

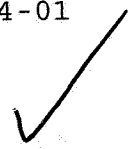
US EPA ARCHIVE DOCUMENT

CBI  
W 512-02

3/17/1999

MRID No. : 445354-01

**DATA EVALUATION RECORD  
AQUATIC INVERTEBRATE LIFE CYCLE TEST  
GUIDELINE 72-4**



1. **CHEMICAL:** Zinc Pyrethione PC Code No.: 088002  
(Zinc 2-pyridinethios-1-oxide)

2. **TEST MATERIAL:** Zinc Omadine<sup>o</sup> Purity: 98.2%

3. **CITATION:**

Authors: R.L. Boeri, J.P. Magazu, and T.J. Ward  
Title: Chronic Toxicity of Zinc Omadine<sup>o</sup> to the Daphnid,  
*Daphnia magna*

Study Completion Date: February 24, 1998

Laboratory: T.R. Wilbury Laboratories, Inc., Marblehead, MA

Sponsor: Olin Corporation, New Haven, CT

Laboratory Report ID: 1312-OL

MRID No.: 445354-01

DP Barcode: D246384

4. **REVIEWED BY:** Max Feken, M.S., Environmental Toxicologist,  
Golder Associates Inc.

**Signature:**

**Date:**

**APPROVED BY:** Pim Kosalwat, Ph.D., Senior Scientist,  
Golder Associates Inc.

**Signature:**

**Date:**

*J. V. Montague*  
OPP/AD/RASSB

3/17/99

REC-30 8.25 PPH  
M 2 211 PPH

5. **APPROVED BY:**

**Signature:**

**Date:**

6. **STUDY PARAMETERS:**

**Age of Test Organism:** < 24 hours  
**Definitive Test Duration:** 21 days  
**Study Method:** Flow-Through  
**Type of Concentrations:** Mean Measured

7. **CONCLUSIONS:** This study is scientifically sound and fulfills the guideline requirements

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for a freshwater invertebrate life-cycle test.

**Results Synopsis:** Most sensitive endpoint: Reproduction and Length

NOEC: 2.7 ppb ai LOEC: 5.8 ppb ai MATC: 4.0 ppb

LOEC's for specific endpoints:

Reproduction: 5.8 ppb ai

Survival: 49 ppb ai

Length: 5.8 ppb ai

Dry Weight: 49 ppb ai

**8. ADEQUACY OF THE STUDY:**

A. **Classification:** Core

B. **Rationale:** Although raw data for growth were not submitted, the statistical analysis using individual measurements would not have changed the NOEC and LOEC values in this study.

C. **Repairability:** N/A

**9. GUIDELINE DEVIATIONS:**

1. Individual lengths and weights of daphnids were not reported. The authors presented mean daphnid length and weight by replicate.

2. Dilution water was deionized tap water.

**10 SUBMISSION PURPOSE:**

**11. MATERIALS AND METHODS:**

A. **Test Organisms/Acclimation:**

Guideline Criteria	Reported Information
Species <i>Daphnia magna</i>	<i>Daphnia magna</i>

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Guideline Criteria	Reported Information
<b>Source</b> Laboratory, commercial, or wild stock.	In-house cultures
<b>Parental Acclimation Conditions</b> Parental stock must be maintained separately from the brood culture in dilution water and under test conditions.	Held in dilution water at a temperature range of 19.6 - 20.6°C
<b>Parental Acclimation Period</b> At least 21 days.	21 days
<b>Age of Parental Stock</b> At least 10-12 days old at the beginning of the acclimation period.	26 days old
<b>Food</b> Synthetic foods (trout chow), algae, or synthetic foods in combination with alfalfa yeast and algae.	Yeast/trout chow suspension and the freshwater algae <i>Selenastrum capricornutum</i>
<b>Food Concentration</b> 5 mg/L (dry wt.) of synthetic food or 10 <sup>8</sup> cells/L of algae is recommended.	Approximately 5 mg/L of yeast/trout chow suspension and <i>S. capricornutum</i> (approx-imately 100,000 cells/mL) twice a day.
<b>Were daphnids in good health during acclimation period?</b>	Yes

**B. Test System:**

Guideline Criteria	Reported Information
<b>Test Water</b> Unpolluted well or spring that has been tested for contamin-ants, or appropriate reconsti-tuted water (see ASTM for details).	Deionized tap water, stored in a polyethylene tank where it was aerated and continuously passed through a particle filter, UV sterilizer, and activated carbon. The hardness was adjusted to 160-180 mg/L as CaCO <sub>3</sub> .

Guideline Criteria	Reported Information
<p><b><u>Water Temperature</u></b> 20°C ±2°C. Must not deviate from 20°C by more than 5°C for more than 48 hours.</p>	20.4 - 21.7°C
<p><b><u>pH</u></b> 7.6 to 8.0 is recommended. Must not deviate by more than one unit for more than 48 hours.</p>	7.6 - 7.9
<p><b><u>Total Hardness</u></b> 160 to 180 mg/L as CaCO<sub>3</sub> is recommended.</p>	164-172 mg/L as CaCO <sub>3</sub>
<p><b><u>Dissolved Oxygen</u></b> <u>Renewal</u>: must not drop below 50% for more than 48 hours. <u>Flow-through</u>: ≥60% throughout test.</p>	≥70% of saturation during the test
<p><b><u>Test Vessels or Compartments</u></b> 1. <u>Material</u>: Glass, No. 316 stainless steel, or perfluorocarbon plastics 2. <u>Size</u>: 250 mL with 200 mL fill volume is preferred; 100 mL with 80 mL fill volume is acceptable.</p>	<p>1. Glass</p> <p>2. 1 L test chambers containing 800 mL of solution.</p>
<p><b><u>Covers</u></b> <u>Renewal</u>: Test vessels should be covered with a glass plate. <u>Flow-through</u>: openings in test compartments should be covered with mesh nylon or stainless steel screen.</p>	Test chambers had Nitex® covered overflows and were loosely covered.
<p><b><u>Type of Dilution System</u></b> Must provide reproducible supply of toxicant. Inter-mittent flow proportional diluters or continuous flow serial diluters should be used.</p>	Intermittent-flow proportional diluter. The flow rate provided 36 volume replacements/day. Measured concentrations of the test compound were consistent throughout the study.
<p><b><u>Renewal Rate</u></b> Three times weekly.</p>	N/A

Guideline Criteria	Reported Information
<p><b><u>Aeration</u></b> Dilution water should be vigorously aerated, but the test tanks should not be aerated.</p>	Dilution water was thoroughly mixed before delivery to the exposure chambers. Exposure chambers were not aerated.
<p><b><u>Photoperiod</u></b> 16 hours light, 8 hours dark</p>	16 hours light, 8 hours dark
<p><b><u>Solvents</u></b> Not to exceed 0.5 mL/L for static tests or 0.1 mL/L for flow-through tests. Acceptable solvents are dimethylformamide, triethylene glycol, methanol, acetone and ethanol.</p>	Solvent: DMF Maximum conc.: 0.1 mL/L

### C. Test Design:

Guideline Criteria	Reported Information
<p><b><u>Duration</u></b> 21 days</p>	21 days
<p><b><u>Nominal Concentrations</u></b> Control(s) and at least 5 test concentrations; dilution factor not less than 50%.</p>	Dilution water control, solvent control, and five nominal concentrations: 3.1, 6.5, 13, 25, and 50 $\mu\text{g ai/L}$
<p><b><u>Number of Test Organisms</u></b> 22 daphnids/level; 7 test chambers should contain 1 daphnid each, and 3 test chambers should contain 5 daphnids each.</p>	40 daphnids/level; 4 test chambers with 10 daphnids each
<p><b>Test organisms randomly or impartially assigned to test vessels?</b></p>	Yes
<p><b><u>Renewal</u></b> Parent daphnids in all beakers must be transferred to containers with fresh test solution (&lt; 4 hours old) three times each week (e.g. every Monday, Wednesday and Friday).</p>	N/A

Guideline Criteria	Reported Information
<p><b><u>Water Parameter Measurements</u></b></p> <p>1. Dissolved oxygen must be measured at each concentration at least once a week.</p> <p>2. pH, alkalinity, hardness, and conductance must be measured once a week in one test concentration and in one control.</p> <p>3. Temperature should be monitored at least hourly throughout the test in one test chamber, and near the beginning, middle and end of the test in all test chambers.</p>	<p>1. DO, temperature, pH, and conductivity were measured in each chamber with live daphnids at test initiation and daily thereafter.</p> <p>2. Hardness and alkalinity were measured weekly in the in the control and highest tested concentration containing live daphnids during the test.</p> <p>3. Temperature was also monitored continuously in one replicate vessel.</p>
<p><b><u>Chemical Analysis</u></b></p> <p>Needed if chemical was volatile, insoluble, or known to absorb, if precipitate formed, if containers were not steel or glass, or if flow-through system was used.</p>	<p>Samples were collected from two replicates of each treatment and control on days 0 (replicate 1 and 2 vessels), 7 (replicate 3 and 4 vessels), 14 (replicate 1 and 2 vessels), and 21 (replicate 3 and 4 vessels), and analyzed by HPLC resulting in mean recoveries ranging from 87 to 98% of nominal. Results were based on the mean measured concentrations.</p>

**12. REPORTED RESULTS:**

**A. General Results:**

Guideline Criteria	Reported Information
<p><b>Quality assurance and GLP compliance statements were included in the report?</b></p>	<p>Yes</p>
<p><b><u>Control Mortality</u></b> ≤ 30%</p>	<p>5% and 7% mortality in the dilution water and solvent control, respectively</p>
<p><b>Did daphnids in each control produce at least 40 young after 21 days?</b></p>	<p>Yes</p>

Guideline Criteria	Reported Information
Were no ehippia produced in any of the controls?	Not reported
<b>Data Endpoints</b> - Survival of first-generation daphnids, - Number of young produced per female, - Dry weight (required) and length (optional) of each first generation daphnid alive at the end of the test, - Observations of other effects or clinical signs.	-Survival of parental daphnids, -Day of first brood, -Number of young produced per surviving adult, -Average dry weight and length of surviving first-generation daphnids, -Clinical signs.
Raw data included?	No, only mean values per replicate were reported.

Effects Data

Toxicant Concentration ( $\mu\text{g ai/L}$ )		No. (%) Dead or Immobile (21 Days)	Mean # Young/ Adult	Mean Total Length (mm)	Mean Dry Weight (mg)
Nominal	Measured				
Control	<0.50	2/40 (5%)	57	3.8	0.62
Solvent Control	<0.50	3/40 (7%)	49	3.8	0.55
3.1	2.7	2/40 (5%)	48	3.8	0.70
6.5	5.8	4/40 (10%)	16 <sup>a</sup>	3.7 <sup>a</sup>	0.62 <sup>a</sup>
13	12	7/40 (17%)	10 <sup>a</sup>	3.6 <sup>a</sup>	0.53 <sup>a</sup>
25	22	7/40 (17%)	4.6 <sup>a</sup>	3.6 <sup>a</sup>	0.56 <sup>a</sup>
50	49	40/40 (100%) <sup>a</sup>	0.0 <sup>b</sup>	-- <sup>b</sup>	-- <sup>b</sup>

IC 50 acute test  
8.25 →

<sup>a</sup>Significantly reduced when compared to the pooled control.  
 present at test termination.

<sup>b</sup>No surviving daphnids were present at test termination.



Toxicity Observations: No sublethal signs of toxicity were observed in surviving daphnids.

**B. Statistical Results:** Growth analysis based on mean values by replicate compared to the pooled controls.

Endpoint	Method	NOEC ( $\mu\text{g ai/L}$ )	LOEC ( $\mu\text{g ai/L}$ )
Survival	Bonferroni's test	22	49
Time to First Brood	Bonferroni's test	12	22
Reproduction	Bonferroni's test	2.7	5.8
Length	Bonferroni's test	2.7	5.8
Dry Weight	Bonferroni's test	22	49

13. **VERIFICATION OF STATISTICAL RESULTS:** Statistical analyses on growth used replicate mean values compared to the solvent control.

Endpoint	Method	NOEC (ppb ai)	LOEC (ppb ai)
Survival	Williams' test	22	49
Reproduction	Williams' test	2.7	5.8
Length	Williams' test	2.7	5.8
Dry Weight	Williams' test	22	49

14. **REVIEWER'S COMMENTS:** Individual growth measurements of the daphnids were not submitted. However, upon examining the replicate means, the reviewer believes that statistical analysis using individual measurements would not change the NOEC and LOEC values.

This study is scientifically sound, fulfills the guideline requirements for a daphnid life-cycle test, and can be classified as Core. Based on the most sensitive endpoints (reproduction and length), the NOEC and LOEC were 2.7 and 5.8 ppb ai, respectively. The MATC was determined to be 4.0 ppb ai.