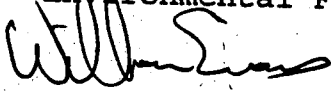



US EPA ARCHIVE DOCUMENT

DATA EVALUATION RECORD  
AQUATIC INVERTEBRATE LIFE CYCLE TEST  
GUIDELINE 72-4(B)

1. **CHEMICAL:** PIRATE™; AC 303,630 PC Code No.: 129093
2. **TEST MATERIAL:** 4-bromo-2-(4-chlorophenyl)-1-(ethoxymethyl)-5-(trifluoromethyl)-1H-pyrrole-3-carbonitrile  
  
Purity: 94.5%
3. **CITATION**  

Authors: Ward, G. Scott, Wisk, Joseph D., Davis, Jay W.  
Title: Chronic Toxicity of AC 393,630 to the Mysid (*Mysidopsis bahia*) Under Flow Test conditions  
Study Completion Date: July 6, 1994  
Laboratory: Toxikon Environmental Sciences, 106 Coastal Way, Jupiter, Florida 33477  
Sponsor: American Cyanamid Company, Agricultural Research Division, P.O. Box 400, Princeton, NJ 08543-0400  
Laboratory Report ID: J9301002  
MRID No.: 434928-21  
DP Barcode: D210808
4. **REVIEWED BY:** William Evans, Biologist  
Ecological Effects Branch  
Environmental Fate and Effects Division  

Signature:  Date: 12/9/96
5. **APPROVED BY:** Ann Stavola, Section Chief, Section 5  
Ecological Effects Branch  
Environmental Fate and Effects Division  

Signature:  Date: 12/9/96
6. **STUDY PARAMETERS**  

Age of Test Organism: <24 hours old at test initiation  
Definitive Test Duration: 28 days  
Study Method: Flow-through  
Type of Concentrations: Mean measured
7. **CONCLUSIONS:** According to study authors', the most sensitive endpoint to mysid shrimp was survival. The NOEC was observed to be 0.172 µm/L. However, this effect level cannot be confirmed since insufficient raw data was provided to verify the statistical test for survival. Specifically, the replicate data for first generation males was missing. In

order to confirm the authors' results, this data must be submitted. Additionally, individual growth data as dry weight by sex was not submitted and statistical analysis could not be performed. This data must also be submitted. This study is, therefore, classified as supplemental. Upon verification of the statistics this study may be classified as core.

#### Results Synopsis

NOEC: 0.172  $\mu\text{g/L}$       LOEC: 0.385  $\mu\text{mg ai/L}$       MATC: 0.257  $\mu\text{g ai/L}$

#### LOEC's for specific effects

Young/Female/Repro. Day: 0.970  $\mu\text{g ai/L}$

Larvae Survival: 0.385  $\mu\text{g ai/L}$

Growth 1) length: Male >3.86  $\mu\text{g ai/L}$

Female >3.86  $\mu\text{g ai/L}$

Male & Female >3.86  $\mu\text{g ai/L}$

2) weight: Male \_\_\_\_  $\mu\text{g ai/L}$

Female \_\_\_\_  $\mu\text{g ai/L}$

Male & Female 3.86  $\mu\text{g ai/L}$

#### 8. ADEQUACY OF THE STUDY

A. Classification: Supplemental

B. Rationale: Insufficient raw data was provided to verify the statistical test for survival. Data for first generation males needs to be submitted. Additionally, individual growth data as dry weight by sex needs to be submitted to perform statistical analysis.

C. Repairability: N/A

#### 8. MAJOR GUIDELINE DEVIATIONS:

1. The age of Parental Stock should be at least 10-12 days old at the beginning of the acclimation period. The age of parental stock was not mentioned.
2. Pairing of mysids should be accomplished on the same day and should be conducted when most of the mysids are sexually mature (usu. 10-14 days after test initiation). It does not appear that the mysids were separated into pairs.
3. Food concentration was not mentioned. Guidelines recommend 5 mg/L (dry wt.) of synthetic food or  $10^8$  cells/L of algae.
4. The water Temperature must not deviate from 20°C by more than 5°C for more than 48 hours. According to

protocol the target water temperature was  $27 \pm 2^{\circ}\text{C}$  with a measured range of  $26.1$  to  $29^{\circ}\text{C}$  which deviates more than  $5^{\circ}\text{C}$  for more than 48 hours.

5. Total hardness of dilution water was not mentioned in the study. A hardness of 160 to 180 mg/L as  $\text{CaCO}_3$  is recommended.
6. A minimum of 15 mysids per compartment, 2 compartments per chamber, 2 chambers per concentration for a total of 60/level should be distributed before pairing. Two replicates each containing 2 screened chambers with 10 mysids/chamber for a total of 40/level were distributed for this test.
7. During the test, difference between highest and lowest measured salinities must be less than 10 g/kg, should be measured daily, and should be between 15 and 30 g/kg. Salinity was between 18-20‰ (parts per thousand) in control measured every 7 days.
8. Mean measured temperature for each chamber at test termination should be within  $1^{\circ}\text{C}$  of selected test temperature. Each individual measured temperature must be within  $3^{\circ}\text{C}$  of the mean of the time-weighted averages. For mysid shrimp,  $27^{\circ}\text{C}$  is recommended. Whenever temp. is measured concurrently in more than one test chamber the highest & lowest temp. must not differ by more than  $2^{\circ}\text{C}$ . As the target temperature was reported to be  $27 \pm 2^{\circ}\text{C}$  with a range of  $26.1$  to  $29^{\circ}\text{C}$ , it can not be determined if all the guideline criteria are met.
9. Meter systems should be calibrated before study and checked twice daily during test period, and renewal must not drop below 50% for more than 48 hours. However, actual exposure concentrations were collected and determined on days 0, 6, 13, 20, and 28.
10. According to guideline criteria the following solvents are acceptable: triethylene glycol, methanol, acetone, ethanol. DMF (Dimethylformamide) was used for this test.
11. According to guideline criteria the average number of young produced by the 1<sup>st</sup> generation females in the control(s) should be more than 3. The average number of live young produced in controls for first generation was 1.10 and 1.03 for the solvent control.

12. Individual dry weight measurements by sex should have been submitted. Only mean dry weight measurements were submitted.

10. MATERIALS AND METHODS:

A. Biological System:

Guideline Criteria	Reported Information
<b>Species:</b> An estuarine shrimp species, preferably <u>Americamysis bahia</u> .	Test species was Mysid ( <u>Mysidopsis bahia</u> )
<b>Duration</b> 28 days/one generation	28 days
<b>Source</b> (or supplier)	Aquatic BioSystems, Inc., Fort Collins, CO
<b>Parental Acclimation</b> 1) Parental stock must be maintained separately from the brood culture in dilution water and under test conditions. 2) Mysids should be in good health.	Salinity: 20-23‰ Temperature: 21.1-62°C lighting: 16h/8h Cultures were maintained separately and were in good health (no diseases were observed).
<b>Parental Acclimation Period</b> At least 14 days	Acclimated for two week period prior to test initiation.
<b>Chamber Location:</b> Treatments should be randomly assigned to test chamber locations.	Treatments were randomly assigned to test chamber locations.
<b>Duration of the Test:</b> A mysid test must not be terminated before 7 days past the median time of 1 <sup>st</sup> brood release in the control treatment.	1 <sup>st</sup> brood released in control and solvent controls on days 12 and 13 respectively. The test duration was 28 days.

Guideline Criteria	Reported Information
<b>Brood Stock:</b> Test started with mysids: 1) from only one brood stock or 2) from 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.	Post-larval mysids from only one brood stock.
<b>Distribution:</b> <b>No. of mysids before pairing:</b> Minimum of 15 mysids per compartment, 2 compartments per chamber, 2 chambers per concentration for a total of 60/level. <b>No. of mysids after pairing:</b> ≥ 20 randomly selected pairs/treatment (excess males should be held in separate compartment to replace paired males).	<b>Before pairing:</b> Two replicates each containing 2 screened chambers with 10 mysids/chamber for a total of 40/level. <b>After pairing:</b> 20 offspring per replicate were transferred into additional retention chambers within the same test chamber.
<b>Pairing:</b> 1) Should be conducted when most of the mysids are sexually mature (usu. 10-14 days after test initiation). 2) Should be paired on the same day	Does not appear that adult mysids were separated into pairs.
<b>Feeding:</b> 1) Mysids should be fed live brine shrimp nauplii at least once daily. 2) 150 live brine shrimp nauplii per mysid per day or 75 twice a day is recommended.	Mysids fed live brine shrimp ( <i>Artemia salina</i> ) nauplii hatched daily from cysts at least once daily throughout test, generally fed 2-3 times/day.

Guideline Criteria	Reported Information
<b>Counts:</b> Live adult mysids should be counted 1) at initiation, 2) at pairing, 3) and daily after pairing. 4) Live young must be counted and removed daily. 5) Missing or impinged animals should be recorded.	Live and dead were monitored daily throughout study according to guidance criteria.
<b>Controls:</b> Survival in any control chamber (between pairing and test termination) must not be less than 70%.	83-94% control survival
<b>Controls:</b> Negative control and carrier control (when applicable) are required.	Solvent: Dimethylformamide (DMF)

Comments: None

**B. Physical System:**

Guideline Criteria	Reported Information
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<p><b>Test Water:</b></p> <ol style="list-style-type: none"><li>1) May be natural (sterilized and filtered) or a commercial mixture;</li><li>2) During the test, difference between highest and lowest measured salinities must be less than 10 g/kg. Should be measured daily.</li><li>3) Salinity should be between 15 and 30 g/kg.</li><li>4) Measured pH should be between 7.6 and 8.2. Must not deviate by more than one unit for more than 48 hours. Should be measured at the beginning, end of test and weekly.</li><li>5) Water must be free of pollutants.</li><li>6) DO must be measured @ each conc. @ least once a wk. (see details in ASTM)</li></ol>	<ol style="list-style-type: none"><li>1) Filtered saltwater from shallow well, aerated, carbon-treated, and adjusted for salinity.</li><li>2&amp;3) Salinity between 18-20‰ in control measured every 7 days.</li><li>4) pH was 7.8 to 8.3.</li><li>5) Water was free of pollutants.</li><li>6) D.O. measured once per week in all test solutions</li></ol>
<p><b>Test Temperature:</b></p> <ol style="list-style-type: none"><li>1) Mean measured temperature for each chamber at test termination should be within 1°C of selected test temperature.</li><li>2) Each individual measured temperature must be within 3°C of the mean of the time-weighted averages.</li><li>3) For mysid shrimp, 27°C is recommended.</li><li>4) Whenever temp. is measured concurrently in more than one test chamber the highest &amp; lowest temp. must not differ by more than 2°C.</li></ol>	<p>Water temperature in dilution water control was monitored and recorded hourly using a data logger and spot checked with a thermocouple thermometer daily. Water bath temperature was monitored continuously with a minimum/maximum thermometer and the diurnal temperature range recorded daily. Target: <math>27 \pm 2^{\circ}\text{C}</math> Range: 26.1 to 29°C</p>
<p><b>Photoperiod:</b> Recommend 16L/8D.</p>	<p>16 hours light, 8 hours dark.</p>



<b>Dosing Apparatus:</b> 1) Intermittent flow proportional diluters or continuous flow serial diluters should be used. 2) A minimum of 5 toxicant concentrations 3) with a dilution factor not greater than 0.5 and controls should be used.	Continuous flow aerial diluters were used on 5 test concentrations. The dilution factor was 50%. Controls were used.
<b>Toxicant Mixing:</b> 1) Mixing chamber is recommended but not required; 2) Aeration should not be used for mixing; 3) It must be demonstrated that the test solution is completely mixed before intro. into the test system; 4) Flow splitting accuracy must be within 10%.	Mixing chambers were used and aeration was not used for mixing. A total volume of 36.46 $\mu$ L of the secondary stock solution was pumped into the 3710 ml chemical mixing chamber during each dilution cycle.
<b>Test Vessels:</b> 1) Material: all glass, No. 316 stainless steel, or perfluorocarbon plastic 2) Size: 250 ml with 200 ml fill volume is preferred; 100 ml with 80 ml fill volume acceptable 3) 90 or 140 mm inside dia. glass Petri dish bottoms with collars made of 200 - 250 $\mu$ m mesh screen.	24 liter glass tanks for the test chambers. Screened retention chambers were 142 mm diameter glass petri dish with a 17 cm high collar of 355 $\mu$ m mesh nitex screen.
<b>Covers</b> 1) Renewal: Test vessels should be covered with a glass plate. 2) Flow-through: Openings in the test compartments should be covered with nylon mesh or stainless steel screen.	Screened retention chambers covered with 17 cm high collar of 355 $\mu$ m-mesh nitex screening.

<b>Flow Rate:</b> 1) Flow rates should provide 5 to 10 volume additions per 24 hr. 2) Flow rate must maintain DO at or above 60% of saturation and maintain the toxicant level. 3) Meter systems calibrated before study and checked twice daily during test period 4) Renewal must not drop below 50% for more than 48 hours.	6.2 volume additions/24 hours. DO was maintained above 60% saturation at toxicant level. Actual exposure concentrations were collected and determined on days 0, 6, 13, 20, and 28.
<b>Aeration:</b> 1) Dilution water should be aerated to insure DO concentration at or near 100% saturation. 2) Test tanks may be aerated.	Dilution water aerated by air stone and the test solutions were gently aerated to maintain d.o. concentrations.

Comments:C. Chemical System:

Guideline Criteria	Reported Information
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<p><b>Concentrations:</b></p> <ol style="list-style-type: none"> <li>1) Minimum of 5 concentrations and a control, all replicated, plus solvent control if appropriate.</li> <li>2) Toxicant conc. must be measured in one tank at each toxicant level every week.</li> <li>3) One concentration must adversely affect a life stage and one concentration must not affect any life stage.</li> <li>4) The measured conc. of the test material of any treatment should be at least 50% of the time-weighted average measured conc. for &gt;10% of the duration of the test.</li> <li>5) The measured conc. for any treatment level should not be more than 30% higher than the time-weighted average measured conc. for more than 5% of the duration of the test.</li> </ol>	<ol style="list-style-type: none"> <li>1) Control &amp; solvent control and 5 test concentrations at 50% dilution factor. The test substance was a 1:1 mixture of <sup>14</sup>C-AC 303,630 and non-radio labeled technical AC 303,630.</li> <li>2) Actual exposure concentrations were collected and determined on days 0, 6, 13, 20, and 28.</li> <li>3) At least one concentration adversely affects a life stage and at least one concentration does not.</li> <li>4) Measured conc. of the test material of any treatment was at least 50% of the time-weighted average measured conc. for &gt;10% of the duration of the test.</li> <li>5) Measured conc. for any treatment level was not more than 30% higher than the time-weighted average measured conc. for more than 5% of the duration of the test.</li> </ol>
<p><b>Solvents:</b></p> <ol style="list-style-type: none"> <li>1) Should not exceed 0.1 ml/L in a flow-through system.</li> <li>2) Following solvents are acceptable: triethylene glycol, methanol, acetone, ethanol.</li> </ol>	<p>Solvent: Dimethylformamide (DMF) Maximum conc.: 0.00983 ml/L.</p>

Comments:**11. REPORTED RESULTS:**

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes

Guideline Criteria	Reported Information
1) At least 75% of the paired 1 <sup>st</sup> generation females in the control produced young or 2) the average number of young produced by the 1 <sup>st</sup> generation females in the control(s) was more than 3.	The average number of live young produced in controls for first generation was 1.10 and 1.03 for the solvent control.
<b>Data Endpoints must include:</b> 1) Survival of first-generation mysids Female Male 2) Number of live young produced per female 3) Dry weight of each first-generation mysid alive at the end of the test Female Male 4) Length of each 1 <sup>st</sup> generation mysid alive at the end of the study Female Male 5) Incidence of pathological or histological effects; 6) Observations of other effects or clinical signs.	1) Yes 2) Yes 3) Individual dry weight measurements by sex were not submitted. 4) Yes 5) Pathological or histological effects was not mentioned 6) Observations and abnormalities in the behavior or physical appearance of time to first brood was recorded.
<b>Raw data included? (Y/N)</b> At a minimum, individual data should be included for: 1) surviving 1st generation ♂ and ♀ mysids. 2) Number of live young produced per female. 3) Individual length measurements of ♂ and ♀ mysids. 4) Individual dry weight measurements for ♂ and ♀ mysids at the end of the test.	1) Data was included. 2) Data was included. 3) Data was included. 4) Data for each sex was not included.

Effects Data:-

Toxicant Conc. ( $\mu\text{g/L}$ ) <sup>1</sup>		Mean # Young/fem. /repro. day <sup>2</sup>	Survival (28 days)			Mean Total Length (mm)			Mean Dry weight (mg) <sup>3</sup>		
Nom.	Meas.		♂	♀	♂ & ♀	♂	♀	♂ & ♀	♂	♀	♂ & ♀
Ctrl	<0.10 <sub>8</sub>	1.08	14	17	31	6.1	6.5	6.3			0.89
Sol	<0.10 <sub>8</sub>	1.03	15	24	39	5.9	6.3	6.1			0.89
0.25	0.172	0.72	12	22	34	6.1	6.9	6.4			0.87
0.50	0.385	0.69	15	17	32	6.0	6.3	6.1			0.82
1.0	0.970	0.31	14	18	32	6.1	6.2	6.2			0.89
2.0	1.89	0.35	13	19	32	6.2	6.4	6.3			0.79
4.0	3.86	0.38	9	5	14	6.3	6.3	6.3			0.66

<sup>1</sup> Measured concentration of PIRATE™ as <sup>14</sup>AC 303,630 equivalents.

<sup>2</sup> Calculated by dividing the cumulative number of young released by the number or reproductive days. For each female, the number of reproductive days is the number of days the female was alive from the day of first brood release by any female in the test to the end of the test. This corrects for varying number of reproductive days per treatment and female mortality.

<sup>3</sup> The test assumed that the first young were produced in 12 days. However, data submitted shows that the observed length of time required for production of young varied from 12 days in control replicates to 23 days at the 3.86  $\mu\text{g/L}$  level.

<sup>4</sup> Individual growth data as dry weight by sex was not presented in this submission.

Toxicity Observations:Statistical Results:

The following results were obtained by the authors.

Most sensitive endpoint: Survival was the most sensitive endpoint.

Endpoint	Method	NOEC (µg/L)			LOEC (µg/L)			MATC (µg/L)		
Survival	Fisher's Exact test	0.172			0.385			0.257		
Reproduction	ANOVA	0.385			0.970			0.611		
		♂	♀	♂ & ♀	♂	♀	♀ & ♂	♂	♀	♀ & ♂
Weight	ANOVA			1.89			3.86			2.70
Length	ANOVA			>3.86			>3.86			>3.86

Comments: Control data were pooled because there was no significant difference between solvent and dilution water control. Statistical analysis for individual data by sex for length and dry weight was not performed.

## 12. Reviewer's Statistical Results:

Most sensitive endpoint:

Endpoint	Method	NOEC (µg/L)			LOEC (µg/L)			MATC (µg/L)		
Survival	N/A	0.172			N/A			N/A		
Reproduction	ANOVA Bonferroni test	0.385			0.970			0.611		
		♂	♀	♂ & ♀	♂	♀	♀ & ♂	♂	♀	♀ & ♂
Weight	ANOVA Bonferroni test			1.89			3.86			2.70
Length	ANOVA Bonferroni test			>3.86			>3.86			>3.86

Comments: Insufficient raw data was provided to run the test for survival. Specifically, the replicate data for first generation males was missing. In order to confirm the authors results this data must be submitted. Additionally, since individual data by sex for dry weight was not submitted statistical analysis could not be performed. Finally, as there was no significant difference between the solvent and dilution control groups, the dilution control was omitted from the analysis. However, the same results were obtained for reproduction, weight, and length of first generation mysids.

mysid chronic repro pooled  
File: mysidrp Transform: NO TRANSFORMATION

t-test of Solvent and Blank Controls

Ho:GRP1 MEAN = GRP2 MEAN

GRP1 (SOLVENT CTRL) MEAN =	1.0275	CALCULATED t VALUE =	-0.2374
GRP2 (BLANK CTRL) MEAN =	1.0950	DEGREES OF FREEDOM =	6
DIFFERENCE IN MEANS =	-0.0675		

---

TABLE t VALUE (0.05 (2), 6) = 2.447 NO significant difference at alpha=0.05  
TABLE t VALUE (0.01 (2), 6) = 3.707 NO significant difference at alpha=0.01

mysid chronic repro pooled  
File: mysidrp Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	2.486	0.497	3.601
Within (Error)	22	3.036	0.138	
Total	27	5.523		

Critical F value = 2.66 (0.05,5,22)  
Since F > Critical F REJECT Ho:All groups equal

mysid chronic repro pooled  
File: mysidrp 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	1.061	1.061		
2	0.172	0.752	0.752	1.357	
3	0.385	0.783	0.783	1.225	
4	0.97	0.313	0.313	3.291	*
5	1.89	0.448	0.448	2.698	*
6	3.86	0.325	0.325	3.236	*

Bonferroni T table value = 2.51 (1 Tailed Value, P=0.05, df=22,5)

mysid chronic repro pooled  
File: mysidrp Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2

Ho:Control<Treatment

chronic mysid repro.

File: mysidr

Transform: NO TRANSFORM

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	1.706	0.341	2.795
Within (Error)	18	2.193	0.122	
Total	23	3.899		

Critical F value = 2.77 (0.05,5,18)

Since F > Critical F REJECT Ho:All groups equal

chronic mysid repro.

File: mysidr

Transform: NO TRANSFORM

BONFERRONI T-TEST

TABLE 1 OF 2

Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	solvent control	1.028	1.028		
2	0.172	0.773	0.773	1.032	
3	0.385	0.783	0.783	0.992	
4	0.97	0.313	0.313	2.895	*
5	1.89	0.448	0.448	2.348	
6	3.86	0.325	0.325	2.844	*

Bonferroni T table value = 2.55 (1 Tailed Value, P=0.05, df=18,5)

chronic mysid repro.

File: mysidr

Transform: NO TRANSFORM

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	solvent control	4			
2	0.172	4			
3	0.385	4	0.631	61.4	0.255
4	0.97	4	0.631	61.4	0.245
5	1.89	4	0.631	61.4	0.715
6	3.86	4	0.631	61.4	0.580
			0.631	61.4	0.702

15



mysid chronic length

File: mysidl

Transform: NO TRANSFORMATION

# ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.228	0.046	1.278
Within (Error)	18	0.650	0.036	
Total	23	0.878		

Critical F value = 2.77 (0.05,5,18)

Since  $F < \text{Critical } F$  FAIL TO REJECT  $H_0$ : All groups equal

mysid chronic length

File: mysidl

Transform: NO TRANSFORMATION

## BONFERRONI T-TEST

TABLE 1 OF 2

$H_0$ : Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	solvent control	6.100	6.100		
2	0.172	6.350	6.350	-1.863	
3	0.385	6.100	6.100	0.000	
4	0.97	6.150	6.150	-0.373	
5	1.89	6.300	6.300	-1.491	
6	3.86	6.250	6.250	-1.118	

Bonferroni T table value = 2.55 (1 Tailed Value,  $P=0.05$ ,  $df=18,5$ )

mysid chronic length

File: mysidl

Transform: NO TRANSFORMATION

## BONFERRONI T-TEST

TABLE 2 OF 2

$H_0$ : Control < Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	solvent control	4			
2	0.172	4	0.343	5.6	-0.250
3	0.385	4	0.343	5.6	0.000
4	0.97	4	0.343	5.6	-0.050
5	1.89	4	0.343	5.6	-0.200
6	3.86	4	0.343	5.6	-0.150

mysid chronic length

mysid chronic length male

File: mysidlm

Transform: NO TRANSFORMATION

# ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.423	0.085	2.576
Within (Error)	18	0.590	0.033	
Total	23	1.013		

Critical F value = 2.77 (0.05,5,18)

Since  $F < \text{Critical } F$  FAIL TO REJECT  $H_0$ : All groups equal

mysid chronic length male

File: mysidlm

Transform: NO TRANSFORMATION

## BONFERRONI T-TEST

TABLE 1 OF 2

$H_0$ : Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	solvent control	5.875	5.875		
2	0.172	6.100	6.100	-1.752	
3	0.385	5.975	5.975	-0.778	
4	0.97	6.075	6.075	-1.557	
5	1.89	6.200	6.200	-2.530	
6	3.86	6.275	6.275	-3.114	

Bonferroni T table value = 2.55 (1 Tailed Value,  $P=0.05$ ,  $df=18,5$ )

mysid chronic length male

File: mysidlm

Transform: NO TRANSFORMATION

## BONFERRONI T-TEST

TABLE 2 OF 2

$H_0$ : Control < Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	solvent control	4			
2	0.172	4	0.328	5.6	-0.225
3	0.385	4	0.328	5.6	-0.100
4	0.97	4	0.328	5.6	-0.200
5	1.89	4	0.328	5.6	-0.325
6	3.86	4	0.328	5.6	-0.400

mysid chronic length females  
File: mysidlf Transform: NO TRANSFORM

# ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.237	0.047	0.516
Within (Error)	18	1.642	0.091	
Total	23	1.880		

Critical F value = 2.77 (0.05,5,18)  
Since  $F < \text{Critical F}$  FAIL TO REJECT  $H_0$ : All groups equal

mysid chronic length females  
File: mysidlf Transform: NO TRANSFORM

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

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	solvent control	6.275	6.275		
2	0.172	6.425	6.425	-0.703	
3	0.385	6.100	6.100	0.820	
4	0.97	6.250	6.250	0.117	
5	1.89	6.350	6.350	-0.352	
6	3.86	6.275	6.275	0.000	

Bonferroni T table value = 2.55 (1 Tailed Value,  $P=0.05$ ,  $df=18,5$ )

mysid chronic length females  
File: mysidlf Transform: NO TRANSFORM

## 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	solvent control	4			
2	0.172	4	0.545	8.7	-0.150
3	0.385	4	0.545	8.7	0.175
4	0.97	4	0.545	8.7	0.025
5	1.89	4	0.545	8.7	-0.075
6	3.86	4	0.545	8.7	0.000

mysid chronic weight

File: mysidw

Transform: NO TRANSFORM

# ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.158	0.032	6.400
Within (Error)	18	0.093	0.005	
Total	23	0.251		

Critical F value = 2.77 (0.05,5,18)

Since  $F > \text{Critical } F$  REJECT  $H_0$ : All groups equal

mysid chronic weight

File: mysidw

Transform: NO TRANSFORM

## BONFERRONI T-TEST - TABLE 1 OF 2

$H_0$ : Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	solvent control	0.887	0.887		
2	0.172	0.870	0.870	0.350	
3	0.385	0.815	0.815	1.450	
4	0.97	0.888	0.888	-0.000	
5	1.89	0.793	0.793	1.900	
6	3.86	0.655	0.655	4.650	*

Bonferroni T table value = 2.55 (1 Tailed Value,  $P=0.05$ ,  $df=18,5$ )

mysid chronic weight

File: mysidw

Transform: NO TRANSFORM

## BONFERRONI T-TEST - TABLE 2 OF 2

$H_0$ : Control < Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	solvent control	4			
2	0.172	4	0.128	14.4	0.017
3	0.385	4	0.128	14.4	0.072
4	0.97	4	0.128	14.4	-0.000
5	1.89	4	0.128	14.4	0.095
6	3.86	4	0.128	14.4	0.232

```
if trtmnt = 'solvent';  
run;  
*/  
title2 ' ';  
proc npar1way wilcoxon data=trout;  
class trtmnt;  
var weight;  
quit;
```