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DATA EVALUATION RECORD

1. CHEMICAL: Methomyl

2. FORMULATION: Technical (>99%)

3. CITATION: Driscoll, R.R. 1982. Early life stage toxicity of methomyl to fathead minnow. Unpublished; Haskell Lab report No. 528-82; submitted by E.I. Dupont de Nemours & Co., Inc., Newark Delaware; in support of Reg. No. 352-342; 352-370; Acc. No. 071268

4. REVIEWED BY: John J. Bascietto
Wildlife Biologist
EEB/HED

5. DATE REVIEWED: 2-3-83

6. TEST TYPE: Early Life Stage - freshwater fish
A.) Test Species: Fathead minnow (Pimephales promelas)

7. REPORTED RESULTS:

"Of the biological responses measured, 28-day larval survival is the most sensitive indicator of methomyl toxicity. The test material significantly reduces (P <0.05) fathead minnow survival at measured test concentrations as low as 117 ug/l. Based upon the results of this study, the Maximum Acceptable Toxicant Concentration (MATC) for fathead minnows exposed to methomyl is estimated to be greater than 57 ug/l but less than 117 ug/l."

8. REVIEWER'S CONCLUSIONS: The study is scientifically sound. Based on the results, the MATC for fathead minnow is >57 ug/l <117 ug/l. Methomyl significantly reduced larval survival at 117 ug/l - (meausred conc.) but did not inhibit embryo hatch at < 491 ug/l (measured conc). Larval growth was first significantly inhibited at 243 ug/l (measured). The study would fulfill the requirement for early life stage study on a freshwater fish if the raw data is submitted.

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9. Materials/Methods

A. Procedures

A Mount and Brungs proportional diluter deliverd the desired toxicant concentrations to aquaria. Dilution water was from a deep well, aerated and filtered (10 u) prior to testing.

Five (5) test concentrations and a control were used (duplicates used). Aquaria were glass, test volume = ml each. Turnover rate per aquaria was 10X per day. 16 hr light/8 hr dark (flourescent) used. Test temperature = 25°C.

Embryo Test

A preliminary 96 hr acute test was performed on fathead larvae which resulted in early life stage test concentrations of:

(control) 0, 500, 250, 125, 62, 31 ppb (ug/l). Test solutions were made from a concentated stock solution of 13.5 mg/l delivered to diluter by FMI Model G50 metering pump.

Embryos were taken from spawn substrate and pooled in glass culture vessel (dish). 50 embryos were randomly placed in each of 12 embryo cups (glass jars with 40 mesh screen bottoms). These were gently agitated.

Daily observations were made. Dead embryos were removed after counting. Percentage hatch was calculated based on number live larvae per cup after hatch was completed - compared to initial number embryos per cup.

28-day larval exposure - initiated by randomly selecting (20) larvae hatched from the above phase; placing them to respective test aquaria (20 per each - same concentrations used for embryo phase). Larvae fed freshly hatched brine shrimp (Artenia salina) nauplii x3 per day (X1 on non-work day). Daily observations included mortality, behavior, appearance. Number of live fry in each was estimated and recorded once per week.

At the end of the post hatch phase (28 days post hatch) the actual number of surviving larvae were recorded per aquarium. Larvae from each were sacrificed and standard length and wet weights of each fish wre recorded

Chemical and Physical Analyses The (EDTA) hardness total alkalinity, and conductivity of the control water were measured X1 per week. D.O. and pH were taken at beginning and at least X3 per week in the control, high, medium, and low test concentrations. Temp. of control tanks measured at 4 hrs interval on work days.

Analytical Chemistry - 200 ml samples were withdrawn from one duplicate for each concentration and control at beginning of embryo exposure, at embryo hatch completion, and weekly thereafter. Chemical analyses by H-P liquid chromatography at Dupont Lab. Arithmetic means calculated for each to determine mean measured concentrations.

B. STATISTICAL ANALYSIS Percentage Embryo hatch and larval survival were Arc sin transformed and subjected to Analysis of variance. If significant differences ($P < 0.05$) between treatments were indicated, the treatment group mean was compared to control response using Dunnett's procedure. Results of this analysis were used to estimate MATC.

10. Results

Range Finder Test -

96-hr LC_{50} estimated at >972 ug/L

TABLE I
96-hr Acute Test Results

Nominal Conc. ug/l	Mean meausred (ug/l)	(HOURS) 24	48	72	96
1000	972	20	20	30	30
		30	40	50	50
300	395	0	0	0	0
		10	10	10	10
250	254	0	0	0	0
		0	0	0	0
125	113	0	10	10	10
		0	0	0	0
62.5	57	0	0	0	0
		0	0	0	0
H_2O control	<1	0	0	0	0
		0	0	0	0

*Number of organisms per duplicate vessel was not reported. No details of this study were given, therefore it could not be considered to fulfill a guidelines requirement for a 96-hr LC_{50}, but is useful to establish range for the chronic study.

Chronic Study -

A.) Embryo Hatch - Table II (Summary Data)

Conc. Nominal (ug/l)	Conc. Mean measured (ug/l)	Embryo hatch (%)
500	491	86 78
250	243	84 84
125	117	76 80
62	57	88 90
31	27	100 90
H_2O control	<1-4	96 94

No statistically signigicant differences were observed.

Table III

B.) Summary of 28-Day Larval Survival

Conc. Nominal ug/l	Conc. Mean Measured ug/l	28-Day Larval survival (%)
500	491	90*
		80
250	243	85*
		90
125	117	90*
		90
62	57	100
		95
31	27	100
		100
H_2O	<1-4	100
		100

* Treatment group is significantly different (P <0.05) than control by Dunnett's procedure.

Table IV

Summary - 28 Day Larval Growth Data

Conc. Nominal ug/l	Mean Measured Conc. ug/l	Mean Standard Length (mm)*	Mean Wet weight (mg)*
500	491	13.2 (2.4)**	29.3 (17.6)**
		13.5 (2.0)	33.0 (17.5)
250	243	14.7 (2.6)**	43.0 (22.4)**
		14.8 (2.2)	38.9 (18.5)
125	117	15.2 (2.3)	44.6 (22.2)
		16.3 (1.9)	50.7 (20.2)
62	57	14.5 (3.2)	41.8 (19.6)
		16.2 (1.5)	49.4 (15.6)
31	27	16.2 (1.2)	55.5 (12.5)
		15.4 (1.4)	42.7 (12.9)
H_2O control	<1-4	17.0 (1.6)	60.9 (20.1)
		15.2 (3.4)	45.5 (21.9)

* ( ) indicates ± standard deviation of the mean.
** Treatment group is significantly different (P <0.05) from control.

Table V

Physical and Chemical Parameters

ACUTE STAGE -

	Nominal conc. (ug/l)			
Parameter	1000	250	62.5	Control
D.O. (ppm)				
0 HR	8.5	8.4	8.4	8.5
48 HR	8.5	8.5	8.5	8.4
96 HR	8.5	8.3	8.4	8.4
PH				
0 HR	7.0	7.0	7.1	7.0
96 HR	7.2	7.2	7.3	7.2
Total Alkalinity (mg/l as $CaCO_3$)				
0 HR	-	-	-	97
EDTA Hardness (mg/l as $CaCO_3$)				
0 HR	-	-	-	84
Conductivity (umhos)				
0 HR	-	-	-	185

Table VI

Chronic Phase - Physical/Chemical Parameters

	Nominal conc. (ug/l)			
Parameter	500	125	31	Control
D.O. (ppm)[a]	8.5 (0.1)	8.5 (0.1)	8.5 (0.1)	8.6 (0.04)
pH[b]	7.6-7.7	7.6-7.7	7.6-7.8	7.6-7.7
Total Alkalinity[a,c]	-	-	-	106.5 (0.8)
EDTA Hardness[a,c]	-	-	-	91.3 (0.8)
Conductivity (umhos)[a]	-	-	-	182.5 (2.7)
Temperature (°C)[a]	-	-	-	24.9 (0.2)

a - mean ± standard deviation
b - range
c - mg/l as $CaCO_3$

11. Reviewer's Evaluation

A. Procedures -

procedures for the chronic phase were acceptable as they were in substantial agreement with protocols currently under development by Committee E-47 of American Society for Testing and Materials (Draft No. 1-2/81 - "Standard Practice for Conducting Toxicity Tests with Early Life Stages of Fishes").

Procedures for the acute phase were not explained.

B. Statistics -

The experimental results were not statistically validated for this review because the raw data on embryo survival, as well as larval survival and growth were not submitted.

C. Results -

The 96-hr LC_{50} result is only "supplementary" information. It should not be used to evaluate hazard to freshwater fish because it does not establish a definitive 96-hr LC_{50} with 95% confidence limits.

The summary chronic results indicate that for methomyl exposure the MATC >57 ug/l <117 ug/l. This exposure results in a significant decrease in 28-day larval survival.

Standard length and wet weight first show a significant decrease at 243 ug/l. Decrease of growth therefore occurs at some point >117 ug/l $\leq$ 243 ug/l.

Results of "behavior and appearance" were not reported.

D. Conclusions

1. Category: Supplemental for chronic study. Supplemental for acute study.

2. Rationale: No raw data submitted for chronic study. Acute study was a "range finder" and did not report details of the procedure, nor did it establish a definitive LC_{50}.

3. Repair: None possible for acute study because they did not test high enough concentrations.

Submit the raw data for the embryo hatch, as well as that for larval survival and growth.