MRID No. 427131-01

DATA EVALUATION RECORD

- CHEMICAL: Acetochlor. 1. Shaughnessey No. 121601.
- TEST MATERIAL: Acetochlor; Batch/Lot/NBR No. QUE-9001-1482-2. T: 92.07% active ingredient; a brown liquid.
- STUDY TYPE: 72-3. Estuarine Shrimp Flow-Through Acute 3. Toxicity Test. Species Tested: Mysid (Mysidopsis bahia).
- 1992. Acetochlor: CITATION: Swigert, J.P. and G.J. Smith. 4. A 96-Hour Flow-Through Acute Toxicity Test with the Saltwater Mysid (Mysidopsis bahia). Project No. 139A-133. Prepared by Wildlife International Ltd., Easton, MD. Submitted by Acetochlor Registration Partnership. EPA MRID No. 427131-01.
- 5. REVIEWED BY:

Louis M. Rifici, M.S. Associate Scientist KBN Engineering and Applied Sciences, Inc.

APPROVED BY: 6.

> Rosemary Graham Mora, M.S. Associate Scientist KBN Engineering and Applied Sciences, Inc.

Henry T. Craven, M.S. Supervisor, EEB/EFED USEPA

Signature: Jours m Reference 5/21/93

Signature: / NUMAN/ SIGNAM / NO.

Date: 5/21/97

Signature: William Rabert 10/29/93

Date: # 7. (2/2/93

- **CONCLUSIONS:** This study is scientifically sound and meets 7. the guideline requirements for an acute estuarine shrimp toxicity study. The 96-hour LC_{50} value for mysids was 2.2 mg a.i./1 mean measured concentration. Therefore acetochlor is classified as moderately toxic to mysids. The NOEC was 0.56 mg a.i./l.
- RECOMMENDATIONS: N/A. 8.
- BACKGROUND: 9.

10. DISCUSSION OF INDIVIDUAL TESTS: N/A.

11. MATERIALS AND METHODS:

- A. <u>Test Animals</u>: Juvenile mysids (<24 hours old) were obtained from in-house cultures. Brooding adults were held under conditions similar to those during testing for at least 14 days before the juveniles were collected. During this period, water temperature was 24.4-25.3°C, salinity was 24-27 parts per thousand (ppt), and the pH was 7.8-8.1.
- Test System: A continuous-flow proportional diluter B. system was used to prepare and deliver the test solutions. The test compartments were 500-ml beakers with two screen-covered holes on each side. The compartments were suspended in Teflon®-lined, 8-1 polyethylene aquaria filled with 6 l of test solution. The solution depth was approximately 17.5 cm. Approximately 9.6 volume additions were delivered to the chambers every 24 hours. The diluter was preconditioned with the test material for 43 hours prior to testing. The aquaria were impartially positioned in a temperature-controlled water bath (25 ±1°C) under a 16-hour light photoperiod with 30-minute dawn and dusk simulations. Light intensity at the test solution surface was 431 lux.

One stock solution was prepared for each of the five concentrations. The primary stock (80.0 mg/ml) was prepared by dissolving the test material in dimethylformamide (DMF). Aliquots of this stock were diluted with DMF to prepare four additional stocks. The stocks were injected into the diluter mixing chambers.

Natural seawater, collected at Indian River Inlet, DE, was diluted with well water, aerated, and filtered before use as test dilution water. The salinity of the dilution water was 24-25 ppt and the pH was 7.9-8.4 during the 4-week period immediately preceding the test.

C. <u>Dosage</u>: Ninety-six-hour, flow-through test. Based on the results of a preliminary testing, five nominal concentrations (0.65, 1.08, 1.8, 3.0, and 5.0 mg/l), a solvent control, and a dilution water control were tested. The nominal test concentrations were mg/l of whole material (i.e., not adjusted for the percentage

active ingredient). The concentration of solvent in the solvent control and exposures was 0.06 ml/l.

D. <u>Design</u>: Mysids were impartially removed from holding tanks using wide-bore, disposable pipettes and distributed to 25-ml plastic containers until each contained 10 individuals. The containers were dipped into the test chambers to release the mysids. Two replicates were used, for a total of 20 individuals per concentration. The mysids were fed live brine shrimp nauplii daily during the test.

Observations of mortality and treatment-related effects were made at 3, 24, 48, 72, and 96 hours. The dissolved oxygen concentration (DO) and pH were measured in alternate replicates of each test level at the beginning of the test and at each 24-hour observation. The temperature was monitored continuously in one of the control chambers and measured at test initiation and termination in each replicate vessel. Salinity of the dilution water control was measured at the beginning of the test.

Test solution samples were collected from each test chamber at 0, 48, and 96 hours. The samples were analyzed for acetochlor using gas chromatography.

- E. Statistics: The median lethal concentration (LC_{50}) values were calculated, if necessary, using a computer program developed by C.E. Stephan.
- 12. <u>REPORTED RESULTS</u>: The mean measured concentrations were 0.61, 1.1, 1.7, 3.0, and 5.0 mg/l (Table 1, attached).

No mortality or sublethal effects were noted in the dilution water control or the 0.61 mg/l test level (Table 3, attached). In replicate B of the solvent control, three mysids were dead at 48 hours. The remaining mysids in the solvent control appeared healthy and normal throughout the test. In test concentrations ≥1.1 mg/l, mortality ranged from 10 to 100%.

During the test, the DO was >60% of saturation (5.1-7.0 mg/l). The pH values ranged from 8.1 to 8.3 and the temperature was 24.3-24.9°C. The salinity of the dilution water control was 25 ppt.

13. STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:
The 96-hour LC₅₀ value for mysids was 2.4 mg/l with a 95% confidence interval of 2.0-2.8 mg/l. The slope of the

concentration-response curve was 5.62. The no mortality concentration and no-observed-effect concentration (NOEC) were 0.61 mg/l.

Quality Assurance and Good Laboratory Practice Statements were included in the report, indicating that the study was conducted in accordance with U.S. EPA Good Laboratory Practice Standards set forth in 40 CFR Part 160. The dates and types of quality assurance audits were reported. Characterization of the test material was the responsibility of the sponsor.

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

A. <u>Test Procedure</u>: The test procedures were generally in accordance with the SEP, except for the following:

Three of ten mysids died in solvent control replicate B leading to 15% mortality overall in the solvent control during the test. The SEP states that no more than 5% control mortality is considered acceptable in flow through mysid studies.

The recommended temperature for mysid toxicity tests is 22 ±1°C. The temperature used in the study was approximately 25°C.

The salinity of the dilution water in the study was 25 ppt. The recommended salinity for estuarine shrimp is 10-17 ppt.

- B. <u>statistical Analysis</u>: The reviewer used mean measured concentrations of active ingredient (Table 1, attached) and EPA's Toxanal program to calculate the 96-hour LC₅₀ value as 2.2 mg a.i./l with a 95% C.I. of 1.9-2.6 mg a.i./l (see attached printout). The slope of the doseresponse curve was 5.6. The NOEC was 0.56 mg a.i./l.
- C. <u>Discussion/Results</u>: Three of ten mysids died in solvent control replicate B leading to 15% mortality overall in the solvent control during the test. However, no mysids died in the dilution water control, solvent control replicate A, or the lowest test concentration. It is likely that the mortality observed in this replicate was some random event and not due to poor health of the test population or solvent toxicity.

This study is scientifically sound and meets the guideline requirements for an acute estuarine shrimp

toxicity study. The 96-hour LC_{50} value for mysids was 2.2 mg a.i./l mean measured concentration. Therefore acetochlor is classified as moderately toxic to mysids. The NOEC was 0.56 mg a.i./l.

D. Adequacy of the Study:

- (1) Classification: Core.
- (2) Rationale: N/A.
- (3) Repairability: N/A.
- 15. COMPLETION OF ONE-LINER FOR STUDY: Yes, 05-17-93.

ACETOCHLOR				
Page is not included in this copy. Pages through are not included.				
The material not included contains the following type of information:				
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RIFICI ACETOCHLOR MYSIDOPSIS BAHIA 05-14-93

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)	
4.57	20	20	100	9.536742E-05	
2.77	20	14	70	5.765915	
1.58	20	2	10	2.012253E-02	
1.02	20	2	10	2.012253E-02	
.56	20	0	0	9.536742E-05	

THE BINOMIAL TEST SHOWS THAT 1.58 AND 4.57 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 2.326997

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN G LC50 95 PERCENT CONFIDENCE LIMITS
4 5.135007E-02 2.123929 1.811626

2.56165

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS G H GOODNESS OF FIT PROBABILITY

5 .1118078 1 .1483432

SLOPE = 5.609276 95 PERCENT CONFIDENCE LIMITS = 3.733664 AND 7.484887

LC50 = 2.178928 95 PERCENT CONFIDENCE LIMITS = 1.861219 AND 2.564871