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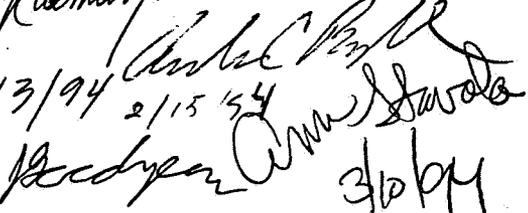
MRID No. 429186-69

DATA EVALUATION RECORD

FILE COPY

1. **CHEMICAL:** MB 46030 (Fipronil).
Shaughnessey No. 129121.
2. **TEST MATERIAL:** M&B 45950; Lot No. JJW2126/79; RM1502; 100% active ingredient; a yellow powder.
3. **STUDY TYPE:** 72-2. Freshwater Invertebrate Acute Flow-Through Toxicity Test. Species Tested: *Daphnia magna*.
4. **CITATION:** McNamara, P.C. 1990. (M & B 45950) - Acute Toxicity to Daphnids (*Daphnia magna*) During a 48-Hour Flow-Through Exposure. Laboratory Report No. 90-1-3203. Prepared by Springborn Laboratories, Inc., Wareham, MA. Submitted by Rhone-Poulenc Ag Company, Research Triangle Park, NC. EPA MRID No. 429186-69.
5. **REVIEWED BY:**

Mark A. Mossler, M.S. Associate Scientist KBN Engineering and Applied Sciences, Inc.	Signature:  Date: 1/13/94
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6. **APPROVED BY:**

Rosemary Graham Mora, M.S. Associate Scientist KBN Engineering and Applied Sciences, Inc.	Signature:  Date: 1/13/94
James J. Goodyear, Ph.D. Project Officer, EEB/EFED USEPA	Signature:  Date: 3 21 94
7. **CONCLUSIONS:** This study is scientifically sound and fulfills the guideline requirements. Based on mean measured concentrations, the 48-hour EC₅₀ for daphnids exposed to M&B 45950 was 100 µg ai/l. Therefore, M&B 45950 is classified as highly toxic to *Daphnia magna*. The NOEC could not be determined.
8. **RECOMMENDATIONS:** N/A.
9. **BACKGROUND:**
10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. MATERIALS AND METHODS:

- A. **Test Animals:** *Daphnia magna* (≤ 24 hours old) were obtained from in-house cultures maintained under a 16-hour light photoperiod at $20 \pm 2^\circ\text{C}$. The culture water was fortified well water filtered through a resin column (Amberlite XAD-7) and a carbon filter. The cultures were fed *Ankistrodesmus falcatus* and a trout food suspension once daily.
- B. **Test System:** An intermittent-flow proportional diluter was used to deliver the test solutions. The test vessels were made of glass and contained a constant solution volume of 1.4 l. The test solution depth was approximately 15 cm. The flow of test solution from the mixing/splitting chambers into the test chambers was restricted using glass capillary tubes to minimize turbulence in the chambers. Test solutions were delivered to each vessel at an approximate rate of six volume replacements per day.

The test area was controlled to maintain the solution temperature at $20 \pm 1^\circ\text{C}$. The area was illuminated at an intensity of 70-120 footcandles using fluorescent tubes on a 16-hour light/8-hour dark photoperiod.

The dilution water was from the same source as that used in culturing. The water had a total hardness of 170-180 mg/l as CaCO_3 , an alkalinity of 120 mg/l as CaCO_3 , a pH of 8.1-8.3, and a specific conductivity of 500 $\mu\text{mhos/cm}$.

A 3.9 mg active ingredient (ai)/ml stock solution was prepared by dissolving 0.195 g of the test material to a final volume of 50 ml in acetone. The test material was injected into the diluter chemical mixing chamber resulting in a high nominal test concentration of 360 $\mu\text{g ai/l}$. This solution was subsequently diluted to provide the remaining treatment solutions. The diluter was pre-conditioned with the test material for ten days prior to test initiation.

- C. **Dosage:** Forty-eight-hour, flow-through test. Based on preliminary testing, five nominal concentrations (47, 78, 130, 220, and 360 $\mu\text{g ai/l}$) were selected for the test. A dilution water and solvent (0.09 ml acetone/l) control were also prepared.
- D. **Design:** Two chambers were used for each treatment or control with ten impartially-selected daphnids per

chamber. The number of immobilized daphnids was recorded daily. Observations of sublethal effects and of the physical characteristics of the test solutions were made at test initiation and every 24 hours thereafter. The daphnids were not fed during the test.

Dissolved oxygen concentration (DO), pH, and temperature were measured once daily in all replicates. At test initiation, hardness, alkalinity, and conductivity were determined in one replicate vessel of each treatment and control group. The temperature of one test vessel was monitored continuously.

Water samples from both replicates of each treatment and control group were taken at test initiation and termination. The concentration of test material was determined using high pressure liquid chromatography.

- E. **Statistics:** The median effective concentration (EC_{50}) and associated 95% confidence interval (C.I.) were calculated using a computer program that employed three methods of analysis. The probit, moving average angle, and binomial probability methods were examined to determine the best-fitting model. The no-observed-effect concentration (NOEC) was defined as the highest concentration tested at and below which there were no toxicant-related mortalities or sublethal effects.

12. **REPORTED RESULTS:** No undissolved test material was noted in the exposure aquaria. The mean measured concentrations were 34, 60, 100, 180, and 320 $\mu\text{g ai/l}$ and averaged 80% of nominal concentrations (Table 3, attached). Measured concentrations between sampling days were generally consistent. Quality control samples averaged 103% of nominal fortification levels.

The response of the daphnids is given in Table 4 (attached). The 48-hour EC_{50} was 100 $\mu\text{g ai/l}$ with a 95% C.I. of 81-130 $\mu\text{g ai/l}$. The NOEC was <34 $\mu\text{g ai/l}$, the lowest concentration tested.

Dissolved oxygen ranged from 8.0 to 9.1 mg/l. The pH values ranged from 8.2 to 8.6. The temperature was 19-21°C. Hardness and alkalinity were 160-180 and 110-120 mg/l as CaCO_3 , respectively. Specific conductance was 500 $\mu\text{mhos/cm}$.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** The author concluded that the test material would be classified as highly toxic to *Daphnia magna*.

Quality Assurance and Good Laboratory Practice Statements were included in the report, indicating that the study was conducted in accordance with all pertinent EPA Good Laboratory Practice Regulations (40 CFR, Part 160). However, the stability, characterization, and verification of the test substance is the responsibility of the study sponsor.

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

- A. Test Procedure:** The test procedures were generally in accordance with the SEP, except for the following:

Observations of the daphnid cultures such as adult mortality, stress, and the presence of ehippia were not reported.

First instar *Daphnia magna* used in tests should be from the fourth or later broods of a given parent. The author did not indicate which brood was the source of the test animals.

- B. Statistical Analysis:** The reviewer used EPA's Toxanal program to calculate the EC₅₀ value and obtained slightly less conservative results (see attached printout).

- C. Discussion/Results:** This study is scientifically sound and fulfills the guideline requirements. Based on mean measured concentrations, the 48-hour EC₅₀ for daphnids exposed to M&B 45950 was 100 µg ai/l. Therefore, M&B 45950 is classified as highly toxic to *Daphnia magna*. The NOEC could not be determined due to increased mortality compared to the controls and sublethal effects at all treatment levels.

- D. Adequacy of the Study:**

- (1) **Classification:** Core.
- (2) **Rationale:** N/A.
- (3) **Repairability:** N/A.

- 15. COMPLETION OF ONE-LINER FOR STUDY:** Yes, 1-13-94.

Fifra/Review

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Pages 5 through 6 are not included in this copy.

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