

## DATA EVALUATION RECORD

1. **CHEMICAL:** N-(phosphonomethyl) glycine *diatom*  
Shaughnessey No. 103601.
2. **TEST MATERIAL:** Technical glyphosate, 96.6% a.i., Lot No. NBP-3594465, CAS No. 1071-83-6, a white solid
3. **STUDY TYPE:** Nontarget area phytotoxicity, aquatic plant growth - Navicula pelliculosa.
4. **CITATION:** Hughes, J.S. 1987. The Toxicity of Glyphosate Technical to Navicula pelliculosa. Project No. 1092-02-1100-2. Prepared by Malcolm Pirnie, Inc. White Plains, NY. Submitted by Monsanto Agricultural Company, Chesterfield, MO. Mrid No. 402369-02.

5. **REVIEWED BY:**

Bruce A. Rabe  
Aquatic Toxicologist  
Hunter/ESE

Signature: *Bruce A. Rabe*Date: *12/6/88*6. **APPROVED BY:**

Prapimpan Kosalwat, Ph.D.  
Staff Toxicologist  
KBN Engineering and  
Applied Sciences, Inc.

Signature: *P. Kosalwat*Date: *Dec. 6, 1988*

Henry Craven  
Supervisor, EEB/HED  
USEPA

Signature: *Henry T. Craven*Date: *1/9/90*

7. **CONCLUSIONS:** This study appears scientifically sound. The 7-day EC50 for Glyphosate Technical was 38.6 mg/L.

8. **RECOMMENDATIONS:** N/A *4-day EC50 39.9 mg/L (36.85 to 43.49 mg/L) CRL 9/18/91*

9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A



11. **MATERIALS AND METHODS:** An algal assay bottle test on Navicula pelliculosa, obtained from stock cultures, was conducted by the laboratory of Malcolm Pirnie, White Plains, New York. The test was conducted for 7 days in a Psycrotherm Controlled Environment Incubator Shaker, Model G-27. The test flasks were continuously shaken at 100 oscillations per minute with continuous illumination of  $4306 \pm 650$  lumens/m<sup>2</sup> provided by cool-white fluorescent lights. Temperature was maintained at  $20 \pm 2$  °C.

Test bottles utilized were sterile 250-mL Erlenmeyer flasks fitted with foam stoppers. Three replicates were used for each concentration.

Nominal tests concentrations of 10, 18, 32, 56, and 100 mg/L were prepared by diluting appropriate volumes of a 5.0 mg a.i./mL stock solution to 50 mL volumes with sterile-filtered AAP medium. Test and control solutions were inoculated with algae from a 7-day old stock culture to give an initial cell count of 3000 cells/mL. Growth, as measured by cell counts, was determined on test days 2, 3, 4, and 7 using a Coulter Counter Model ZBI equipped with a C-1000 Channelyzer and MHR Computer. Three counts per replicate were made. All counts were multiplied by the appropriate conversion factors (for sample dilution and volume counted) to yield cells/mL.

Samples were analyzed by Monsanto Company, Chesterfield, MO for actual concentrations of glyphosate on test days 0 and 7. Samples on day 0 before inoculation and samples passed through a 0.8-micron membrane filter on day 7 were placed in polyethylene bottles and frozen prior to shipment to Monsanto Company. Samples were analyzed by a high pressure liquid chromatograph (HPLC) equipped with an o-phthalaldehyde (OPA) post-column reactor (PCR) and fluorescence detector.

The EC25 and EC50 values for glyphosate were calculated by plotting the log of average measured concentration (x-axis) against the percent inhibition expressed as probit (y-axis). Inverse estimation least squares linear regression was used to determine the line of best fit, the concentrations corresponding to 25 and 50 percent inhibition and associated 95% confidence limits. Parameters of the regression line were determined using the SAS statistical package. The values for the test concentrations that were stimulatory were omitted from the regression analysis.

12. **REPORTED RESULTS:** Mean standing crop (cells/mL) and Percent Change, Relative to Control, for Navicula pelliculosa Exposure to Glyphosate Technical

Mean Measured Percent Concentration <sup>a</sup> mg/L	Day 2	Day 3	Day 4 ↓	Day 7	Change <sup>b</sup>	
<0.05 (0)	35000	90333	750667	3020000	--	Day 4
10.6 (10)	34000	101000	794667	2933333	-2.9	0
19.1 (18)	31000	100000	645333	3080000	+2.0	14%
33.6 (32)	36000	92667	734667	3253333	+7.7	2%
56.1 (56)	8667	11000	13667	63500	-97.9	98%
103 (100)	9333	11333	8667	8000	-99.7	99%

<sup>a</sup> The nominal concentrations are given in parentheses

<sup>b</sup> The percent change is based on day 7 values

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:**

Based on mean standing crop, the 7-day EC25 was 18.0 mg/L and the 7-day EC50 was 24.9 mg/L. The 95% confidence limits for these EC values could not be determined from the data since an error condition arises in the calculations as a result of an attempt to take the square root of a negative number. The measured concentrations on day 7 yielded an average of 100.6% of the nominal concentrations.

The study was conducted following the intent of the Good Laboratory Practice Regulations and the final report was reviewed by Malcolm Pirnie's Quality Assurance Unit. A Quality Assurance Statement was included and signed by the Quality Assurance Officer.

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

A. Test Procedure: The procedures were generally in accordance with protocols recommended by the Guidelines, but deviated from the SEP as follows:

- o Growth observations were only taken on days 2, 3, 4, and 7 instead of daily as recommended.
- o The test was conducted at  $20 \pm 2$  °C, instead of the recommended  $24 \pm 2$  °C.

B. Statistical Analysis: The reviewer used linear regression analysis to calculate the EC25 and EC50 values of 27.7 mg/L and 38.6 mg/L, respectively. These calculations are attached. The calculated values are higher than the reported values (i.e., EC25 18.0 mg/L and EC50 24.9 mg/L). The reported values were calculated with the omission of the two stimulatory concentrations. This omission results in artificially low EC values. The reviewer believes that the EC25 and EC50 values calculated by linear regression are a more representative set of values because stimulatory as well as inhibitory values are included in the EC determinations.

C. Discussion/Results: The study results appear to be scientifically valid. The 7-day EC50 value based upon measured concentrations was estimated to be 38.6 mg/L.

D. Adequacy of the Study:

- (1) Classification: ~~(to be added)~~ CORE 9/19/91 CR
- (2) Rationale: N/A
- (3) Repairability: N/A

15. Completion OF ONE-LINER FOR STUDY: Yes, 11-30-88

No. \_\_\_\_\_

Chemical Name Glyphosate Chemical Class \_\_\_\_\_  
Technical

Page 1 of \_\_\_\_\_

Study/Species/Lab/  
Succession

Chemical  
No. 9 a.1

Results

Reviewer/  
Date

Validation  
Status

14-Day Single Dose Oral LD<sub>50</sub>

LD<sub>50</sub> = \_\_\_\_\_ mg/kg ( 95% C.L. ) Contr. Mort.(%) = \_\_\_\_\_

Species \_\_\_\_\_

Slope = \_\_\_\_\_ # Animals/Level = \_\_\_\_\_ Age (Days) = \_\_\_\_\_  
Sex = \_\_\_\_\_

Lab \_\_\_\_\_

14-Day Dose Level mg/kg/(% Mortality)  
( ), ( ), ( ), ( ), ( )

Acc. \_\_\_\_\_

Comments:

14-Day Single Dose Oral LD<sub>50</sub>

LD<sub>50</sub> = \_\_\_\_\_ mg/kg ( 95% C.L. ) Contr. Mort.(%) = \_\_\_\_\_

Species \_\_\_\_\_

Slope = \_\_\_\_\_ # Animals/Level = \_\_\_\_\_ Age (Days) = \_\_\_\_\_  
Sex = \_\_\_\_\_

Lab \_\_\_\_\_

14-Day Dose Level mg/kg/(% Mortality)  
( ), ( ), ( ), ( ), ( )

Acc. \_\_\_\_\_

Comments:

8-Day Dietary LC<sub>50</sub>

LC<sub>50</sub> = \_\_\_\_\_ ppm ( 95% C.L. ) Contr. Mort.(%) = \_\_\_\_\_

Species \_\_\_\_\_

Slope = \_\_\_\_\_ # Animals/Level = \_\_\_\_\_ Age (Days) = \_\_\_\_\_  
Sex = \_\_\_\_\_

Lab \_\_\_\_\_

8-Day Dose Level ppm/(Mortality)  
( ), ( ), ( ), ( ), ( )

Acc. \_\_\_\_\_

Comments:

8-Day Dietary LC<sub>50</sub>

LC<sub>50</sub> = \_\_\_\_\_ ppm ( 95% C.L. ) Contr. Mort.(%) = \_\_\_\_\_

Species \_\_\_\_\_

Slope = \_\_\_\_\_ # Animals/Level = \_\_\_\_\_ Age (Days) = \_\_\_\_\_  
Sex = \_\_\_\_\_

Lab \_\_\_\_\_

8-Day Dose Level ppm/(Mortality)  
( ), ( ), ( ), ( ), ( )

Acc. \_\_\_\_\_

Comments:

8-Day Dietary LC<sub>50</sub>

LC<sub>50</sub> = \_\_\_\_\_ ppm ( 95% C.L. ) Contr. Mort.(%) = \_\_\_\_\_  
Sol. Contr. Mort.(%) = \_\_\_\_\_

Species \_\_\_\_\_

Slope = \_\_\_\_\_ # Animals/Level = \_\_\_\_\_ Temperature = \_\_\_\_\_

Lab \_\_\_\_\_

96-Hour Dose Level ppm/(Mortality)  
( ), ( ), ( ), ( ), ( )

Acc. \_\_\_\_\_

Comments:

7-day EC<sub>50</sub>

EC<sub>50</sub> = 38.6 ppm ( 95% C.L. )  
NA ( NA ) Con. Mort.(%) = NA  
Sol. Con. Mort.(%) = NA

\*=mean measured  
Concentration

Species Navicula pelliculosa

Slope = NA # Animals/Level = NA

Lab Malcolm Pirnie

96.6

7-Day Dose Level ppm/% Inhibition Temp. = 20±2°C

BAR

11/30/88

10.6(2.9)19.1(2.0)33.6(7.7)56.1(97.9)103(99.7)

Comments: Underlined inhibition values are % stimulation

96-Hour LC<sub>50</sub>

LC<sub>50</sub> = \_\_\_\_\_ ppm ( 95% C.L. ) Contr. Mort.(%) = \_\_\_\_\_  
Sol. Con. Mort.(%) = \_\_\_\_\_

Species \_\_\_\_\_

Slope = \_\_\_\_\_ # Animals/Level = \_\_\_\_\_ Temperature = \_\_\_\_\_

Lab \_\_\_\_\_

96-Hour Dose Level ppm/(Mortality)  
( ), ( ), ( ), ( ), ( )

Acc. \_\_\_\_\_

Comments:

Acc. No. 402369-02

## Linear Regression

Glyphosate technical

Navicula pelliculosa

Measured conc.	$\log$	% inhibition
<math>0.05</math>		
@ * 10.6	1.025	2.9
19.1	1.281	- 2.0
33.6	1.526	- 7.7
@ * 56.1	1.7489	97.9
* 103	2.0128	99.7

\* w/ 3 conc.  
 (omitting (-1))

EC 25 = 16.6 mg/L  
 EC 50 = 27.9 mg/L

Reported

EC 25 = 18.0 mg/L  
 50 = 24.9 mg/L

@ w/ 2 conc.

EC 25 = 15.6 mg/L  
 EC 50 = 24.2 mg/L

all conc.

EC 25 = 27.7 mg/L  
 EC 50 = 38.6 mg/L

Bruce A. Paul  
 11/29/88

Lewis glyphosate navicula 4-day

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*****
CONC.      NUMBER      NUMBER      PERCENT      BINOMIAL
          EXPOSED      DEAD        DEAD        PROB. (PERCENT)
103       100           99          99          0
56.1      100           98          98          0
33.6      100           2           2           0
19.1      100           14          14          0
10.6      100           0           0           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 43.41611

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

```
SPAN      G      LC50      95 PERCENT CONFIDENCE LIMITS
4         1.286186E-02      39.93036      36.85304
43.48886
```

RESULTS CALCULATED USING THE PROBIT METHOD

```
ITERATIONS      G      H
GOODNESS OF FIT PROBABILITY
7              2.364793      33.66671
```

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 = 5.876824  
 95 PERCENT CONFIDENCE LIMITS = -3.160486 AND 14.91413

LC50 = 39.35394  
 95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

LC10 = 23.92682  
 95 PERCENT CONFIDENCE LIMITS = 0 AND 49.98271

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