

## Data Evaluation Record

1. Chemical: Di-Syston Technical (Disulfoton)  
Shaughnessy No.: 032501
2. Test Material: Disulfoton (o,o-diethyl s-(2-(ethylthio)ethyl) phosphorodithioate), 98%, batch #7030185 (CAS #298-04-4); a clear liquid
3. Study type: Aquatic Invertebrate Life <sup>Cycle</sup> Stage (72-4)  
Test Species: Daphnia magna
4. Study ID: Blakemore, Greg and Alan Forbis. Chronic toxicity of Di-Syston technical to Daphnia magna under flow-through test conditions. Performed by Analytical Bio-Chemistry Laboratories, Inc., 7200 East ABC La., P.O. Box 1097, Columbia, MO 65205, for Mobay Corporation, Agricultural Chemicals Division, Research and Development Department, P.O. Box 4913, Kansas City, MO 64120. Study Id #33896. MRID #419358-02.
5. Reviewed by: Kathryn Valente  
Biologist  
EEB/EFED  
Signature: *Kathryn Valente*  
Date: 11/27/91
6. Approved by: Allen Vaughan  
Acting Head, Section II  
EEB/EFED  
Signature: *Allen W. Vaughan*  
Date: 12.04.91
7. Conclusions: The study is supplemental due to the lack of dry weight measurements. The 21-day EC<sub>50</sub> was 0.55 ug/L. The NOEC was 0.037 ug/L, the LOEC was 0.070 ug/L and the MATC was 0.051 ug/L.
8. Recommendations: N/A
9. Background information: This study was submitted in support of reregistration for disulfoton.
10. Discussion of Individual Tests: N/A
11. Materials and Methods:
  - a. Test animals: Adult daphnids were obtained from a laboratory culture at ABC Laboratories. Daphnids were kept on a 16-hr light/8-hr dark photoperiod. First instar daphnids (<24 hours old) were used for the test.
  - b. Test system: Tests were conducted in 1 liter glass beakers. A proportional diluter was used to deliver control water of test substance to each beaker. The beakers were kept in a water bath which held a temperature of 20 +/- 1°C.



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c. Study design: A 13-day preliminary range-finding study was conducted. The results from this range-finding study indicated 30% mortality at 0.8 ug/L, 2.5% at 0.4 ug/L, 5% at 0.2 ug/L and 5% at .096 ug/L. For the definitive test, 10 daphnids were placed in each of four beakers per treatment, giving a total of 40 daphnids per concentration. Observations of survival, abnormal effects and reproduction were made daily throughout the study. Reproductive success was measured by counting and discarding the number of offspring produced in each concentration every Monday, Wednesday and Friday. At the end of the study, surviving adult daphnids were measured from the apex of the helmet to the base of the posterior spine as an index of growth. Temperature, dissolved oxygen and pH were measured on days 0, 4, 7, 14 and 21 in 2 replicates of control, middle and high concentrations. These water parameters were measured daily in the dilution water (hard blended water made from ABC well water and reverse osmosis water). Dimethyl formamide was used as a solvent. The nominal exposure levels of Di-syston were control, solvent control, 0.048, 0.096, 0.20, 0.40 and 0.80 ug/L. Analytical samples were collected on days 0, 4, 7, 11, 14 and 21, in order to determine the actual concentrations of Di-syston at each treatment level. Mean measured concentrations of Di-syston were: 0.037, 0.07, 0.12, 0.28 and 0.64 ug/L.

d. Statistics: Survival data were analyzed using frequency analysis with Chi Square or Fisher's exact tests. Growth, time to first brood and number young/adult reproduction day data were analyzed using 1-way analysis of variance (ANOVA) with Dunnett's mean separation. The control and solvent control were pooled if a t-test showed no significant difference between the means of the two groups. In addition, an EC<sub>50</sub> was calculated using the computer program of Stephan. The NOEC, LOEC and MATC were also determined.

12. Reported Results: Water quality parameters were as follows; DO ranged from 7.8-8.9 mg/L (92-103% saturation); temperature ranged from 17.7-21.3°C; pH ranged from 7.9-8.4; conductivity, hardness and alkalinity were not measured.

Survival: Day 21 survival percentages were:

pooled controls:	95%	0.12 ug/L:	100%
0.037 ug/L:	100%	0.28 ug/L:	95%
0.07 ug/L :	90%	0.64 ug/L:	37.5%

There was a statistically significant reduction in survival between the pooled controls and 0.64 ug/L.

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<sup>1</sup>The total number of adult reproduction days during the study was based on the number of days daphnids were reproducing and the number of adult daphnids alive on each day. The number of reproduction days was counted from the first day neonate production was observed until the end of the study.

Length: Mean standard lengths at 21 days were:

pooled controls:	4.34 mm	0.12 ug/L:	4.25 mm
0.037 ug/L:	4.33 mm	0.28 ug/L:	3.84 mm
0.07 ug/L :	4.29 mm	0.64 ug/L:	3.33 mm

ANOVA with Dunnett's mean separation showed a significant reduction in growth for 0.28 ug/L (0.64 ug/L was not included in the statistical analysis due to the statistically significant reduction in survival at this level).

Time to first brood: The mean number of days for production of the first brood for each group was as follows:

pooled controls:	9.8	0.12 ug/L:	7.0
0.037 ug/L:	7.5	0.28 ug/L:	7.25
0.07 ug/L:	7.5	0.64 ug/L:	8.5

ANOVA with Dunnett's mean separation showed no significant differences for any level compared to the controls (p=0.05).

Number of young per adult reproduction days: The mean number of young produced per adult reproduction days for each treatment were:

pooled controls:	9.8	0.12 ug/L:	7.90
0.037 ug/L:	9.15	0.28 ug/L:	4.97
0.07 ug/L:	7.72	0.64 ug/L:	2.45

ANOVA with Dunnett's mean separation showed a significant difference at 0.07, 0.12 and 0.28 ug/L compared to the pooled controls (p=0.05). (0.64 ug/L was not included in the statistical analysis due to the statistically significant reduction in survival at this level).

13. Study Author's Conclusions/Quality Assurance Report: The NOEC was 0.037 ug/L and the LOEC was 0.070 ug/L. The MATC point estimate (geometric mean of the NOEC and LOEC) was 0.051 ug/L.

Quality Assurance and Good Laboratory Practice statements were included in the report.

14. Reviewer's Discussion and Interpretation of the Results:

a. Test Procedure: The test design and procedure were scientifically sound, but due to the lack of dry weight data, growth could not be accurately assessed, and the study is therefore classified as supplemental. Also, there was a reported problem with the analytical samples collected on day 7, but no explanation was given for the problem; the concentration values for this day were not included in the calculation of the mean measured concentrations.

b. Statistical Analysis: All statistics were verified using EPA's TOXSTAT and TOXANAL computer programs. All values were in agreement with the reported results (see attachment A).

c. Discussion/Results: The study is scientifically sound, but is classified as supplemental for the reasons described under

Section 14(a) above.

d. Adequacy of the study:

- (1) Classification: Supplemental.
- (2) Rationale: No dry weight measurements taken to assess growth.
- (3) Repairability: N/A

NOTE: THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE  
OF THE LOWER CONCENTRATIONS HAD ZERO MORTALITY.  
THEREFORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

VALENTE DI-SYSTON DAPHNIA CHRONIC

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
.64	40	25	62.5	0
.28	40	2	5	0
.12	40	0	0	0
.07	40	4	10	0
.037	40	0	0	0

BECAUSE THE NUMBER OF ORGANISMS USED WAS SO LARGE, THE 95 PERCENT  
CONFIDENCE INTERVALS CALCULATED FROM THE BINOMIAL PROBABILITY ARE  
UNRELIABLE. USE THE INTERVALS CALCULATED BY THE OTHER TESTS.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS .5485008

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
1	.1087706	.5485008	.4760297	.6732599

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H
5	2.764711	9.151921

0

A PROBABILITY OF 0 MEANS THAT IT IS LESS THAN 0.001.

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED  
USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE = 2.189987  
95 PERCENT CONFIDENCE LIMITS = -1.451397 AND 5.831371

LC50 = .644465  
95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

LC10 = .1695459  
95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

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