068102

Data Evaluation Report
Ecological Effects Branch

1. Chemical: Methylene bis thiocyanate

2. Test Material: MBT, "tested as 100% active ingredient" by the laboratory.

3. Study Type: 48 Hour Static Acute Toxicity Test with Daphnia magna

4. Study Identification:

   Study Director: Surprenant, Donald C.
   Study Laboratory: Springborn Bionomics, Inc. Wareham, Mass.
   Study Dates: December 26-28, 1985
   Study Identification: Study No. 11,192-1185-6101-110
   Sponsor: Bromine Compounds LTD, Beer-Sheva, Israel
   EPA Identification: MRID 405186-10 and Summary Report MRID 92116006

5. Reviewed by: Brian Montague, Fisheries Biologist
   Ecological Effects Branch
   Environmental Fate and Effects Division 10/9/90

6. Approved by: Ray Matheny, Supervisory Biologist
   Ecological Effects Branch
   Environmental Fate and Effects Division(H7507C) 10/9/90

7. Conclusions: The study has demonstrated Methylene bisthiocyanate to be very highly toxic to freshwater aquatic invertebrate life with an estimated LC_{50} value of 0.061 mg/L(C.L.s 0.036-0.10mg/L). This study is acceptable for registration purposes.

8. Recommendations: N/A

9. Submission Purpose: Submitted to satisfy registration guideline requirements for toxicity testing of a freshwater invertebrate.

10. Study Design and Protocol: The test protocol was Springborn's own entitled Static Acute Toxicity Test with the Water Flea(Daphnia magna), December 1983. This protocol was modeled after 1980 ASTM guidelines for conducting acute toxicity testing on aquatic organisms.

Test Organisms: Daphnia were obtained from laboratory cultures and were ≤24 hours old at test initiation. The cultures were maintained in reformulated wellwater according to ASTM specifications for hardwater. The water was filtered through carbon and resin filtration systems to remove potential contaminants. The daphnids were fed algal cultures and yeast once daily. A 16D/8N photoperiod regime was maintained.

Test Solution Preparation: A 1000 ppm stock solution was prepared by addition of 0.10 gms. of MBT to dimethyl formamide as a solvent. Individual test concentrations were prepared to 1000 ml aliquots by addition of the appropriate volume of stock solution. This preparation was stirred and then added to three replicate 250 ml beakers(200 ml/beaker). The remaining 400 ml was discarded. The test levels included a control group, a solvent control group, and 0.0080, 0.013, 0.022, 0.036, 0.060, and 0.10 mg/L concentration groups.

Test Procedures and Materials: Fifteen daphnids were randomly selected and added to three 250 ml glass beakers containing 200 ml of solution for each test concentration level(five daphnids per replicate) within fifteen minutes of the test solution preparation. The daphnids were not fed during the test period and test solutions were not aerated. Observations of behavior abnormality, mortality, and temperature level(one control replicate) were made every 24 hours. Measurement of pH and dissolved oxygen were made at 0 and 48 hours in one replicate of the control, solvent control, 0.10, 0.036, and 0.0080 ppm test groups. Temperature was maintained by room temperature at 21±1°C.

11. Reported Test Results: No mortality was seen after 24 hours in any of the test groups. After 48 hours 100% mortality(immobilization) was observed at the 0.10 ppm concentration and a mean mortality of 47% was seen at the next lower concentration of 0.060 ppm. No mortality or abnormal behavior was seen in any other concentrations.

Water quality parameters remained acceptable with pH and

dissolved oxygen remaining well above required levels. Temperature also remained stable in the test vessel it was measured in.

12. **Study Author's Conclusions:** " The 48 hour LC_{50} for D. magna exposed to methylene bis thiocyanate was calculated by non-linear interpolation to be 0.061 mg/L(95% confidence limit, calculated by binomial probability, =0.036-0.10 mg/L)."

13. **Reviewer's Discussion:** The test has followed acceptable testing methods and results appear to support the author's conclusions. One discrepancy was the fact the laboratory has assumed the test material to be of 100% purity, but has not really stated that it was labeled as such. Past testing with MBT technical(see review by Ken Clark,1/10/86) demonstrated similar toxicity results. Based on this fact, it would appear that the test material used in this study was also of technical grade. The history of the culture is not really expanded upon in the report, however the summary report mentions that the culture was at least 3 generations old. Though fewer than twenty organisms are utilized, as recommended by present Agency evaluation procedures, ASTM recommendations consider over 10 organisms as acceptable for static tests. Water hardness and alkalinity were above recommended levels, however this did not appear to affect the test results. No abnormalities are mentioned in the original report, however the summary report mentions lethargy and flared carapaces as symptoms observed. The NOEL based on these observations was felt to be 0.036 ppm. The LC_{50} of 0.061 ppm(61 ppb) would classify MBT as very highly toxic to freshwater invertebrate life.

Adequacy of Study:
Classification: Core
Rationale: The study was conducted acceptably and results appear to confirm the study author's conclusions.
Repairability: N/A