

MRID No. 415651-33

DATA EVALUATION RECORD

1. **CHEMICAL:** Acetochlor.
Shaughnessey Number: 121601.
2. **TEST MATERIAL:** Acetochlor technical; 2-chloro-N-(ethoxymethyl)-N-(2-ethyl-6-methylphenyl) acetamide; 89.7% active ingredient w/w; a brown liquid
3. **STUDY TYPE:** Freshwater Fish Static Toxicity Test. Species Tested: Bluegill Sunfish (*Lepomis macrochirus*).
4. **CITATION:** Tapp, J.F., S.A. Sankey, J.E. Caunter, and B.J. Harland. 1989. Acetochlor: Determination of Acute Toxicity to Bluegill Sunfish (*Lepomis macrochirus*). Brixham Study No. R1072/B. Study performed by Imperial Chemical Industries PLC, Brixham Laboratory, Freshwater Quarry, Brixham, Devon, U.K. Submitted by ICI Americas, Inc. EPA MRID No. 415651-33.
5. **REVIEWED BY:**

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Signature: *Rosemary Graham Mora*
Date: *9/20/91*
6. **APPROVED BY:**

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Date: *9/20/91*

Henry T. Craven, M.S.
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Signature: *Michael Darry*
Date: *12/12/91*

Dan Bice 2-4-92
7. **CONCLUSIONS:** This study is scientifically sound and meets the guideline requirements for an acute toxicity study using warmwater fish. The 96-hour LC₅₀ of acetochlor technical for *Lepomis macrochirus* was 1.6 mg/l based on mean measured concentrations, which classifies acetochlor technical as moderately toxic to *Lepomis macrochirus*. The NOEC was 0.89 mg/l based on mean measured concentrations.
8. **RECOMMENDATIONS:** N/A
9. **BACKGROUND:**

10. DISCUSSION OF INDIVIDUAL TESTS: N/A

11. MATERIALS AND METHODS:

- A. Test Animals: The test organisms (*Lepomis macrochirus*) were obtained from Monkfield Aquatics, Monkfield, Bourn, U.K. The fish were treated with formalin approximately 14 days prior to test initiation. During the 3 days prior to test initiation, less than 1% mortality was observed. The fish were held in glass aquaria under daylight and artificial lighting, and for 18 days prior to the beginning of the test, were held at $22 \pm 1^\circ\text{C}$. The pre-test diet was the Aquarian flaked food®.

At test termination, the control fish had a mean weight of 0.63 g (range of 0.36-0.88 g), and a mean length of 32 mm (range of 27-35 mm).

- B. Test System: The glass test vessels (460 X 305 X 385 mm) had a holding capacity of 50 l and contained 40 l of solution. The test was performed in a temperature-controlled room at $22 \pm 1^\circ\text{C}$. The photoperiod was 16 hours of light and 8 hours of darkness.

The dilution water was tap water supplied from a reservoir (retention time of 24 hours). This supply was dechlorinated with sodium thiosulfate, carbon-filtered, filtered to remove particulate, held in a second reservoir, and delivered to the test via a temperature-controlled header tank (22°C). The dilution water had a total hardness of 67.3 mg/l as CaCO_3 , a specific conductivity of $191 \mu\text{S/cm}$, and a pH of 7.8. Free and combined residual chlorine were not detected in the water.

- C. Dosage: Ninety-six-hour static acute test. Seven nominal concentrations were chosen for this study (0.18, 0.32, 0.56, 1.0, 1.8, 3.2, and 5.6 mg/l). In addition, a dilution water control was also used.

The test solutions were prepared by adding appropriate amounts of test substance to each test vessel, solutions were then stirred thoroughly. For this study, 0.55936 g of the test material was used.

- D. Design: Ten fish were used in each test concentration and control (one vessel/treatment and control). The fish were not fed during the test.

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

Dissolved oxygen concentration, pH, and temperature were measured daily.

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

- E. **Statistics:** The LC_{50} values were calculated with the moving average method (Stephan, 1977) using a Brixham Laboratory computer program.

12. **REPORTED RESULTS:** Measured concentrations of acetochlor technical were 0.15, 0.28, 0.50, 0.89, 1.5, 2.8, and 4.5 mg/l (Table 1, attached). These measurements represent 80-89% of nominal concentrations.

No sublethal effects or mortality were observed in the control or the three middle concentrations (0.28, 0.50, and 0.89 mg/l based on mean measured concentrations) (Tables 2 and 3, attached). A single fish mortality was noted in the lowest test concentration (0.15 mg/l based on mean measured concentrations) and was considered of no significance since the next three highest test concentrations showed no mortality or signs of sublethal effects. Forty-percent mortality was observed at the 1.5 mg/l, based on mean measured concentrations, and total mortality was observed at the remaining test concentrations (2.8 and 4.5 mg/l based on mean measured concentrations). The 96-hour LC_{50} and 95% confidence interval for bluegill exposed to acetochlor technical are 1.5 mg/l and 1.2-2.0 mg/l based on mean measured concentrations, respectively. The NOEC is 0.89 mg/l based on mean measured concentrations.

During the study, the pH was 7.0-7.7, the temperature was 21.5-22.2°C, and the dissolved oxygen concentration was 5.6-7.8 mg/l.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:**
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

indicated that this study satisfied the requirements of 40 CFR Part 160.

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

- A. Test Procedure:** The test procedures were generally in accordance with guidelines and protocols recommended by the guidelines, except for the following deviations:

The dilution water was chlorinated tap water which was dechlorinated prior to use. The SEP discourages the use of dechlorinated water because removal of chlorine is rarely complete and residual chlorine can be toxic to aquatic organisms. Since levels of residual and combined chlorine were below the detection limit, and no control mortality was observed, the use of dechlorinated tap water probably did not affect the results of this study.

The age of the test fish was not given.

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 recommended photoperiod for a freshwater fish acute toxicity study is 16-hour light/8-hour dark with 15- to 30-minute transitions. The report did not indicate whether transition periods were employed.

The light intensity 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.

- 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's LC₅₀ value (1.6 mg/l based on mean measured concentrations) and 95% confidence interval (0.89-2.8 mg/l based on mean measured concentrations) were similar to those of the authors (printout, attached).

C. Discussion/Results: 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 warmwater fish. The 96-hour LC₅₀ of acetochlor technical to *Lepomis macrochirus* was 1.6 mg/l based on mean measured concentrations. The NOEC was 0.89 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 18, 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:

- ☐ Identity of product inert ingredients.
 - ☐ Identity of product impurities.
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Rosemary Graham Mora Acetochlor Lepomis macrochirus 9-17-91

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB.(PERCENT)
4.5	10	10	100	9.765625E-02
2.8	10	10	100	9.765625E-02
1.5	10	4	40	37.69531
.89	10	0	0	9.765625E-02
.5	10	0	0	9.765625E-02
.28	10	0	0	9.765625E-02
.15	10	1	10	1.074219

THE BINOMIAL TEST SHOWS THAT .89 AND 2.8 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 1.623542

THE MOVING AVERAGE METHOD CANNOT BE USED WITH THIS DATA SET
BECAUSE NO SPAN WHICH PRODUCES MOVING AVERAGE ANGLES THAT
BRACKET 45 DEGREES ALSO USES TWO PERCENT DEAD BETWEEN 0 AND
100 PERCENT.

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
7	6.623071	23.55155	0

A PROBABILITY OF 0 MEANS THAT IT IS LESS THAN 0.001.

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED
USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE = 3.167605
95 PERCENT CONFIDENCE LIMITS = -4.98433 AND 11.31954

LC50 = 1.452881
95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

LC10 = .57716
95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY
