DATA EVALUATION RECORD
AQUATIC INVERTEBRATE LIFE CYCLE TEST
§ 72-4(c)

1. **CHEMICAL:** Propazine PC Code No.: 080808

2. **TEST MATERIAL:** Propazine Purity: 98%

3. **CITATION:**

Authors: Boeri, R. L., P. L. Kowalski and T. J. Ward

Title: Chronic Toxicity of Propazine To The Mysid, *Mysidopsis bahia*

Study Completion Date: October 11, 1995

Laboratory: T. R. Wilbury Laboratories, Inc.
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Marblehead, Massachusetts 01945

Sponsor: Griffin Corporation
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Laboratory Report ID: 573-AB

MRID No.: 441848-03

DP Barcode: D237791

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5. **APPROVED BY:** Anita Pease, OPP/EFED/ERB III

Signature: **Date:**

6. STUDY PARAMETERS:

Scientific Name of Test Organisms: *Mysidopsis bahia*

Age of Test Organism: Neonates (≤ 24 hours old)

Definitive Test Duration 28 days

Study Method: Flow-through

Type of Concentrations: Mean-measured

7. CONCLUSIONS:

In a 28-day life cycle test, *Mysidopsis bahia* neonates were exposed under flow-through conditions to Propazine at nominal concentrations of 0 (negative and solvent controls), 0.31, 0.65, 1.3, 2.5 and 5.0 ppm a.i. Mean-measured concentrations were <0.157 (<LOQ, controls), 0.269, 0.706, 1.20, 2.51 and 5.12, respectively. The high:low ratios for all measured concentrations were <1.5.

Prior to sexual maturity and pairing, there were 60 mysids/level, indiscriminately and equally distributed between two replicates. Within each test vessel, the 30 mysids were evenly subdivided into two retention chambers. Male:female pairs (10 pair/vessel, 20 pair/level) were isolated for individual mating on Day 10 for the negative and solvent controls and mean-measured 0.269 and 0.706 ppm a.i. treatment groups, on Day 11 in the mean-measured 1.20 ppm a.i. treatment group and Day 15 in the mean-measured 2.51 ppm a.i. treatment group. Mysids in the mean-measured 5.12 ppm a.i. treatment group failed to reach a size in which sexual differentiation could be determined, consequently, pairing was not conducted. First-generation mysids were observed for mortality and signs of abnormal behavior at test initiation and at 24-hour intervals thereafter. Second-generation mysids were recorded and removed daily beginning on Day 13 (the day that offspring were first observed). Data endpoints included terminal percent survival of first-generation mysids (Day 28; combined sexes), the day of first brood, reproduction (number of young produced per female per reproductive day), and dry weight and total length of surviving first-generation mysids (Day 28; gender-specific). No effects on behavior or appearance (other than a visual difference in size which was analyzed as length and weight) were observed at any test concentration.

Reproduction was significantly reduced at the mean-measured 0.706-5.12 ppm a.i. treatment levels. The number of offspring/female averaged 17.1 and 22.1 for the negative and solvent controls, respectively, and 18.0, 10.9, 7.2, 1.0 and 0.0 for the mean-measured 0.269, 0.706, 1.20, 2.51 and 5.12 ppm a.i. treatment levels, respectively.

Dry weight was significantly reduced in both males and females at the mean-measured 0.706-5.12 ppm a.i. treatment groups; wet weight was significantly reduced in males at the 2.51 and 5.12 ppm a.i. treatment groups (no differences were observed for females) and length was significantly reduced in both males and females in the mean-measured 2.51 and 5.12 ppm a.i. treatment groups. In males, dry weight averaged 0.85-0.91 mg in the controls and mean-measured 0.269 ppm a.i. treatment group and 0.32-0.77 mg for the 0.709-5.12 ppm a.i. treatment groups; wet weight averaged 3.9-4.3 mg for the controls and mean-measured 0.269-1.20 ppm a.i. treatment groups and 1.5-3.5 mg for the mean-measured 2.51 and 5.12 ppm a.i. treatment groups; and length averaged 7.9-8.4 mm in the controls and mean-measured 0.269-1.20 ppm a.i. treatment groups and 4.8-7.4 in the mean-measured 2.51 and 5.12 ppm a.i. treatment groups. In females, dry weight averaged 1.08-1.17 mg in the controls and mean-measured 0.269 ppm a.i. treatment group and 0.67-0.88 mg for the 0.709-2.51 ppm a.i. treatment groups; wet weight averaged 3.1-5.5 mg for the controls and mean-measured 0.269-2.51 ppm a.i. treatment groups; and length averaged 8.3-8.5 mm in the controls and mean-measured 0.269-1.20 ppm a.i. treatment groups and 6.9-7.3 in the mean-measured 2.51 ppm a.i. treatment group. Length and weight measurements were not taken for females in the mean-measured 5.12 ppm a.i. treatment group due to 100% mortality.

Based on significant effects on growth (dry weight; combined and separate sexes) and reproduction, the NOEC and LOEC are 0.269 and 0.706 ppm a.i., respectively.

This study is scientifically sound. However, since second-generation mysids were not maintained for at least 4 days and observed for survival, development, and behavior, this study does not fulfill the guideline requirements for an aquatic invertebrate life-cycle toxicity test using *Mysidopsis bahia* (§72-4c), and is classified as **Supplemental**.

Results Synopsis:
Survival (Day 28)
NOEC: 0.706 ppm a.i.
LOEC: 1.20 ppm a.i.

Reproductive Success (no. young/female)
NOEC: 0.269 ppm a.i.
LOEC: 0.706 ppm a.i.

Day of First Brood
NOEC: 1.20 ppm a.i.
LOEC: 2.51 ppm a.i.

Male Length
NOEC: 1.20 ppm a.i.

Female Length
NOEC: 1.20 ppm a.i.

LOEC: 2.51 ppm a.i.

LOEC: 2.51 ppm a.i.

Male Dry Weight
NOEC: 0.269 ppm a.i.
LOEC: 0.706 ppm a.i.

Female Dry Weight
NOEC: 0.269 ppm a.i.
LOEC: 0.706 ppm a.i.

Male Wet Weight
NOEC: 1.20 ppm a.i.
LOEC: 2.51 ppm a.i.

Female Wet Weight
NOEC: 2.51 ppm a.i.
LOEC: >2.51 ppm a.i.

Endpoints affected: Survival, growth (total lengths, dry weights and wet weights) of both sexes, day of first brood and reproduction
Most sensitive endpoint: Dry weight (separate and combined sexes) and Reproduction

8. ADEQUACY OF THE STUDY:

A. Classification: Supplemental

B. Rationale: Second-generation mysids were not observed daily for at least 4 days for survival, development, and behavior.

C. Repairability: Since the second-generation mysids were counted and then discarded, this study is not repairable.

9. GUIDELINE DEVIATIONS:

I. The measured pH values during the definitive test (7.4-8.0) ranged lower than recommended (7.6-8.2).

II. The size of the test vessels (20 L) was slightly smaller than recommended (20.25 L) and the reported test solution depth (4-10 cm) ranged lower than recommended (10 cm).

III. The size of the mesh screen and the fill volume of the test compartments were not reported.

IV. It was not reported whether or not the diluter system was checked twice daily during the definitive test period.

V. All pairing was not conducted on the same day.

VI. The quantity of live brine shrimp fed to the mysids was not specified.

VII. The flow splitting accuracy was not reported.

VIII. Gender-specific daily survival data were not provided.

IX. Live offspring were counted and discarded. Survival, development, and behavior of second-generation mysids were not monitored for at least 4 days.

10. SUBMISSION PURPOSE: This study was submitted to provide data on the chronic toxicity of Propazine to an estuarine/marine shrimp for the purpose of new use chemical registration.

11. MATERIALS AND METHODS:

A. Test Organisms/Acclimation

Guideline Criteria	Reported Information
Species An estuarine shrimp species, preferably *Americamysis bahia*	*Mysidopsis bahia*
Source/Supplier	In-house cultures maintained by T. R. Wilbury Laboratories (culture originally procured from Aquatic Research Organisms, Inc., Hampton, New Hampshire).
Age at Beginning of Test <24 hours old	≤24 hours old
Parental Acclimation Parental stock must be maintained separately from the brood culture in dilution water and under test conditions. Mysids should be in good health.	The culture stock was maintained in natural seawater collected from the Atlantic Ocean at T. R. Wilbury Laboratories in Marblehead, Massachusetts. Culture mysids were reported to be free of disease, injury and abnormalities.

Guideline Criteria	Reported Information
Parental Acclimation Period At least 14 days	Continuous
Brood Stock Test started with mysids from: - one brood stock, or - brood stock which has not obtained sexual maturity or had been maintained for >14 days in a laboratory with same food, water, temperature, and salinity used in the test.	At test initiation, juvenile mysids were collected from the laboratory culture stock. The in-house culture was maintained with the same food, water, temperature, salinity, and pH as used in the definitive test.

B. Test System

Guideline Criteria	Reported Information
Source of Dilution Water May be natural (sterilized and filtered) or a commercial mixture; water must be free of pollutants.	Water used for culturing and testing was natural seawater collected from the Atlantic Ocean at T. R. Wilbury Laboratories in Marblehead, Massachusetts. Salinity was adjusted to 16-17 ‰ with deionized tap water. Dilution water was then aerated, passed through a particle-filter, an activated carbon filter and an unltraviolet sterilizer prior to use. Periodic analyses of pesticides, PCB's, and toxic metals in the dilution water (collected during February 1995 as part of a routine water quality monitoring program) indicated that none of these compounds were measured to be above the detection limit (0.5-2 ppb).

Guideline Criteria	Reported Information
	Analysis was performed by Pace, Inc., Hampton, New Hampshire.
Does water support test animals without observable signs of stress?	Yes
Water Temperature 27°C for mysids - At test termination, mean-measured temperature for each chamber should be within 1°C of selected test temperature. - Must be within 3°C of the mean of the time-weighted averages. - Must not differ by >2°C between chambers during the same interval.	Target: 25 ± 2°C Actual range: 24.6-26.8°C. - All criteria were met.
Salinity 15-30 ‰ - The difference between highest and lowest measured salinities should be less than 5‰.	16-17‰ Criteria met.
pH 7.6-8.2	7.4-8.0
Dissolved Oxygen 60-100% saturation	Mean: 6.4 (5.3-8.0) mg/L; ≥70%. (saturation= 7.6 mg/L at 25°C and 16‰ salinity)
Photoperiod 16-hr light/8-hr dark (14-hr light/10-hr dark also acceptable)	16 hours light, 8 hours dark, with an intensity of 25 foot candles. A 15-minute transition period was provided between the light and dark periods.
Test Chambers 1. Material: All glass, No. 316 stainless steel, or	1. Glass

Guideline Criteria	Reported Information
perflorocarbon plastic 2. Size: Typically 30 x 45 x 15 cm (20.25 L) 3. Fill depth: 10 cm 4. Were chambers identical and covered during the test?	2. 20 L 3. Test vessels contained up to 8 L of test solution and were equipped with self-starting siphons to insure adequate flow of test media to the mysids (test media depth ranged from 4-10 cm). 4. The test chambers were identical and were loosely covered
Test Compartments (within chambers) - 250-mL glass beakers with side cutouts covered with nylon mesh or stainless steel screen, or - 90- or 140-mm id glass Petri dish bottoms with collars made of 200-250 µm mesh screen	Test compartments were glass petri dishes (10-cm diameter) with 12-cm high collars of Nitex® screen attached by silicone adhesive. The size of the mesh screen and the fill volume were not reported. Reproductive compartments (beginning on Days 10-15) were glass petri dishes (6-cm diameter) with 12-cm high collars of Nitex® screen attached by silicone adhesive. The size of the mesh screen and the fill volume were not reported.
Type of Dilution System Intermittent flow proportional diluters or continuous flow serial diluters should be used.	A intermittent-flow proportional diluter.
Toxicant Mixing 1. Mixing chamber is recommended but not required; aeration should not be used for mixing. 2. If a mixing chamber was not employed, was it demonstrated that the test solution was completely mixed before introduction into the test system? 3. Was flow splitting accuracy within 10%?	1. A high shear pump was used. 2. N/A 3. Not reported.

Guideline Criteria	Reported Information
Flow Rate 1. 5-10 volume additions per 24 hours. 2. Did the flow rate maintain the toxicant level and the DO at ≥60% of saturation? 3. Were the meter systems calibrated before study and checked twice daily during test period?	1. 8.7 volume additions/24 hours 2. Yes 3. The diluter was calibrated before and after the test and was in operation for 48-hours prior to the addition of the mysids, however it was not reported whether or not the diluter system was checked twice daily during the definitive test period.
Solvents - Acceptable solvents include triethylene glycol, methanol, acetone, and methanol. - Solvent should not exceed 0.1 mL/L in a flow-through system.	DMF, 0.1 mL/L
Aeration Dilution water should be vigorously aerated, but the test tanks should not be aerated.	The dilution water was aerated prior to use. The test chambers were not aerated.

C. Test Design

Guideline Criteria	Reported Information
Duration of the Test Approximately 28 days. Was the test terminated within 7 days of the median time of first brood release in the controls?	28 days No, the study duration was adequate as the first brood release in the controls occurred on Day 13.

Guideline Criteria	Reported Information
Nominal Concentrations Negative control, a solvent control (when applicable), and at least five treatment levels, one of which must adversely affect a life stage and one must not affect any life stage. The dilution factor should not be >50%.	0 (negative and solvent controls), 0.31, 0.65, 1.3, 2.5 and 5.0 ppm a.i. (concentrations were reported based on active ingredient) Dilution factor was 50%.
Distribution Number of mysids before pairing: Minimum of 15 mysids per compartment, 2 compartments per chamber, 2 chambers per concentration for a total of 60/treatment level. Number of mysids after pairing: ≥20 randomly selected pairs/treatment (excess males should be held in separate compartment in same treatment to replace paired males).	60 mysids/level: 15 mysids/compartment, 2 compartments/aquarium, and 2 replicate test aquaria/level. 20 pair/level: 1 pair/compartment, 10 compartments/aquarium, and 2 replicate test aquaria/level. Extra, unpaired mysids were sexually differentiated and placed in chambers 11 and 12.
Pairing Should be conducted when most of the mysids are sexually mature, usually 10-14 days after test initiation. All pairing should occur on the same day.	Pairs were isolated on Day 10 for the negative and solvent controls and mean-measured 0.269 and 0.706 ppm a.i. treatment groups, on Day 11 in the mean-measured 1.20 ppm a.i. treatment group and Day 15 in the mean-measured 2.51 ppm a.i. treatment group. Mysids in the mean-measured 5.12 ppm a.i. treatment group failed to reach a size in which sexual differentiation could be determined, consequently, pairing was not conducted
Test organisms randomly or impartially assigned to test vessels?	Yes

Guideline Criteria	Reported Information
Were treatments randomly assigned to individual test chamber locations?	Yes
Feeding Mysids should be fed live brine shrimp nauplii at least once daily. 150 live brine shrimp nauplii per mysid per day or 75 twice a day is recommended.	Mysids were fed live brine shrimp (*Artemia salina*) nauplii (≤48 hours old) two or three times daily (amount not reported) except for the 24-hour period prior to test termination.
Counts Live adult mysids should be counted at initiation, at pairing, and daily after pairing.	Yes
Live young must be counted and removed daily.	Yes
Missing or impinged animals should be recorded.	Yes
Controls Negative control and carrier control (when applicable) are required.	A negative (saltwater) control and solvent control were used.

Guideline Criteria	Reported Information
Water Parameter Measurements 1. Temperature should be monitored daily in one chamber and at least three times in all chambers. 2. Salinity should be measured daily in at least one test vessel. 3. pH should be measured at the beginning, the end, and at least weekly during the test in the control vessels and highest test level. 4. Dissolved oxygen must be measured at each concentration at least once a week.	1. Temperature was measured daily in each replicate and continuously in the control test vessel. 2-4. Salinity, pH, and DO were measured daily in each replicate test vessel.
Chemical Analysis Toxicant concentration must be measured in one chamber at each toxicant level every week.	Toxicant concentration was measured in each replicate test vessel on Days 0, 7, 14, 21, and 28 as long as living organisms were present. The nominal 0.65 ppm a.i. concentration was measured in duplicate on Day 14. Each batch of samples was accompanied by fortified QC samples at 0.31, 1.3 and 5.0 ppm a.i. as well as a sample of the primary stock solution (50,000 mg/L) and secondary stock solution (5.0 mg/L). All samples were analytically quantified by ABC Laboratories, Columbia, Missouri.

12. REPORTED RESULTS

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Chemical Analysis For all test groups, a) the measured concentration of the test material should not be <50% of the time-weighted average measured concentration for >10% of the duration of the test, and b) the measured concentration should not be >30% of the time-weighted average measured concentration for >5% of the duration of the test.	Mean-measured concentrations were <0.157 (<LOQ, controls), 0.269, 0.706, 1.20, 2.51 and 5.12 ppm a.i., which represents 87-109% of the nominal concentrations. Mean-measured recoveries were 103% for the primary stock solution, 106% for the secondary stock solution and 102-107% for the QC fortifications. Time-weighted average concentrations were not determined.
Controls - Survival of the paired first-generation controls must be ≥70%. - ≥75% of the paired first-generation female controls produced young, or - The average number of young produced by the first-generation female controls was ≥3.	All criteria met. Survival of all (i.e., paired and unpaired) first-generation mysids was ≥90% for each replicate of the negative and solvent controls. The average number of young produced by the first-generation female negative and solvent controls was 17.1 and 22.1 respectively.
Data Endpoints Must Include 1. Survival of first-generation mysids, gender specified 2. Number of live young produced per female 3. Dry weight and length of each first generation mysid alive at the end of the test, gender specified **Data Endpoints Should Also Include**	1. Survival of first-generation mysids at study termination (Day 28; combined sexes) 2. Number of live young produced per female per reproductive day. 3. Dry weight, wet-weight and total length of each first generation living at end of test, gender specific (analyses of growth were conducted using combined sexes).

Guideline Criteria	Reported Information
4. Incidence of morphological findings. 5. Survival, development, and behavior of second-generation mysids for at least 4 days.	4. Abnormal appearance or behavior of first-generation mysids. 5. Criteria NOT met. Toxic effects on second-generation mysids were not addressed in the study.
Raw data must include 1. Survival of first-generation mysids, gender specified 2. Number of live young produced per female 3. Terminal weight and length measurements, individual and gender specified	1. Criteria NOT met. Summarized data were provided for survival (Days 1-28); data were not gender-specific. 2. Criteria met. 3. Criteria met.

Effects Data

Concentration (ppm a.i.)		Survival, Day 28[1]		
Nominal	Mean Measured (% nominal)	No. _	No. _	_ and _ Percentage (and ratio)
Control	<0.157	29	27	93(56/60)
Solvent Control	<0.157	29	27	93(56/60)
0.31	0.269 (87)	25	30	92(55/60)
0.65	0.706 (109)	24	33	95(57/60)
1.3	1.20 (92)	25	16	68(41/60)
2.5	2.51 (100)	20	23	72(43/60)
5.0	5.12 (102)	20	0	33(20/60)

[1] Individual gender data reviewer-summed per sex from male and female length raw data tables.

Concentration (ppm a.i.)		Cumulative Number of Young[1]	Mean Number of Young/Female/Day[1]	Mean Number of Young/Female Day 28
Nominal	Mean Measured (% nominal)			
Control	<0.157	342	1.06	17.1
Solvent Control	<0.157	437	1.37	22.1
0.31	0.269 (87)	363	1.13	18.0
0.65	0.706 (109)	220	0.775	10.9*
1.3	1.20 (92)	146	0.55	7.2*
2.5	2.51 (100)	21	0.141	1.0*
5.0	5.12 (102)	0	0.0**	0.0**

[1] Reviewer-summed from reproduction raw data tables.
* Statistically-significant reduction compared to pooled control (based on Kruskal-Wallis results).
** Value was assumed to be different from the pooled controls (these data were excluded from statistical calculation).

Concentration (ppm a.i.)		Growth, Day 28					
Nominal	Mean Measured (% nominal)	Mean Length, mm		Mean Wet Weight, mg		Mean Dry Weight, mg	
		–	–	–	–	–	–
Control	<0.157	8.0	8.3	4.2	5.3	0.91	1.15
Solvent Control	<0.157	8.3	8.5	4.1	4.7	0.88	1.11
0.31	0.269 (87)	8.1	8.4	4.0	5.2	0.88	1.12
0.65	0.706 (109)	8.1	8.3	4.0	5.0	0.70	0.82
1.3	1.20 (92)	8.2	8.4	4.2	5.0	0.68	0.72
2.5	2.51 (100)	7.2*	7.1*	3.0	3.6	0.56	0.72
5.0	5.12 (102)	4.8*	--	1.7	--	0.32	--

* Statistically-significant reduction compared to pooled control (based on Kruskal-Wallis results).
– All females were dead by test termination.

Toxicity Observations: First-generation mysids were reportedly observed daily for abnormal appearance and behavior. No effects on behavior or appearance (other than a visual difference in size which was analyzed as length and weight) were observed at any test concentration.

Overall survival of first-generation mysids (combined sexes) was significantly different in the mean measured 2.51 and 5.12 ppm a.i. treatment groups which had percent survivals of 71.7 and 33.4%, respectively. Percent survival was 93.4 and 93.3% in the negative and solvent controls, respectively, and ranged from 85.0-95.0% in the mean-measured 0.269-1.20 ppm a.i. treatment groups.

Brood appearance (i.e., gravid females) was observed in the controls, and mean-measured 0.269 ppm a.i. treatment groups on Day 13, on Day 15 in the 0.706 ppm a.i. treatment group, on Day 16 in the 1.20 ppm a.i. treatment group and on Day 22 in the 2.51 ppm a.i. treatment group. No second-generation mysids were produced in the 5.12 ppm a.i. treatment group. Treatment-related effects on reproduction (assessed as the number of offspring/female/reproductive day) was observed in the 0.706-2.51 ppm a.i. treatment groups upon comparison to the pooled control. Significant effects were assumed in the 5.12 ppm a.i. treatment group, however the data was not analyzed due to the absence of second-generation mysids. The number of offspring/female/repro. day averaged 17.1 and 22.1 for the negative and solvent control groups, respectively, and 18.0, 10.9, 7.2, 1.0 and 0.0 for the mean-measured 0.269, 0.706, 1.20, 2.51 and 5.12 ppm a.i. treatment groups,

respectively.

Growth was affected (combined sexes) in the mean-measured 2.51 and 5.12 ppm a.i. treatment groups, with statistically significant reductions in total length of combined sexes compared to pooled control. Total body length averaged 8.2 and 8.4 mm in the negative and solvent controls, respectively, 8.2-8.3 mm in the 0.269-1.20 ppm a.i. treatment groups, 7.1 mm in the 2.51 ppm a.i. treatment group and 4.8 mm in the 5.12 ppm a.i. treatment group; dry weight averaged 1.03 and 0.99 mg for the negative and solvent controls, respectively, 1.01 mg in the 0.269 ppm a.i. treatment group, 0.65-0.77 mg in the 0.706-2.51 ppm a.i. treatment groups and 0.32 mg in the 5.12 ppm a.i. treatment group; and wet weight averaged 4.7 and 4.4 mg in the negative and solvent controls, respectively, 4.6-4.7 mg in the 0.269-1.20 ppm a.i. treatment groups, 3.3 mg in the 2.51 ppm a.i. treatment group and 1.7 mg in the 5.12 ppm a.i. treatment group. All surviving mysids were observed to be visually smaller than the controls at 1.20 ppm a.i. from Day 7 through Day 22, at 2.51 ppm a.i. from Day 7 through Day 28 and at 5.12 ppm a.i. from Day 3 through Day 28.

B. Statistical Results:

Statistical Method(s): Statistical analyses were performed on terminal (Day 28) survival of the first-generation mysids (combined sexes), the number of young released per female per reproductive day, day of first brood release, terminal length and wet and dry weight of each surviving first-generation mysid (combined sexes). Data were analyzed by standard statistical techniques using a computer program (Gulley *et al*, 1990). A *t*-Test was conducted for each endpoint to compare the negative and solvent control responses. No significant differences were observed for survival, reproduction, length and wet weight, and these groups were therefore pooled prior to subsequent comparisons. A significant difference was observed for dry weight, and therefore treatment data were compared to the solvent control data.

The Shapiro-Wilk's Test was used to determine that data were normally distributed, and Bartlett's Test was used to determine that variances were homogeneous. If variances homogenous and normally distributed, a parametric one-way analysis of variance (ANOVA) and Bonferroni's test were used to compare treatment and control means (survival, reproduction, and dry weight). If variances were heteroscedastic or data were not normally distributed, a non-parametric ANOVA (Kruskal and Wallis Test, Steel and Torrie, 1960) was used to compare control and treatment means (wet weight, day of first brood, and total length). Analyses were conducted at the 95% level of certainty, except for the Bartlett's and Shapiro-Wilk's Tests, in which the 99% level of certainty was applied. The MATC was calculated as the geometric mean of the NOEC and LOEC. Mean-measured values were used in all analyses.

Results Synopsis

Endpoint	Method	NOEC	LOEC	MATC
Adult Survival (Day 28)	Bonferroni's Test	1.20 ppm a.i.	2.51 ppm a.i.	1.74 ppm a.i
Reproduction (young/female)	Bonferroni's Test	0.269 ppm a.i.	0.706 ppm a.i.	0.436 ppm a.i.
Day of First Brood	Kruskal Wallis Test	1.20 ppm a.i.	2.51 ppm a.i.	1.74 ppm a.i
Total Length (mm)	Kruskal Wallis Test	1.20 ppm a.i.	2.51 ppm a.i.	1.74 ppm a.i
Mean Dry Weight (mg)	Bonferroni's Test	5.12 ppm a.i.	>5.12 ppm a.i.	>5.12 ppm a.i.
Mean Wet Weight (mg)	Kruskal Wallis Test	5.12 ppm a.i.	>5.12 ppm a.i.	>5.12 ppm a.i.

Most sensitive endpoint(s): Reproduction (young/female)

13. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method(s): Percent survival, reproduction (mean number of young/female) growth, separate and combined sexes (terminal length, wet weight and dry weight) negative and solvent control data were pooled for all statistical analyses since a *t*-test indicated no significant differences ($p>0.05$). The negative and solvent control data for time to first brood release were identical. After confirming normality and homogeneity of variances, the treatment response data for the above endpoints were statistically compared to the pooled control using ANOVA and Bonferroni's multiple comparison test. If the assumptions of ANOVA were not met (normality and homogeneity) then the analysis was conducted using the non-parametric Kruskal-Wallis test. The above analyses were performed using TOXSTAT statistical software. Additionally, the reviewer calculated percent reductions from control, particularly for endpoints subject to non-parametric statistical analysis. NOEC and LOEC values for each endpoint were determined based on the results of the statistical analyses and the visual interpretation of percent reduction data. At the highest mean-measured treatment level, 5.12 ppm a.i., no offspring were produced; therefore, that treatment level was excluded from the statistical analyses of time to first brood release, reproduction and female growth (length, wet weight and dry weight). All toxicity values were determined in terms of the mean-measured treatment concentrations.

Results Synopsis

Endpoint	Method	NOEC	LOEC
Adult Survival (Day 28)	William's Test	0.706 ppm a.i.	1.20 ppm a.i.
Reproduction (young/female)	William's Test	0.269 ppm a.i.	0.706 ppm a.i.
Day of First Brood	Kruskal Wallis Test	1.20 ppm a.i.	2.51 ppm a.i.
Total Male Length (mm)	Kruskal Wallis Test; % Reduction	1.20 ppm a.i.	2.51 ppm a.i.
Total Female Length (mm)	William's Test	1.20 ppm a.i.	2.51 ppm a.i.
Total Length (Combined Sexes; mm)	Kruskal-Wallis Test; % Reduction	1.20 ppm a.i.	2.51 ppm a.i.
Mean Male Dry Weight (mg)	William's Test	0.269 ppm a.i.	0.706 ppm a.i.
Mean Female Dry Weight (mg)	William's Test	0.269 ppm a.i.	0.706 ppm a.i.
Total Dry Weight (Combined Sexes; mg)	Kruskal-Wallis Test; % Reduction	0.269 ppm a.i.	0.706 ppm a.i.
Mean Male Wet Weight (mg)	William's Test	1.20 ppm a.i.	2.51 ppm a.i.
Mean Female Wet Weight (mg)	Kruskal-Wallis Test	2.51 ppm a.i.	>2.51 ppm a.i.
Total Wet Weight (Combined Sexes; mg)	Kruskal-Wallis Test; % Reduction	1.20 ppm a.i.	2.51 ppm a.i.

Most sensitive endpoint(s): Reproduction (mean young/female) and Dry Weight (for separate and combined sexes).

14. REVIEWER'S COMMENTS:

Results of the reviewer's statistical analyses were identical to those of the study authors for reproduction (mean number of young/female) and total wet and dry weight (combined sexes). The reviewer's toxicity values for adult survival were more conservative than the study authors and less conservative than the study authors for day to first brood release and total length (combined sexes). The reviewer calculated and considered the biological relevance of percent reductions from control when statistical tests (particularly non-

parametric tests) revealed no differences. Based on these considerations, the reviewer concluded that the 15% reduction in total length at the mean-measured 2.51 ppm a.i. treatment level, the 28% reduction in wet weight at the mean-measured 2.51 ppm a.i. treatment level and the 24% reduction in dry weight at the mean-measured 0.706 ppm a.i. treatment level were biologically significant, even though the statistical analyses did not detect differences for these endpoints at these respective levels. The reviewer's and the study authors' statistical analyses presumably did not detect these biologically significant reductions because the study design only employed two replicates per treatment level, which greatly reduced the statistical power and, thus, ability to detect differences from control. Based on the reviewer's interpretation of the results, the NOEC and LOEC values were determined to be 1.20 and 2.51 ppm a.i., respectively, for length, 1.20 and 2.51 ppm a.i., respectively for wet weight and 0.269 and 0.706 ppm a.i., respectively, for dry weight (combined sexes).

Screening tests were attempted on October 13 and 25, and November 7, 1994. These tests were terminated after 4 to 8 days due to either poor control survival or erratic survival throughout the test. A final screening test was conducted under static-renewal conditions for 18 days from November 14 to December 2, 1994. Nominal concentrations of Propazine were 0.010, 0.10, 0.50, 1.0 and 5.0 ppm a.i. Mysids were also ezposed to a dilution water control and a solvent control (0.1 mL/L dimethylformamide). After 18 days of exposure there was at least 50% survival in the control, solvent control, 0.010, 0.10 and 0.50 ppm a.i. test vessels, 30% survival at 1.0 ppm a.i. and 0% survival at 5.0 ppm a.i.

The study was conducted using estuarine salinity (16 ‰). If salinity were to be found to affect the activity of Propazine, a study reflecting marine salinity (30-35 ‰) would be necessary to address the salinity difference between marine and estuarine habitats.

Test solutions containing Propazine were analytically quantified by ABC Laboratories, Columbia, Missouri, with samples collected from each replicate test vessel on Days 0, 7, 14, 21 and 28, as long as living organisms were present. Approximately 60-ml samples were collected from each test vessel, replicates were pooled, and transferred to a 125 ml amber glass bottle.

Each batch of samples was accompanied by fortified QC samples at 0.31, 1.3 and 5.0 ppm a.i. prepared at T. R. Wilbury Laboratories, Inc. A sample of the primary stock solution (50,000 ppm a.i.) and secondary stock solution (5.0 ppm a.i.) was collected into a 125 ml glass bottle and provided with each set of samples. Samples were shipped to ABC Laboratories in coolers with ice packs by overnight courier.

The analyses of the QC samples yielded mean recoveries of 103, 102 and 107%, respectively; the primary and secondary stock solutions had mean percent recoveries of 106 and 103%, respectively; and the laboratory control spikes (0.258 and 5.16 ppm a.i.)

yielded mean percent recoveries of 111 and 110%, respectively.

All samples during the definitive test were analyzed for a.i. using gas-liquid chromatography (GLC) by ABC Laboratories, Columbia, Missouri.
The stability of the test material under exposure conditions was assumed but not verified.

Throughout the definitive exposure period, no visible sign of undissolved test substance was observed in the mixing chamber, the chemical cells of the diluter system, or in any of the exposure solutions.

Pesticide and PCB data were collected during February, 1995 as part of a routine water quality monitoring program. Analysis was performed by Pace, Inc., Hampton, New Hampshire.

This study was performed according to U.S. EPA (FIFRA) Good Laboratory Practice Standards (40 CFR, Part 160) with the exception of the collection for the water and food contaminant screening analyses. A signed and dated Quality Assurance Statement was provided.

15. REFERENCES:

ASTM. 1990. Guide for Conducting Life-Cycle Toxicity Tests with Saltwater Mysids. Designation D 1191-90.

Gulley, D. D., A. M. Boelter and H. L. Bergman. 1990. Toxstat Version 3.3. Fish Physiology and Toxicology Laboratory, University of Wyoming, Laramie, Wyoming.

Steel, R. G. D. and J. H. Torrie. 1960. Principles and Procedures of Statistics. McGraw-Hill. New York.

Stephan, C. E. 1983. Computer Program for the Calculation of LC_{50} Values. U.S. EPA. Duluth, MN. Personal Communication.

U.S. EPA. 1985. Standard Evaluation Procedure. Fish Early Life Stage. Hazard Evaluation Division, Office of Pesticide Programs. Washington, DC.

U.S. EPA. 1988. Pesticide Assessment Guidelines. Subdivision E, Hazard Evaluation: Wildlife and Aquatic Organisms. 72-4. Fish Early Life Stage and Aquatic Invertebrate Life-Cycle Studies. Ecological Effect Branch, Hazard Evaluation Division, Office of Pesticide Programs, Washington, DC. Draft, March 1988.

U.S. EPA. 1992. 40 CFR Part 160. Federal Insecticide, Fungicide and Rodenticide Act (FIFRA); Good Laboratory Practice Standards; Final Rule.

16. RESULTS OF STATISTICAL VERIFICATION:

Percent Survival (%) Adults_Day 28
File: 4803as Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	5970.446	1194.089	85.628
Within (Error)	8	111.562	13.945	
Total	13	6082.009		

Critical F value = 3.69 (0.05,5,8)
Since F > Critical F **REJECT Ho:All groups equal**

Percent Survival (%) Adults_Day 28
File: 4803as Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	93.325	93.325		
2	0.269	91.650	91.650	0.518	
3	0.706	95.000	95.000	-0.518	
4	1.20	85.000	85.000	2.574	
5	2.51	71.650	71.650	6.702	*
6	5.12	33.350	33.350	18.545	*

Bonferroni T table value = 2.90 (1 Tailed Value, P=0.05, df=8,5)

Percent Survival (%) Adults_Day 28
File: 4803as Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.269	2	9.369	10.0	1.675
3	0.706	2	9.369	10.0	-1.675
4	1.20	2	9.369	10.0	8.325
5	2.51	2	9.369	10.0	21.675
6	5.12	2	9.369	10.0	59.975

Percent Survival (%) Adults_Day 28
File: 4803as Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	93.325	93.325	93.325
2	0.269	2	91.650	91.650	93.325
3	0.706	2	95.000	95.000	93.325
4	1.20	2	85.000	85.000	85.000
5	2.51	2	71.650	71.650	71.650
6	5.12	2	33.350	33.350	33.350

Percent Survival (%) Adults_Day 28
File: 4803as Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE WILLIAMS P=.05	DEGREES OF FREEDOM
GRPS 1&2 POOLED	93.325				
0.269	93.325	0.000		1.86	k= 1, v= 8
0.706	**93.325**	**0.000**		**1.96**	**k= 2, v= 8**
1.20	85.000	2.574	*	2.00	k= 3, v= 8
2.51	71.650	6.702	*	2.01	k= 4, v= 8
5.12	33.350	18.545	*	2.02	k= 5, v= 8

s = 3.734
Note: df used for table values are approximate when v > 20.

Reproduction (mean number of young/female)_Day 28
File: 4803yf Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	4	586.097	146.524	30.311
Within (Error)	7	33.840	4.834	
Total	11	619.937		

Critical F value = 4.12 (0.05,4,7)
Since F > Critical F **REJECT Ho:All groups equal**

Reproduction (mean number of young/female)_Day 28
File: 4803yf Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	19.550	19.550		
2	0.269	18.000	18.000	0.814	
3	0.706	10.850	10.850	4.569	*
4	1.20	7.150	7.150	6.512	*
5	2.51	1.000	1.000	9.742	*

Bonferroni T table value = 2.84 (1 Tailed Value, P=0.05, df=7,4)

Reproduction (mean number of young/female)_Day 28
File: 4803yf Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.269	2	5.411	27.7	1.550
3	0.706	2	5.411	27.7	8.700
4	1.20	2	5.411	27.7	12.400
5	2.51	2	5.411	27.7	18.550

Reproduction (mean number of young/female)_Day 28
File: 4803yf Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	19.550	19.550	19.550
2	0.269	2	18.000	18.000	18.000
3	0.706	2	10.850	10.850	10.850
4	1.20	2	7.150	7.150	7.150
5	2.51	2	1.000	1.000	1.000

Reproduction (mean number of young/female)_Day 28
File: 4803yf Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE WILLIAMS P=.05	DEGREES OF FREEDOM
GRPS 1&2 POOLED	19.550				
0.269	**18.000**	**0.814**		**1.89**	**k= 1, v= 7**
0.706	10.850	4.569	*	2.00	k= 2, v= 7
1.20	7.150	6.512	*	2.04	k= 3, v= 7
2.51	1.000	9.742	*	2.06	k= 4, v= 7

s = 2.199
Note: df used for table values are approximate when v > 20.

Days Until First Brood Release
File: 4803br Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	solvent control	13.000	13.000	7.000
2	neg control	13.000	13.000	7.000
3	0.269	13.000	13.000	7.000
4	0.706	15.500	15.500	16.000
5	1.20	16.000	16.000	18.000
6	**2.51**	**23.000**	**23.000**	**23.000**

Calculated H Value = 10.777 Critical H Value Table = 11.070
Since Calc H < Crit H **FAIL TO REJECT Ho:All groups are equal.**

Days Until First Brood Release
File: 4803br Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP 0 0 0 0 0 0 2 3 1 4 5 6
2	neg control	13.000	13.000	\
3	0.269	13.000	13.000	. \
1	solvent control	13.000	13.000	. . \
4	0.706	15.500	15.500	. . . \
5	1.20	16.000	16.000	. . . . \
6	**2.51**	**23.000**	**23.000**	**. . . . . **

* = significant difference (p=0.05) . = no significant difference
Table q value (0.05,6) = 2.936 SE = 3.351

Length (mm); Males_Day 28

File: 4803ml Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	GRPS 1&2 POOLED	8.125	8.125	37.000
2	0.269	8.100	8.100	18.000
3	0.706	8.100	8.100	18.000
4	1.20	8.200	8.200	22.000
5	2.51	7.150	7.150	7.000
6	**5.12**	**4.800**	**4.800**	**3.000**

Calculated H Value = 8.971 Critical H Value Table = 11.070
Since Calc H < Crit H **FAIL TO REJECT Ho:All groups are equal.**

Length (mm); Males_Day 28
File: 4803ml Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP 6 5 2 3 1 4
6	**5.12**	**4.800**	**4.800**	****
5	2.51	7.150	7.150	. \
2	0.269	8.100	8.100	. . \
3	0.706	8.100	8.100	. . . \
1	GRPS 1&2 POOLED	8.125	8.125	. . . . \
4	1.20	8.200	8.200	. . . . . \

* = significant difference (p=0.05) . = no significant difference
Table q value (0.05,6) = 2.936 Unequal reps - multiple SE values

Length (mm); Female_Day 28
File: 4803fl Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	4	2.692	0.673	33.650
Within (Error)	7	0.137	0.020	
Total	11	2.829		

Critical F value = 4.12 (0.05,4,7)
Since F > Critical F **REJECT Ho:All groups equal**

Length (mm); Female_Day 28
File: 4803fl Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	8.375	8.375		
2	0.269	8.400	8.400	-0.204	
3	0.706	8.350	8.350	0.204	
4	1.20	8.350	8.350	0.204	
5	2.51	7.100	7.100	10.410	*

Bonferroni T table value = 2.84 (1 Tailed Value, P=0.05, df=7,4)

Length (mm); Female_Day 28
File: 4803fl Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.269	2	0.348	4.2	-0.025
3	0.706	2	0.348	4.2	0.025
4	1.20	2	0.348	4.2	0.025
5	2.51	2	0.348	4.2	1.275

Length (mm); Female_Day 28
File: 4803fl Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	8.375	8.375	8.383
2	0.269	2	8.400	8.400	8.383
3	0.706	2	8.350	8.350	8.350
4	1.20	2	8.350	8.350	8.350
5	2.51	2	7.100	7.100	7.100

Length (mm); Female_Day 28
File: 4803fl Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE WILLIAMS P=.05	DEGREES OF FREEDOM
GRPS 1&2 POOLED	8.383				
0.269	8.383	0.069		1.89	k= 1, v= 7
0.706	8.350	0.206		2.00	k= 2, v= 7
1.20	**8.350**	**0.206**		**2.04**	**k= 3, v= 7**
2.51	7.100	10.505	*	2.06	k= 4, v= 7

s = 0.140
Note: df used for table values are approximate when v > 20.

Total Length (mm)_Combined Sexes_Day 28
File: 4803tl Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	GRPS 1&2 POOLED	8.250	8.250	36.000
2	0.269	8.300	8.300	21.000
3	0.706	8.250	8.250	17.500
4	1.20	8.300	8.300	20.500
5	2.51	7.100	7.100	7.000
6	**5.12**	**4.850**	**4.850**	**3.000**

Calculated H Value = 8.819 Critical H Value Table = 11.070
Since Calc H < Crit H **FAIL TO REJECT Ho:All groups are equal.**

Total Length (mm)_Combined Sexes_Day 28
File: 4803tl Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP 0 0 0 0 0 0 6 5 3 1 2 4
6	**5.12**	**4.850**	**4.850**	****
5	2.51	7.100	7.100	. \
3	0.706	8.250	8.250	. . \
1	GRPS 1&2 POOLED	8.250	8.250	. . . \
2	0.269	8.300	8.300	. . . . \
4	1.20	8.300	8.300	. . . . . \

* = significant difference (p=0.05) . = no significant difference
Table q value (0.05,6) = 2.936 Unequal reps - multiple SE values

Dry Weight (mg); Males_Day 28

File: 4803dm Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.538	0.108	54.000
Within (Error)	8	0.014	0.002	
Total	13	0.553		

Critical F value = 3.69 (0.05,5,8)
Since F > Critical F **REJECT Ho:All groups equal**

Dry Weight (mg); Males_Day 28
File: 4803dm Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	0.890	0.890		
2	0.269	0.880	0.880	0.258	
3	0.706	0.700	0.700	4.906	*
4	1.20	0.675	0.675	5.551	*
5	2.51	0.560	0.560	8.521	*
6	5.12	0.320	0.320	14.717	*

Bonferroni T table value = 2.90 (1 Tailed Value, P=0.05, df=8,5)

Dry Weight (mg); Males_Day 28
File: 4803dm Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.269	2	0.112	12.6	0.010
3	0.706	2	0.112	12.6	0.190
4	1.20	2	0.112	12.6	0.215
5	2.51	2	0.112	12.6	0.330
6	5.12	2	0.112	12.6	0.570

Dry Weight (mg); Males_Day 28

File: 4803dm Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	0.890	0.890	0.890
2	0.269	2	0.880	0.880	0.880
3	0.706	2	0.700	0.700	0.700
4	1.20	2	0.675	0.675	0.675
5	2.51	2	0.560	0.560	0.560
6	5.12	2	0.320	0.320	0.320

Dry Weight (mg); Males_Day 28
File: 4803dm Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS		SIG TABLE P=.05 WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	0.890				
0.269	**0.880**	**0.271**		**1.86**	**k= 1, v= 8**
0.706	0.700	5.157	*	1.96	k= 2, v= 8
1.20	0.675	5.835	*	2.00	k= 3, v= 8
2.51	0.560	8.957	*	2.01	k= 4, v= 8
5.12	0.320	15.471	*	2.02	k= 5, v= 8

s = 0.043
Note: df used for table values are approximate when v > 20.

Dry Weight (mg); Females_Day 28
File: 4803df Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	4	0.426	0.107	26.750
Within (Error)	7	0.026	0.004	
Total	11	0.452		

Critical F value = 4.12 (0.05,4,7)
Since F > Critical F **REJECT Ho:All groups equal**

Dry Weight (mg); Females_Day 28
File: 4803df Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	1.135	1.135		
2	0.269	1.120	1.120	0.274	
3	0.706	0.820	0.820	5.751	*
4	1.20	0.730	0.730	7.394	*
5	2.51	0.725	0.725	7.486	*

Bonferroni T table value = 2.84 (1 Tailed Value, P=0.05, df=7,4)

Dry Weight (mg); Females_Day 28
File: 4803df Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.269	2	0.156	13.7	0.015
3	0.706	2	0.156	13.7	0.315
4	1.20	2	0.156	13.7	0.405
5	2.51	2	0.156	13.7	0.410

Dry Weight (mg); Females_Day 28
File: 4803df Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	GRPS 1&2 POOLED	4	1.135	1.135	1.135
2	0.269	2	1.120	1.120	1.120
3	0.706	2	0.820	0.820	0.820
4	1.20	2	0.730	0.730	0.730
5	2.51	2	0.725	0.725	0.725

Dry Weight (mg); Females_Day 28
File: 4803df Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

GROUP	IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG TABLE WILLIAMS P=.05	DEGREES OF FREEDOM

GRPS 1&2 POOLED	1.135				
0.269	**1.120**	**0.283**		**1.89**	**k= 1, v= 7**
0.706	0.820	5.948	*	2.00	k= 2, v= 7
1.20	0.730	7.647	*	2.04	k= 3, v= 7
2.51	0.725	7.741	*	2.06	k= 4, v= 7

s = 0.061
Note: df used for table values are approximate when v > 20.

Total Dry Weight (mg)_Combined Sexes_Day 28
File: 4803td Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	GRPS 1&2 POOLED	1.008	1.008	45.500
2	0.269	1.010	1.010	23.500
3	0.706	0.775	0.775	14.000
4	1.20	0.700	0.700	11.500
5	2.51	0.645	0.645	7.500
6	**5.12**	**0.320**	**0.320**	**3.000**

Calculated H Value = 11.778 Critical H Value Table = 11.070
Since Calc H > Crit H **REJECT Ho:All groups are equal.**

Total Dry Weight (mg)_Combined Sexes_Day 28
File: 4803td Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP 0 0 0 0 0 0 6 5 4 3 1 2
6	**5.12**	**0.320**	**0.320**	****
5	2.51	0.645	0.645	. \
4	1.20	0.700	0.700	. . \
3	0.706	0.775	0.775	. . . \
1	GRPS 1&2 POOLED	1.008	1.008	. . . . \
2	0.269	1.010	1.010	. . . . . \

* = significant difference (p=0.05) . = no significant difference
Table q value (0.05,6) = 2.936 Unequal reps - multiple SE values

Wet Weight (mg); Males_Day 28
File: 4803wm Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	10.698	2.140	22.526
Within (Error)	8	0.762	0.095	
Total	13	11.460		

Critical F value = 3.69 (0.05,5,8)
Since F > Critical F **REJECT Ho:All groups equal**

Wet Weight (mg); Males_Day 28
File: 4803wm Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	GRPS 1&2 POOLED	4.125	4.125		
2	0.269	4.050	4.050	0.281	
3	0.706	4.050	4.050	0.281	
4	1.20	4.200	4.200	-0.281	
5	2.51	2.950	2.950	4.402	*
6	5.12	1.700	1.700	9.085	*

Bonferroni T table value = 2.90 (1 Tailed Value, P=0.05, df=8,5)

Wet Weight (mg); Males_Day 28
File: 4803wm Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	GRPS 1&2 POOLED	4			
2	0.269	2	0.773	18.7	0.075
3	0.706	2	0.773	18.7	0.075
4	1.20	2	0.773	18.7	-0.075
5	2.51	2	0.773	18.7	1.175
6	5.12	2	0.773	18.7	2.425

Wet Weight (mg); Males_Day 28
File: 4803wm Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	ORIGINAL	TRANSFORMED	ISOTONIZED

	IDENTIFICATION	N	MEAN	MEAN	MEAN
1	GRPS 1&2 POOLED	4	4.125	4.125	4.125
2	0.269	2	4.050	4.050	4.100
3	0.706	2	4.050	4.050	4.100
4	1.20	2	4.200	4.200	4.100
5	2.51	2	2.950	2.950	2.950
6	5.12	2	1.700	1.700	1.700

Wet Weight (mg); Males_Day 28
File: 4803wm Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG	TABLE P=.05 WILLIAMS	DEGREES OF FREEDOM
GRPS 1&2 POOLED	4.125				
0.269	4.100	0.094		1.86	k= 1, v= 8
0.706	4.100	0.094		1.96	k= 2, v= 8
1.20	**4.100**	**0.094**		**2.00**	**k= 3, v= 8**
2.51	2.950	4.395	*	2.01	k= 4, v= 8
5.12	1.700	9.070	*	2.02	k= 5, v= 8

s = 0.309
Note: df used for table values are approximate when v > 20.

Wet Weight (mg); Females_Day 28
File: 4803wf Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN ORIGINAL UNITS	MEAN CALCULATED IN RANK	SUM
1	GRPS 1&2 POOLED	5.050	5.050	32.000
2	0.269	5.150	5.150	16.000
3	0.706	5.000	5.000	12.000
4	1.20	5.050	5.050	15.000
5	**2.51**	**3.550**	**3.550**	**3.000**

Calculated H Value = 5.149 Critical H Value Table = 9.490
Since Calc H < Crit H **FAIL TO REJECT Ho:All groups are equal.**

Wet Weight (mg); Females_Day 28
File: 4803wf Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

	GROUP
TRANSFORMED	ORIGINAL 0 0 0 0 0

GROUP	IDENTIFICATION	MEAN	MEAN	5 3 4 1 2
5		**2.51**	**3.550**	**3.550 **
3		0.706	5.000	5.000 . \
4		1.20	5.050	5.050 . . \
1	GRPS 1&2 POOLED		5.050	5.050 . . . \
2		0.269	5.150	5.150 . . . . \

* = significant difference (p=0.05) . = no significant difference
Table q value (0.05,5) = 2.807 Unequal reps - multiple SE values

Total Wet Weight (mg)_Combined Sexes_Day 28
File: 4803tw Transform: NO TRANSFORMATION

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	GRPS 1&2 POOLED	4.550	4.550	36.000
2	0.269	4.700	4.700	23.000
3	0.706	4.600	4.600	16.000
4	1.20	4.650	4.650	20.000
5	2.51	3.300	3.300	7.000
6	**5.12**	**1.700**	**1.700**	**3.000**

Calculated H Value = 9.315 Critical H Value Table = 11.070
Since Calc H < Crit H **FAIL TO REJECT Ho:All groups are equal.**

Total Wet Weight (mg)_Combined Sexes_Day 28
File: 4803tw Transform: NO TRANSFORMATION

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP 0 0 0 0 0 0 6 5 1 3 4 2
6		**5.12**	**1.700**	**1.700 **
5		2.51	3.300	3.300 . \
1	GRPS 1&2 POOLED		4.550	4.550 . . \
3		0.706	4.600	4.600 . . . \
4		1.20	4.650	4.650 . . . . \
2		0.269	4.700	4.700 . . . . . \

* = significant difference (p=0.05) . = no significant difference
Table q value (0.05,6) = 2.936 Unequal reps - multiple SE values