

DATA EVALUATION RECORD
§ 72-1 - ACUTE LC₅₀ TEST WITH A WARMWATER FISH

1. **CHEMICAL:** Suttocide A **PC Code No.:** 128972

2. **TEST MATERIAL:** Sodium Hydroxymethylglycinate **Purity:** 49.53%

3. **CITATION:**

Authors: W.C. Graves and J.P. Swigert
Title: Suttocide A (Integra 44): A 96-Hour Flow-Through Acute Toxicity Test with the Bluegill (*Lepomis macrochirus*)

Study Completion Date: August 23, 1996

Laboratory: Wildlife International Ltd., Easton, MD

Sponsor: ISP Sutton Laboratories, Chatham, NJ

Laboratory Report ID: 300A-108

MRID No.: 441058-03

DP Barcode: D230022 & D230023

4. **REVIEWED BY:** Mark Mossler, M.S., Toxicologist,
KBN Engineering and Applied Sciences, Inc.,

Signature: 

Date: 6/18/97

APPROVED BY: Pim Kosalwat, Ph.D., Senior Scientist,
KBN Engineering and Applied Sciences, Inc.,

Signature: P. Kosalwat

Date: 6/18/97

5. **APPROVED BY:** Allen W. Vaughan 6/27/97

Signature:

Date:

Handwritten note: N. Cairns 01/06/98

6. **STUDY PARAMETERS:**

Age or Size of Test Organism: 19-27 mm
Definitive Test Duration: 96 hours
Study Method: Flow-through
Type of Concentrations: Mean measured

7. **CONCLUSIONS:** This study is scientifically sound and fulfills the guideline requirements. A 96-hour LC₅₀ of 75 ppm ai classifies Suttocide A as slightly toxic to the bluegill.

Results Synopsis:

LC₅₀: 75 ppm ai
NOEC: 40 ppm ai

95% C.I.: 59 - 109 ppm ai
Probit Slope: N/A

8. ADEQUACY OF THE STUDY:

- A. **Classification:** Core for a formulated product.
- B. **Rationale:** N/A.
- C. **Repairability:** N/A.

9. GUIDELINE DEVIATIONS: The pH of the test solutions (up to 9.2) was greater than recommended (7.2-7.6).

10. SUBMISSION PURPOSE:

11. MATERIALS AND METHODS:

A. **Test Organisms**

Guideline Criteria	Reported Information
<u>Species</u> Preferred species is the bluegill sunfish (<i>Lepomis macrochirus</i>)	<i>Lepomis macrochirus</i>
<u>Mean Weight</u> 0.5-5 g	0.28 g
<u>Mean Standard Length</u> Longest not > 2x shortest	Mean: 22 mm Range: 19-27 mm
<u>Supplier</u>	Northeastern Biologists Inc., Rhinebeck, NY
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	Held under similar conditions to testing for 85 days and under testing conditions for 51 hours prior to testing
Wild caught organisms were quarantined for 7 days?	N/A

Guideline Criteria	Reported Information
Were there signs of disease or injury?	No signs of disease or stress
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	Last fed 51 hours prior to testing
<u>Pretest Mortality</u> < 3% mortality 48 hours prior to testing	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	Moderately-hard well water, filtered and aerated
Does water support test animals without observable signs of stress?	Yes
<u>Water Temperature</u> 17 or 22°C	22.0-22.5°C
<u>pH</u> Prefer 7.2 to 7.6	8.2-9.2
<u>Dissolved Oxygen</u> Static: ≥ 60% during 1 st 48 hrs and ≥ 40% during 2 nd 48 hrs, flow-through: ≥ 60%	≥92% of saturation during the test
<u>Total Hardness</u> Prefer 40 to 200 mg/L as CaCO ₃	140 mg/L as CaCO ₃

Guideline Criteria	Reported Information
Test Aquaria 1. Material: Glass or stainless steel 2. Size: Volume of 18.9 L (5 gal) or 30 x 60 x 30 cm 3. Fill volume: 15-30 L of solution	Teflon®-lined polyethylene 25-L 15 L
Type of Dilution System Must provide reproducible supply of toxicant	Continuous-flow diluter
Flow Rate Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period	6 volume replacements every 24 hours
Biomass Loading Rate 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	0.031 g/L/day
Photoperiod 16 hours light, 8 hours dark	16 hours light, 8 hours dark
Solvents Not to exceed 0.5 mL/L for static tests or 0.1 mL/L for flow-through tests	Solvent: none Maximum conc.: N/A

D. Test Design

Guideline Criteria	Reported Information
Range Finding Test If $LC_{50} > 100$ mg/L with 30 fish, then no definitive test is required.	Yes, but results were not reported

Guideline Criteria	Reported Information
<p><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</p>	<p>Control, 16, 26, 43, 72, and 120 mg active ingredient (ai)/L</p>
<p><u>Number of Test Organisms</u> Minimum 10/level, may be divided among containers</p>	<p>20, 10 per replicate</p>
<p>Test organisms randomly or impartially assigned to test vessels?</p>	<p>Yes</p>
<p>Biological observations made every 24 hours?</p>	<p>Yes</p>
<p><u>Water Parameter Measurements</u> 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 ever 48 h in the high, medium, and low doses and in the control</p>	<p>Temperature measured constantly in one negative control chamber and at test initiation and termination in each test chamber DO and pH measured daily in alternating chambers that contained live fish</p>
<p><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</p>	<p>Samples were collected from one replicate chamber at 0, 48, and 96 hours after test initiation and analyzed using HPLC coupled with pulsed electrochemical detection.</p>

12. REPORTED RESULTS:

A. General Results

Guideline Criteria	Reported Information
<p>Quality assurance and GLP compliance statements were included in the report?</p>	<p>Yes</p>

5

Guideline Criteria	Reported Information
<u>Recovery of Chemical</u>	16 mg/L nominal group - 36%, all other groups - 63-91%
<u>Control Mortality</u> Not more than 10% control organisms may die or show abnormal behavior.	0% mortality in the control group
Raw data included?	Yes
Signs of toxicity (if any) were described?	Yes

Mortality

Concentration (ppm ai)		Number of Fish	Cumulative Number Dead			
Nominal	Mean Measured		Hour of Study			
			24	48	72	96
Control	<0.26	20	0	0	0	0
16	5.8	20	0	0	0	0
26	16	20	0	0	0	0
43	40	20	0	0	0	0
72	59	20	0	0	2	2
120	109	20	10	17	19	20

Other Significant Results: Lethargy, surfacing, and lying quiescently on the bottom were signs of toxicity noted among fish exposed at the highest-concentration exposure level.

B. Statistical Results

Method: Binomial

96-hr LC₅₀: 75 ppm ai
Probit Slope: N/A

95% C.I.: 59 - 109 ppm ai
NOEC: 40 ppm ai

13. VERIFICATION OF STATISTICAL RESULTS:

Parameter	Result
Binomial Test LC ₅₀ (C.I.)	75 (59 - 109) ppm ai
Moving Average Angle LC ₅₀ (95% C.I.)	N/A
Probit LC ₅₀ (95% C.I.)	N/A
Probit Slope	N/A
NOEC	40 ppm ai

- 14. REVIEWER'S COMMENTS:** No test material was detected in the 96-hour samples collected from the 16 mg/L solution. This accounts for the low overall mean recovery for this group (36%). However, if this group is omitted from the analysis, the results are the same.

This study is scientifically sound, fulfills the guideline requirements, and can be classified as **Core for a formulated product**. The 96-hour LC₅₀ for bluegill sunfish exposed to Suttocide A was determined to be 75 ppm ai (151 ppm of whole product), which classifies this material as slightly toxic to the bluegill.

Mossler Suttocide A Lepomis macrochirus 6-18-97

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
109	20	20	100	9.536742E-05
59	20	2	10	2.012253E-02
40	20	0	0	9.536742E-05
16	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 59 AND 109 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 75.03147

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.
