MRID No. 415651-37

DATA EVALUATION RECORD

- 1. CHEMICAL: Acetochlor.
 - Shaughnessey Number: 121601.
- 2. <u>TEST MATERIAL</u>: Acetochlor technical; 2-chloro-N-(ethoxymethyl)-N-(2-ethyl-6-methylphenyl) acetamide; 89.7% active ingredient w/w; a brown liquid.
- 3. <u>STUDY TYPE</u>: Marine Fish Acute Static Toxicity Test. Species Tested: Sheepshead minnow (*Cyprinodon variegatus*).
- 4. <u>CITATION</u>: Tapp, J.F., S.A. Sankey, J.E. Caunter, and B.J. Harland. 1989. Acetochlor: Determination of Acute Toxicity to Sheepshead Minnow (*Cyprinodon variegatus*). Brixham Study No. R1072/F. Study performed by Imperial Chemical Industries PLC, Brixham Laboratory, Freshwater Quarry, Brixham, Devon, U.K. Submitted by ICI Americas, Inc. EPA MRID No. 415651-37.
- 5. REVIEWED BY:

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

6. APPROVED BY:

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

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

Signature

Date:

signature: P. Kosalwa

10/3/91

Signature:

Date: Manuel X 2004

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- 7. CONCLUSIONS: This study is scientifically sound and meets the guideline requirements for an acute toxicity study using marine fish. The 96-hour LC₅₀ of acetochlor technical for Cyprinodon variegatus was 3.9 mg/l based on mean measured concentrations. Based on the results of this study, acetochlor technical is considered moderately toxic to Cyprinodon variegatus. The NOEC was 0.91 mg/l based on mean measured concentrations.
- 8. RECOMMENDATIONS: N/A
- 9. BACKGROUND:

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9. BACKGROUND:

10. DISCUSSION OF INDIVIDUAL TESTS: N/A

11. MATERIALS AND METHODS:

A. Test Animals: The test organisms (Cyprinodon variegatus) were hatched at the Brixham Laboratory. No mortalities and no abnormalities were observed during the 5 days prior to the test. The fish were held for 16 days at 22 ±2°C prior to test initiation. The fish were maintained in glass aquaria under daylight and artificial lighting. The pre-test diet was Promin®.

Thirteen days prior to test initiation, the fish were treated with malachite green. At test termination, the control fish had a mean weight of 0.89 g (range of 0.5-1.28 g), and a mean length of 29 mm (range of 23-34 mm).

mm) had a holding capacity of 28 l and contained 20 l of solution. The test was performed in a temperature-controlled room at 22 ±1°C. The test vessels were gently aerated from 24 hours.

The dilution water was seawater collected from Tor Bay, Devon, and filtered. The dilution water was introduced to the test vessels 72 hours prior to test initiation to equilibrate to the test temperature of 22°C.

C. <u>Dosage</u>: Ninety-six-hour static acute test. Seven nominal concentrations were chosen (0.32, 0.56, 1.0, 1.8, 3.2, 5.6, and 10.0 mg/l). In addition, a dilution water control was also included.

The test solutions were prepared by adding appropriate amounts of test substance to each test vessel, solutions were then stirred thoroughly.

D. <u>Design</u>: Ten fish were used in each test concentration and control. The fish were not fed during the test.

Mortality and signs of toxicity were noted every 24 hours during the study.

The salinity was measured at test initiation. Daily measurements of pH, temperature, and dissolved oxygen concentration were recorded daily.

Chemical analysis of each concentration was performed using gas chromatography on samples collected at test initiation, 48 hours, and test termination.

- E. <u>Statistics</u>: The LC₅₀ values were calculated with the moving average method (Stephan, 1977) using a Brixham Laboratory computer program.
- 12. REPORTED RESULTS: Measured concentrations were 0.26, 0.45, 0.91, 1.6, 2.9, 5.1, and 9.6 mg/l (Table 1, attached). These measurements represent 80-96% of nominal concentrations. "All test solutions were clear and colorless though a small amount of test material could be seen on the base of the nominal 10 mg/l exposure concentration test vessel."

No mortality was observed in the control or the five lowest test concentrations (0.26-2.9 mg/l) (Table 2, attached). Total mortality was observed in the two highest test concentrations. The 96-hour LC₅₀ value and 95% confidence interval for sheepshead minnow exposed to acetochlor technical are 3.9 mg/l and 3.0-5.0 mg/l based on mean measured concentrations, respectively. The NOEC is 0.91 mg/l, since no sublethal effects were demonstrated at this concentration (Table 3, attached).

At test initiation, the salinity was 35.11 parts per thousand. During the study, the pH was 7.9-8.3, the temperature was 21.8-22.5°C, and the dissolved oxygen concentration was 3.6-6.8 mg/l.

13. <u>STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:</u>
The authors made no conclusions in the report.

A GLP compliance statement, signed by the study director, the project manager, and a representative of the sponsor company, was included in the report indicating that the study conducted in accordance with the principles of Good Laboratory Practice of the United Kingdom Department of Health Compliance programme (1989). This statement also indicates that the study satisfies the requirements of 40 CFR 160.

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

A. <u>Test Procedure</u>: The test procedures were generally in accordance with protocols recommended by the quidelines, except for the following deviations:

Each test concentration was approximately 56% of the next highest concentration; 60% is recommended.

On day 1, the dissolved oxygen concentration (3.6-4.4 mg/l; 51-62% of saturation) in the test vessels was lower than recommended. The SEP requires that, during the first 48 hours, the dissolved oxygen concentration must be between 60% and 100% of saturation.

The recommended photoperiod is 16-hour light/8-hour dark with 15- to 30-minute transitions. The photoperiod was not reported.

The light intensity during the study was not reported.

The method of exposing the fish to the test solutions (i.e., fish introduced into test solutions within 30 minutes of solution preparation, or fish added first to dilution water then test material added) was not reported.

The report did not indicate whether the fish were randomly assigned to the test chambers as required by the SEP.

Temperature was measured daily; continuous temperature (hourly) measurement is recommended.

Test solutions were aerated during the test. However, this is considered acceptable since test concentrations were measured and appeared to be stable.

- B. Statistical Analysis: EPA's Toxanal computer program was used to verify the LC₅₀ value and 95% confidence interval presented by the authors. The reviewer did not include the data from the highest test concentration (9.6 mg/l), since undissolved test material was evident at this concentration. The reviewer's value (3.9 mg/l based on mean measured concentrations) and 95% confidence interval (2.9-5.1 mg/l) are similar to those of the authors (printout, attached).
- C. <u>Discussion/Results</u>: The deviations listed above probably did not affect the results of this test. This study is scientifically sound and meets the guideline requirements for an acute static toxicity study using marine fish. The 96-hour LC₅₀ of acetochlor technical for Cyprinodon variegatus was 3.9 mg/l based on mean

measured concentrations. Based on the results of this study, acetochlor technical is considered moderately toxic to *Cyprinodon variegatus*. The NOEC was 0.91 mg/l, based on mean measured concentrations.

D. Adequacy of the Study:

- (1) Classification: Core.
- (2) Rationale: N/A.
- (3) Repairability: N/A.
- 15. COMPLETION OF ONE-LINER FOR STUDY: Yes, September 23, 1991.

ACETOCHLOR				
Page is not included in this copy. Pages 6 through 8 are not included.				
The material not included contains the following type of information:				
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Rosemary Graham Mora Acetochlor technical Cyprinodon variegatus 9-23-91

CONC.	NUMBER	NUMBER	PERCENT	BINOMIAL
	EXPOSED	DEAD	DEAD	PROB. (PERCENT)
5.1	10	10	100	9.765625E-02
2.9	10	0	0	9.765625E-02
1.6	10	0	0	9.765625E-02
.91	10	0	0	9.765625E-02
.45	10	0	0	9.765625E-02
.26	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT 2.9 AND 5.1 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 3.845777

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.
