Accession No. 410735-06

DATA EVALUATION RECORD

1. CHEMICAL: RH-7592 Technical.
Shaughnessey Number: Not available. 129011

2. TEST MATERIAL: RH-7592 Technical; Lot No. BPP-3-1786R; 96.7% active ingredient; a white solid.

3. STUDY TYPE: Freshwater Fish Flow-through Acute Toxicity Test. Species Tested: Bluegill (Lepomis macrochirus).

4. CITATION: Swigert, J.P. 1988. Acute Flow-through Toxicity of RH-7592 Technical to Bluegill Sunfish (Lepomis macrochirus). Prepared by Analytical Bio-Chemistry Laboratories, Inc., Columbia, Missouri. Report No. 88RC-0024. Submitted by Rohm and Haas Company, Spring House, PA. Accession No. 410735-06.

5. REVIEWED BY:

Kimberly Rhodes
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KBN Engineering and
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Signature: Kimberly Rhodes
Date: June 13, 1989

6. APPROVED BY:

Prapimpan Kosalwat, Ph.D.
Staff Toxicologist
KBN Engineering and
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Signature: P. Kosalwat
Date: June 14, 1989

Henry T. Craven
Supervisor, EEB/HED
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Signature: Henry T. Craven
Date: 11/2 7/89
11-27-89

7. CONCLUSIONS: This study appears scientifically sound and fulfills the Guideline requirements for an acute 96-hour flow-through toxicity test using a warmwater fish species. The 96-hour LC50 of RH-7592 Technical to bluegill (Lepomis macrochirus) was 0.68 mg a.i./L based on mean measured concentrations. Therefore, RH-7592 Technical is classified as highly toxic to bluegill. The NOEC was determined to be 0.42 mg a.i./L after 96 hours.

8. RECOMMENDATIONS: N/A

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9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A

11. **MATERIALS AND METHODS:**

A. **Test Animals:** Bluegill (Lepomis macrochirus) were obtained from a commercial supplier in Missouri. The fish were maintained at the testing facility in well water and were fed newly hatched brine shrimp or a commercially available fish food daily. Bluegill were removed from the culture and placed in the temperature acclimation unit 48 hours prior to test initiation. During this time, the fish were held without food. The bluegill used as the control group during this study had a mean weight of 0.21 (± 0.042) grams and a mean length of 22 (± 1.3) millimeters at test termination.

B. **Test System:** A proportional diluter system described by Mount and Brungs, utilizing a Hamilton Micro Lab 420 syringe dispenser, was used for the intermittent introduction of RH-7592 Technical test solutions and diluent water into each test chamber. The proportional diluter system used for the project was set to provide test levels approximately 50 percent dilutions of each other.

The diluter delivered 0.5 liter of test solution or control water to the test vessels at an average rate of 8.0 times per hour over the course of the study. This flow rate was sufficient to replace the 15-liter volume of each replicate within the test chambers 6.4 times per day. The test chambers were immersed in a temperature controlled water bath held at 22 ± 1°C. The laboratory environment was maintained on a 16-hour daylight photoperiod.

Dilution water for the bluegill test was a blend of reverse osmosis water and ABC well water characterized as having a pH of 7.1 - 7.9, total hardness of 40 - 48 mg/L as $CaCO_3$, total alkalinity of 44 - 56 mg/L $CaCO_3$ and specific conductance of 100 - 160 umhos/cm.

C. **Dosage:** 96-hour flow-through acute test.

D. **Design:** Based on the results of a preliminary test, a control, solvent control, and five nominal RH-7592 concentrations of 0.25, 0.50, 1.0, 2.0, and 4.0 mg a.i./L were tested. Each test concentration and

control was replicated twice with ten fish per test vessel. The test was initiated when fish were impartially distributed to each test vessel after the flow-through system had been running for approximately 24 hours. The concentration of acetone in the solvent control (0.1 mL of acetone in 1 liter of water) was equivalent to that received by the highest test concentration.

All concentrations were observed once every 24 hours for mortality and abnormal effects. The water quality parameters (temperature, dissolved oxygen and pH) were measured in each concentration and control at 0, 48, and 96 hours of testing. Analytical samples were collected from each test level and the diluter stock at 0 and 96 hours of the exposure.

E. **Statistics:** The concentration of toxicant lethal to 50% of the population (LC50's) and 95% confidence intervals was determined at 24-, 48-, 72-, and 96-hour exposure periods by the computer program developed by Stephan et al. (1978).

12. **REPORTED RESULTS:** Mortality and behavioral observations during the acute flow-through toxicity test of RH-7592 to bluegill are shown in Table 5 (attached). The mean measured concentrations of RH-7592 Technical were 0.13, 0.18, 0.42, 0.92 and 2.2 mg a.i./L. The mean measured concentrations ranged from 36% to 79% of the nominal concentrations. A white precipitate was noted in the diluter mixing cell and in the three highest test concentrations at 0-hour and in all concentrations after 24-hours indicating that not all of the RH-7592 Technical was going into solution.

Water chemistry parameters measured at 0, 48, and 96 hours were within the specified limits for conducting aquatic toxicity tests. Temperature ranged from 21 to 23°C, dissolved oxygen ranged from 7.4 to 9.3 mg/L (representing 87 and 109% saturation at 21°C), and pH ranged from 7.3 to 8.1.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** The 24-, 48-, 72-, and 96-hour LC50 values for RH-7592 Technical were 0.92, 0.80, 0.70 and 0.68 mg a.i./L, respectively, based upon mean measured concentrations. The slope of the 96-hour dose-response line was 7.5. Behavioral/sublethal effects (e.g., loss of equilibrium, fish on bottom of test chamber, surfacing, and quiescence) were noted among the fish in the 0.92 and 2.2 mg a.i./L test

concentrations. Given these behavioral/sublethal effects, a no-observed effect concentration (NOEC) of RH-7592 Technical to bluegill was determined to be 0.42 mg a.i./L. This conclusion is supported by the lack of mortality or behavior/sublethal effects at the test concentrations of 0.13, 0.18 and 0.42 mg a.i./L.

Quality Assurance and Good Laboratory Practice Regulation Statements were included in the report, indicating that the study was conducted in accordance with the FIFRA Good Laboratory Practice Standards set forth in 40 CFR Part 160.

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

A. Test Procedure: The test procedures were generally in accordance with protocols recommended by the Guidelines, but deviated from the SEP as follows:

o The SEP states that "test vessels should be described as to construction material and size, as well as depth and volume of solution." The test vessels were not described in this report.

o The ASTM (1980 and 1988) standard protocols state that "the concentration of toxicant in the test chambers should be measured as often as practical during the test. At a minimum, the concentration of toxicant must be measured in (a) each chamber concurrently at least once during the test, preferably near the beginning of the test; (b) except for the control treatment, each test chamber (especially for those toxicant concentrations closest to the LC_{50} or EC_{50}) at least one additional time during the test, on a schedule designed to give reasonable confidence in the concentration of toxicant in the test chambers, . . . and (c) at least one appropriate chamber whenever a malfunction is detected in any part of the metering system. In this study the concentrations of toxicants were measured only in the "A" replicates (control A, 1A, 2A, etc.) at the beginning of the test and only in the "B" replicates (control B, 1B, 2B, etc.) at the end of the test. At no time were the concentrations in all chambers measured concurrently.

o The SEP recommends that fish be acclimated to study conditions for at least two weeks prior to testing. The bluegill were removed from the culture tank and placed in the temperature acclimation unit 48 hours prior to test initiation.

o The SEP states that individual fish should weigh between 0.5 and 5 grams. The fish used in this study weighed between 0.132 and 0.319 grams.

o The SEP states that each designated treatment group should be exposed to a concentration of toxicant that is at least 60% of the next highest concentration. Each designated treatment group for the test was only 50% of the next highest concentration.

The toxicity report did not provide the following information required by the SEP:

o Percent of mortality 48 hours prior to test initiation.

o The SEP recommends a 16-hour light and an 8-hour dark photoperiod with a 15- to 30-minute transition period between light and dark. The report did not state whether a 15- to 30-minute transition period between light and dark was maintained.

B. **Statistical Analysis:** The reviewer used EPA's Toxanal computer program to calculate the LC50 values. These calculations are attached. The binomial test provides a 96-hour LC50 value of 0.68 mg a.i./L with a 95 percent confidence interval of 0.42 to 0.92 mg a.i./L which is the same as that reported by the author.

C. **Discussion/Results:** Although the study appears to be scientifically valid, the 96-hour LC50 value is based upon the mean of measured RH-7592 Technical concentrations. Due to the fact that using the mean of the measured concentration is improper (since measurements were not made in accordance with established standards), the LC_{50} value thus calculated is invalid. The reviewer used EPA's Toxanal computer program to calculate the LC_{50} values using 1) the mean concentration values as used in the report, 2) the measured concentration values of the "A" replicates only, and 3) the measured concentration values of "B" replicates only. The 96-hour LC_{50} value, based on the

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mean measured RH-7592 Technical concentrations, was estimated to be 0.68 mg a.i./L with a 95 percent confidence interval of 0.42 to 0.92 mg a.i./L which is the same as that reported by the author. The 96-hour LC_{50} value, based on the actual measured RH-7592 Technical concentrations of the "A" replicates, was estimated to be 0.62 mg a.i./L with a 95 percent confidence interval of 0.39 to 0.83 mg a.i./L. The 96-hour LC50 value, based on the actual measured RH-7592 Technical concentrations of the "B" replicates, was estimated to be 0.74 mg a.i./L with a 95 percent confidence interval of 0.46 to 1.0 mg a.l./L. In light of the similarity of the calculated LC_{50} values, the study author's value, LC_{50} of 0.68 mg a.i./L, will be accepted. Therefore, RH-7592 Technical is classified as highly toxic to bluegill (Lepomis macrochirus). The no-observed effect concentration (NOEC) was determined to be 0.42 mg a.i./L mean measured concentration.

D. **Adequacy of the Study:**

(1) Classification: Core.

(2) Rationale: Although the test procedures deviated from the guidelines, the reviewer does not believe they significantly affected the toxicity results.

(3) Repairability: N/A.

15. **COMPLETION OF ONE-LINER FOR STUDY:** Yes, 06-06-89.

Shaughnessey No. Not available | Chemical Name BH-7592 Technical | Chemical Class ____ | Page ____ of ____

Study/Species/Lab/ Accession	Chemical % a.i.	Results	Reviewer/ Date	Valid Stx
14-Day Single Dose Oral LD_{50}		LD50 = . mg/kg (95% C.L.) Contr. Mort.(%)=		
Species ____		Slope= # Animals/Level= Age(Days)= Sex =		
Lab		14-Day Dose Level mg/kg/(% Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		
14-Day Single Dose Oral LD_{50}		LD50 = mg/kg. (95% C.L.) Contr. Mort.(%)=		
Species ____		Slope= # Animals/Level= Age(Days)= Sex =		
Lab		14-Day Dose Level mg/kg/(% Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		
8-Day Dietary LC_{50}		LC50 = ppm (95% C.L.) Contr. Mort.(%)=		
Species		Slope= # Animals/Level= Age(Days)= Sex =		
Lab ____		8-Day Dose Level ppm/(%Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		
8-Day Dietary LC_{50}		LC50 = ppm (95% C.L.) Contr. Mort.(%)=		
Species		Slope= # Animals/Level= Age(Days)= Sex =		
Lab ____		8-Day Dose Level ppm/(%Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		
48-Hour LC_{50}		LC50 = pp__ (95% C.L.) Contr. Mort.(%)= Sol. Contr. Mort.(%)=		
Species		Slope= # Animals/Level= Temperature =		
Lab ____		48-Hour Dose Level pp /(%Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		
96-Hour LC_{50}		LC50 = 0.68 ppm (95% C.L. 0.42 - 0.92) Binomial method Con. Mor.(%)= 0 Sol. Con. Mor.(%)= 0	H.K. 06/06/89 Core	
Species Lepomis macrochirus 96.7%		Slope= N/A # Animals/Level= 20 Temp.= 22 ± 1°C		
Lab Analytical Bio-Chemistry Laboratory		96-Hour Dose Level ppm/(%Mortality) 0.15 (0), 0.18 (0), 0.42 (0), 0.92 (90), 2.2 (100)		
Acc. 410735-06		Comments: Based on mean measured concentrations		
96-Hour LC_{50}		LC50 = pp__ (95% C.L.) Con. Mort.(%)= Sol. Con. Mort.(%)=		
Species		Slope= # Animals/Level= Temp.=		
Lab ____		96-Hour Dose Level pp /(%Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		

RIN 3477-95

EEB FENBUCONAZOLE REVIEW

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Pages _____ through _____ are not included.

The material not included contains the following type of information:

____ Identity of product inert ingredients.

____ Identity of product impurities.

____ Description of the product manufacturing process.

____ Description of quality control procedures.

____ Identity of the source of product ingredients.

____ Sales or other commercial/financial information.

____ A draft product label.

____ The product confidential statement of formula.

____ Information about a pending registration action.

✓ FIFRA registration data.

____ The document is a duplicate of page(s) _____.

____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

KIMBERLY RHODES RH-7592 LEPOMIS MACROCHIRUS 06-06-89

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
2.2	20	20	100	9.536742E-05
.92	20	18	90	2.012253E-02
.42	20	0	0	9.536742E-05
.18	20	0	0	9.536742E-05
.13	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT .42 AND .92 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 .6767549

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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Harry A. Winnik LC50 BASED ON "A" REPLICATE MEASURED CONCENTRATIONS

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB.(PERCENT)
1.9	10	10	100	9.765625E-02
.83	10	9	90	1.074219
.39	10	0	0	9.765625E-02
.18	10	0	0	9.765625E-02
.13	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT .39 AND .83 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 .6161175

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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Harry A. Winnik LC50 BASED ON "B" REPLICATE MEASURED CONCENTRATIONS

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB.(PERCENT)
2.6	10	10	100	9.765625E-02
1	10	9	90	1.074219
.46	10	0	0	9.765625E-02
.19	10	0	0	9.765625E-02
.13	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT .46 AND 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 .7361127

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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