DATA EVALUATION RECORD

1. **CHEMICAL:** Tetramethrin  
   Shaughnessey No. 69003

2. **TEST MATERIAL:** Neo-Pynamin; Lot No. 90304; 95.3% active ingredient; a white powder.

3. **STUDY TYPE:** Freshwater Fish Acute Flow-Through Toxicity Test. Species Tested: *Lepomis macrochirus*


5. **REVIEWED BY:**
   Louis M. Rifici, M.S.  
   Associate Scientist II  
   KBN Engineering and Applied Sciences, Inc.  
   Signature:  
   Date: 2/22/91

6. **APPROVED BY:**
   Pim Kosalwat, Ph.D.  
   Senior Scientist  
   KBN Engineering and Applied Sciences, Inc.  
   Signature:  
   Date: 2/22/91

   Henry T. Craven, M.S.  
   Supervisor, EEB/HED USEPA  
   Signature:  
   Date: 3/20/92

7. **CONCLUSIONS:** This study is scientifically sound and meets the guideline requirements for an acute flow-through toxicity test for freshwater fish. Based on mean measured concentrations, the 96-hour LC₅₀ was 15.9 µg/L. Therefore, Neo-Pynamin is classified as very highly toxic to bluegill. The NOEC could not be determined due to the presence of sublethal effects at all test concentrations.

8. **RECOMMENDATIONS:** N/A

9. **BACKGROUND:**

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10. DISCUSSION OF INDIVIDUAL TESTS: N/A

11. MATERIALS AND METHODS:

A. Test Animals: Bluegill sunfish (Lepomis macrochirus) were obtained from Osage Catfisheries in Osage Beach, MO. The fish were maintained in well water and fed newly hatched brine shrimp or a commercially available fish food daily. Fish were acclimated to the test temperature for 48 hours and feeding was discontinued. Mean weight and length of the control fish were 2.03 (±0.05) g and 41 (±3) mm. Biomass loading rate in the control was 0.20 g/L/day.

B. Test System: A 4 mg/mL diluter stock solution was prepared using 1.049 g Neo-Pyramid in 250 mL of dimethyl formamide (DMF). The stock was delivered to the diluter using a syringe pump. The proportional diluter delivered 1 liter of test solution or control water to individual 30-liter aquaria at an average rate of 8.5 times per hour (or 6.8 volume replacements per day) over the course of the study. Soft blended water with the characteristics listed in Table 1 (attached) was used as diluent. The laboratory environment was maintained on a 16-hour daylight photoperiod. The test aquaria were immersed in a temperature-controlled water bath set to 22°±1°C. The test was initiated after allowing the diluter to run for 48 hours.

C. Dosage: Ninety-six-hour static test. Based on a preliminary test, five nominal concentrations (25, 50, 100, 200, and 400 µg/L), a dilution water control and a solvent control (0.1 mL DMF/L) were used. The concentrations made were corrected for the purity of the test material.

D. Design: Twenty bluegill were impartially distributed to each aquarium; one aquarium per concentration. Observations of mortality and sublethal responses were made every 24 hours. Dead fish were removed from the containers. The temperature, dissolved oxygen (D.O.), and pH were measured in the control, solvent control and the low, middle and high concentrations every 48 hours.
Neo-Pyamin concentrations were measured by gas-liquid chromatography from samples taken at test initiation and termination.

The fish were not fed during the test.

E. **Statistics:** The median lethal concentration (LC\textsubscript{50}) and associated 95\% confidence interval (C.I.) for each 24-hour interval were calculated using a computer program developed by Stephan et al. (1978).

12. **REPORTED RESULTS:** The mean measured concentrations were 12, 23, 41, 84, and 190 \( \mu \text{g/L} \). These values represent 45 \( \pm 3.3\% \) of nominal concentrations (Table 2, attached). Measured concentrations were about the same at test initiation and termination.

The responses of bluegill are given in Table 6 (attached). The 96-hour LC\textsubscript{50} based on mean measured concentrations was 16 \( \mu \text{g/L} \) (95\% C.I. = 12–23 \( \mu \text{g/L} \)). The slope of the dose-response curve was given as 14 (calculated by least squares regression analysis). Sublethal or lethal effects were seen in all test concentrations except the controls. The no-observed-effect concentration (NOEC) could not be calculated but was given as less than 12 \( \mu \text{g/L} \).

Dissolved oxygen ranged from 98 to 105\%. The pH values ranged from 7.7 to 8.0. The temperature was 22°–23°C throughout the test.

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

The authors presented no conclusions.

Quality Assurance and Good Laboratory Practice Regulation Statements were included in the report, indicating that the study was conducted in accordance with FIFRA Good Laboratory Practice Standards set forth in 40 CFR Part 160.

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

A. **Test Procedure:** The test procedures were generally in accordance with protocols recommended by the guidelines, but deviated from the SEP as follows:

Oxygen saturation was greater than 100\% (the recommended range is 60–100\% at initiation) in all solutions at test initiation (104–105\%).
The NOEC could not be determined because lethal or sublethal (behavioral) effects were observed at all toxicant concentrations.

A 30-minute transition period between light and dark is recommended in the SEP. A transition period was not used in the study.

Each selected nominal concentration was approximately 50% of the next highest concentration. The SEP recommends that each concentration be 60% of the next concentration.

An acclimation period to the test water was not described in the report. The fish were held in well water prior for holding and placed in soft blended water for testing. Previous reviews of reports generated at ABC laboratories (e.g. MRID Nos. 250-147865 and 416851-01) indicate that the well water used was very hard (225-275 mg/L as CaCO₃). The SEP recommends that fish be acclimated to the test water and conditions for at least 2 weeks.

B. **Statistical Analysis:** The reviewer used EPA's Toxanal program to calculate the EC₅₀ value and obtained similar results (see attached printout).

C. **Discussion/Results:** Judging from the response of the control organisms (Table 6, attached), oxygen supersaturation did not modify the response of the bluegill in the test.

The loss of Neo-Pynamin may be the result of temperature related decomposition. A memo from the manufacturer to ABC Laboratories in the Appendix of this report (p. 170) indicated that Neo-Pynamin decomposed under static range-finding conditions. It is unlikely that rapid decomposition would take place under flow-through conditions. However, the three flow-through, acute studies reviewed experienced 29 to 87% reductions from nominal test concentrations. The greatest decreases are found in tests using higher temperatures. The largest decrease (87% reduction) is found at 19°-20°C and 5.0 volume replacements per day in the daphnid test (MRID No. 416096-09). The temperature in the bluegill test was 22°-23°C with 6.8 tank volume replacements per day but the loss of active ingredient averaged only 55%. In any future tests of this product, increasing the number of volume
replacements to near ten per day may be helpful in keeping measured concentrations close to nominal levels.

The test concentrations, measured at 0 and 96 hours, were fairly consistent between sampling times indicating the concentration of the active ingredient was constant throughout the test. The mean measured concentrations given are probably good approximations of the actual concentrations the fish were exposed to.

This study is scientifically sound and meets the guideline requirements for a flow-through freshwater fish toxicity study. The 96-hour LC$_{50}$ of 15.9 µg/L (based on mean measured concentrations) classifies Neo-Pynamin as very highly toxic to bluegill. The NOEC could not be calculated.

D. Adequacy of the Study:

(1) Classification: Core
(2) Rationale: N/A
(3) Repairability: N/A

15. COMPLETION OF ONE-LINER FOR STUDY: Yes, 02-05-91.
LOUIS M. RIFICI NEO-PYRAMIN LEPOMIS MACROCHIRUS 2-4-91

<table>
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<th>CONC.</th>
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</table>

The binomial test shows that 12 and 23 can be used as statistically sound conservative 95 percent confidence limits, because the actual confidence level associated with these limits is greater than 95 percent.

An approximate LC50 for this set of data is 15.91819

When there are less than two concentrations at which the percent dead is between 0 and 100, neither the moving average nor the probit method can give any statistically sound results.
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The material not included contains the following type of information:

___ Identity of product inert ingredients.
___ Identity of product impurities.
___ Description of the product manufacturing process.
___ Description of quality control procedures.
___ Identity of the source of product ingredients.
___ Sales or other commercial/financial information.
___ A draft product label.
___ The product confidential statement of formula.
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