	12.15	<0.005 *
4	0.651		

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC₅₀

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err.	Lower Bound /Estimate
EC1	0.0021	0.00045	0.0094	0.32	0.22
EC5	0.0098	0.0030	0.032	0.25	0.30
EC25	0.090	0.043	0.19	0.16	0.47
EC50	0.42	0.26	0.68	0.10	0.62

Slope = 1.01 Std.Err. = 0.105

Goodness of fit: $p = 0.070$ based on DF= 3.0 18.
 EMWT-CUC.DAT : EMERGENCE - CUCUMBER DRY WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. - Pred.	%Control	%Change
0.00	4.00	3.48	3.60	-0.114	100.	0.00
0.0490	4.00	3.31	2.97	0.339	82.7	17.3
0.150	4.00	2.19	2.43	-0.234	67.5	32.5
0.440	4.00	1.82	1.77	0.0418	49.3	50.7
1.30	4.00	1.03	1.12	-0.0965	31.2	68.8
4.00	4.00	0.651	0.587	0.0635	16.3	83.7

!!!Warning: EC1 not bracketed by doses evaluated.

!!!Warning: EC5 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	1.0208	21.000	0.048611
Means Lack-of-Fit	0.32427	3.0000	0.10809
Within Groups	0.69655	18.000	0.038697

Directly fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std. Err.	Correlations
Control Mean	3.5951	0.20087	1.0000
Log10 EC50	-0.37312	0.099776	-0.82943
Bruce&V. SIGMA	0.99451	0.10422	0.55388
Starting Values	3.4813	-0.39239	1.0699

Program: NUTHATCH
Path : C:\NUTHATCH
Date : 1/25/95

Toxicity measurement for continuous endpoints, using weighted nonlinear regression, weighting proportional to predicted means.

Reference

R.D. Bruce and D.J. Versteeg. 1992. A statistical procedure for modeling continuous toxicity data. Env. Tox. and Chem. 11:1485-1494.

EMHT-ONI.DAT : EMERGENCE - ONION HEIGHT

Raw data:

EMERGENCE - ONION HEIGHT

6
4
4
4
4
4
3
3
0
116
101
116
129
0.049
90
107
120
142
0.15
66
53
74
62
0.44
32
28
54
51
1.3
22
29
20
19
4.0
19
56
27

One-Sided Test for Decrease

Dose Isotone T-bar P-value Significance

0 116

0.049 115 0.07498 N.S.
0.15 63.8 5.173 <0.005 *
0.44 41.3 7.423 <0.005 *
1.3 27.4 8.804 <0.005 *
4 27.4 8.151 <0.005 *

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of ECx

Parameter	Estimate	95% Bounds Lower Upper	Std.Err.	Lower Bound /Estimate
EC1	0.00039	6.1E-06 0.025	0.87	0.016
EC5	0.0026	0.00010 0.067	0.68	0.039
EC25	0.038	0.0053 0.28	0.41	0.14
EC50	0.25	0.072 0.87	0.26	0.29

Slope = 0.829 Std.Err. = 0.200

!!!Poor fit: P = 0.0028 based on DF= 3.0 17.
EMHT-ONI.DAT : EMERGENCE - ONION HEIGHT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Change XControl
0.00	4.00	116	123	-7.75	100
0.049	4.00	115	88.9	25.8	72.1
0.150	4.00	63.8	70.6	-6.87	57.3
0.440	4.00	41.3	51.7	-10.4	41.9
1.30	4.00	22.5	34.1	-11.6	27.6
4.00	3.00	34.0	19.6	14.4	15.9

!!!Warning: EC1 not bracketed by doses evaluated.

!!!Warning: EC5 not bracketed by doses evaluated.

!!!Warning: EC25 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	163.63	20.000	8.1813
Means Lack-of-Fit	90.499	3.0000	30.166
Within Groups	73.127	17.000	4.3016

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	123.25	15.686	1.0000
Log10 EC50	-0.60192	0.25942	-0.81402
Bruce&V. SIGMA	1.2064	0.29119	0.45469
Starting Values	115.50	-0.82082	1.6257

EMHT-RYE.DAT : EMERGENCE - RYEGRASS HEIGHT

Raw data:

EMERGENCE - RYEGRASS HEIGHT

6

4

68

Observed vs. Predicted Treatment Group Means	
Dose	#Reps.
Obs. Mean	Pred. Mean
%Change	%Control
0.00	4.00
0.00310	4.00
0.00610	4.00
0.0120	4.00
0.0250	4.00
0.0490	4.00

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	55.562	21.000	2.6449
Means Lack-of-Fit	10.417	3.0000	3.4724
Within Groups	45.125	18.000	2.5069

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	141.83	5.8205	1.0000
log10 EC50	-1.4528	0.037047	-0.60867
Bruce&V. SIGMA	0.33397	0.064787	0.47579
Starting Values	150.50	1.5074	0.44907

EMHT-LET.DAT : EMERGENCE - LETTUCE HEIGHT

Raw data:

EMERGENCE - LETTUCE HEIGHT

6
4
4
4
4
4
4
4
0
102
94
99
86
0.0031
90
93
86
79
0.0061
88
88
91
92
0.012
92
90
101
89
0.025
90

EMHT-RYE.DAT : EMERGENCE - RYEGRASS HEIGHT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	Significance
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0	151			
0.0031	145	0.5239	N.S.	
0.0061	134	1.241	N.S.	
0.012	119	2.728	0.008	*
0.025	104	4.104	<0.005	*
0.049	45.3	9.189	<0.005	*

***=Significant (p L/E 0.05) **N.S.=Not Significant

Estimates of ECx

Parameter	Estimate	95% Bounds	Std.Err.	Lower Bound	Upper Bound
EC1	0.0059	0.0026	0.013	0.17	0.45
EC5	0.010	0.0054	0.018	0.13	0.55
EC25	0.021	0.015	0.029	0.067	0.72
EC50	0.035	0.030	0.042	0.037	0.84

Slope = 2.99 Std.Err. = 0.581

Goodness of fit: p = 0.28 based on DF= 3.0 18.

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Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	= 95.031	2.9765	1.0000
log10 EC50	= 4.1466	6.1627	-0.21464
Bruce&V. SIGMA	= 4.0135	4.3380	0.34516
Starting Values	95.250	1.4190	2.0964

EMMT-LET.DAT : EMERGENCE - LETTUCE HEIGHT

Williams Test

(One-Sided Test for Decrease)

Dose	Isotone Means	T-bar	P-value	Significance
0	95.3			
0.0031	89.9	1.369	N.S.	
0.0061	89.9	1.369	N.S.	
0.012	89.9	1.369	N.S.	
0.025	89.5	1.476	N.S.	
0.049	84	2.887	0.0058	*

***Significant (p L/E 0.05) **N.S. = Not Significant.

Estimates of ECx

Parameter	Estimate	95% Bounds	Std.Err.	Lower Bound	Upper Bound	/Estimate
EC1	6.5E-06	1.9E-14	2.2E+03	4.1	2.9E-09	
EC5	0.0035	4.4E-06	2.8	1.4	0.0012	
EC25	28	3.8E-06	2.0E+08	3.3	1.4E-07	
EC50	1.4E+04	2.1E-09	9.2E+16	6.2	1.5E-13	

Slope = 0.249 Std.Err. = 0.269

Goodness of fit: p = 0.19 based on DF= 18.

EMMT-LET.DAT : EMERGENCE - LETTUCE HEIGHT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	95.3	95.0	0.219	100	0.00
0.00310	4.00	87.0	90.4	-3.41	95.1	4.86
0.00610	4.00	89.8	89.7	0.0868	94.4	5.65
0.0120	4.00	93.0	88.8	4.17	93.5	6.53
0.0250	4.00	89.5	87.8	1.69	92.4	7.60
0.0490	4.00	84.0	86.8	-2.76	91.3	8.70

!!!Warning: EC1 not bracketed by doses evaluated.

!!!Warning: EC25 not bracketed by doses evaluated.

!!!Warning: EC50 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	7.8402	21.000	0.37335
Means Lack-of-Fit	1.7841	3.0000	0.59470
Within Groups	6.0561	18.000	0.33645

13

0.049	3.96	0.01294	N.S.	
0.15	3.52	1.432	N.S.	
0.44	3.52	1.432	N.S.	
1.3	2.86	3.553	<0.005	*
	1.49	7.977	<0.005	*

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Parameter	Estimates of EC ₅₀			Lower Bound /Estimate
	Estimate	95% Bounds	Std.Err.	
		Lower	Upper	
C1	0.19	0.047	0.75	0.29
C5	0.42	0.15	1.2	0.21
C25	1.3	0.79	2.2	0.60
C50	2.9	2.2	3.9	0.75

Slope = 1.95 Std.Err. = 0.443

Goodness of fit: p = 0.36 based on DF= 3.0 18.

GWT-SOY.DAT : VEGETATIVE VIGOR - PLANT DRY WEIGHT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	3.96	3.79	0.167	100.	0.00
0.0490	4.00	3.96	3.79	0.164	100.	0.0262
0.150	4.00	3.55	3.77	-0.419	99.4	0.588
0.440	4.00	3.68	3.59	0.0948	94.6	5.41
1.30	4.00	2.86	2.86	0.000866	75.4	24.6
4.00	4.00	1.49	1.50	-0.00543	39.5	60.5

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	1.5839	21.000	0.075425
Means Lack-of-Fit	0.25413	3.0000	0.084709
Within Groups	1.3298	18.000	0.073877

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	3.7931	0.15558	1.0000
log10 EC50	0.46603	0.059574	-0.54037
Force & V. SIGMA	0.51200	0.11626	0.46536

tarting Values 3.9598 0.41425 0.72442

GWT-OAT.DAT : VEG VIG - OAT DRY WT

aw data:

EG VIG - OAT DRY WT

4	4	4	4	4
0	1.640			
1	2.114			
2	2.145			
3	1.763			
4	0.049			
5	2.024			
6	2.052			
7	1.753			
8	1.761			
9	0.15			
10	1.885			
11	2.206			
12	1.441			
13	1.22			
14	0.44			
15	1.488			
16	1.09			
17	0.973			
18	0.979			
19	1.3			
20	0.491			
21	0.552			
22	0.374			
23	0.596			
24	4.0			
25	0.408			
26	0.597			
27	0.228			
28	0.597			

VGWT-OAT.DAT : VEG VIG - OAT DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	Significance
0	1.92	0.09967	N.S.	
0.049	1.9	1.26	N.S.	
0.15	1.69	4.336	<0.005	*
0.44	1.13	7.682	<0.005	*
1.3	0.528	8.073	<0.005	*
4	0.457			

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC₅₀

Parameter	Estimate	95% Bounds	Std.Err.	Lower Bound /Estimate
		Lower	Upper	
EC1	0.0059	0.00066	0.053	0.46
EC5	0.023	0.0042	0.13	0.36
EC25	0.16	0.056	0.47	0.22
EC50	0.63	0.32	1.2	0.14

Slope = 1.15 Std.Err. = 0.200

Goodness of fit: p = 0.093 based on DF= 3.0 18.

VGWT-OAT.DAT : VEG VIG - OAT DRY WT

666

Program: NUTHATCH
Path: C:\NUTHATCH
Date: 1/16/95

Toxicity measurement for continuous endpoints, using weighted nonlinear regression, weighting proportional to predicted means.

Reference

R.D. Bruce and D.J. Versteeg. 1992. A statistical procedure for modeling continuous toxicity data. Env. Tox. and Chem. 11:1485-1494.

VGWT-CAB.DAT : VEG VIG - CABBAGE DRY WT

Raw data:

VEG VIG - CABBAGE DRY WT

6
4
4
4
4
4
4
0
1.116
1.654
1.550
2.579
0.049
1.506
1.565
2.502
2.261
0.15
2.056
2.79
2.184
2.134
0.44
2.031
2.785
1.776
2.154
1.3
1.939
2.007
1.952
2.836
4.0
0.358
0.481
0.282
0.455

VGWT-CAB.DAT : VEG VIG - CABBAGE DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose Isotone T-bar P-value Significance
Means

0 2.07 -1.125 N.S.
0.049 2.07 -1.125 N.S.
0.15 2.07 -1.125 N.S.
0.44 2.07 -1.125 N.S.
1.3 2.07 -1.125 N.S.
4 0.394 4.349 <0.005 *

***Significant (p L/E 0.05) "N.S."=Not Significant.

!!!Failure #3: Data not suitable for probit model fit.
Criterion is 3 or more distinct isotone means.

VGWT-SOY.DAT : VEGETATIVE VIGOR - PLANT DRY WEIGHT

Raw data:

VEGETATIVE VIGOR - PLANT DRY WEIGHT

6
4
4
4
4
4
4
0
4.359
4.017
4.297
3.166
0.049
3.865
4.563
3.616
3.779
0.15
3.493
3.781
2.7
3.432
0.44
3.615
3.733
3.783
3.6
1.3
3.142
2.857
2.355
3.092
4.0
2.226
0.969
1.136
1.644

VGWT-SOY.DAT : VEGETATIVE VIGOR - PLANT DRY WEIGHT

Williams Test

[One-Sided Test for Decrease]

Statistical Analysis:

Method: Regression Analysis

Most sensitive dicot: cucumber Parameter: dry weight

Most sensitive monocot: ryegrass Parameter: dry weight

Phytotoxicity:

Symptoms: stunting, leaf chlorosis, leaf necrosis, leaf deformity, leaf curling, restriction of leaf expansion, and leaf desiccation

NOECs (lb ai/acre): soybean, lettuce, cabbage = 1.3;
radish, corn, onion = 0.44; tomato, oat = 0.15; cucumber = 0.049; ryegrass = 0.012

Mortality: Significant mortality (65%) occurred in ryegrass at a treatment rate of 1.3 lb ai/acre and in radish, tomato, and cabbage (20-85%) at 4.0 lb ai/acre.

11. REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:

Test Procedures: Test procedures met guideline criteria.

Statistical Analysis:

Method: EPA Nuthatch program

Most sensitive monocot: ryegrass Parameter: dry weight

EC₂₅: 0.016 lb ai/acre 5% C.L.: 0.012-0.020 lb ai/acre

NOEC: 0.0031 lb ai/acre

Most sensitive dicot: cucumber Parameter: dry weight

EC₂₅: 0.030 lb ai/acre 95% C.L.: 0.020-0.046 lb ai/acre

NOEC: 0.025 lb ai/acre

Discussion: The study is scientifically sound and fulfills the guideline requirement for a Tier 2 vegetative vigor phytotoxicity study.

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

Guideline Criteria	Reported Information
Controls: negative and solvent (y/n)	Yes
Replicates per dose: (3)	4
Maximum Labeled Rate:	Not specified
Weekly Observations (y/n)	Yes

10. **REPORTED RESULTS:**

Guideline Criteria	Reported Information
NOEC Observed?	Yes
Measured Initial Chemical Concentrations? (optional)	Yes
Raw data included?	Yes
Quality Assurance Measures?	Yes

Plant dry weight and height EC_{25} values: The most sensitive EC_{25} value (lb ai/acre) is tabulated for each plant species. Values were recorded 21 days posttreatment.

species	Parameter	EC_{25}	NOEC
corn	dry weight	0.65	0.44
oat	dry weight	0.31	0.15
ryegrass	dry weight	0.02	0.012
onion	dry weight	0.58	0.44
soybean	dry weight	1.7	0.44
lettuce	dry weight	1.2	0.44
radish	dry weight, height	1.5	0.44
tomato	dry weight	1.2	0.44
cucumber	dry weight	0.055	0.025
cabbage	dry weight	2.1	1.3

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A. Test Organisms:

Guideline Criteria	Reported Information
Species: 6 dicots in 4 families including soybean and rootcrop; 4 monocots in 2 families including corn	Monocots: corn, ryegrass, oat, onion Dicots: soybean, lettuce, radish, tomato, cucumber, cabbage
Number of plants/rep: (5)	5
Source of Seed: (y/n)	Yes

B. Test System:

Guideline Criteria	Reported Information
Solvent used:	50% acetone
Site of test:	Greenhouse
Soil type:	Sterilized sandy loam (1.1% organic matter), containing perlite (1:4) to facilitate drainage
Pot size:	7.5 x 7.5 x 6.0 cm
Fertilizer:	17:9:13
Planting Method: species/pot	1 seed/pot
Method of Application:	Spray booth equipped with an overhead nozzle (TeeJet 8001-E)
Watering Method: under foliage	By hand to prevent wetting of leaves during first 48 h; thereafter, by low volume mist nozzles
No. Days After Application: (14)	21
Growth Stage/Application: Past first true leaf stage (y/n)	Yes

C. Test Design:

Guideline Criteria	Reported Information
Dose range: (2X-3X)	3X
Doses: (≥ 5)	5

(2)

**DATA EVALUATION RECORD
VEGETATIVE VIGOR EC₂₅ TEST
GUIDELINE 123-1b (TIER II)**

1. **CHEMICAL:** Metolachlor
Shaughnessey #: 108801

2. **TEST MATERIAL:** Metolachlor
Purity: 97.3%

3. **CITATION:**

Author: Chetram, R.S.

Date: 1994

Title: Tier 2 vegetative vigor nontarget
phytotoxicity study using metolachlor

Sponsor: Ciba Crop Protection, Greensboro, NC

Laboratory: Pan-Agricultural Labs, Inc., Madera, CA

Lab. Report #: 94350

MRID No.: 434871-08

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:** This study is scientifically sound and satisfies the guideline requirement for a Tier 2 vegetative vigor test. Ryegrass, with an EC₂₅ value of 0.016 lb ai/acre for dry plant weight was the most sensitive monocot. Cucumber was the most sensitive dicot; the EC₂₅ value was 0.030 lb ai/acre for dry plant weight. The NOEC values were 0.0031 and 0.025 lb ai/acre for ryegrass and cucumber, respectively.

7. **ADEQUACY OF THE STUDY:** Core.

8. **MAJOR GUIDELINE DEVIATIONS:** None.

9. **MATERIALS AND METHODS:**

Residual	1.3007	21.000	0.061940
Means Lack-of-Fit	0.41357	3.0000	0.13786
Within Groups	0.88718	18.000	0.049288

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
-----------	----------	----------	--------------

Control Mean	=	2.8339	0.16377	1.0000	-0.65315	0.62128
log10 EC50	=	0.39739	0.099409	-0.65315	1.0000	-0.24457
BrucellaV. SIGMA	=	0.82666	0.19444	0.62128	-0.24457	1.0000

Starting values	2.8443	0.39654	1.0751
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VGUT-CU2.DAT : VEG VIG - CUCUMBER DRY WT (2ND TEST)

Raw data:

VEG VIG - CUCUMBER DRY WT (2ND TEST)

2.891
3.060
3.850
3.781
0.0031
3.338
2.958
3.541
3.818
0.0061
3.121
2.22
3.177
3.393
0.012
2.591
2.814
3.254
2.908
0.025
2.22
2.793
3.173
2.484
0.049
3.316
2.985
3.342
1.114

GH1-CH2 DAT : VEG VIG : CILIMBER DRY HT (2ND TEST)

Williams Test

One-Sided Test for Decrease

Isotone	T-bar	p-value	Significance
100	100	0.0001	*
101	101	0.0001	*
102	102	0.0001	*
103	103	0.0001	*
104	104	0.0001	*
105	105	0.0001	*
106	106	0.0001	*
107	107	0.0001	*
108	108	0.0001	*
109	109	0.0001	*
110	110	0.0001	*
111	111	0.0001	*
112	112	0.0001	*
113	113	0.0001	*
114	114	0.0001	*
115	115	0.0001	*
116	116	0.0001	*
117	117	0.0001	*
118	118	0.0001	*
119	119	0.0001	*
120	120	0.0001	*
121	121	0.0001	*
122	122	0.0001	*
123	123	0.0001	*
124	124	0.0001	*
125	125	0.0001	*
126	126	0.0001	*
127	127	0.0001	*
128	128	0.0001	*
129	129	0.0001	*
130	130	0.0001	*
131	131	0.0001	*
132	132	0.0001	*
133	133	0.0001	*
134	134	0.0001	*
135	135	0.0001	*
136	136	0.0001	*
137	137	0.0001	*
138	138	0.0001	*
139	139	0.0001	*
140	140	0.0001	*
141	141	0.0001	*
142	142	0.0001	*
143	143	0.0001	*
144	144	0.0001	*
145	145	0.0001	*
146	146	0.0001	*
147	147	0.0001	*
148	148	0.0001	*
149	149	0.0001	*
150	150	0.0001	*
151	151	0.0001	*
152	152	0.0001	*
153	153	0.0001	*
154	154	0.0001	*
155	155	0.0001	*
156	156	0.0001	*
157	157	0.0001	*
158	158	0.0001	*
159	159	0.0001	*
160	160	0.0001	*
161	161	0.0001	*
162	162	0.0001	*
163	163	0.0001	*
164	164	0.0001	*
165	165	0.0001	*
166	166	0.0001	*
167	167	0.0001	*
168	168	0.0001	*
169	169	0.0001	*
170	170	0.0001	*
171	171	0.0001	*
172	172	0.0001	*
173	173	0.0001	*
174	174	0.0001	*
175	175	0.0001	*
176	176	0.0001	*
177	177	0.0001	*
178	178	0.0001	*
179	179	0.0001	*
180	180	0.0001	*
181	181	0.0001	*
182	182	0.0001	*
183	183	0.0001	*
184	184	0.0001	*
185	185	0.0001	*
186	186	0.0001	*
187	187	0.0001	*
188	188	0.0001	*
189	189	0.0001	*
190	190	0.0001	*
191	191	0.0001	*
192	192	0.0001	*
193	193	0.0001	*
194	194	0.0001	*
195	195	0.0001	*
196			

Means

Model	Log-likelihood	AIC	BIC	Bayes factor
Model 1	0	3.43	-0.1158	N.S.
Model 2	0.0031	3.43	-0.1158	N.S.
Model 3	0.0061	3.43	1.697	N.S.
Model 4	0.012	2.9	1.697	N.S.
Model 5	0.025	2.9	5.036	<0.005
Model 6	0.049	1.94		

"#="Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC%

Parameter	Estimate	95% Bounds Lower Upper	Std.Err.	Lower Bound /Estimate
EC1	0.0053	0.0011	0.025	0.33
EC5	0.011	0.0038	0.032	0.21
EC25	0.030	0.020	0.046	0.34
EC50	0.062	0.043	0.089	0.66
			0.076	0.69

Slope = 2.19 Std.Err. = 0.755

Goodness of fit: $\chi^2 = 0.37$ based on DF = 3.0

Goodness of fit: $\chi^2 = 0.57$ based on 1 df =
VGWT-CU2-DAT : VEG VIG - CUCUMBER DRY WT (2ND TEST)

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	Pred. %Control	%Change
0.00	4.00	3.40	3.39	0.00134	100.	0.00
0.00310	4.00	3.41	3.39	0.0273	99.8	0.226
0.00610	4.00	3.48	3.35	0.131	98.6	1.40
0.0120	4.00	2.89	3.19	-0.298	94.0	6.01
0.0250	4.00	2.92	2.73	0.188	80.4	19.6
0.0490	4.00	1.94	1.99	-0.0500	58.6	41.4

!!!Warning: EC50 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	1.2211	21.000	0.058146
Means Lack-of-Fit	0.18988	3.0000	0.063294
Within Groups	1.0312	18.000	0.057288

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	= 3.3942	0.1404	1.0000
Log10 EC50	= -1.2104	0.076100	-0.055538
Bruce&V. SIGMA	= 0.45715	0.15774	0.55972
Starting Values	3.4290	-1.1926	0.59490

WEIGHT-ON-TAT : VEG VIG - ONION DRY WT

Raw data:

VEG VIC - UNION DEP. LT.

0111

EC25 0.96 1.9 0.14 0.50
EC50 3.1 4.6 0.080 0.58

Slope = 1.31 Std.Err. = 0.291

Goodness of fit: p = 0.65 based on DF= 3.0 18.
VGWT-LET.DAT : VEG VIG - LETTUCE DRY WT

Observed vs. Predicted Treatment Group Means						
Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	2.82	2.76	0.0563	100	0.00
0.0490	4.00	2.75	2.74	0.00959	99.1	0.904
0.150	4.00	2.60	2.65	-0.0432	95.8	4.20
0.440	4.00	2.28	2.40	-0.118	86.8	13.2
1.30	4.00	2.05	1.91	0.138	69.1	30.9
4.00	4.00	1.19	1.23	-0.0421	44.4	55.6

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	0.91062	21.000	0.043363
Means Lack-of-Fit	0.076779	3.000	0.025593
Within Groups	0.83384	18.000	0.046324

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	2.7644	0.12450	1.0000
log10 EC50	0.49537	0.079932	-0.53451
Bruce&V. SIGMA	0.76360	0.16964	0.59632
Starting Values	2.8207	0.46854	0.80996

VGWT-CRN.DAT : VEG VIG - CORN DRY WT

Raw data:

VEG VIG - CORN DRY WT

6 2.636
4 2.865
4 2.947
4 2.266
4 0.049
4 2.751
4 3.115
4 3.041
4 2.209
4 0.15
4 2.63
4 3.078
4 3.405
4 3.188

0.44
2.537
2.133
1.689
2.646
1.3
2.133
1.56
1.358
1.492
4.0
1.344
1.154
1.072
1.368

VGWT-CRN.DAT : VEG VIG - CORN DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	Significance
0	2.84			
0.049	2.84	-0.6884	N.S.	
0.15	2.84	-0.6884	N.S.	
0.44	2.25	1.775	N.S.	
1.3	1.64	4.331	<0.005	*
4	1.23	5.998	<0.005	*

***=Significant (p L/E 0.05) **N.S.=Not Significant.

Estimates of ECx

Parameter	Estimate	95% Bounds	Std.Err.	Lower Bound	Upper Bound	/Estimate
EC1	0.030	0.0029	0.31	0.49	0.097	
EC5	0.11	0.020	0.61	0.36	0.18	
EC25	0.69	0.29	1.7	0.18	0.42	
EC50	2.5	1.6	4.0	0.099	0.62	

Slope = 1.21 Std.Err. = 0.286

Goodness of fit: p = 0.070 based on DF= 3.0 18.
VGWT-CRN.DAT : VEG VIG - CORN DRY WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	2.68	2.86	-0.185	100	0.00
0.0490	4.00	2.78	2.81	-0.0299	98.1	1.92
0.150	4.00	3.08	2.67	0.410	93.1	6.93
0.440	4.00	2.25	2.35	-0.0933	82.0	18.0
1.30	4.00	1.64	1.82	-0.181	63.4	36.6
4.00	4.00	1.23	1.15	0.0832	40.2	59.8

Warning: EC1 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
--------	----	----	----

13

.69
.3
538
588
331
636
0
355
473

Parameter Estimate Std. Err. Correlations
Control Mean = 4.0368 0.22541 1.0000 -0.44608 0.27289
log10 EC50 = 0.18388 0.05471 -0.44608 1.0000 -0.58871
Bruce&V. SIGMA = 0.26112 0.044823 0.27289 -0.58871 1.0000
Starting Values 4.4390 0.018526 0.49675

GWT-TOM.DAT : VEG VIG - TOMATO DRY WT

Williams Test

One-Sided Test for Decrease

ose Isotone Means T-bar P-value Significance

.049 4.44 0.7573 N.S.
.15 4.04 0.7573 N.S.
.44 3.48 1.83 N.S.
.3 2.52 3.668 <0.005 *
0.207 8.103 <0.005 *

***Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC%

Parameter	Estimate	95% Bounds Lower	Upper	Std. Err.	Lower Bound /Estimate
1	0.38	0.19	0.75	0.14	0.50
5	0.57	0.33	0.98	0.11	0.58
25	1.0	0.71	1.5	0.076	0.69
50	1.5	1.2	2.0	0.054	0.77

Slope = 3.83 Std.Err. = 0.657

Goodness of fit: p = 0.54 based on DF= 3.0 18.

GWT-TOM.DAT : VEG VIG - TOMATO DRY WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	4.44	4.04	0.402	100.	0.00
0.0490	4.00	3.92	4.04	-0.115	100.	5.32e-07
0.150	4.00	4.17	4.04	0.128	100.	0.00568
0.440	4.00	3.48	3.96	-0.476	98.1	1.92
1.30	4.00	2.52	2.44	0.0786	60.6	39.4
4.00	4.00	0.207	0.221	-0.0135	5.46	94.5

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	3.9294	21.000	0.18711
Means Lack-of-Fit	0.43168	3.0000	0.14389
Within Groups	3.4977	18.000	0.19432

Directly-fitted parameters and Asymptotic Covariances

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Observed vs. Predicted Treatment Group Means						
Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	Pred. %Control	%Change
0.00	4.00	0.514	0.517	-0.00295	100.	0.00
0.0490	4.00	0.510	0.509	0.000706	98.4	1.58
0.150	4.00	0.485	0.489	-0.00468	94.6	5.37
0.440	4.00	0.465	0.446	0.0195	86.2	13.8
1.30	4.00	0.352	0.370	-0.0175	71.5	28.5
4.00	4.00	0.269	0.264	0.00492	51.0	49.0

!!!Warning: EC1 not bracketed by doses evaluated.

!!!Warning: EC50 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	0.16165	21.000	0.0076974
Means Lack-of-Fit	0.0073505	3.0000	0.0024502
Within Groups	0.15430	18.000	0.0085720

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	= 0.51720	0.024586	1.0000 -0.36731 0.63555
Log10 EC50	= 0.62487	0.10281	-0.36731 1.0000 0.21710
Bruce&V. SIGMA	= 0.89982	0.22896	0.63555 0.21710 1.0000
Starting Values	0.51425	0.62791	0.90753

VGWT-TOM.DAT : VEG VIG - TOMATO DRY WT

Raw data:

VEG VIG - TOMATO DRY WT

6	3.778
4	5.286
4	4.090
4	4.602
4	0.049
4	3.629
4	5.326
4	3.012
4	3.721
4	0.15
4	3.603
4	3.112
4	4.419
4	5.526
4	0.44
4	3.715
4	3.163
4	3.366

VGWT-ONI.DAT : VEG VIG - ONION DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose Isotone T-bar P-value Significance

Dose	Isotone Means	T-bar	P-value	Significance
0	0.514	0.1116	N.S.	
0.049	0.51	0.7314	N.S.	
0.15	0.485	1.209	N.S.	
0.44	0.465	4.017	<0.005	*
1.3	0.352	6.087	<0.005	*
4	0.269			

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err.	Lower Bound /Estimate
EC1	0.034	0.0028	0.41	0.52	0.083
EC5	0.14	0.024	0.81	0.37	0.17
EC25	1.0	0.47	2.3	0.17	0.45
EC50	4.2	2.6	6.9	0.10	0.61

Slope = 1.11 Std.Err. = 0.283

Goodness of fit: p = 0.83 based on DF= 3.0 18.

VGWT-ONI.DAT : VEG VIG - ONION DRY WT

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File:A:\NUTATCH.OUT Page 4
 Residual 1144.5 117.00 9.7820
 Means Lack-of-Fit 19.664 3.0000 6.5546
 Within Groups 1124.8 114.00 9.8670

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	= 402.44	7.5795	1.0000
Log10 EC50	= 0.95209	0.14212	0.25759
Bruce&V. SIGMA	= 0.52565	0.19285	0.45736
Starting Values	406.50	1.0501	0.69183

VGHT-LET.DAT : VEG VIG - LETTUCE HT

Raw data:

VEG VIG - LETTUCE HT

6
20
20
20
20
20
20
0
156
165
203
166
202
125
130
130
129
120
125
133
144
165
151
138
152
130
134
133
0.049
130
135
130
134
158
184
165
198
149
148
120
149
145
146
150
135
134
140

350
0
341
320
334
205
210
404
384
291
280
351
329
319
300

VGHT-SOY.DAT : VEG VIG - SOYBEAN HT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	Significance
0	407			
0.049	406	0.04694	N.S.	
0.15	406	0.04694	N.S.	
0.44	386	1.108	N.S.	
1.3	384	1.199	N.S.	
4	300	5.705	<0.005	*

***Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC₅₀

Parameter	Estimate	95% Bounds	Lower	Upper	Std.Err.	Lower Bound	Upper Bound
EC1	0.54	0.12	2.3	0.32	0.23		
EC5	1.2	0.51	3.0	0.19	0.41		
EC25	4.0	3.1	5.1	0.054	0.78		
EC50	9.0	4.7	17.	0.14	0.52		

Slope = 1.90 Std.Err. = 0.698

Goodness of fit: p = 0.58 based on DF= 3.0 1.1e+02

VGHT-SOY.DAT : VEG VIG - SOYBEAN HT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. - Pred.	%Control	%Change
0.00	20.0	407.	402.	4.06	100.	0.00
0.0490	20.0	397.	402.	-5.18	100.	0.00842
0.150	20.0	414.	402.	11.7	100.	0.0364
0.440	20.0	386.	400.	-14.0	99.4	0.639
1.30	20.0	384.	380.	4.01	94.5	5.54
4.00	20.0	300.	301.	-0.577	74.7	25.3

!!!Warning: EC50 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
146			
150			
135			
134			
140			

77

File:A:\NUTHATCH.OUT Page 6

File
97 121 149 130 142 135 136 132 145 143
99 0 100 78 121 122 101

VGHT-LET.DAT : VEG VIG - LETTUCE HT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	Significance
0	148			
0.049	148	-0.1517	N.S.	
0.15	148	-0.1517	N.S.	
0.44	143	-0.5445	N.S.	
1.3	137	1.47	N.S.	
4	114	4.871	<0.005	*

##=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC%

Parameter	Estimate	95% Bounds	Std.Err.	Lower Bound /Estimate
		Lower Upper		
EC1	0.29	0.045	1.9	0.41
EC5	0.91	0.30	2.7	0.24
EC25	4.5	3.1	6.6	0.69
EC50	14.	5.2	37.	0.37

slope = 1.39 Std.Err. = 0.506

Goodness of fit: $p = 0.96$ based on $Df = 3.0$ $1.1e+02$
 VGH-T-LET.DAT : VEG VIG - LETTUCE HT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	Pred. %Control	%Change
0.00	20.0	147.	147.	-0.821	100.	0.00
0.090	20.0	148.	147.	0.177	100.	0.0325
0.150	20.0	149.	147.	1.74	99.7	0.313
0.2440	20.0	143.	145.	-1.73	98.1	1.86
1.30	20.0	137.	136.	0.587	92.4	7.64
20.0	20.0	114.	114.	-0.115	77.4	22.6

!!!Warning: EC25 not bracketed by doses evaluated.

Warning: EC50 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	2357.3	117.00	20.148
Means Lack-of-Fit	41.141	3.0000	13.714
Within Groups	2316.1	114.00	20.317

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	281.69	8.5878	1.0000
log10 EC50	0.36752	0.034985	-0.48161
Bruce&V. SIGMA	0.27779	0.038055	0.34440

Starting Values 299.65 0.21373 0.49928

VGHT-RAD.DAT : VEG VIG - RADISH HT

Raw data:

VEG VIG - RADISH HT

6
20
20
20
20
20
20
0
120
126
134
130
110
110
144
131
110
120
110
105
119
120
149
151
136
121
139
137
0.049
110
115
120
141
136
140
136
137
131
120
120
129
135
163

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	2357.3	117.00	20.148
Means Lack-of-Fit	41.141	3.0000	13.714
Within Groups	2316.1	114.00	20.317

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	281.69	8.5878	1.0000
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Starting Values 299.65 0.21373 0.49928

VGHT-RAD.DAT : VEG VIG - RADISH HT

Raw data:

VEG VIG - RADISH HT

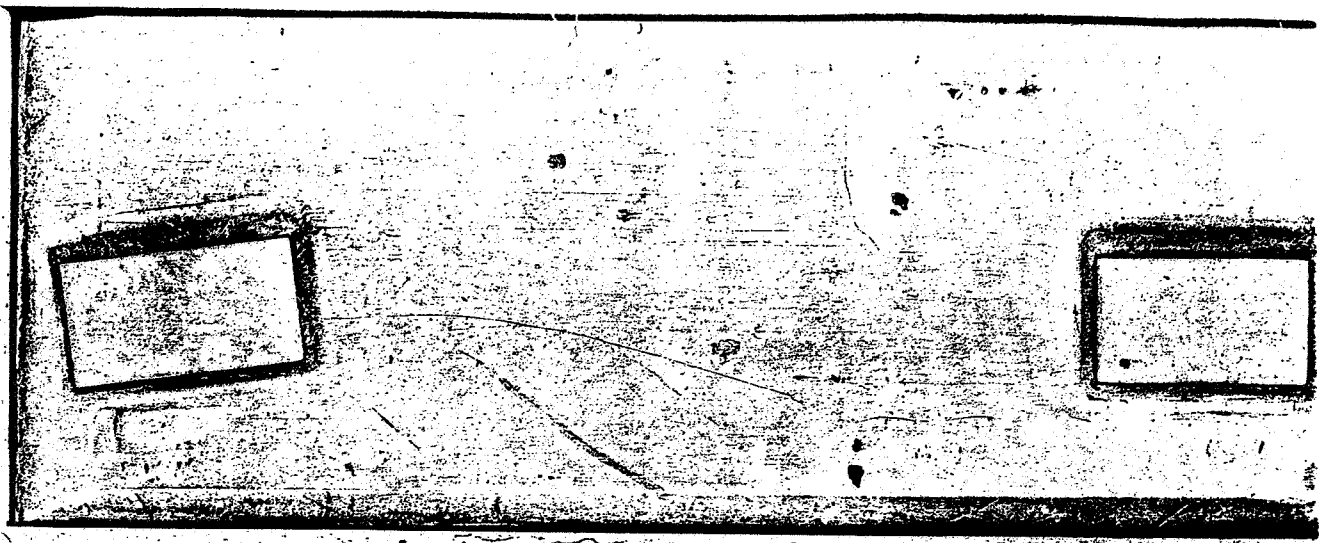
6
20
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105
119
120
149
151
136
121
139
137
0.049
110
115
120
141
136
140
136
137
131
120
120
129
135
163

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	1994.2	117.00	17.045
Means Lack-of-Fit	8.9394	3.0000	2.9798
Within Groups	1985.3	114.00	17.415

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	125.51	5.2414	1.0000
EC50	0.31341	0.044746	-0.45442
sigma	0.24874	0.058728	0.31944
Starting Values	127.95	0.27598	0.27501



Observed vs. Predicted Treatment Group Means						
Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	Pred. %Control	%Change
0.00	4.00	1.92	2.02	-0.107	100.	0.00
0.0490	4.00	1.90	1.82	0.0801	89.9	10.1
0.150	4.00	1.69	1.54	0.146	76.3	23.7
0.440	4.00	1.13	1.15	-0.0211	57.0	43.0
1.30	4.00	0.528	0.724	-0.196	35.8	64.2
4.00	4.00	0.457	0.559	0.0985	17.8	82.2

!!!Warning: EC1 not bracketed by doses-evaluated.

!!!Warning: EC5 not bracketed by doses evaluated.

Analysis of Weighted Variance				
Source	SS	DF	MS	
Residual	1.4101	21.000	0.067146	
Means Lack-of-Fit	0.41340	3.0000	0.13780	
Within Groups	0.99667	18.000	0.055370	

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	2.0226	0.16857	1.0000
log10 EC50	-0.20235	0.13934	-0.82432
Bruce&V. SIGMA	0.86975	0.15161	0.59862
Starting Values	1.9155	-0.21160	0.94123

VGWT-RAD.DAT : VEG VIG -RADISH DRY WT

Raw data:

VEG VIG -RADISH DRY WT

6
4
4
4
4
4
4
0
1.308
1.279
1.216
1.217
0.049
1.089
1.294
1.324
0.15
1.372
1.278
1.257
1.288
0.44
1.405
1.287
1.131
1.188

VGWT-RAD.DAT : VEG VIG -RADISH DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar	P-value	Significance
0	1.26			
0.049	1.26	-0.0265	N.S.	
0.15	1.26	-0.0265	N.S.	
0.44	1.25	0.03111	N.S.	
1.3	1.17	1.206	N.S.	
4	0.111	15.82	<0.005	*

***=Significant (p L/E 0.05) "N.S."=Not significant.

Estimates of EC₅₀

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err.	Lower Bound /Estimate
EC1	0.93	0.48	1.8	0.14	0.52
EC5	1.2	0.71	2.1	0.11	0.59
EC25	1.8	1.2	2.6	0.076	0.69
EC50	2.3	1.8	3.0	0.053	0.78

Slope = 5.79 Std.Err. = 1.26

Goodness of fit: p = 0.96 based on DF= 3.0 18.

VGWT-RAD.DAT : VEG VIG -RADISH DRY WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	Pred. %Control	%Change
0.00	4.00	1.25	1.26	-0.00880	100.	0.00
0.0490	4.00	1.22	1.26	-0.0389	100.	1.77e-16
0.150	4.00	1.30	1.26	0.0429	100.	2.51e-10
0.440	4.00	1.25	1.26	-0.00311	100.	0.00334
1.30	4.00	1.17	1.17	2.58e-06	93.0	7.02
4.00	4.00	0.111	0.111	-1.21e-07	8.82	91.2

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	0.61764	21.000	0.029411
Means Lack-of-Fit	0.010702	3.0000	0.0035672
Within Groups	0.60694	18.000	0.033719

Directly-fitted parameters and Asymptotic Covariances

4.00 4.00 1.26 1.47 -0.204 71.3

Warning: EC1 not bracketed by doses evaluated.

Warning: EC5 not bracketed by doses evaluated.

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	1.4838	21.000	0.070658
Means Lack-of-Fit	0.43995	3.0000	0.14665
Within Groups	1.0439	18.000	0.057993

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	5.1126	0.29672	1.0000 -0.84553 0.49420
Log10 EC50	-0.20948	0.13770	-0.84553 1.0000 -0.50270
Bruce&V. SIGMA	1.4435	0.19774	0.49420 -0.50270 1.0000
Starting Values	5.1815	-0.24948	1.4942

VGWT-RY2.DAT : VEG VIG - RYEGRASS DRY WT (TEST 2)

Raw data:

VEG VIG - RYEGRASS DRY WT (TEST 2)

6	0.582
4	0.506
4	0.526
4	0.593
4	0.0031
4	0.517
4	0.489
4	0.452
4	0.583
4	0.0061
4	0.409
4	0.534
4	0.436
4	0.533
4	0.012
4	0.453
4	0.525
4	0.404
4	0.474
4	0.025
4	0.227
4	0.206
4	0.37
4	0.256
4	0.049
4	0.115
4	0.086
4	0.1
4	0.125

4.27
3.781
3.672
0.15
2.736
3.36
3.447
2.933
0.44
2.69
2.776
2.949
3.538
1.3
2.007
1.965
2.716
2.846
4.0
1.269
1.801
0.841
1.143

VGWT-CUC.DAT : VEG VIG - CUCUMBER DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose Isotone T-bar P-value Significance

Dose	Isotone Means	T-bar	P-value	Significance
0	5.18			
0.049	3.88	4.823	<0.005	*
0.15	3.12	7.654	<0.005	*
0.44	2.99	8.139	<0.005	*
1.3	2.38	10.38	<0.005	*
4	1.26	14.54	<0.005	*

***Significant (p L/E 0.05) **N.S.=Not Significant.

Estimates of EC%

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err.	Lower Bound /Estimate
EC1	0.00027	2.0E-05	0.0036	0.54	0.074
EC5	0.0026	0.00036	0.019	0.41	0.14
EC25	0.066	0.021	0.20	0.23	0.32
EC50	0.62	0.32	1.2	0.14	0.52

Slope = 0.693 Std.Err. = 0.0949

Goodness of fit: p = 0.090 based on DF= 3.0 18.

VGWT-CUC.DAT : VEG VIG - CUCUMBER DRY WT

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	5.18	5.11	0.0689	100.	0.00
0.0490	4.00	3.88	3.97	-0.0910	77.7	22.3
0.150	4.00	3.12	3.40	-0.280	66.5	33.5
0.440	4.00	2.76	2.76	0.224	54.1	45.9
1.30	4.00	2.38	2.10	0.280	41.1	58.9

VGWT-RY2.DAT : VEG VIG - RYGRASS DRY WT (TEST 2)

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar Means	P-value	Significance
0	0.552	1.095	N.S.	
0.0031	0.51	1.946	0.039	*
0.0061	0.478	2.316	0.019	*
0.012	0.464	7.574	<0.005	*
0.025	0.265	11.75	<0.005	*
0.049	0.107			

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC%

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err.	Lower Bound /Estimate
EC1	0.0045	0.0025	0.0080	0.12	0.56
EC5	0.0075	0.0048	0.012	0.094	0.64
EC25	0.016	0.012	0.020	0.056	0.77
EC50	0.026	0.022	0.030	0.034	0.85

Slope = 3.04 Std.Err. = 0.388

Goodness of fit: p = 0.48 based on DF= 3.0 18.

VGWT-RY2.DAT : VEG VIG - RYGRASS DRY WT (TEST 2)

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	%Control	%Change
0.00	4.00	0.552	0.524	0.0281	100.	0.00
0.00310	4.00	0.510	0.522	-0.0121	99.7	0.252
0.00610	4.00	0.478	0.509	-0.0310	97.2	2.80
0.0120	4.00	0.464	0.443	0.0211	84.6	15.4
0.0250	4.00	0.265	0.272	-0.00740	52.0	48.0
0.0490	4.00	0.107	0.105	0.00135	20.1	79.9

Analysis of Weighted Variance

Source	SS	DF	MS
Residual	0.15665	21.000	0.0074593
Means Lack-of-Fit	0.019611	3.0000	0.0065369
Within Groups	0.13703	18.000	0.0076130

Directly-fitted parameters and Asymptotic Covariances

Parameter	Estimate	Std.Err.	Correlations
Control Mean	0.52369	0.019858	1.0000
log10 EC50	-1.5858	0.033446	-0.66639
Bruce&V. SIGMA	0.32906	0.041976	0.49115

Starting Values	0.55175	-1.6453	0.41290
Control Mean	0.52369	0.019858	1.0000
log10 EC50	-1.5858	0.033446	-0.66639
Bruce&V. SIGMA	0.32906	0.041976	0.49115

Starting Values

VGWT-LET.DAT : VEG VIG - LETTUCE DRY WT

Raw data:

VEG VIG - LETTUCE DRY WT

6
4
4
4
4
4
4
0
2.722
2.698
3.192
2.671
0.049
2.65
3.171
2.275
2.9
0.15
2.661
2.426
2.808
2.525
0.44
2.311
2.259
2.301
2.251
1.3
1.968
1.806
2.194
2.23
4.0
1.752
1.267
0.645
1.082

VGWT-LET.DAT : VEG VIG - LETTUCE DRY WT

Williams Test

[One-Sided Test for Decrease]

Dose	Isotone Means	T-bar Means	P-value	Significance
0	2.82	2.75	0.3568	N.S.
0.049	2.75	2.6	1.073	N.S.
0.15	2.28	2.687	0.0088	*
0.44	2.05	3.835	<0.005	*
1.3	1.19	8.127	<0.005	*

***=Significant (p L/E 0.05) "N.S."=Not Significant.

Estimates of EC%

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err.	Lower Bound /Estimate
EC1	0.052	0.0075	0.37	0.41	0.14
EC5	0.17	0.042	0.71	0.29	0.24

Starting Values