19 6-13-00

MRID No. 449017-02

DATA EVALUATION RECORD
§ 72-1 - ACUTE LC_{50} TEST WITH A COLDWATER FISH

1. **CHEMICAL:** Mesotrione PC Code No.: 122990

2. **TEST MATERIAL:** 2-amino-4-methylsulfonylbenzoic acid (AMBA)
A metabolite of mesotrione - 99% purity

3. **CITATION:**

Authors:	S.E. Magor and N. Shillabeer
Title:	R044276 (AMBA): Acute Toxicity to Rainbow Trout (*Oncorhynchus mykiss*)
Study Completion Date:	April 10, 1998
Laboratory:	Brixham Environmental Laboratory, Devon, England
Sponsor:	ZENECA Ag Products, Wilmington, DE
Laboratory Report ID:	BL6391/B
MRID No.:	449017-02
DP Barcode:	D259964

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

Signature: [signature] **Date:** 12/21/99

APPROVED BY: Max Feken, M.S., Environmental Toxicologist,
Golder Associates Inc.

Signature: [signature] **Date:** 12/21/99

5. **APPROVED BY:** James Goodyear, USEPA

Signature: [signature] **Date:** 6/13/00

6. **STUDY PARAMETERS:**

Age or Size of Test Organism:	39-49 mm
Definitive Test Duration:	96 hours
Study Method:	Static
Type of Concentrations:	Nominal

7. **CONCLUSIONS:** This study is FOR AMBA scientifically sound and fulfills the guideline requirements. A 96-hour LC_{50} of 150 ppm classifies AMBA as practically non-toxic to the rainbow trout.

Results Synopsis:

LC_{50}: 150 ppm 95% C.I.: 130-180 ppm
NOEC: 100 ppm Probit Slope: N/A

8. **ADEQUACY OF THE STUDY:**

   A. Classification: Core

   B. Rationale: N/A

   C. Repairability: N/A

9. **GUIDELINE DEVIATIONS:** Water temperature (14.3-15.2°C) was higher than recommended (12°C).

10. **SUBMISSION PURPOSE:**

11. **MATERIALS AND METHODS:**

   A. Test Organisms

Guideline Criteria	Reported Information
Species Preferred species is the rainbow trout (*Oncorhynchus mykiss*)	*Oncorhynchus mykiss*
Mean Weight 0.5-5 g	Range: 0.60-1.7 g Mean: 1.1 g
Mean Standard Length Longest not > 2x shortest	Range: 39-49 mm Mean: 44 mm
Supplier	Houghton Springs Fish Farm, Dorset, England
All fish from same source?	Yes
All fish from the same year class?	Not reported

   B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period Minimum 14 days	Held under similar conditions to testing for 9 weeks prior to testing
Wild caught organisms were quarantined for 7 days?	N/A

Guideline Criteria	Reported Information
Were there signs of disease or injury?	Fish reported to be in good condition
If treated for disease, was there no sign of the disease remaining during the 48 hours prior to testing?	Last treated with medicine five weeks prior to testing
Feeding No feeding during the study	Last fed 24 hours prior to testing
Pretest Mortality < 3% mortality 48 hours prior to testing	No mortality in the seven days prior to testing

C. Test System

Guideline Criteria	Reported Information
Source of dilution water Soft reconstituted water or water from a natural source, **not** dechlorinated tap water	Dechlorinated (sodium thiosulphate) city water, particle and carbon filtered, and UV irradiated
Does water support test animals without observable signs of stress?	Yes
Water Temperature 12°C	14.3-15.2°C
pH Prefer 7.2 to 7.6	4.3-7.9
Dissolved Oxygen Static: ≥ 60% during 1st 48 hrs and ≥ 40% during 2nd 48 hrs, flow-through: ≥ 60%	≥88% of saturation during the test
Total Hardness Prefer 40 to 200 mg/L as $CaCO_3$	44 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	 Glass 16.5-L 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 ≤ 17°C, ≤ 0.5 g/L at > 17°C; flow-through: ≤ 1 g/L/day	0.73 g/L
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.	No range finding test reported

Guideline Criteria	Reported Information
Nominal Concentrations of Definitive Test Control & 5 treatment levels; dosage should be 60% of the next highest concentration; concentrations should be in a geometric series	Control, 18, 32, 56, 100, and 180 ppm
Number of Test Organisms Minimum 10/level, may be divided among containers	10
Test organisms randomly or impartially assigned to test vessels?	Yes
Biological observations made every 24 hours?	Yes
Water Parameter Measurements 1. Temperature Measured constantly or, if water baths are used, every 6 hrs, may not vary > 1°C 2. DO and pH Measured at beginning of test and ever 48 h in the high, medium, and low doses and in the control	Temperature was measured constantly in one test chamber and daily in each test chamber DO and pH were measured daily in each test chamber
Chemical Analysis 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	Samples were collected from each vessel at 0, 48, and 96 hours after test initiation and analyzed using HPLC. Analyses were conducted before and after a centrifugation step.

12. REPORTED RESULTS:

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes, but compliance was to UK GLPs

Guideline Criteria	Reported Information
Recovery of Chemical Percent of nominal, procedural recovery, limit of detection (LOD)	99-102% of nominal, procedural recovery not reported, LOD = 0.044 ppm
Control Mortality Not more than 10% control organisms may die or show abnormal behavior.	0% mortality
Raw data included?	Yes
Signs of toxicity (if any) were described?	Yes

Analytical results

Nominal concentration (ppm)	Measured concentration (ppm)*		
	Hour of Study		
	0	48	96
Control	<LOD	<LOD	<LOD
18	18	18	18
32	32	32	32
56	57	58	58
100	100	100	100
180	180	180	180

*Only post-centrifugation results are reported in the table.

Mortality

Concentration (ppm)		Number of Fish	Cumulative Number Dead			
			Hour of Study			
Nominal	Mean Measured		24	48	72	96
Control	<LOD	10	0	0	0	0
18	18	10	0	0	0	0
32	32	10	0	0	0	0
56	58	10	0	0	0	0
100	100	10	0	0	0	0
180	180	10	0	0	2	8

Other Significant Results: Signs of test material toxicity included surfacing, quiescence, erratic swimming, spiralling, loss of equilibrium, abnormal respiration, and darkened pigmentation.

B. Statistical Results

Method: moving average angle

96-hr LC_{50}: 150 ppm
Probit Slope: N/A
95% C.I.: 130-180 ppm
NOEC: 100 ppm

13. VERIFICATION OF STATISTICAL RESULTS:

Method: binomial

96-hr LC_{50}: 150 ppm
Probit Slope: N/A
95% C.I.: not determined
NOEC: 100 ppm

14. REVIEWER'S COMMENTS: This study is scientifically sound, fulfills the guideline requirements, and is classified as **Core.** Based on mean measured concentrations, the 96-hour LC_{50} of 150 ppm classifies AMBA as practically non-toxic to the rainbow trout. The NOEC was determined to be 100 ppm.

MOSSLER MESOTRIONE ONCORHYNCHUS MYKISS 11-9-99

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
180	10	8	80	5.46875
100	10	0	0	9.765625E-02
58	10	0	0	9.765625E-02
32	10	0	0	9.765625E-02
18	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT 0 AND +INFINITY 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 149.5979

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.

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