

**Data Evaluation Report
Ecological Effects Branch**

1. **Chemical:** Thiabendazole
2. **Test Material:** Lot No. PRM-029 received on 8/25/88 from Merck and Co. 99.6% purity.
3. **Study Type:** Acute Toxicity under Flow Through Conditions with Mysidopsis bahia

4. **Study Identification:**

Study Author: Surprenant, Donald
Laboratory: Springborn Life Sciences, Inc, Wareham, Mass.
Study Dates: March 2-6, 1989
Laboratory Identification: Study No. 359.0888.6105.515
Sponsor: Merck Sharp and Dohme Research Laboratories
EPA Identification: MRID 411920-02

5. **Reviewed by:** Brian Montague, Fisheries Biologist
Ecological Effects Branch
Environmental Fate and Effects Division

6. **Approved by:** Ray Matheny, Supervisory Biologist
Ecological Effects Branch, Section I
Environmental Fate and Effects Division (H7507C)

7. **Conclusion:** The study has fulfilled guideline requirements for acute toxicity testing under flowthrough conditions with mysid shrimp. The LC_{50} value of 0.34 mg/L (C.L. 0.25-0.42) categorizes this chemical as highly toxic to Mysidopsis bahia. The no effect level established by the study is 0.25 mg/L.

8. **Recommendations:** N/A

9. **Submission Purpose:** To satisfy registration guideline requirements for estuarine studies.

10. **Test Methods and Protocol:** Springborn used its own protocol (no. 102387) entitled "Protocol for Conducting a Flow Through Acute Toxicity Test with Mysid Shrimp (Mysidopsis bahia). Protocol was stated to closely follow EPA/OTS guidelines dated 1985.

Test Organisms: The mysid shrimp were obtained from continuously maintained laboratory cultures. Culture and dilution water were of the same source. The temperature was maintained at $25 \pm 1^{\circ}\text{C}$ under at 16D/8N photoperiod at 70-110 footcandles of intensity.

Dilution Water and Test Solution: Dilution water was natural seawater obtained from Cape Cod Canal in Bourne, Mass. The seawater was circulated through 20 and 5 micron polypropylene core filters and held in an epoxy-lined concrete reservoir. Prior to use in the study the seawater was passed through an activated carbon filter system and a polypropylene heat exchanger. Quality of the seawater for life support was confirmed by successful reproduction, survival and growth of mysid shrimp. Analysis revealed no pesticide or PCB contamination.

Test solutions were prepared with the use of triethylene glycol as the solvent at a maximum concentration level of 0.1 ml/L. Stock solutions of 10.1 mg AI/L were prepared by addition of 2.5278 gms a.i. of thiabendazole to 250ml of triethylene glycol. The highest treatment level, 1.0 mg/L, was prepared by addition of 0.252 ml of stock solution to 2.52 Liters of dilution water. The solution was sonicated for 14 minutes in an ultrasonic bath before passage to the diluter. This procedure was performed to enhance solubility of the test material in the dilution water.

Test Materials and Design: Glass aquaria were used to house the test organisms. Five mysid shrimp were housed in glass petri dish retention chambers covered with nitrex screen collars. Two of these retention chambers (10 organisms) were placed in each of the 14 test aquaria. The diluter delivered test concentrations of 1.0, 0.65, 0.42, 0.27 and 0.18 mg/L. Solvent and dilution water controls were also maintained and all levels were replicated once.

At test initiation ten mysid shrimp under 24 hours old were

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introduced to each replicate aquaria (20 per concentration level) by random distribution. The aquaria were allowed to fluctuate from 4 to 7 L by use of a self starting siphon thus insuring an even exchange of solution within the retention dishes. Aquaria were impartially positioned within a constant control water bath equipped with a thermoregulator. Temperature was maintained at $25 \pm 1^{\circ}\text{C}$.

The aquaria were monitored every 24 hours for water quality parameters and organism response. Mortalities were removed at this time. Live brine shrimp nauplii were added twice per day for the first 3 days. Temperature was continuously monitored in one replicate at the 1.0 mg/L level. Prior to the definitive test measured samples of test solutions at the control, .018, .042, and 1.0 ppm test levels were analyzed to confirm proper diluter operation. During the definitive test samples were removed from mid level on day 0 and day 4 for all test replicates. Light intensity ranged from 20-90 footcandles during the test phase.

Analysis of the samples was made with HPLC procedures. Recovery percentage was generally above 88% of nominal test concentration.

11. **Reported Test Results:** Water quality parameters remained within acceptable limits during the 96 hour study with Temperature range between 24 to 26°C , pH from 7.7 to 7.8, and dissolved oxygen at 6.8 to 7.6 mg/L. Salinity remained constant at 32 ppt.

Measured concentration levels fell relatively close to nominal concentration estimates and there was little inter-replicate variation. Mean measured concentrations were 0.88, 0.64, 0.42, 0.25, and 0.15 mg/L with standard deviations of .025 mg/L at the lowest concentration to 0.16 mg/L at the 1.0 mg/L nominal concentration level.

No mortality was seen in the controls, 0.15, or 0.25 ppm test levels. At 0.42 ppm mortality began after the 48 hour point with 20% and 30% at 72 hours and total mortality recorded at the 96 hour point. Mortality at 48 hours was 35% at the 0.64 mg/L level and 70 % at the 0.88 mg/L concentration level. Both of these levels experienced 100% mortality at the termination of the study.

12. **Reviewer's Discussion:** The study has generally followed EPA and ASTM guidelines for the acute toxicity testing under flowthrough conditions for mysid shrimp.

Some difficulty was experienced in the analysis of actual concentrations in 2 of the 3 quality assurance samples. Despite these difficulties no apparent difficulty was experienced in the analysis of samples from the test vessels.

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Statistical analysis of the mean average measured concentrations confirmed the LC_{50} values obtained by the laboratory. As no precipitate or film was noted throughout the test period it is assumed that the test material remained in solution and that the measured concentrations accurately reflect the actual levels of the test concentration.

Adequacy of Study:

Classification: Core

Rationale: The study has satisfactorily followed guideline requirements and established an LC_{50} level useable in risk analysis procedures.

Repairability: N/A

Montague Thiabendazole Acute flowthrough mysid shrimp

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CONC.      NUMBER      NUMBER      PERCENT      BINOMIAL
          EXPOSED      DEAD        DEAD        PROB.(PERCENT)
      .88         20         20         100         9.536742E-05
      .64         20         20         100         9.536742E-05
      .42         20         18          90         2.012253E-02
      .25         20          0          0         9.536742E-05
      .15         20          0          0         9.536742E-05
```

THE BINOMIAL TEST SHOWS THAT .25 AND .42 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 .3427813

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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.88         20         20         100         9.536742E-05
.64         20         20         100         9.536742E-05
.42         20         18          90         2.012253E-02
.25         20          0          0         9.536742E-05
.15         20          0          0         9.536742E-05
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DO YOU WISH TO RUN ANOTHER DATA SET?

ENTER Y OR N.

?

1LIST 2RUN 3LOAD" 4SAVE" 5CONT 6,"LPT1 7TRON 8TROFF 9KEY 10SCREE

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.88         20         20         100         9.536742E-05
.64         20         20         100         9.536742E-05
.42         20         18          90         2.012253E-02
.25         20          0          0         9.536742E-05
.15         20          0          0         9.536742E-05
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