UNDATED

DATA EVALUATION RECORD

1. **CHEMICAL:** Molinate
Shaughnessey No. 041402

2. **TEST MATERIAL:** Molinate technical; S-ethyl hexahydro-1H -azepine-1-carbothioate; Test substance No. T256; 99% w/w active i straw-colored liquid.

3. **STUDY TYPE:** Growth and Reproduction of Aquatic Plants -- Tier 2 Tested: Anabaena flos-aquae.

4. **CITATION:** Smyth, D.V., J.F. Tapp, S.A. Sankey and R.D. Stanley. 1 Molinate: Determination of Toxicity to the Blue-green Alga Anabaen Laboratory Project No. T256/B. Conducted by Imperial Chemical Indu Brixham, Devon, UK. Submitted by ICI Agrochemicals, Fernhurst, S EPA MRID No. 417027-01.

5. **REVIEWED BY:**

Mark A. Mossler, M.S. **Signature:**
Associate Scientist II
KBN Engineering and **Date:**
Applied Sciences, Inc.

6. **APPROVED BY:**

Louis M. Rifici, M.S. **Signature:**
Associate Scientist II
KBN Engineering and **Date:**
Applied Sciences, Inc.

Henry T. Craven, M.S. **Signature:**
Supervisor, EEB/HED
USEPA **Date:**

7. **CONCLUSIONS:** This study is scientifically sound but does not meet guideline requirements for a Tier 2 non-target plant growth and rep The initial cell densities were higher than recommended and rates w fold progression rather than a two-fold. The 5-day EC_{25} and EC_{50} v molinate for A. flos-aquae were 1.64 and 9.48 mg ai/l, respective was determined to be 0.95 mg ai/l.

8. **RECOMMENDATIONS:** N/A

9. **BACKGROUND:**

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

11. **MATERIALS AND METHODS:**

A. **Test Species:** The alga used in the test, Anabaena flos-aquae, from the Culture Centre of Algae and Protozoa, Freshwater Association, the Ferry House, Ambleside, Cumbria, UK. Stock were maintained in synthetic nutrient medium (Miller et al., 2870 lux illumination, and a temperature of 24 ±1°C, with orbi 100 rpm. Cool white fluorescent tubes and a continuous photop used. Cultures that were growing logarithmically were used for the test.

B. **Test System:** Test vessels used were glass 250 ml conical flas with foam stoppers. The test medium was the same as that culturing, with a pH between 7.2 and 7.3.

The test vessels were kept in an incubator with environmental like those employed in culturing.

C. **Dosage:** Results from the preliminary range-finding test demon that rates between 0.1 and 100 mg ai/l would be appropriate to definitive test. Nominal rates of 0.1, 0.32, 1.0, 3.2, 10, 3 medium control, and a blank (no algal inoculum) were used definitive test.

D. **Test Design:** The two highest nominal rates (100 and 32 mg ai/ prepared by direct addition of the test material to sterile Aliquots of the stock were added to sterile culture medium t lower nominal concentrations. All solutions were clear an One-hundred milliliters of the test solution were placed in replicate 250 ml flasks (3 per treatment level). The contr replicated six times.

An inoculum volume of 1.490 ml used to provide 2.0×10^4 cells/ml per flask. Cell counts were performed ever for 5 days using a spectro-photometer. The instrument was ze the blank flask solution. After counting, the flasks were rows within the incubator.

At the start of the test, samples were taken of each test solu excess remaining after filling the test vessels, and were an concentra-tion of test substance by gas chromatography (GC). hours, each blank solution was sampled and analyzed in th

manner.

The pH of the test solutions were measured at test init termination. Light intensity was measured once during the exp temperature was monitored continuously.

E. Statistics: For each nominal concentration, the mean of the m concentration from the blanks on day 0 and day 4 was calcula mean measured concentrations were then used as the basis for analysis. The area under the growth curve and growth r examined as a function of time. Probit and Dunnett's analy were conducted on both of these parameters at day 4 and day 5.

12. **REPORTED RESULTS:** Algal cell absorbances for the control and the e concentrations throughout the test are given in Table 2 (attached).

Measured concentrations on day 0 were 94% to 105% of nominal whi measured concentrations were between 94% and 100%. The means o measured concentrations were 0.095, 0.32, 0.95, 3.3, 9.7, 31, and 9

Increasing concentrations of molinate had increasingly inhibitory e growth and reproduction of Anabaena flos-aquae.

By day 5, the effect of the test material on the area under the relative to the control, ranged between 0% and 86% inhibition attached). The EC_{50} was 21 mg ai/l with confidence limits of 2.1 a ai/l.

By day 5, the effect of the test material on the growth rate, rela ranged between 0% and 34% inhibition (Table 4, attached). The EC determined to be greater than 97 mg ai/l. No confidence limits wer

Results from Dunnett's analysis indicated that the areas under the on day 5 at the five highest concentrations were significantly controls. The lowest rate of 0.095 mg ai/l was also significantly control, but since the amount of inhibition was 14% and the next hi not significantly different, the NOEC was determined to be 0.32 results from the growth rate data indicated that by day 5, the rat was not significantly different from the control. The NOEC was re mg ai/l.

The pH in the control and the exposure concentrations were 7.9 to 8 9.5, respectively, by test termination. The temperature ranged 24.8°C.

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

No conclusions were made by the authors.

Good laboratory practice and Quality Assurance Unit statements were the report indicating compliance with EPA Good Laboratory Practic under the Federal Insecticide, Fungicide, and Rodenticide Act.

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

A. **Test Procedure:** The test procedure and the report were genera accordance with the SEP and Subdivision J guidelines, excep following deviations:

The dissolved oxygen and conductivity of the test solution measured.

The age of the organisms used for inoculum was not stated.

The light intensity was 2.87 klux. The recommended intensity

The test rates were three-fold progressions. Two-fold di recommended.

The initial cell density was greater than the recommended d 20,000 vs. 3000).

Although the raw data were submitted for cell density data, t raw data for the area under the growth curve and growth rate w

No subtoxic (EC_{25}) values were reported.

B. **Statistical Analysis:** The reviewer used a computer program to statistical analysis (attached) of the 5 day absorbance data the NOEC, the area under the growth curve was used to determin $_{50}$ value. Moving average and Dunnett's tests were used to de EC and NOEC values, respectively. The results from Dunnett' were in agreement with the authors'. The reviewer obtained an value that was the same as the authors', however, the confid were smaller.

C. **Discussion/Results:** This study is scientifically sound but do the guideline requirements for a Tier 2 non-target aquatic Growth of Anabaena flos-aquae was increasingly inhibited by amounts of molinate.

D. **Adequacy of the Study:**

(1) **Classification:** Supplemental.

(2) **Rationale:** Initial cell density and test concentrations the guideline requirements.

(3) **Repairability:** No.

15. **COMPLETION OF ONE-LINER:** Yes, 6/20/91.