

US EPA ARCHIVE DOCUMENT

(10-13-98)

MRID No. 445234-13

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

- 1. CHEMICAL: Methyl isothiocyanate PC Code No.: 068103
- 2. TEST MATERIAL: Methyl isothiocyanate Purity: 94.9%
- 3. CITATION:

Authors: J.K. Schupner and B.J. Stachura
Title: W150 MITC: The Acute Toxicity of MITC Technical to Rainbow Trout, *Oncorhynchus mykiss*, in a Flow Through System
Study Completion Date: September 12, 1991
Laboratory: NOR-AM Chemical Company, Pikeville, NC
Sponsor: AgrEvo USA Company, Wilmington, DE
Laboratory Project ID: 502AF
MRID No.: 445234-13
DP Barcode: D248126

- 4. REVIEWED BY: Mark Mossler, M.S., Toxicologist, Golder Associates Inc.

Signature: *[Handwritten Signature]* Date: 10/6/98

- APPROVED BY: Pim Kosalwat, Ph.D., Senior Scientist, Golder Associates Inc.

Signature: *P. Kosalwat* Date: 10/6/98

- 5. APPROVED BY:

Signature: *Thomas M. Steyer* Date: 10/13/98

- 6. STUDY PARAMETERS:

Age or Size of Test Organism: 34 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 94 ppb classifies methyl isothiocyanate as very highly toxic to the rainbow trout.

Results Synopsis:

LC₅₀: 94 ppb 95% C.I.: 78 - 131 ppb
 NOEC: 40 ppb Probit Slope: N/A

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LC₅₀: 94 ppb
 NOEC: 40 ppb

95% C.I.: 78 - 131 ppb
 Probit Slope: N/A

8. ADEQUACY OF THE STUDY:

A. Classification: Core

B. Rationale: N/A

C. Repairability: N/A

9. GUIDELINE DEVIATIONS: The pH of the dilution water (6.2-6.5) was less 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 rainbow trout (<i>Oncorhynchus mykiss</i>)	<i>Oncorhynchus mykiss</i>
<u>Mean Weight</u> 0.5-5 g	Mean: 0.64 g
<u>Mean Standard Length</u> Longest not > 2x shortest	Mean: 34 mm
<u>Supplier</u>	Aquatic Research Organisms, Hampton, NH
All fish from same source?	Yes
All fish from the same year class?	Yes

B. Source/Acclimation

Guideline Criteria	Reported Information
<u>Acclimation Period</u> Minimum 14 days	52 hours
Wild caught organisms were quarantined for 7 days?	N/A
Were there signs of disease or injury?	No signs reported
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 52 hours prior to testing
<u>Pretest Mortality</u> < 3% mortality 48 hours prior to testing	No mortality in the 52 hours prior to testing

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	Mixture of well water and deionized water
Does water support test animals without observable signs of stress?	Yes
<u>Water Temperature</u> 12°C	11.3-12.6°C
<u>pH</u> Prefer 7.2 to 7.6	6.2-6.5
<u>Dissolved Oxygen</u> Static: ≥ 60% during 1 st 48 hrs and ≥ 40% during 2 nd 48 hrs, flow-through: ≥ 60%	≥76% of saturation during the test

Guideline Criteria	Reported Information
Total Hardness Prefer 40 to 200 mg/L as CaCO ₃	44 mg/L as CaCO ₃
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	Glass 12-L 10 L
Type of Dilution System Must provide reproducible supply of toxicant	Intermittent-flow proportional diluter
Flow Rate Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period	8.6 volume additions 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	0.64 g/L 0.074 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: acetone Maximum conc.: 0.1 mL/L

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, five fish exposed at 0, 32, 90, and 250 ppb, 48-hour mortality of 0, 0, 0, and 100%, respectively

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>	Control, solvent control, 32, 54, 90, 150, and 250 ppb
<p><u>Number of Test Organisms</u> Minimum 10/level, may be divided among containers</p>	20, 10 per replicate
<p>Test organisms randomly or impartially assigned to test vessels?</p>	Yes
<p>Biological observations made every 24 hours?</p>	Yes
<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 the water bath and at 0, 48, and 96 hours after test initiation in each chamber</p> <p>DO and pH measured at 0, 48, and 96 hours after initiation</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>	Samples were collected from each group at 0 and 96 hours after test initiation and analyzed using GC.

12. REPORTED RESULTS:

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Recovery of Chemical	74-87% of nominal
Control Mortality Not more than 10% control organisms may die or show abnormal behavior.	0% mortality in either control group
Raw data included?	Yes
Signs of toxicity (if any) were described?	Yes

Mortality

Concentration (ppb)		Number of Fish	Cumulative Number Dead			
Nominal	Mean Measured		Hour of Study			
			24	48	72	96
Control	<10	20	0	0	0	0
Sol. Con.	<10	20	0	0	0	0
32	26	20	0	0	0	0
54	40	20	0	0	0	0
90	78	20	0	0	1	3
150	131	20	0	10	19	20
250	210	20	3	20	20	20

Other Significant Results: Lethargy, discoloration, reduced/labored breathing, reduced swimming, loss of equilibrium, spinal curvature, and peritoneal lump were signs of toxicity noted among fish exposed at the three highest-concentration exposure levels.

B. Statistical Results

Method: binomial probability

96-hr LC₅₀: 94 ppb.

Probit Slope: N/A

95% C.I.: 78 - 131 ppb

NOEC: 40 ppb

13. VERIFICATION OF STATISTICAL RESULTS:

Parameter	Result
Binomial Test LC ₅₀ (95% C.I.)	94 (78 - 131) ppb
Moving Average Angle LC ₅₀ (95% C.I.)	N/A
Probit LC ₅₀ (95% C.I.)	N/A
Probit Slope	N/A
NOEC	40 ppb

14. **REVIEWER'S COMMENTS:** This study is scientifically sound, fulfills the guideline requirements, and can be classified as Core. The 96-hour LC₅₀ for trout exposed to methyl isothiocyanate was determined to be 94 ppb, which classifies this material as very highly toxic to the rainbow trout.

Mossler MITC Oncorhynchus mykiss 10-5-98

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
210	20	20	100	9.536742E-05
131	20	20	100	9.536742E-05
78	20	3	15	.1288414
40	20	0	0	9.536742E-05
26	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 78 AND 131 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 93.58413

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.
