

MRID No.: 436781-61

**DATA EVALUATION RECORD
ALGAE OR DIATOM EC₅₀ TEST
GUIDELINE 123-2 (TIER II)**

1. **CHEMICAL:** Azoxystrobin **PC Code No.:** 128810

2. **TEST MATERIAL:** ICIA5504 technical **Purity:** 96.2%

3. **CITATION**

Authors: D.V. Smyth, S.J. Kent, S.A. Sankey, J.M. Shearing

Title: ICIA5504: Toxicity to the Green Alga (*Selenastrum capricornutum*)

Study Completion Date: February 26, 1994

Laboratory: Brixham Environmental Laboratory, Brixham Devon, UK

Sponsor: Zeneca Ag Products, Wilmington, DE

Laboratory Report ID: BL5101/B

MRID No.: 436781-61

4. **REVIEWED BY:**

William Erickson
Biologist
EEB/EFED/EPA

Signature:

W. Erickson

Date:

4/01/96

5. **APPROVED BY:**

Harry Craven
Section Head 4
EEB/EFED/EPA

Signature:

H. Craven
6/21/96

Date:

6. **STUDY PARAMETERS:**

Definitive Test Duration: 120 hours

Type of Concentrations: Nominal

7. **CONCLUSIONS:** This study is scientifically sound and fulfills the guideline requirement for an algal toxicity test.

Results Synopsis

EC₅₀: 106 ppb

EC₀₅: 23 ppb

95% C.I.: 92 - 121 ppb

Probit Slope: N/A

8. **ADEQUACY OF THE STUDY:**

A. Classification: Core

B. Rationale: N/A

①

C. Repairability: N/A

9. GUIDELINE DEVIATIONS: None noted.10. SUBMISSION PURPOSE:11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
<u>Species</u> <i>Skeletonema costatum</i> <i>Anabaena flos-aquae</i> <i>Selenastrum capricornutum</i> <i>Navicula pelliculosa</i>	<i>Selenastrum capricornutum</i>
<u>Initial Number of Cells</u> 3,000 - 10,000 cells/ml	3,000 cells/ml
<u>Nutrients</u> Standard formula, e.g. 20XAAP	ATCC 22662

B. Test System

Guideline Criteria	Reported Information
<u>Solvent</u>	Acetone (0.1 mL/L)
<u>Temperature</u> <i>Skeletonema</i> : 20°C Others: 24-25°C	24°C
<u>Light Intensity</u> <i>Anabaena</i> : 2.0 KLux ($\pm 15\%$) Others: 4.0-5.0 KLux ($\pm 15\%$)	4.38 KLux
<u>Photoperiod</u> <i>Skeletonema</i> : 14 h light, 10 h dark or 16 h light, 8 h dark Others: Continuous	Continuous
<ph </ph <i>Skeletonema</i> : approx. 8.0 Others: approx. 7.5	Initial 7.3 - 7.4 Final 7.4 - 9.2

C. Test Design

MRID No.: 436781-61

DATA EVALUATION RECORD
ALGAE OR DIATOM EC₅₀ TEST
GUIDELINE 123-2 (TIER II)

1. **CHEMICAL:** ~~Sulfentrazone~~ ^{Azoxystrobin}

PC Code No.: ~~129081~~ ¹²⁸⁸¹⁰

2. **TEST MATERIAL:** ICIA5504 technical

Purity: 96.2%

3. **CITATION**

Authors: D.V. Smyth, S.J. Kent, S.A. Sankey, J.M. Shearing

Title: ICIA5504: Toxicity to the Green Alga
(*Selenastrum capricornutum*)

Study Completion Date: February 26, 1994

Laboratory: Brixham Environmental Laboratory, Brixham
Devon, UK

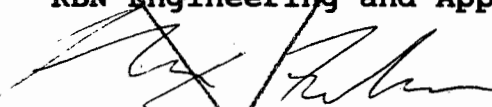
Sponsor: Zeneca Ag Products, Wilmington, DE

Laboratory Report ID: BL5101/B

DP Barcode: ~~D217072, D217078~~

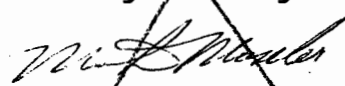
MRID No.: 436781-61

4. **REVIEWED BY:** Max Feken, M.S., Environmental Toxicologist,
KBN Engineering and Applied Sciences, Inc.

Signature: 

Date: 12/5/95

APPROVED BY: Mark Mossler, M.S., Toxicologist,
KBN Engineering and Applied Sciences, Inc.

Signature: 

Date: 12/5/95

5. **APPROVED BY:**

Signature:

Date:

6. **STUDY PARAMETERS**

Definitive Test Duration: 120 hours

Type of Concentrations: Nominal

7. **CONCLUSIONS:** This study is scientifically sound and fulfills
the guideline requirements for an algal toxicity test.

Results Synopsis

EC₅₀: 105 ppb

NOEC: 23 ppb

95% C.I.: 93 - 119 ppb

Probit Slope: 2.5

8. **ADEQUACY OF THE STUDY**

A. Classification: Core

3

Guideline Criteria	Reported Information
Dose range 2X or 3X progression	2X
Doses at least 5	8
Controls negative and/or solvent	Negative and solvent control
Replicates per dose 3 or more	3
Duration of test 120 hours	120 hours
Daily observations were made?	Yes
Method of Observations	Cellular counts
Maximum Labeled Rate	Not reported

12. REPORTED RESULTS

Guideline Criteria	Reported Information
Initial and 120 h cell densities were measured?	Yes
Control cell count at 120 hr $\geq$ 2X initial count?	Yes
Initial chemical concentrations measured? (Optional)	Yes
Raw data included?	Yes

Dose Response

Nominal Dose (μ g/L)	Avg. Cell Density ($\times 10^4$ cells/ml)	% reduction in area under growth curve	120-Hour pH
Control	436	-	9.1
Solvent Control	437	-	9.2
25	412	4	9.0
50	348	25	8.9

Nominal Dose ($\mu\text{g/L}$)	Avg. Cell Density ($\times 10^4$ cells/ml)	% reduction in area under growth curve*	120-Hour pH
100	217	57	8.2
200	110	78	7.9
400	33.5	92	7.6
800	17.3	95	7.5
1600	12.3	97	7.5
3200	8.36	97	7.4

*compared to the pooled control

Other Significant Results: Area under growth curve was the most sensitive end-point measured in the study.

Statistical Results for Areas Under the Growth Curve

Statistical Method: Probit analysis and Dunnett's method for pooled control comparisons.

EC₅₀: 120 ppb

95% C.I.: 17 - 290 ppb

Probit Slope: Not reported

NOEC: 25 ppb

Statistical Results for Growth Rate

Statistical Method: Probit analysis and Dunnett's method for pooled control comparisons.

EC₅₀: 1400 ppb

95% C.I.: 14 - >3200 ppb

Probit Slope: Not reported

NOEC: 25 ppb

13. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: Moving average and Williams' test
for pooled control comparisons. Results
based on nominal concentrations.

EC₅₀: 106 ppb

95% C.I.: 92 - 121 ppb

EC₀₅: 23 ppb

Probit slope: N/A

14. REVIEWER'S COMMENTS: This study is scientifically sound and fulfills the guideline requirement for an algal toxicity test. Based on nominal concentrations, the 120-hour EC₅₀ and EC₀₅ for *S. capricornutum* exposed to ICIA5504 were 105 and 23 ppb, respectively. This study is categorized as Core.

FEKEN ~~SULFENTRASON~~ SELENASTRUM 11-28-95

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
3200	100	98	98	0
1600	100	97	97	0
800	100	96	96	0
400	100	92	92	0
200	100	75	75	0
100	100	50	50	0
50	100	20	20	0
25	100	6	6	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 99.99999

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
5	1.573445E-02	106.2915	92.31609	121.2704

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
5	.1213309	5.16995	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 = 2.012943
95 PERCENT CONFIDENCE LIMITS = 1.311783 AND 2.714103

LC50 = 111.5149
95 PERCENT CONFIDENCE LIMITS = 71.87549 AND 164.609

LC10 = 26.08651
95 PERCENT CONFIDENCE LIMITS = 9.6854 AND 44.7551

from cell Densities

ADDER =

SULFENTRAZONE (ALGA)

File: 43678161

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	436.500	436.500	436.500
2		25	412.333	412.333	412.333
3		50	348.000	348.000	348.000
4		100	217.000	217.000	217.000
5		200	109.667	109.667	109.667
6		400	33.533	33.533	33.533
7		800	17.300	17.300	17.300
8		1600	12.300	12.300	12.300
9		3200	8.363	8.363	8.363

SULFENTRAZONE (ALGA)

File: 43678161

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	436.500				
25	412.333	2.543	*	1.72	k= 1, v=21
50	348.000	9.313	*	1.80	k= 2, v=21
100	217.000	23.097	*	1.83	k= 3, v=21
200	109.667	34.392	*	1.84	k= 4, v=21
400	33.533	42.403	*	1.85	k= 5, v=21
800	17.300	44.111	*	1.85	k= 6, v=21
1600	12.300	44.638	*	1.85	k= 7, v=21
3200	8.363	45.052	*	1.86	k= 8, v=21

s = 13.440

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

NOEC - could not be determined
 NOEC = EC₅ - (next pg) = 23 ppb

Selenastrum cell density

Estimated EC Values and Confidence Limits

Point	Conc.	Lower 95% Confidence	Upper Limits
EC 1.00	12.1082	8.1812	16.3046
EC 5.00	22.8268	17.0538	28.5868
EC10.00	32.0082	25.1539	38.6795
EC15.00	40.2116	32.6278	47.5382
EC50.00	105.4821	93.2364	119.4403
EC85.00	276.6981	233.2028	342.8525
EC90.00	347.6128	286.5067	444.8884
EC95.00	487.4291	387.5259	656.4232
EC99.00	918.9192	679.2305	1368.7682

y - 2.4778 x 0.01

N252 - 23.40