Data Evaluation Report on the acute toxicity of Penoxsulam metabolite on the Freshwater Alga, *Pseudokirchneriella subcapitata*

PMRA Submission #:{.....} EPA MRID #: 45831117

Data Requirement:	PMRA DATA CODE	{.....}
	EPA DP Barcode	D288160
	OECD Data Point	{.....}
	EPA MRID	45831117
	EPA Guideline	123-2

Test material:	Penoxsulam	Purity: 100%
Common name:	XDE-638 Metabolite (BST)	
Chemical name:	IUPAC: Not reported	
	CAS name: Not reported	
	CAS No.: Not reported	
	Synonyms: Not reported	

Primary Reviewer: Rebecca Bryan
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Signature: Rebecca Bryan
Date: 12/29/03

QC Reviewer: Dana Worcester
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Signature: Dana Worcester
Date: 12/29/03

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Date: {.....} Goodyear

Secondary Reviewer(s): {.....}
{EPA/OECD/PMRA}

Date: {.....}

Company Code {.....} [For PMRA]
Active Code {.....} [For PMRA]
EPA PC Code ~~199031~~ 119031
Date Evaluation Completed: {dd-mmm-yyyy}

CITATION: Hoberg, J.R. 2002. XDE-638 Metabolite (BST): Toxicity to the Freshwater Green Alga, *Pseudokirchneriella subcapitata*. Unpublished study performed by Springborn Laboratories, Inc., Wareham, Massachusetts. Laboratory Project Identification No. 12550.6169/Dow Study No. 011235. Study submitted by The Dow Chemical Company for Dow AgroSciences, LLC Midland, Michigan. Experimental start date December 20, 2001 and experimental termination date December 24, 2001. The final report issued January 29, 2002.

EXECUTIVE SUMMARY:

Acute toxicity of Penoxsulam metabolite to the Freshwater Alga, *Pseudokirchneriella subcapitata* MRID 45831117
In a 96-hour acute toxicity study, cultures of *Pseudokirchneriella subcapitata* were exposed to Penoxsulam, as XDE-638 Metabolite (BST), under static conditions. The nominal concentrations were 0 (negative and solvent controls), 0.10, 0.26, 0.64, 1.6, 4.0, and 10 mg a.i./L. The mean measured concentrations were <0.011-0.012 (LOQ, negative and solvent controls), 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L. The 96-hour cell density percent inhibitions were 4, 4, -4, 2, 5, and 7% for the 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L treatment groups, respectively. The 72-hour growth rate percent inhibitions were 0, 2, 1, 2, -1, and 12% for the 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L treatment groups, respectively. The 72-hour area under the growth curve (biomass) percent inhibitions were 3, 10, 4, 6, -4, and 36% for the 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L treatment groups, respectively. There were significant effects on growth rate and biomass in the 9.6 mg a.i./L treatment group; however, no reductions exceeded 50% for any endpoint. Based on these results, the EC_{50} was >9.6 mg a.i./L and the NOAEC was 3.9 mg a.i./L (based on biomass and growth rate). The EC_{05}: could not be determined.

The study is scientifically sound and satisfies the U.S. EPA Guideline Subdivision J, §123-2 for an aquatic nonvascular plant study with *Pseudokirchneriella subcapitata*. This study is classified as Core.

Results Synopsis

Test Organism: *Pseudokirchneriella subcapitata*
Test Type: Static

Cell Density:
NOAEC: 9.6 mg a.i./L
EC_{05}: 8.1 mg a.i./L 95% C.I.: 0.44-150 mg a.i./L
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A
Slope: 0.903±2.27

Growth rate:
NOAEC: 3.9 mg a.i./L
EC_{05}: could not determine 95% C.I.: N/A
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A
Slope: N/A

Area Under the Growth Curve (Biomass):
NOAEC: 3.9 mg a.i./L
EC_{05}: could not determine 95% C.I.: N/A
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A
Slope: N/A

Endpoints Affected: Growth rate and biomass

I. MATERIALS AND METHODS

GUIDELINES FOLLOWED: OECD Guideline for Testing of Chemicals #201, Alga, Growth Inhibition Test (OECD, 1984); The EC Guideline Annex V-Method C.3., Algal Inhibition Test (EC, 1997); and U.S. EPA FIFRA Subdivision J Guideline, §123-2 (U.S. EPA, 1982). There were no notable deviations from U.S. EPA Guideline, §123-2.

COMPLIANCE: Signed and dated GLP, Quality Assurance and No Data Confidentiality statements were provided.

A. MATERIALS:

1. Test Material Penoxsulam, XDE-638 Metabolite (BST)

Description: Not reported

Lot No./Batch No. : E1167-37

Purity: 100%

Stability of Compound
Under Test Conditions: The mean measured concentrations of XDE-638 Metabolite (BST) were 81-98% of nominal at hour 0 and 81-100% of nominal at hour 96 (Table 3, p. 25).

(OECD requires water solubility, stability in water and light, pKa, Pow, vapor pressure of test compound)

Storage conditions of test chemicals: The test substance was stored at room temperature.

2. Test organism:

Name: *Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum*)

EPA requires a nonvascular species: For tier I testing, only one species, S. capricornutum, *to be tested; for tier II testing, S. costatum,* A. flos-aquae, S. capricornutum, *and a freshwater diatom is tested*

OECD suggests the following species are considered suitable: S. capricornutum, S. subspicatus, *and* C. vulgaris. *If other species are used, the strain should be reported*

Strain: 1648
Source: Originally from Carolina Biological Supply Company, Burlington, NC. Current in-house laboratory cultures.
Age of inoculum: 3 days old
Method of cultivation: Algal Assay Procedure (AAP) medium (Table 1, p. 23).

B. STUDY DESIGN:

a) Range-finding Study: No range-finding study was reported.

b) Definitive Study

Table 1 . Experimental Parameters

Parameter	Details	Remarks *Criteria*
Acclimation period: culturing media and conditions: (same as test or not)	Continuous Algal Assay Procedure (AAP) medium (Table 1, p. 23); same as test.	Inoculum used in test was taken from stock culture and transferred to fresh medium three days before testing. *EPA recommends two week acclimation period.*
health: (any toxicity observed)	Not reported	*OECD recommends an amount of algae suitable for the inoculation of test cultures and incubated under the conditions of the test and used when still exponentially growing, normally after an incubation period of about 3 days. When the algal cultures contain deformed or abnormal cells, they must be discarded.*
Test system static/static renewal: renewal rate for static renewal:	Static	
Incubation facility	Environmental chamber	
Duration of the test	96 hours	*EPA requires: 96 - 120 hours* *OECD: 72 hours*
Test vessel material: (glass/polystyrene) size: fill volume:	Glass Erlenmeyer flasks with stainless steel caps 250 ml 100 ml	*OECD recommends 250 ml conical flasks are suitable when the volume of the test solution is 100 ml or use a culturing apparatus.*
Details of growth medium name: pH at test initiation: pH at test termination: Chelator used: Carbon source:	Algal Assay Procedure (AAP) medium 6.8-7.1 9.3-9.8 Yes $NaHCO_3$	*OECD recommends the medium pH after equilibration with air is ~8 with less than .001 mmol/l of chelator if used.*

Parameter	Details	Remarks *Criteria*
Salinity **(for marine algae):**	N/A	*EPA recommends 20X-AAP medium.*
If non-standard nutrient medium was used, detailed composition provided (Yes/No)	N/A	
Dilution water source: type: pH: salinity **(for marine algae):** water pretreatment (if any): Total Organic Carbon: particulate matter: metals: pesticides: chlorine:	 Deionized water Sterilized 7.5 ± 0.1 N/A None 1.0 mg a.i./L Not reported Not detected Not detected Not reported	*EPA pH:* Skeletonema costatum= *~8.0 Others = ~7.5 from beginning to end of the test. EPA salinity: 30-35 ppt. EPA is against the use of dechlorinated water.* *OECD: pH is measured at beginning of the test and at 72 hours, it should not normally deviate by more than one unit during the test.*
Indicate how the test material is added to the medium (added directly or used stock solution)	Stock solutions	
Aeration or agitation	Agitation, 100 rpm	*EPA recommends agitation only for* Selenastrum *at 100 cycles per min and* Skeletonema *at ~60 cycles per min. Aeration is not recommended.*
Initial cells density	Approximately 10,000 cells/ml	*EPA requires an initial number of 3,000 - 10,000 cells/ml. For* Selenastrum capricornutum, *cell counts on day 2 are not required.* *OECD recommends that the initial cell concentration be approximately 10,000 cells/ml for* S. capricornutum *and* S. subspicatus. *When other species are used the biomass should be comparable.*
Number of replicates control: solvent control: treated ones:	 3 3 3	*EPA requires a negative and/or solvent control with 3 or more*

Parameter	Details	Remarks *Criteria*
		replicates per doses. Navicula *sp. tests should be conducted with four replicate.* *OECD preferably three replicates at each test concentration and ideally twice that number of controls. When a vehicle is used to solubilize the test substance, additional controls containing the vehicle at the highest concentration used in the test cultures should be included in the test.*
Test concentrations nominal: measured:	0 (negative and solvent controls), 0.10, 0.26, 0.64, 1.6, 4.0, and 10 mg a.i./L <0.011-0.012 (LOQ, negative and solvent controls), 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L	*EPA requires at least 5 test concentrations, with each at least 60% of the next higher one.* *OECD recommends at least five concentrations arranged in a geometric series, with the lowest concentration tested should have no observed effect on the growth of the algae. The highest concentration tested should inhibit growth by at least 50% relatively to the control and, preferably, stop growth completely.*
Solvent (type, percentage, if used)	Dimethylformamide (DMF), 0.10 ml/L	
Method and interval of analytical verification	HPLC; 0 and 96 hours	
Test conditions temperature: photoperiod: light intensity and quality:	23-25°C Continuous 3900-4300 lux, fluorescent lighting	*EPA temperature:* Skeletonema*: 20°C, Others: 24-25°C; EPA photoperiod:* S. costatum *14 hr light/ 10 hr dark, Others: Continuous; EPA light:* Anabaena*: 2.0 Klux (±15%), Others: 4 - 5 Klux (±15%)* *OECD recommended the temperature in the range of 21*

Parameter	Details	Remarks
		Criteria
		to25°C maintained at ± 2°C and continuous uniform illumination provided at approximately 8000 Lux measured with a spherical collector.
Reference chemical {if used) name: concentrations:	N/A	
Other parameters, if any	None	

2. Observations:

Table 2: Observation parameters

Parameters	Details	Remarks/Criteria
Parameters measured including the growth inhibition/other toxicity symptoms	Cell count, area under the growth curve (biomass), and growth rate	Biomass and growth rates were determined for up to 72 hours of exposure. *EPA recommends the growth of the algae expressed as the cell count per ml, biomass per volume, or degree of growth as determined by spectrophotometric means.*
Measurement technique for cell density and other end points	Haemocytometer with a compound microscope	*EPA recommends the measurement technique of cell counts or chlorophyll a* *OECD recommends the electronic particle counter, microscope with counting chamber, fluorimeter, spectrophotometer, and colorimeter. (note: in order to provide useful measurements at low cell concentrations when using a spectrophotometer, it may be necessary to use cuvettes with a light path of at least 4 cm).*

Parameters	Details	Remarks/Criteria
Observation intervals	Every 24 hours	*EPA and OECD: every 24 hours.*
Other observations, if any	None	
Indicate whether there was exponential growth in the control	Yes, dilution water and solvent control group cell densities at test termination were 93X and 92X greater, respectively than the dilution water and solvent control group cell densities at test initiation.	*EPA requires control cell count at termination to be ≥2X initial count or by a factor of at least 16 during the test.* *OECD: cell concentration in control cultures should have increased by a factor of at least 16 within three days.*
Were raw data included?	Yes	

II. RESULTS and DISCUSSION:

A. INHIBITORY EFFECTS:

The 96-hour cell density percent inhibitions were 4, 4, -4, 2, 5, and 7% for the 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L treatment groups, respectively. The 72-hour growth rate percent inhibitions were 0, 2, 1, 2, -1, and 12% for the 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L treatment groups, respectively. The 72-hour area under the growth curve (biomass) percent inhibitions were 3, 10, 4, 6, -4, and 36% for the 0.093, 0.22, 0.58, 1.4, 3.9, and 9.6 mg a.i./L treatment groups, respectively. The growth rate and area under the growth curve were significantly reduced in the 9.6 mg a.i./L treatment group after 72 hours.

Table 3: Effect of Penoxsulam, XDE-638 Metabolite (BST), on freshwater alga (*Pseudokirchneriella subcapitata*)

Treatment mean measured and nominal concentrations [a] (mg a.i./L)	Initial cell density (cells/ml)	Mean Cell density (cells/ml) at		
		24 hours	96 hours	
			cell count	% inhibition[b]
Dilution water control	~10,000	42,500	930,000	--
Solvent control	~10,000	38,300	920,000	--
0.10 (0.093)	~10,000	25,000	880,000	4
0.27 (0.22)	~10,000	25,800	880,000	4
0.64 (0.58)	~10,000	26,700	960,000	-4
1.7 (1.4)	~10,000	30,800	900,000	2
4.2 (3.9)	~10,000	31,700	870,000	5

10 (9.6)	~10,000	22,500	860,000	7
Reference chemical (if used)	N/A	N/A	N/A	N/A

[a] The nominal test concentrations are presented in parentheses.
[b] The % inhibition was based on pooled controls.

Table 4: Effect of Penoxsulam, XDE-638 Metabolite (BST), on freshwater alga *(Pseudokirchneriella subcapitata)*

Mean Measured and Nominal Treatment Concentrations [a] (mg a.i./L)	Initial cell density (cells/ml)	Mean Growth Rate per day	% inhibition (Mean Growth Rate per day)[b]	Mean Area Under Growth Curve	% inhibition (Mean Area Under Growth Curve)[b]
Dilution water control	~10,000	1.26	--	403,000	--
Solvent control	~10,000	1.33	--	484,000	--
0.10 (0.093)	~10,000	1.30	0	432,000	3
0.27 (0.22)	~10,000	1.27	2	400,000	10
0.64 (0.58)	~10,000	1.29	1	427,000	4
1.7 (1.4)	~10,000	1.27	2	417,000	6
4.2 (3.9)	~10,000	1.31	-1	461,000	-4
10 (9.6)	~10,000	1.15*	12	282,000	36*
Reference chemical (if used)	Not reported	Not reported	Not reported	Not reported	Not reported

[a] The nominal test concentrations are presented in parentheses.
[b] The percent inhibitions for growth rates and biomass were compared to the pooled control.
* Significantly reduced compare to the pooled control (Williams' Test)

Table 5: Statistical endpoint values.

Statistical Endpoint	Biomass [a]	Growth rate[a]	Cell density[b]
NOAEC (mg a.i./L)	3.9	3.9	9.6
EC_{05} (mg a.i./L)	ND	ND	8.1
EC_{50} (mg a.i./L)	>9.6	>9.6	>9.6
Reference chemical, if used	N/A	N/A	N/A

NOAEC IC_{50}/EC_{50}			

N/A = Not applicable.
[a] The biomass and growth values based on 72 hour data.
[b] The cell density values based on 96 hour data.

B. REPORTED STATISTICS:

Statistical Method: The formulas used to calculate the area under the growth curve and growth rates are presented on pages 15-16. A t-test was used to compare the dilution water (negative) and solvent controls. The controls were pooled for all statistical analyses. The data was analyzed for normality using the Shapiro-Wilk's Test and homogeneity of variance using Bartlett's Test. The Williams' test was used to compare the treatment groups to the pooled control. The cell density values based on 96 hour data. The biomass and growth values based on 72 hour data. The reported statistics were based on the mean measured test concentrations.

Cell Density:
NOAEC/EC_{05}: 9.6 mg a.i./L
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A

Growth rate:
NOAEC/EC_{05}: 3.9 mg a.i./L
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A

Area Under the Growth Curve (Biomass):
NOAEC/EC_{05}: 3.9 mg a.i./L
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A

Endpoint(s) Affected: Growth rates and biomass.

C. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: Cell density, biomass, and growth rate data satisfied the assumptions of ANOVA. The NOAEC values for these endpoints were determined using ANOVA (cell density), followed by Bonferroni's (biomass and growth rate) multiple comparison test via TOXSTAT statistical software. The EC_{05} value for cell density was determined using the Probit method via Nuthatch statistical software; the Probit method could not be used to determine the EC_{05} values for biomass or growth rate because of the non-linear pattern of these responses. The reviewer used the mean measured concentrations to calculate toxicity values.

Cell Density:
NOAEC: **9.6** mg a.i./L
EC_{05}: **8.1 mg a.i./L** **95% C.I.**: 0.44-150 mg a.i./L
EC_{50}: >9.6 mg a.i./L **95% C.I.: N/A**
Slope: 0.903±2.27

Growth rate:
NOAEC: **3.9** mg a.i./L
EC_{05}: **could not determine** **95% C.I.**: N/A
EC_{50}: >9.6 mg a.i./L **95% C.I.: N/A**

Slope: N/A

Area Under the Growth Curve (Biomass):
NOAEC: 3.9 mg a.i./L
EC_{05}: could not determine 95% C.I.: N/A
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A
Slope: N/A

Endpoints Affected: Growth rate and biomass

D. STUDY DEFICIENCIES:

N/A

E. REVIEWER'S COMMENTS:

The reviewer's conclusions were identical to the study author's.

The study followed the U.S. EPA Good Laboratory Practice with the exception of the collection of samples for routine water contaminant screening analyses.

F. CONCLUSIONS: The study is scientifically sound and satisfies the guidelines for an aquatic nonvascular plant study with *Pseudokirchneriella subcapitata* [§123-2]. This study is classified as Core. There were significant effects on growth rate and biomass in the 9.6 mg a.i./L treatment group; however, no reductions exceeded 50% for any endpoint. Based on these results, the EC_{50} was >9.6 mg a.i./L and the NOAEC of Penoxsulam metabolite (BST) was 3.9 mg a.i./L (based on biomass and growth rate).

Cell Density:
NOAEC: 9.6 mg a.i./L
EC_{05}: 8.1 mg a.i./L 95% C.I.: 0.44-150 mg a.i./L
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A
Slope: 0.903±2.27

Growth rate:
NOAEC: 3.9 mg a.i./L
EC_{05}: could not determine 95% C.I.: N/A
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A
Slope: N/A

Area Under the Growth Curve (Biomass):
NOAEC: 3.9 mg a.i./L
EC_{05}: could not determine 95% C.I.: N/A
EC_{50}: >9.6 mg a.i./L 95% C.I.: N/A
Slope: N/A

Endpoints Affected: Growth rate and biomass

III. REFERENCES:

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APPENDIX I. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:
cell density
File: 1117cd Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	6	240.500	40.083	0.394
Within (Error)	17	1729.333	101.725	
Total	23	1969.833		

Critical F value = 2.70 (0.05,6,17)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

cell density
File: 1117cd Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	92.333	92.333		
2	0.093	87.667	87.667	0.654	
3	0.22	87.667	87.667	0.654	
4	0.58	96.000	96.000	-0.514	
5	1.4	90.000	90.000	0.327	
6	3.9	87.000	87.000	0.748	
7	9.6	86.333	86.333	0.841	

Bonferroni T table value = 2.65 (1 Tailed Value, P=0.05, df=17,6)

cell density
File: 1117cd Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	6			
2	0.093	3	18.935	20.5	4.667
3	0.22	3	18.935	20.5	4.667
4	0.58	3	18.935	20.5	-3.667
5	1.4	3	18.935	20.5	2.333

6	3.9	3	18.935	20.5	5.333
7	9.6	3	18.935	20.5	6.000

cell density
File: 1117cd Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	6	92.333	92.333	92.333
2	0.093	3	87.667	87.667	90.444
3	0.22	3	87.667	87.667	90.444
4	0.58	3	96.000	96.000	90.444
5	1.4	3	90.000	90.000	90.000
6	3.9	3	87.000	87.000	87.000
7	9.6	3	86.333	86.333	86.333

cell density
File: 1117cd Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	92.333				
0.093	90.444	0.265		1.74	k= 1, v=17
0.22	90.444	0.265		1.82	k= 2, v=17
0.58	90.444	0.265		1.85	k= 3, v=17
1.4	90.000	0.327		1.87	k= 4, v=17
3.9	87.000	0.748		1.87	k= 5, v=17
9.6	86.333	0.841		1.88	k= 6, v=17

s = 10.086
Note: df used for table values are approximate when v > 20.

Estimates of EC%

Parameter	Estimate	95% Bounds Lower	Upper	Std.Err. /Estimate	Lower Bound
EC5	**8.1**	**0.44**	**1.5E+02**	**0.61**	**0.055**
EC10	20.	0.12	3.5E+03	1.1	0.0057
EC25	96.	0.00029	3.2E+07	2.7	3.0E-06
EC50	5.3E+02	2.3E-07	1.2E+12	4.5	4.3E-10

Slope = 0.903 Std.Err. = 2.27

Goodness of fit: p = 0.80 based on DF= 4.0 17.

1117CD : cell density

Observed vs. Predicted Treatment Group Means

Dose	#Reps.	Obs. Mean	Pred. Mean	Obs. -Pred.	Pred. %Control	%Change
0.00	6.00	92.3	91.0	1.29	100.	0.00
0.0930	3.00	87.7	91.0	-3.35	100.	0.0343
0.220	3.00	87.7	90.9	-3.28	99.9	0.112
0.580	3.00	96.0	90.7	5.29	99.6	0.372
1.40	3.00	90.0	90.1	-0.149	99.0	0.987
3.90	3.00	87.0	88.6	-1.60	97.3	2.68
9.60	3.00	86.3	85.8	0.521	94.2	5.75

!!!Warning: EC10 not bracketed by doses evaluated.
!!!Warning: EC25 not bracketed by doses evaluated.

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

biomass
File: 1117b Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	6	665.906	110.984	2.704
Within (Error)	17	697.733	41.043	
Total	23	1363.640		

Critical F value = 2.70 (0.05,6,17)
Since F > Critical F REJECT Ho:All groups equal

biomass
File: 1117b Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	44.367	44.367		
2	0.093	43.167	43.167	0.265	

3	0.22	39.967	39.967	0.971
4	0.58	42.667	42.667	0.375
5	1.4	41.733	41.733	0.581
6	3.9	46.133	46.133	-0.390
7	9.6	28.167	28.167	3.576 *

Bonferroni T table value = 2.65 (1 Tailed Value, P=0.05, df=17,6)

biomass
File: 1117b Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	6			
2	0.093	3	12.027	27.1	1.200
3	0.22	3	12.027	27.1	4.400
4	0.58	3	12.027	27.1	1.700
5	1.4	3	12.027	27.1	2.633
6	3.9	3	12.027	27.1	-1.767
7	9.6	3	12.027	27.1	16.200

biomass
File: 1117b Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	6	44.367	44.367	44.367
2	0.093	3	43.167	43.167	43.167
3	0.22	3	39.967	39.967	42.625
4	0.58	3	42.667	42.667	42.625
5	1.4	3	41.733	41.733	42.625
6	3.9	3	46.133	46.133	42.625
7	9.6	3	28.167	28.167	28.167

biomass
File: 1117b Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

ISOTONIZED	CALC.	SIG	TABLE	DEGREES OF

IDENTIFICATION	MEAN	WILLIAMS	P=.05	WILLIAMS	FREEDOM
GRPS 1&2 POOLED	44.367				
0.093	43.167	0.265		1.74	k= 1, v=17
0.22	42.625	0.384		1.82	k= 2, v=17
0.58	42.625	0.384		1.85	k= 3, v=17
1.4	42.625	0.384		1.87	k= 4, v=17
3.9	42.625	0.384		1.87	k= 5, v=17
9.6	28.167	3.576	*	1.88	k= 6, v=17

s = 6.406
Note: df used for table values are approximate when v > 20.

EC_x

!!!Failure#1: near-singular matrix, model possibly unsuitable.

growth rate
File: 1117g Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	6	0.059	0.010	3.333
Within (Error)	17	0.044	0.003	
Total	23	0.103		

Critical F value = 2.70 (0.05,6,17)
Since F > Critical F REJECT Ho:All groups equal

growth rate
File: 1117g Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	1.298	1.298		
2	0.093	1.303	1.303	-0.129	
3	0.22	1.270	1.270	0.732	
4	0.58	1.293	1.293	0.129	
5	1.4	1.273	1.273	0.645	
6	3.9	1.307	1.307	-0.215	
7	9.6	1.147	1.147	3.916	*

Bonferroni T table value = 2.65 (1 Tailed Value, P=0.05, df=17,6)

growth rate
File: 1117g Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	6			
2	0.093	3	0.103	7.9	-0.005
3	0.22	3	0.103	7.9	0.028
4	0.58	3	0.103	7.9	0.005
5	1.4	3	0.103	7.9	0.025
6	3.9	3	0.103	7.9	-0.008
7	9.6	3	0.103	7.9	0.152

growth rate
File: 1117g Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	6	1.298	1.298	1.300
2	0.093	3	1.303	1.303	1.300
3	0.22	3	1.270	1.270	1.286
4	0.58	3	1.293	1.293	1.286
5	1.4	3	1.273	1.273	1.286
6	3.9	3	1.307	1.307	1.286
7	9.6	3	1.147	1.147	1.147

growth rate
File: 1117g Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	1.300				
0.093	1.300	0.046		1.74	k= 1, v=17
0.22	1.286	0.349		1.82	k= 2, v=17
0.58	1.286	0.349		1.85	k= 3, v=17
1.4	1.286	0.349		1.87	k= 4, v=17

3.9	1.286	0.349		1.87	k= 5, v=17
9.6	1.147	4.231	*	1.88	k= 6, v=17

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s = 0.051
Note: df used for table values are approximate when v > 20.

EC_x

!!!Failure#1: near-singular matrix, model possibly unsuitable.