

US EPA ARCHIVE DOCUMENT

DP Barcode: D231056

MRID No.: 44125704

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DATA EVALUATION RECORD

§ 72-1(b) -- ACUTE LC₅₀ TEST WITH A WARMWATER FISH

1. CHEMICAL: Glyphosate S# No.: 417300
2. TEST MATERIAL: Typical End-Use Product: Glygran WDG
Purity: 80%
3. CITATION
Authors: Ward, T. J.; Magazu, J. P.; Boeri, R.L.
Title: Acute Toxicity of Glygran WDG To The
Fathead Minnow, *Pimephales promelas*
Study Completion Date: August 2, 1996
Laboratory: T.R. Wilbury Laboratories, Inc.
Sponsor: Lewis and Harrison for
I. Pi. Ci. Industria Prodotti Chimici,
S.p.A.
Laboratory Report ID: 1008-LP
MRID No.: 44125704
DP Barcode: D231056
4. REVIEWED BY: Dennis J. McLane, Wildlife Biologist, EEB, EFED
Signature: *Dennis McLane* Date: 4-14-97
5. APPROVED BY: Les W. Touart, Head of Section 1, EEB, EFED
Signature: *Les W. Touart* Date: 4/24/97
6. STUDY PARAMETERS
Length of Test Organism: 29 mm
Definitive Test Duration: 96 hours
Study Method: Static
Type of Concentrations: Mean measured

7. CONCLUSIONS:

Results Synopsis

The study fulfills the guideline requirements. This active ingredient is practically nontoxic to fathead minnows. Notice that as the concentration of active ingredient increases the water becomes more acidic (see table on last page).



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LC₅₀: 67.9 ppm a.i. 95% C.I.: 59.1-98.9 ppm a.i.
NOEC: 35.9 ppm a.i. Probit Slope: N/A

8. ADEQUACY OF THE STUDY

A. Classification: Core

B. Rationale: The intent of the guidelines have been met.

C. Repairability: N/A

9. GUIDELINE DEVIATIONS

The following items were not reported:

1. length of the fish
2. year class of fish
3. raw data
4. source of water well or public water system

10. SUBMISSION PURPOSE:

Second registration of an "old chemical".

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
Species Preferred species is the bluegill sunfish (<i>Lepomis macrochirus</i>)	Fathead Minnow, <i>Pimephales promelas</i>
Mean Weight 0.5-5 g	0.19 g
Mean Standard Length Longest not > 2x shortest	Mean: 29 mm Range:???
Supplier	Aquatic Biosystems, Fort Collins, Colorado
All fish from same source?	Yes

Guideline Criteria	Reported Information
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?	No
If treated for disease, was there no sign of the disease remaining during the 48 hours prior to testing?	N/A)
<u>Feeding</u> No feeding during the study	"...48 hours immediately preceding the test initiation" "Fish were not fed during the test."
<u>Pretest Mortality</u> No more than 3% mortality 48 hours prior to testing	"Mortality during the 48 hours prior to the test initiation was less than <3 %."

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	The source of the water was not reported only that it came from T.R. Wilbury Laboratories.
Does water support test animals without observable signs of stress?	Yes
<u>Water Temperature</u> 17°C or 22°C	21.9 to 22.1°C

Guideline Criteria	Reported Information
pH Prefer 7.2 to 7.6	Control pH ranged from 7.3 to 7.9, However the higher the concentration the lower the pH, For example the 13.3 conc. lowest pH is 6.8 and the 98.9 conc. is 3.7. (See attached table)
Dissolved Oxygen Static: $\geq 60\%$ during 1 st 48 hrs and $\geq 40\%$ during 2 nd 48 hrs, flow-through: $\geq 60\%$	The control and first two levels show low DO levels however they did not fall below 60% The control's lowest was 63.3% and the 21.9 level lowest was 69%. (report lowest % DO & hour)
Total Hardness Prefer 40 to 48 mg/L as CaCO_3	44 mg/L as CaCO_3 collected at the end of the test control vessel.
Test Aquaria 1. Material: Glass or stainless steel 2. Size: Volume of 19 L (5 gal) or 30 x 60 x 30 cm 3. Fill volume: 15-30 L of solution	1. glass 2. 20 L 3. 15 L
Type of Dilution System Must provide reproducible supply of toxicant	N/A
Flow Rate Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period	N/A
Biomass Loading Rate Static: ≤ 0.8 g/L at $\leq 17^\circ\text{C}$, ≤ 0.5 g/L at $> 17^\circ\text{C}$;	0.13 g/L
Photoperiod 16 hours light, 8 hours dark	16 hr L & 8 hr D

Guideline Criteria	Reported Information
<u>Solvents</u> Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests	Water

D. Test Design

Guideline Criteria	Reported Information
<u>Range Finding Test</u> If $LC_{50} > 100$ mg/L with 30 fish, then no definitive test is required.	Nominal conc. 0, 13, 50, and 100 mg/L; After 96 hours there was 100% survival at 0mg/L, 13 and 50 mg/L and 0% survival at 100 mg/L
<u>Nominal Concentrations of Definitive Test</u> Control & 5 treatment levels; dosage should be 60% of the next highest concentration; concentrations should be in a geometric series	13, 22, 36, 60 and 100 mg/L $13/22 = 59 \times 100 = 59\%$
<u>Number of Test Organisms</u> Minimum 10/level, may be divided among containers	20/level divided into two containers
<u>Test organisms randomly or impartially assigned to test vessels?</u>	"indiscriminately and equally distributed among two replicates"
<u>Biological observations made every 24 hours?</u>	every 24 hours
<u>Water Parameter Measurements</u> 1. <u>Temperature</u> Measured constantly or, if water baths are used, every 6 hrs, may not vary $> 1^{\circ}C$ 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	1. "...in one test vessel was recorded daily in each test chamber during the test." range: 21.9 to $22.1^{\circ}C$ 2. "... were measured and recorded daily in each test chamber that contained live animals."

Guideline Criteria	Reported Information
<u>Chemical Analysis</u> 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	"Aeration was initiated at 72 hours to maintain dissolved oxygen conc. above acceptable levels."

12. 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>	98-102%
<u>Control Mortality</u> Not more than 10% control organisms may die or show abnormal behavior.	0%
Raw data included?	No
Signs of toxicity (if any) were described?	Yes, mortality, Sublethal effects: lack of equilibrium and/or lethargy

Mortality

Concentration (ppm)		Number of Fish	Cumulative Number Dead			
Nominal	Mean Measured		Hour of Study			
			24	48	72	96
Control	0	21	0	0	0	0
13	13.3	20	0	0	0	0
22	21.9	20	0	0	0	0
36	35.9	20	0	0	0	0
60	59.1	20	0	1	3	5

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Concentration (ppm)		Number of Fish	Cumulative Number Dead			
Nominal	Mean Measured		Hour of Study			
			24	48	72	96
100	98.9	20	0	20	20	20

B. Statistical Results

Method: Not reported

96-hr LC₅₀: 67.9 ppm ai

95% C.I.: 59.1-98.9 ppm ai

Probit Slope: Not reported

NOEC: 35.9 ppm ai

13. VERIFICATION OF STATISTICAL RESULTS

Parameter	Result
Binomial Test LC ₅₀ (C.I.)	67.9 (59.1-98.9) ppm a.i.
Moving Average Angle LC ₅₀ (95% C.I.)	N/A
Probit LC ₅₀ (95% C.I.)	N/A
Probit Slope	N/A
NOEC	35.9 ppm a.i.

14. REVIEWER'S COMMENTS:

The length of the fish, year class of fish and raw data were not provided.

The pH varied greatly as shown in the following table:

Mean Measured Conc. of Glygran WDG (mg/L)	pH					
	Rep.	0 hr.	24 hr.	48 hr.	72 hr.	96 hr.
Control	1	7.7	7.6	7.4	7.3	7.9
	2	7.8	7.6	7.4	7.3	7.5

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13.3	1	6.8	7.0	7.0	6.8	7.4
	2	6.7	6.9	7.0	6.8	7.4
21.9	1	6.3	6.5	6.5	6.7	7.1
	2	6.3	6.5	6.5	6.6	7.3
35.9	1	5.7	5.9	6.0	6.1	6.7
	2	5.7	5.9	5.9	6.1	6.3
59.1	1	4.9	4.9	5.0	5.1	5.1
	2	4.8	4.9	5.0	5.1	5.1
98.9	1	3.7	3.7	---	---	---
	2	3.7	3.6	---	---	---

In addition to the fathead the laboratory reported results for range finding and definitive tests for the bluegill. The range finding result were after 96 hours there was 100% survival at 0 mg/L (control), 0.47, 4.9, 9.8, and 98 mg/L, and 0% survival at 490 mg/L. The definite study with control, 100, 170, 290, 480, and 800 mg/L. "The test was terminated on the first day because of toxic effect at all but the lowest concentration".

MCLANE GLYGRAN WDG FATHEAD MINNOW 96 HOUR LC50

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
98.9	20	20	100	9.536742E-05
59.1	20	5	25	2.069473
35.9	20	0	0	9.536742E-05
21.9	20	0	0	9.536742E-05
13.3	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 59.1 AND 98.9 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 67.87987

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.
