TENNESSEE VALLEY AUTHORITY
TOXICITY TEST REPORT

INTRODUCTION / EXECUTIVE SUMMARY

Report Date: May 13, 2003

1. Facility / Discharger: Watts Bar Nuclear Plant / TVA
2. County / State: Rhea / Tennessee
3. NPDES Permit #: TN 0020168
4. Type of Facility: Nuclear-Fueled Electric Generating Plant
5. Average Flow (MGD): 0.255
6. Receiving Stream: Unnamed tributary to Yellow Creek
7. 1Q10: 0.0
8. Outfall Tested: 112
9. Dates Sampled: April 13 – 18, 2003
10. Average Flow on Days Sampled (MGD): 0.5690, 0.3895, 0.6537
11. Pertinent Site Conditions: No unusual conditions reported. (Production / operation data will be provided upon request).
12. Test Dates: April 15 - 22, 2003
13. Test Type: Short-term Chronic Definitive
14. Test Species: Fathead Minnows (*Pimephales promelas*)
    Daphnids (*Ceriodaphnia dubia*)
15. Concentrations Tested (%):
    Outfall 112: 6.25, 12.5, 25, 50, 100
    Pimephales promelas: UV treated Outfall 112: 6.25, 12.5, 25, 50, 100
16. Permit Limit Endpoint (%): Outfall 112: $IC_{25} = 100\%$
17. Test Results: Outfall 112: *Pimephales promelas:* $IC_{25} = 82.5\%$
    Ceriodaphnia dubia: $IC_{25} > 100$
    UV treated Outfall 112: *Pimephales promelas:* $IC_{25} > 100$

18. Facility Contact: Martha I. Ervin
Phone #: (423) 365-3585

19. Consulting / Testing Lab: Environmental Testing Solutions, LLC

20. Lab Contact: Jim Sumner
Phone #: (828) 350-9364

21. TVA Contact: Cynthia L. Russell
Phone #: (256) 386-2755

22) Notes: Outfall 112 samples collected April 13 - 18, 2003, showed no toxic effects to daphnids. The resulting IC_{25} value was > 100 percent.

Bacterial growth was observed on the gills of minnows exposed to non-treated effluent from Outfall 112. This pathogenic growth was likely the cause of mortality in the effluent. The resulting IC_{25} value was 82.5 percent.

Fathead minnows were also exposed to UV treated Outfall 112 samples since fish pathogens present in upstream water have been the suspected cause of interference (anomalous dose response and high variability among replicates) in previous toxicity testing at Watts Bar Nuclear Plant. Survival was 100% in all effluent concentrations with two-minute exposure to UV light prior to introduction of test organisms. These results suggest that fish pathogen interference is a likely cause of minnow mortality in the effluent. The resulting IC_{25} value for UV-treated effluent was > 100 percent for fathead minnows.

METHODS SUMMARY

Samples:

1. Sampling Point: Outfall 112

2. Sample Type: Composite

3. Sample Information:

Sample ID	Date (MM/DD/YY)/ Time (EST) Collected	Date (MM/DD/YY)/ Time (EST) Received	Arrival Temp. (°C)	Initial TRC* (mg/L)	Date (MM/DD/YY)/ Time (EST) Used
112	04/13/03 0830 to 04/14/03 0730	04/15/03 0936	0.5†	<0.10	04/15/03 1420 0416/03 1432
112	04/15/03 0930 to 04/16/03 0830	04/17/03 0942	0.2†	<0.10	04/17/03 1426 04/18/03 1326
112	04/17/03 0828 to 04/18/03 0728	04/19/03 0958	0.4†	<0.10	04/19/03 1337 04/20/03 1350 04/21/03 1356

*TRC = Total Residual Chlorine

†Collected in 2.5 and 5-gallon cubitainers. Temperature was measured in each cubitainer upon arrival

4. Sample Manipulation: Samples from Outfall 112 were warmed to test temperature (25.0 ± 1.0°C) in a warm water bath.

Aliquots of Outfall 112 were UV-treated through a 40-watt Rainbow Lifeguard® UV Sterilizer for 2 minutes.

	Pimephales promelas	*Ceriodaphnia dubia*
Test Organisms:		
1. Source:	Aquatic BioSystems, Inc.	In-house Cultures
2. Age:	23-26 hours old	< 24-hours old
Test Method Summary:		
1. Test Conditions:	Static, Renewal	Static, Renewal
2. Test Duration:	7 days	Until at least 60% of control females have 3 broods
3. Control / Dilution Water:	Moderately Hard Synthetic	Moderately Hard Synthetic
4. Number of Replicates:	4	10
5. Organisms per Replicate:	10	1
6. Test Initiation: (Date/Time)		
Outfall 112	04/15/03-1420 EDT	04/15/03-1319 EDT
UV Treated Outfall 112	04/15/03-1403 EDT	
7. Test Termination: (Date/Time)		
Outfall 112	04/22/03-1333 EDT	04/22/03-1236 EDT
UV Treated Outfall 112	04/22/03-1314 EDT	
8. Test Temperature: Outfall 112:	Mean = 25.3°C (24.9-25.7°C)	Mean = 24.9°C (24.3-25.8°C)
Test Temperature: UV-Treated Outfall 112:	Mean = 25.2°C (24.6-25.8°C)	

9. Physical / Chemical Measurements: Alkalinity, hardness, total residual chlorine, and conductivity were measured at the laboratory in each 100% sample. Daily temperatures were measured in one replicate for each test concentration. Pre- and post-exposure test solutions were analyzed daily for pH and dissolved oxygen.

10. Statistics: Statistics were performed according to methods prescribed by EPA using ToxCalc version 5.0 statistical software (Tidepool Scientific Software, McKinneyville, CA).

TOXICITY TEST RESULTS (see Appendix B for Bench Sheets)

1. Results of a *Pimephales promelas* (Genus species) Chronic/ 7-day Toxicity Test. (Type / Duration)

Conducted April 15 - 22, 2003 using effluent from Outfall 112.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
6.25%	100	100	98	98	98	98	98
12.5%	100	100	100	98	90	90	90
25%	100	100	100	85	83	80	80
50%	100	100	95	88	88	88	88
100%	100	95	65	65	63	60	60

96-hour LC_{50} Value: ≥ 100%	Calculated TU Estimates: < 1.0 TUa* Permit Limit: NA

Test Solutions (% Effluent)	Mean Dry Weight (mg) (replicate number)				
	1	2	3	4	Mean
Control	0.917	0.902	0.827	0.733	0.845
6.25%	0.887	0.738	0.720	0.957	0.825
12.5%	0.782	0.823	0.987	0.573	0.791
25%	0.767	0.532	0.761	0.767	0.707
50%	0.960	0.598	0.655	0.869	0.770
100%	0.494	0.490	0.522	0.802	0.577

IC_{25} Value: 82.5% Permit Limit: 100%	Calculated TU Estimates: 1.2 TUc* Permit Limit: 1.0 TUc

*TUa = $100/LC_{50}$: TUc = $100/ IC_{25}$

TOXICITY TEST RESULTS (see Appendix B for Bench Sheets)

2. Results of a *Ceriodaphnia dubia* (Genus species) Chronic/ 7-day Toxicity Test. (Type / Duration)

Conducted April 15 - 22, 2003 using effluent from Outfall 112.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
6.25%	100	100	100	100	100	100	100
12.5%	100	100	100	100	100	100	100
25%	100	100	100	100	100	100	100
50%	100	100	100	100	100	100	100
100%	100	100	100	100	100	100	100

96-hour LC_{50} Value: > 100%	Calculated TU Estimates: < 1.0 TUa* Permit Limit: NA

Test Solutions (% Effluent)	Reproduction (#young/female/7 days) Data (replicate number)										
	1	2	3	4	5	6	7	8	9	10	Mean
Control	26	32	32	30	34	34	28	33	30	27	30.6
6.25%	32	34	36	36	33	34	35	30	31	31	33.2
12.5%	33	33	29	38	34	35	33	40	34	34	34.3
25%	38	39	31	34	34	38	39	38	37	34	36.2
50%	39	38	36	36	37	38	39	38	41	35	37.7
100%	37	39	41	39	34	37	37	40	37	38	37.9

IC_{25} Value: 82.5% Permit Limit: 100%	Calculated TU Estimates: ≤1.0 TUc* Permit Limit: 1.0 TUc

*TUa = $100/LC_{50}$: TUc = $100/ IC_{25}$

TOXICITY TEST RESULTS, UV-TREATED (see Appendix B for Bench Sheets)

3. Results of a *Pimephales promelas* Chronic/ 7-day Toxicity Test.
(Genus species) (Type / Duration)

Conducted April 15 -22, 2003 using effluent from UV Treated Outfall 112.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
6.25%	100	100	100	100	100	100	100
12.5%	100	100	100	100	100	100	100
25%	100	100	100	100	100	100	100
50%	100	100	100	100	100	100	100
100%	100	100	100	100	100	100	100

96-hour LC_{50} Value: > 100%

Calculated TU Estimates: < 1.0 TUa*

Permit Limit: NA

Test Solutions (% Effluent)	Mean Dry Weight (mg) (replicate number)				
	1	2	3	4	Mean
Control	0.884	0.763	0.873	0.731	0.813
6.25%	0.904	0.807	0.773	0.815	0.825
12.5%	0.769	0.855	0.825	0.764	0.803
25%	0.734	0.806	0.774	1.111	0.856
50%	0.717	0.879	0.802	0.932	0.832
100%	1.137	0.830	1.119	1.063	1.037

IC_{25} Value: > 100%

Calculated TU Estimates: < 1.0 TUc*

Permit Limit: NA

*TUa = $100/LC_{50}$: TUc = $100/ IC_{25}$

REFERENCE TOXICANT TEST RESULTS (see Appendix A and C)

Species	Date	Time	Duration	Toxicant	Results (IC_{25})
Pimephales promelas	April 15, 2003	1337	7-days	KCl	562.1 mg/L
Ceriodaphnia dubia	April 08, 2003	1526	7-days	NaCl	1025.5 mg/L

SUMMARY / CONCLUSIONS

Outfall 112 samples collected April 13 - 18, 2003, showed no toxic effects to daphnids. The resulting IC_{25} value was > 100 percent.

Bacterial growth was observed on the gills of minnows exposed to non-treated effluent from Outfall 112. This pathogenic growth was likely the cause of mortality in the effluent. The resulting IC_{25} value was 82.5 percent.

Fathead minnows were also exposed to UV treated Outfall 112 samples since fish pathogens present in upstream water have been the suspected cause of interference (anomalous dose response and high variability among replicates) in previous toxicity testing at Watts Bar Nuclear Plant. Survival was 100% in all effluent concentrations with two-minute exposure to UV light prior to introduction of test organisms. These results suggest that fish pathogen interference is a likely cause of minnow mortality in the effluent. The resulting IC_{25} value for UV-treated effluent was > 100 percent for fathead minnows.

Appendix A

ADDITIONAL TOXICITY TEST INFORMATION

SUMMARY OF METHODS

1. *Pimephales promelas*

   Tests were conducted according to EPA-821-R-02-013 (October 2002) using four replicates, each containing ten test organisms, per treatment. Test vessels consisted of 400-mL polypropylene beakers, each containing 250-mL of test solution.

2. *Ceriodaphnia dubia*

   Tests were conducted according to EPA-821-R-02-013 (October 2002) using ten replicates, each containing one test organism, per treatment. Test vessels consisted of 30-mL polypropylene cups, each containing 15-mL of test solution.

DEVIATIONS / MODIFICATIONS TO TEST PROTOCOL

1. *Pimephales promelas*

   None

2. *Ceriodaphnia dubia*

   None

DEVIATIONS / MODIFICATIONS TO PRETEST CULTURE OR HOLDING OF TEST ORGANISMS

1. *Pimephales promelas*

   None

2. *Ceriodaphnia dubia*

   None

PHYSICAL AND CHEMICAL METHODS

1. Regents, Titrants, Buffers, etc.: All chemicals were certified products used before expiration dates (where applicable).

2. Instruments: All identification, service, and calibration information pertaining to laboratory instruments is recorded in calibration and maintenance logbooks.

3. Temperature was measured using EPA Method 170.1.

4. Dissolved oxygen was measured using EPA Method 360.1.

5. The pH was measured EPA Method 150.1.

6. Conductance was measured EPA Method 120.1.

7. Alkalinity was measured using EPA Method 310.1.

8. Total Hardness was measured EPA Method 130.2.

9. Total residual chlorine was measured using EPA Method 330.5.

QUALITY ASSURANCE

Toxicity Test Methods: All phases of the study including, but not limited to, sample collection, handling and storage, glassware preparation, test organism culturing/acquisition and acclimation, test organism handling during test, and maintaining appropriate test conditions were conducted according to the protocol as described in this report and EPA-821-R-02-013. Any known deviations were noted during the study and are reported herein.

REFERENCE TOXICANT TESTS (See Appendix C for control chart information)

1. Test Type: 7-day chronic tests with results expressed as IC_{25} values in g KCl or NaCl.

2. Standard Toxicant: Potassium Chloride (KCl crystalline) for *Pimephales promelas.*
   Sodium Chloride (NaCl crystalline) for *Ceriodaphnia dubia.*

3. Dilution Water Used: Moderately hard synthetic water.

4. Statistics: ToxCalc software Version 5.0 was used for statistical analyses.

REFERENCES

1. NPDES Permit No. TN 0020168

2. USEPA. Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, EPA-821-R-02-013 (October 2002).

3. Methods for Chemical Analysis of Water and Wastes, EPA/600/4-79/020 (March 1983).

Watts Bar Nuclear Plant Biomonitoring
April 15 - 22, 2003

Appendix B

Initial and Final Chemistry for Fathead Minnow 7-day and
Daphnid 3-brood Chronic Tests

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for *Pimephales promelas* and *Ceriodaphnia dubia* Tests, Watts Bar Nuclear Plant (WBN), Outfall 112, April 15-22, 2003.

Test	Sample ID	Temperature (°C)		Dissolved Oxygen (mg/L)		pH (S.U.)		Conductance	Alkalinity	Hardness	Total Residual
		Initial	Final	Initial	Final	Initial	Final	(µmhos/cm)	(mg/L $CaCO_3$)	(mg/L $CaCO_3$)	Chlorine (mg/L)
Pimephales promelas	Control	25.5 25.2 - 25.7	25.1 24.9 - 25.4	7.9 7.7 - 8.2	7.1 6.4 - 7.8	7.93 7.72 - 8.01	7.59 7.49 - 7.73	306 288 - 319	64 64 - 64	88 88 - 88	- - -
	6.25%	25.5 25.2 - 25.7	25.1 24.9 - 25.4	8.0 7.7 - 8.2	6.8 6.3 - 7.8	7.97 7.84 - 8.03	7.55 7.44 - 7.71	308 295 - 329	- - -	- - -	- - -
	12.5%	25.4 25.2 - 25.7	25.1 24.9 - 25.4	8.1 7.8 - 8.2	6.8 6.1 - 7.9	7.97 7.83 - 8.03	7.57 7.45 - 7.73	300 289 - 310	- - -	- - -	- - -
	25%	25.4 25.2 - 25.7	25.1 24.9 - 25.4	8.1 7.8 - 8.3	6.9 6.3 - 7.6	7.96 7.83 - 8.02	7.61 7.49 - 7.79	291 282 - 300	- - -	- - -	- - -
	50%	25.4 25.0 - 25.7	25.1 24.9 - 25.4	8.1 7.8 - 8.3	7.0 6.2 - 7.6	7.94 7.83 - 8.04	7.68 7.53 - 7.81	273 267 - 284	- - -	- - -	- - -
	100%	25.4 24.9 - 25.7	25.1 24.9 - 25.4	8.2 7.8 - 8.4	7.2 6.3 - 7.7	7.91 7.81 - 8.05	7.80 7.57 - 7.93	240 221 - 254	93 89 - 98	123 110 - 130	< 0.10 < 0.10 - < 0.10
Ceriodaphnia dubia	Control	25.3 24.7 - 25.7	24.5 24.3 - 24.8	7.9 7.7 - 8.2	8.0 7.7 - 8.2	7.93 7.72 - 8.01	7.97 7.84 - 8.03	306 288 - 319	64 64 - 64	88 88 - 88	- - -
	6.25%	25.3 24.7 - 25.7	24.5 24.3 - 24.8	8.0 7.7 - 8.2	8.1 7.8 - 8.2	7.97 7.84 - 8.03	7.99 7.84 - 8.07	308 295 - 329	- - -	- - -	- - -
	12.5%	25.2 24.7 - 25.6	24.5 24.3 - 24.8	8.1 7.8 - 8.3	8.1 7.7 - 8.2	7.97 7.83 - 8.03	7.99 7.85 - 8.09	300 289 - 310	- - -	- - -	- - -
	25%	25.2 24.7 - 25.7	24.5 24.3 - 24.8	8.1 7.8 - 8.3	8.1 7.8 - 8.3	7.96 7.83 - 8.02	8.03 7.89 - 8.15	291 282 - 300	- - -	- - -	- - -
	50%	25.2 24.7 - 25.8	24.5 24.3 - 24.8	8.1 7.8 - 8.3	8.1 7.9 - 8.2	7.94 7.83 - 8.04	8.07 7.95 - 8.16	273 267 - 284	- - -	- - -	- - -
	100%	25.1 24.6 - 25.8	24.5 24.3 - 24.8	8.2 7.8 - 8.4	8.1 7.8 - 8.3	7.91 7.81 - 8.05	8.15 8.04 - 8.25	240 221 - 254	93 89 - 98	123 110 - 130	< 0.10 < 0.10 - < 0.10

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for the *Pimephales promelas* Test, Watts Bar Nuclear Plant (WBN), UV Treated Outfall 112, April 15-22 2002.

Test	Sample ID	Temperature (°C)		Dissolved Oxygen (mg/L)		pH (S.U.)		Conductance	Alkalinity	Hardness	Total Residual
		Initial	Final	Initial	Final	Initial	Final	(µmhos/cm)	(mg/L $CaCO_3$)	(mg/L $CaCO_3$)	Chlorine (mg/L)
Pimephales promelas	Control	25.4 25.2 - 25.8	24.9 24.6 - 25.1	8.0 7.7 - 8.2	6.9 6.1 - 7.6	7.93 7.80 - 8.03	7.63 7.47 - 7.76	305 299 - 310	64 64 - 64	88 88 - 88	- - -
	6.25%	25.4 25.2 - 25.8	24.9 24.6 - 25.1	8.0 7.8 - 8.3	6.9 6.1 - 7.6	7.93 7.78 - 8.03	7.59 7.38 - 7.78	306 301 - 311	- - -	- - -	- - -
	12.5%	25.4 25.2 - 25.8	24.9 24.6 - 25.1	7.9 7.6 - 8.3	6.9 6.0 - 7.7	7.92 7.77 - 8.03	7.58 7.39 - 7.72	301 296 - 308	- - -	- - -	- - -
	25%	25.5 25.3 - 25.8	24.9 24.6 - 25.1	7.9 7.6 - 8.2	6.9 6.1 - 7.6	7.91 7.76 - 8.03	7.59 7.44 - 7.71	291 285 - 300	- - -	- - -	- - -
	50%	25.4 25.3 - 25.8	24.9 24.6 - 25.1	7.8 7.5 - 8.2	6.8 6.0 - 7.6	7.92 7.78 - 8.05	7.63 7.43 - 7.84	273 261 - 281	- - -	- - -	- - -
	100%	25.4 25.2 - 25.8	24.9 24.6 - 25.1	7.8 7.6 - 8.2	7.2 6.6 - 7.6	7.93 7.79 - 8.11	7.79 7.65 - 7.94	238 216 - 250	93 89 - 98	123 110 - 130	< 0.10 < 0.10 - < 0.10

Watts Bar Nuclear Plant Biomonitoring
April 15 - 22, 2003

Appendix C

Chain of Custody Records and
Toxicity Test Bench Sheets

Sample Receipt Log

Date Received	Time Received	Received By	Received From	Sample Temperature (°C)	Project Number	Sample Number	Sample Name and Description	State	Comments
04-15-03	0936	Jlumsden	Fedex	0.5°C	665	030415.01	TVA WATTS BAR NUCLEAR PLANT	TN	
04-15-03	0942	Jlumsden	UPS	0.2°C	666	030415-02	PROGRESS ENERGY CAROLINAS CAPE FEAR OUTFALL 007	NC	
04-15-03	0942	Jlumsden	UPS	0.2°C	666	030415.03	PROGRESS ENERGY CAROLINAS CAPE FEAR- UPSTREAM	NC	
04-16-03	0808	Jlumsden	D.TRAHMEL	0.2°C	667	030416.01	TEST AMERICA- EARTH ENV. SERVICES MAGGIE VALLEY	NC	
04-16-03	1200	KEKeenan	R. Pate	3.8°C	668	030416.02	ENVIRONMENTAL INC. BRYSON CITY WWTP	NC	
04-17-03	0942	Jlumsden	Fedex	0.2°C	665	030417.01	TVA WATTS BAR NUCLEAR PLANT	TN	
04-18-03	0930	KEKeenan	UPS	0.1°C	666	030418.01	PROGRESS ENERGY CAROLINAS CAPE FEAR 007	NC	
04-18-03	0930	KEKeenan	UPS	0.1°C	666	030418.02	PROGRESS ENERGY CAROLINAS CAPE FEAR UPSTREAM	NC	
04-18-03	1345	KEKeenan	C. LADD	3.8°C	667	030418.03	TEST AMERICA- EARTH ENV. SERVICES MAGGIE VALLEY	NC	
04-19-03	0958	KEKeenan	Fedex	0.4°C	665	030419.01	TVA- WATTS BAR NUCLEAR PLANT	TN	
04-19-03	1155	KEKeenan	A. Wike	1.7°C	668	030419.02	ENVIRONMENTAL, INC. BRYSON CITY WWTP	NC	
04-22-03	1638	KEKeenan	D. Eledge	3.7°C	669	030422.01	PROGRESS ENERGY CAROLINAS ASHEVILLE ASH POND	NC	
04-23-03	0811	KEKeenan	GREYHOUND	0.1°C	670	030422.01,02	NC OUTWARD BOUND SCHOOL	NC	
04-23-03	0928	KEKeenan	UPS	0.1°C	671	030423.02	TRITEST, INC. ELLERBE	NC	
04-23-03	0928	KEKeenan	UPS	0.1°C	672	030423.03	TRITEST, INC. FRANKLIN Co. DEPT. WATER & SEWER	NC	
04-23-03	0928	KEKeenan	UPS	0.1°C	673	030423.04	TRITEST, INC. HAMLET	NC	
04-23-03	1323	KEKeenan	Fedex	1.8°C	674	030423.05	PRISM LABS. EXPRESS FOOD MART	NC	
04-26-03	1010	L. Keenan	GREYHOUND	0.1°C	670	030426.01	NC OUTWARD BOUND SCHOOL	NC	
04-26-03	1016	L. Keenan	Fedex	0.1°C	674	030426.02	PRISM LABS. EXPRESS FOOD MART	NC	
04-26-03	1020	L. Keenan	Fedex	0.1°C	671	030426.03	TRITEST, INC. ELLERBE	NC	
04-26-03	1020	L. Keenan	Fedex	0.1°C	672	030426.04	TRITEST, INC. FRANKLIN Co. DEPT. WATER & SEWER	NC	

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: WBNP BIOTOXICITY

P.O. Number: N/A

Facility Sampled: Watts Bar Nuclear Plant

NPDES Number: TN0026168

Collected By: Del Clark
Lassy Brown

Environmental Testing Solution, Inc.
351 Depot Street.
Asheville, NC
28801
Phone: 828-350-9364
Fax: 828-350-9368

Delivered By (Circle One):
FedEx UPS Bus Client
Other (specify):

General Comments: Contact WBNP personnel at (423) 365-3575 if any problems are encountered. Paper number 1-800-323-4853 then dial 90040 and number to be dialed.

* Custody seals intact. Samples received in good condition. JFurman.

Field Identification / Sample Description	Grab/ Comp.	Ship. Temp. (°C)	Collection Date/Time: Date	Collection Date/Time: Time	Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate): Yes	If Yes, Inches	No	Trace	Laboratory Use: BTS Log Number	Arrival Temp. (°C)	By	Time	Appearance
WBN-OSN-112- 1	Comp	start	4/13/03	0830	112 , 3 gal	0.569			X		030415.01	0.5°C	JF	0936	* See Comments
		end	04-14-03	0730											
TRC = <0.02															

Present 665

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
~~Del Clark~~	~~4/14/0836~~ 1015	Calvin Nichols ~~FedEx~~	4-14-03 @ 1015
Calvin Nichols	4-14-03 @ 1311		
~~FedEx~~		M. Merriman	4-14-03 @ 13 11

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 4°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

Fedex 04-15-03 0936 → JFurman 04-15-03 0936

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: WBNP BIOTOXICITY

P.O. Number: N/A

Facility Sampled: Watts Bar Nuclear Plant

NPDES Number: TN0020168

Collected By: Lanny Brown
Calvin Nichols

Environmental Testing Solution, Inc.
351 Depot Street.
Asheville, NC
28801
Phone: 828-350-9364
Fax: 828-350-9368

Delivered By (Circle One):
(FedEx) UPS Bus Client
Other (specify):_____

General Comments: Contact WBNP personnel at (423) 365-3575 if any problems are encountered. Power number, 1-800-323-4853 then dial 90040 and number to be dialed.

Field Identification / Sample Description	Grab/ Comp.	Ship. Temp. (°C)	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Laboratory Use Preserved WS				
			Date	Time			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
WBN-OSN-112.2	Comp	Start	4/15/03	09:30	112-, 3 gal	0.3895			X		680417-01	0.2°C	JH	0942	
		end	4/16/03	08:30											
TRC = 20.00 ppm															

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Calvin Nichols M Merriman 4-16-03	4/16 1215	FedEx	
FedEx		M Merriman	4-16 12-15

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 4°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

FedEx 04-17-03 0942 → M Merriman 04-17-03 0942

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: WBNP BIOTOXICITY

P.O. Number: N/A

Facility Sampled: Watts Bar Nuclear Plant

NPDES Number: TN0020168

Collected By: Martha Ervin
Pat Clark

Environmental Testing Solution, Inc.
351 Depot Street.
Asheville, NC
28801
Phone: 828-350-9364
Fax: 828-350-9368

Delivered By (Circle One):
(FedEx) UPS Bus Client
Other (specify):

General Comments: Contact WBNP personnel at (423) 365-3575 if any problems are encountered. Pager number 1-800-323-4853 then dial 90040 and number to be dialed.

→ Custody seals intact. Samples received in good condition - 4°C

Field Identification / Sample Description	Grab/ Comp.	Ship. Temp. (°C)	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Laboratory Use Present at 0.5				
			Date	Time			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appropriate
WBN-OSN-112-3	Comp	Start	4/17/03	08:28	112- 2 Gal	0.4537	X	0.96			03099.01	0.4°C	MG	0958	Y SOP
		end	4/18/03	07:28	more 4/18/03										Comments
TRC = <0.02 ppm															

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Pat Clark	4/18/03 @ 1200	FedEx	04-18-03 1200
FedEx		Katherine ETS	04-19-03 0958

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 4°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013 Method 1000.0)
Species: *Pimephales promelas*

Client: TVA
Facility: Watts Bar Nuclear Plant
NPDES #: TN-0020168
Project #: 665
NONTREATED

Dilution preparation information:						*Comments:*
Dilution prep (%)	6.25	12.5	25	50	100	
Effluent volume (mL)	125	250	500	1000	2000	
Diluent volume (mL)	1875	1750	1500	1000	0	
Total volume (mL)	2000	2000	2000	2000	2000	

Test organism information:		*Test information:*	
Organism age:	27 - 30 HOURS	Randomizing template:	PURPLE
Date and times organisms were born between:	04-14-03 1030 TO 1330 HST	Incubator number:	3 D
Organism source:	ABS BATCH 04-14-03	*Artemia* lot number:	BG18090
Transfer bowl information:	pH = 7.80 Temperature = 24.2 °C	Total drying time:	19-HOURS
		Date / Time in: 04-22-03	1500
Average transfer volume:	10.4 ml	Date / Time out: 04-23-03	0800
		Oven temperature:	100°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Control water batch used	Sample numbers used	Analyst
0	04-15-03	—	1430	1420	04-11-03	030415.01	JH
1	04-16-03	0900	1504	1432	04-11-03	030415.01	JH
2	04-17-03	0903	1510	1426	04-11-03	030417.01	JH
3	04-18-03	0900	1500	1326	04-11-03	030417.01	JH
4	04-19-03	0856	1503	1337	04-11-03	030419.01	JH
5	04-20-03	0902	1500	1350	04-11-03	030419.01	JH
6	04-21-03	0847	1500	1356	04-11-03	030419.01	JH
7	04-22-03			1333	—	—	JH

Control information:		Acceptance criteria
% Mortality:	0%	≤ 20%
Average weight per initial larvae:	0.8448	
Average weight per surviving larvae:	0.8448	≥ 0.25 mg/larvae

Summary of test endpoints:	
7-day LC_{50}	>100%
NOEC	50%
LOEC	100%
ChV	70.7%
IC_{25}	82.5%

Species: *Pimephales promelas*

Client: TVA · Watts Bar ~~2~~ NONTREATED **Date:** 04-15-03

Survival and Growth Data

Day	CONTROL				6.25 %				12.5 %			
	A	B	C	D	E	F	G	H	I	J	K	L
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	9 id ~~10~~	10	10	10	10	10	10
4	10	10	10	10	10	9	10	10	10	10	9 id	10
5	10	10	10	10	10	9	10	10	10	10	9	7 3d
6	10	10	10	10	10	9	10	10	10	10	9	7
7	10	10	10	10	10	9	10	10	10	10	9	7
A = Pan weight (mg) BLACK	14.850	14.879	14.943	15.188	15.165	14.665	14.952	15.184	14.764	14.795	14.938	15.048
B = Pan + Larvae weight (mg)	24.02	23.90	23.21	22.52	24.03	22.04	22.15	24.75	22.58	23.02	24.81	20.78
Larvae weight (mg) = A – B	9.170	9.021	8.267	7.332	8.865	7.375	7.198	9.566	7.816	8.225	9.872	5.732

Calculations and data reviewed: JH

To minimize pathogen spread:

Comments: - Test solutions were renewed from lowest concentration to highest, using separated pipettes & decanting vessels for each concentration. Pipettes were discarded daily. Test vessels with mortality in each concentration were renewed last, using different (new) pipettes for each test vessel.

Species: *Pimephales promelas*

Client: TVA - Watts Bar NONTREATED **Date:** 04-15-03

Survival and Growth Data

Day	25·1.				50·1.				100·1.			
	M	N	O	P	Q	R	S	T	U	V	W	X
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	8 2d	10	10	10
3	10	10	10	10	10	10	8 2d	10	5 2d	5 2d	8 2d	8 2d
4	8 2d	8 2d	8 2d	10	10	8 2d	7 1d	10	5	5	8	8
5	8	7 1d	8	10	10	8	7	10	5	5	7 1d	8
6	8	6 1d	8	10	10	8	7	10	5	5	6 1d	8
7	8	6	8	10	10	8	7	10	5	5	6	8
A = Pan weight (mg)	14.952	14.961	14.707	14.656	15.025	15.149	14.679	14.701	15.043	14.789	14.642	14.646
B = Pan + Larvae weight (mg)	22.62	20.28	22.32	22.33	24.62	21.13	21.23	23.39	19.98	19.69	19.86	22.67
Larvae weight (mg) = A – B	7.668	5.319	7.613	7.674	9.595	5.981	6.551	8.689	4.937	4.901	5.218	8.024

Calculations and data reviewed: JH

Comments: * Pathogenic growth observed on minnows before death. Dead minnows were heavily fungused. Mortality did not appear to be due to a toxic effect, but due to a pathogenic interference.

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1000.0)
Species: *Pimephales promelas*

Quality Control
Verification of Data Entry, Calculations, and Statistical Analyses

Client: TVA Watts Bar Nuclear Plant, Nontreated

Test dates: April 15-22, 2003

Project number: 665

Reveiwed by:

Concentration (%)	Replicate	Initial number of larvae	Final number of larvae	A = Pan weight (mg)	B = Pan + Larvae weight (mg)	Larvae weight (mg) = A - B	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight (mg)	Coefficient of variation (%)	Percent reduction from control (%)
Control	A	10	10	14.850	24.020	9.170	0.9170	100.0	0.8448	10.0	Not applicable
	B	10	10	14.879	23.900	9.021	0.9021				
	C	10	10	14.943	23.210	8.267	0.8267				
	D	10	10	15.188	22.520	7.332	0.7332				
6.25%	E	10	10	15.165	24.030	8.865	0.8865	97.5	0.8251	14.0	2.3
	F	10	9	14.665	22.040	7.375	0.7375				
	G	10	10	14.952	22.150	7.198	0.7198				
	H	10	10	15.184	24.750	9.566	0.9566				
12.5%	I	10	10	14.764	22.580	7.816	0.7816	90.0	0.7911	21.5	6.3
	J	10	10	14.795	23.020	8.225	0.8225				
	K	10	9	14.938	24.810	9.872	0.9872				
	L	10	7	15.048	20.780	5.732	0.5732				
25%	M	10	8	14.952	22.620	7.668	0.7668	80.0	0.7069	16.5	16.3
	N	10	6	14.961	20.280	5.319	0.5319				
	O	10	8	14.707	22.320	7.613	0.7613				
	P	10	10	14.656	22.330	7.674	0.7674				
50%	Q	10	10	15.025	24.620	9.595	0.9595	87.5	0.7704	22.3	8.8
	R	10	8	15.149	21.130	5.981	0.5981				
	S	10	7	14.679	21.230	6.551	0.6551				
	T	10	10	14.701	23.390	8.689	0.8689				
100%	U	10	5	15.043	19.980	4.937	0.4937	60.0	0.5770	26.2	31.7
	V	10	5	14.789	19.690	4.901	0.4901				
	W	10	6	14.642	19.860	5.218	0.5218				
	X	10	8	14.646	22.670	8.024	0.8024				

Outfall 112:

Dunnett's MSD value: 0.2269

PMSD: 26.9

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, LLC chronic toxicity tests when a toxicant reduces Pimephales growth by 17.2% from the control (determined through reference toxicant testing).

Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.

Upper PMSD bound determined by USEPA (90th percentile) = 35%.

The lower and upper bounds were calculated by the USEPA using 205 tests conducted from 19 laboratories for Pimephales growth in chronic reference toxicant tests.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Environmental Testing Solutions, LLC

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Survival

Start Date:	4/15/03	Test ID:	PpFRCR	Sample ID:	TVA, Watts Bar Nuclear Plant
End Date:	4/22/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	1.0000	1.0000	1.0000	1.0000
6.25	1.0000	0.9000	1.0000	1.0000
12.5	1.0000	1.0000	0.9000	0.7000
25	0.8000	0.6000	0.8000	1.0000
50	1.0000	0.8000	0.7000	1.0000
100	0.5000	0.5000	0.6000	0.8000

			Transform: Arcsin Square Root					Rank	1-Tailed	Number	Total
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	Sum	Critical	Resp	Number
D-Control	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4			0	40
6.25	0.9750	0.9750	1.3713	1.2490	1.4120	5.942	4	16.00	10.00	1	40
12.5	0.9000	0.9000	1.2661	0.9912	1.4120	15.696	4	14.00	10.00	4	40
25	0.8000	0.8000	1.1281	0.8861	1.4120	19.154	4	12.00	10.00	8	40
50	0.8750	0.8750	1.2306	0.9912	1.4120	17.454	4	14.00	10.00	5	40
*100	0.6000	0.6000	0.8910	0.7854	1.1071	17.027	4	10.00	10.00	16	40

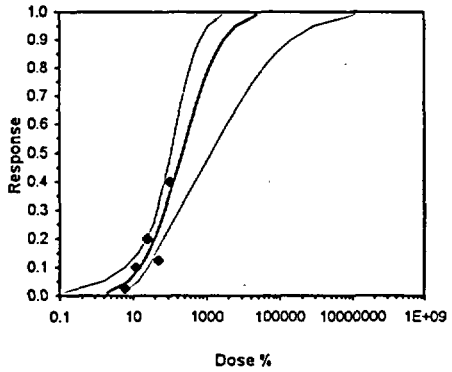
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.958816528	0.884	-0.05269485	-0.28522014
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	50	100	70.71067812	2
Treatments vs D-Control				

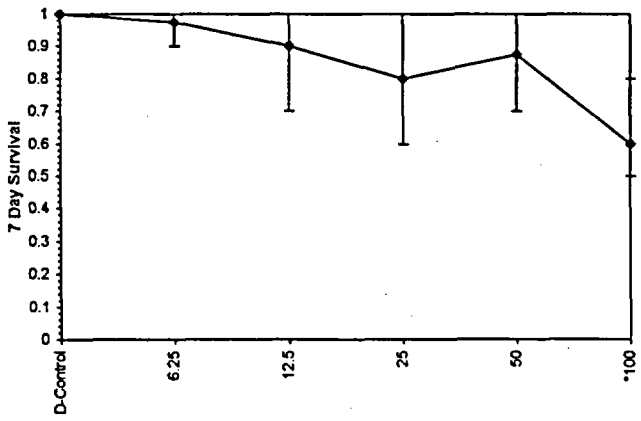
Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	1.131861458	0.281191724	0.580725667	1.682997248	0	4.593949792	7.814724922	0.2	2.337351358	0.883500355	3
Intercept	2.354442085	0.450091547	1.472262636	3.236621535							
TSCR											

Point	Probits	%	95% Fiducial Limits	
EC01	2.674	1.914353665	0.111692883	5.332413295
EC05	3.355	7.658127441	1.575977221	14.31693756
EC10	3.718	16.03614984	6.10884209	25.63937663
EC15	3.964	26.403538	14.02130033	41.28681839
EC20	4.158	39.24449263	24.31553572	67.28716706
EC25	4.326	55.13652923	35.46727229	112.4842601
EC40	4.747	129.8730719	73.74262067	511.2524416
EC50	5.000	217.4459738	108.1783041	1345.880836
EC60	5.253	364.0689745	156.3991526	3595.053085
EC75	5.674	857.5575817	284.3866848	18683.77736
EC80	5.842	1204.825203	359.5369677	36035.78381
EC85	6.036	1790.773682	471.9631622	77586.46628
EC90	6.282	2948.509267	663.7235278	203910.5135
EC95	6.645	6174.190572	1098.028801	855617.0651
EC99	7.326	24699.08416	2812.297807	12655627.49



Dose-Response Plot



Environmental Testing Solutions, LLC

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	4/15/03	Test ID:	PpFRCR	Sample ID:	TVA, Watts Bar Nuclear Plant
End Date:	4/22/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	PP-Pimephales promelas
Comments:					

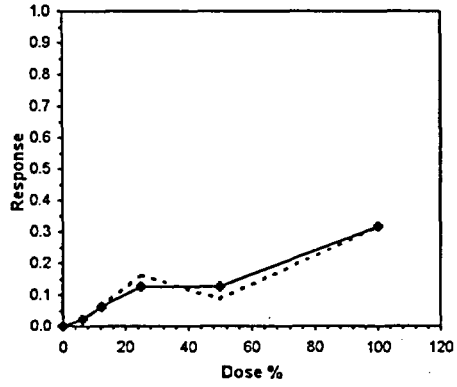
Conc-%	1	2	3	4
D-Control	0.9170	0.9021	0.8267	0.7332
6.25	0.8865	0.7375	0.7198	0.9566
12.5	0.7816	0.8225	0.9872	0.5732
25	0.7668	0.5319	0.7613	0.7674
50	0.9595	0.5981	0.6551	0.8689
100	0.4937	0.4901	0.5218	0.8024

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.8448	1.0000	0.8448	0.7332	0.9170	9.970	4				0.8448	1.0000
6.25	0.8251	0.9767	0.8251	0.7198	0.9566	13.964	4	0.204	2.360	0.2269	0.8251	0.9767
12.5	0.7911	0.9365	0.7911	0.5732	0.9872	21.527	4	0.558	2.360	0.2269	0.7911	0.9365
25	0.7069	0.8368	0.7069	0.5319	0.7674	16.505	4	1.434	2.360	0.2269	0.7386	0.8744
50	0.7704	0.9120	0.7704	0.5981	0.9595	22.287	4	0.773	2.360	0.2269	0.7386	0.8744
100	0.5770	0.6830	0.5770	0.4901	0.8024	26.158	4				0.5770	0.6830

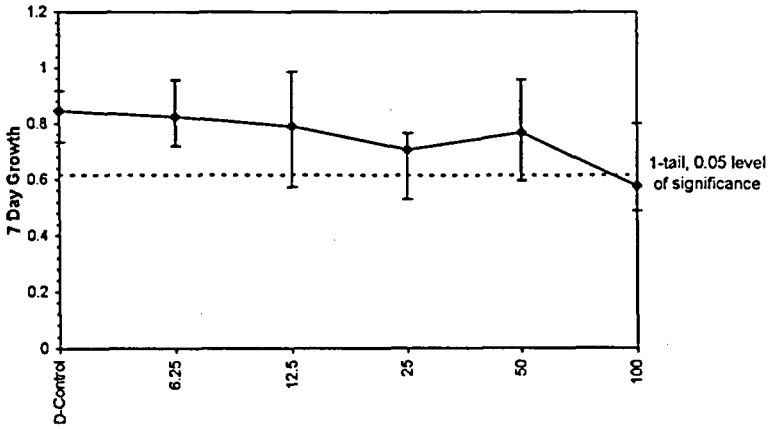
Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.94618905		0.868		-0.2197947	-0.8870032
Bartlett's Test indicates equal variances (p = 0.77)					1.79969084		13.2766981			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	50	>50		2	0.22693221	0.26863831	0.01150119	0.01849262	0.65389222	4, 15
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

Point	%	SD	95% CL(Exp)		Skew
IC05	10.405	13.784	0.000	78.970	2.5927
IC10	19.845				
IC15	56.369				
IC20	69.435				
IC25	82.502				
IC40	>100				
IC50	>100				



Dose-Response Plot



Environmental Testing Solutions, LLC

Statistical Analyses

Determination if 100% concentration is significant for growth.

Start Date:	4/15/03	Test ID:	PpFRCR	Sample ID:	TVA, Watts Bar Nuclear Plant
End Date:	4/22/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.9170	0.9021	0.8267	0.7332
6.25	0.8865	0.7375	0.7198	0.9566
12.5	0.7816	0.8225	0.9872	0.5732
25	0.7668	0.5319	0.7613	0.7674
50	0.9595	0.5981	0.6551	0.8689
100	0.4937	0.4901	0.5218	0.8024

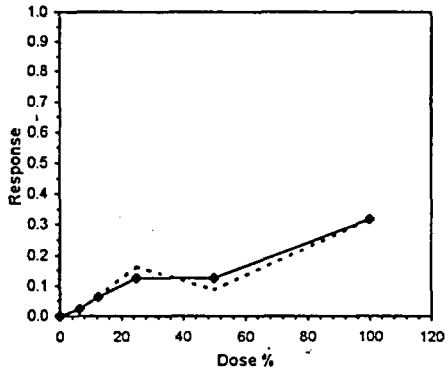
			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.8448	1.0000	0.8448	0.7332	0.9170	9.970	4				0.8448	1.0000
6.25	0.8251	0.9767	0.8251	0.7198	0.9566	13.964	4	0.201	2.410	0.2362	0.8251	0.9767
12.5	0.7911	0.9365	0.7911	0.5732	0.9872	21.527	4	0.547	2.410	0.2362	0.7911	0.9365
25	0.7069	0.8368	0.7069	0.5319	0.7674	16.505	4	1.407	2.410	0.2362	0.7386	0.8744
50	0.7704	0.9120	0.7704	0.5981	0.9595	22.287	4	0.759	2.410	0.2362	0.7386	0.8744
*100	0.5770	0.6830	0.5770	0.4901	0.8024	26.158	4	2.732	2.410	0.2362	0.5770	0.6830

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.963630021	0.884	0.087991444	-0.80591261
Bartlett's Test indicates equal variances (p = 0.87)	1.855975628	15.08631706		

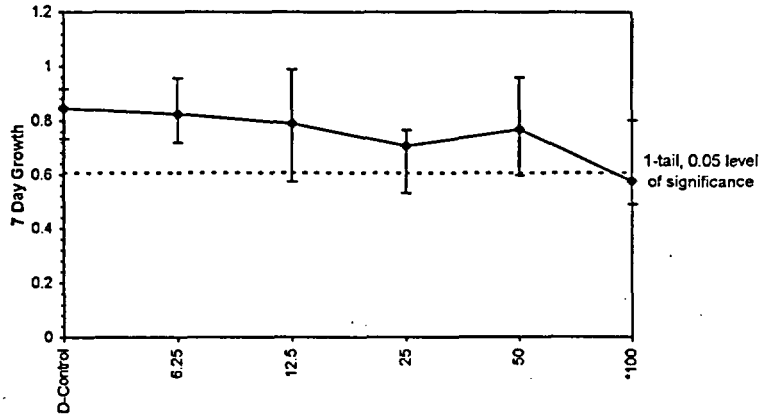
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	50	100	70.71067812	2	0.236175885	0.279580805	0.03878183	0.019207329	0.124426588	5, 18
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

Point	%	SD	95% CL(Exp)		Skew
IC05	10.405	10.467	0.000	75.168	2.7301
IC10	19.845				
IC15	56.369				
IC20	69.435				
IC25	82.502				
IC40	>100				
IC50	>100				



Dose-Response Plot



Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013 Method 1002.0)
Species: *Ceriodaphnia dubia*

Client: TVA
Facility: WATTS BAR NUCLEAR PLANT
NPDES #: TN-0020168
Project #: ~~6666~~ 665

Dilution preparation information:						*Comments:*
Dilution prep (%)	6.25	12.5	25	50	100	* Daphnia spp present in effluent. (removed prior to transferring organisms with a pipette.)
Effluent volume (mL)	125	250	500	1000	2000	
Diluent volume (mL)	1875	1750	1500	1000	0	
Total volume (mL)	2000	2000	2000	2000	2000	

Test organism information:		*Test information:*	
Organism age:	< 24-HOURS OLD	Randomizing template:	ORANGE
Date and times organisms were born between:	04-15-03 0800 TO 1150	Incubator number and shelf location:	2 D
Organism source:	04-08-03 A-D	YCT batch: ABS	03-25-03
Transfer bowl information:	pH = 8.02 Temperature = 24.8C	*Selenastrum* batch: ABS	03-25-03

Daily renewal information:

Day	Date	Test initiation, renewal, or termination time	Control water batch used	Sample numbers used	Analyst
0	04-15-03	1319	04-11-03	030415.01	JL
1	04-16-03	1322	04-11-03	030415.01	JL
2	04-17-03	1316	04-11-03	030417.01	JL
3	04-18-03	1240	04-11-03	030417.01	JL
4	04-19-03	1300	04-11-03	030419.01	JL
5	04-20-03	1248	04-11-03	030419.01	JL
6	04-21-03	1252	04-11-03	030419.01	JL
7	04-22-03	1236			JL

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	≤ 20%	7-day LC50	>100%
% Adults having 3rd Broods:	100%	≥ 80%	NOEC	100%
% Mortality:	0%	≤ 20%	LOEC	>100%
Mean Offspring/Female:	30.6	≥ 15.0 offspring/female	ChV	>100%
% CV:	9.4%	< 40.0 %	IC25	>100%

Species: *Ceriodaphnia dubia*

Client: TVA-WATTS BAR

Date: 04-15-03

CONTROL

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	6	5	6	4	6	4	6	5	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	10	10	13	11	15	11	10	11	10	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	12	16	14	13	15	17	14	16	15	13
Total young produced		26	32	32	30	34	34	28	33	30	27
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L
X for 3rd Broods		X	X	X	X	X	X	X	X	X	X

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	30.6

CONCENTRATION: 6.25%

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	5	6	7	6	4	5	5	4	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	13	12	14	12	10	11	12	0	0	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	3*	0	0	10	12	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	14	17	16	17	14	19	18	15	15	15
Total young produced		32	34	36	36	33	34	35	30	31	31
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

* SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	33.2
% Reduction from Control:	-8.5%

Species: *Ceriodaphnia dubia*

Client: TVA- WATTS BAR **Date:** 04-15-03

CONCENTRATION: 12.5%

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	6	5	7	5	5	5	7	5	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	12	10	14	14	12	11	14	13	15
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	18	15	14	17	15	18	17	19	16	14
Total young produced		33	33	29	38	34	35	33	40	34	34
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	34.3
% Reduction from Control:	-12.1%

CONCENTRATION: 25%

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	6	6	7	5	5	7	7	7	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	15	15	11	10	16	15	12	15	15	13
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	4 *
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	19	18	14	17	13	18	20	16	15	12
Total young produced		38	39	31	34	34	38	39	38	37	34
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

* SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	36.2
% Reduction from Control:	-18.8%

Species: *Ceriodaphnia dubia*

Client: TVA-WATTS BAR Date: 04-15-03

CONCENTRATION: 50% *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	7	5	7	6	6	5	5	7	6
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	13	0	15	10	12	15	15	1*	16	*3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	15	0	0	0	0	0	15	0	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	21	16	16	19	19	17	19	+9=17	18	14
Total young produced		39	38	36	36	37	38	39	38	41	35
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

* carry over

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	37.7
% Reduction from Control:	-23.2%

CONCENTRATION: 100% *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	6	6	7	7	4	5	6	6	6	7
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	14	15	16	14	12	12	14	15	15	13
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	2*	0	2*	1*	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	17	18	18	18	16	20	15	18	16	18
Total young produced		37	39	41	39	34	37	37	40	37	38
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

* SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	37.9
% Reduction from Control:	-23.9%

Environmental Testing Solutions, LLC

Verification of *Ceriodaphnia* Reproduction Totals

Control

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	6	5	6	4	6	4	6	5	4	50
5	10	10	13	11	15	11	10	11	10	0	101
6	0	0	0	0	0	0	0	0	0	10	10
7	12	16	14	13	15	17	14	16	15	13	145
Total	26	32	32	30	34	34	28	33	30	27	306

25%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	6	6	7	5	5	7	7	7	5	59
5	15	15	11	10	16	15	12	15	15	13	137
6	0	0	0	0	0	0	0	0	0	4	4
7	19	18	14	17	13	18	20	16	15	12	162
Total	38	39	31	34	34	38	39	38	37	34	362

6.25%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	5	6	7	6	4	5	5	4	5	52
5	13	12	14	12	10	11	12	0	0	11	95
6	0	0	0	0	3	0	0	10	12	0	25
7	14	17	16	17	14	19	18	15	15	15	160
Total	32	34	36	36	33	34	35	30	31	31	332

50%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	7	5	7	6	6	5	5	7	6	59
5	13	0	15	10	12	15	15	1	16	3	100
6	0	15	0	0	0	0	0	15	0	12	42
7	21	16	16	19	19	17	19	17	18	14	176
Total	39	38	36	36	37	38	39	38	41	35	377

12.5%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	6	5	7	5	5	5	7	5	5	54
5	11	12	10	14	14	12	11	14	13	15	126
6	0	0	0	0	0	0	0	0	0	0	0
7	18	15	14	17	15	18	17	19	16	14	163
Total	33	33	29	38	34	35	33	40	34	34	343

100%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	6	6	7	7	4	5	6	6	6	7	60
5	14	15	16	14	12	12	14	15	15	13	140
6	0	0	0	0	2	0	2	1	0	0	5
7	17	18	18	18	16	20	15	18	16	18	174
Total	37	39	41	39	34	37	37	40	37	38	379

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1002.0)
Species: Ceriodaphnia dubia

Quality Control
Verification of Data Entry, Calculations, and Statistical Analyses

Client: TVA Watts Bar Nuclear Plant, Nontreated

Test dates: April 15-22, 2003

Project number: 665

Reveiwed by: _____

Concentration (%)	Replicate number										Survival (%)	Average reproduction (offspring/female)	Coefficient of variation (%)	Percent reduction from control (%)
	1	2	3	4	5	6	7	8	9	10				
Control	26	32	32	30	34	34	28	33	30	27	100	30.6	9.4	Not applicable
6.25%	32	34	36	36	33	34	35	30	31	31	100	33.2	6.5	-8.5
12.5%	33	33	29	38	34	35	33	40	34	34	100	34.3	8.7	-12.1
25%	38	39	31	34	34	38	39	38	37	34	100	36.2	7.6	-18.3
50%	39	38	36	36	37	38	39	38	41	35	100	37.7	4.7	-23.2
100%	37	39	41	39	34	37	37	40	37	38	100	37.9	5.2	-23.9

Outfall 112:

Dunnett's MSD value: 2.515

PMSD: 8.2

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, LLC chronic toxicity tests when a toxicant reduces Ceriodaphnia reproduction by 9.9% from the control.

Lower PMSD bound determined by USEPA (10^{th} percentile) = 11%.

Upper PMSD bound determined by USEPA (90^{th} percentile) = 37%.

The lower and upper bounds were calculated by the USEPA using 393 tests conducted from 33 laboratories for *Ceriodaphnia* reproduction in chronic reference toxicant tests.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Environmental Testing Solutions, LLC

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	4/15/03	Test ID:	CdFRCR	Sample ID:	TVA, Watts Bar Nuclear Plant
End Date:	4/22/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	CD-Ceriodaphnia dubia
Comments:					

Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	26.000	32.000	32.000	30.000	34.000	34.000	28.000	33.000	30.000	27.000
6.25	32.000	34.000	36.000	36.000	33.000	34.000	35.000	30.000	31.000	31.000
12.5	33.000	33.000	29.000	38.000	34.000	35.000	33.000	40.000	34.000	34.000
25	38.000	39.000	31.000	34.000	34.000	38.000	39.000	38.000	37.000	34.000
50	39.000	38.000	36.000	36.000	37.000	38.000	39.000	38.000	41.000	35.000
100	37.000	39.000	41.000	39.000	34.000	37.000	37.000	40.000	37.000	38.000

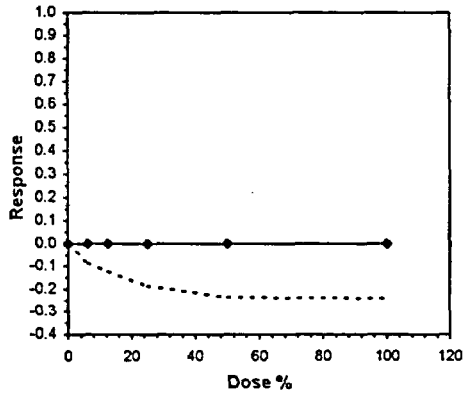
			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	30.600	1.0000	30.600	26.000	34.000	9.396	10				34.983	1.0000
6.25	33.200	1.0850	33.200	30.000	36.000	6.476	10	-2.364	2.287	2.515	34.983	1.0000
12.5	34.300	1.1209	34.300	29.000	40.000	8.698	10	-3.364	2.287	2.515	34.983	1.0000
25	36.200	1.1830	36.200	31.000	39.000	7.571	10	-5.091	2.287	2.515	34.983	1.0000
50	37.700	1.2320	37.700	35.000	41.000	4.687	10	-6.455	2.287	2.515	34.983	1.0000
100	37.900	1.2386	37.900	34.000	41.000	5.196	10	-6.636	2.287	2.515	34.983	1.0000

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.39449021	1.035	-0.1467813	-0.2224922
Bartlett's Test indicates equal variances (p = 0.55)	3.97715425	15.0863171		

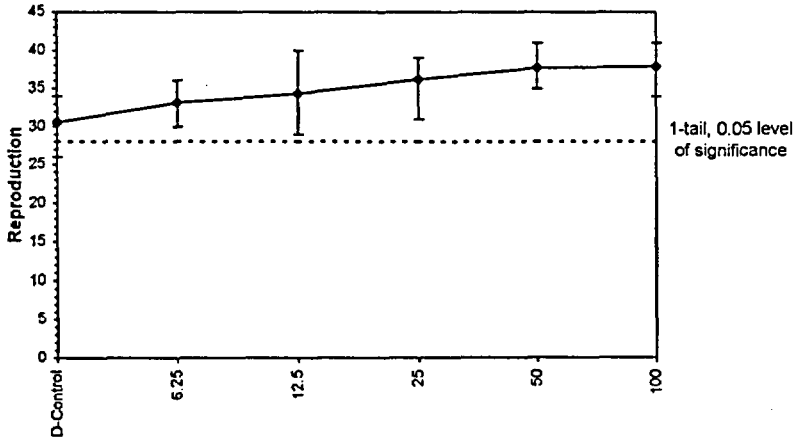
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	100	>100		1	2.51533329	0.08220043	80.4566667	6.05	1.9E-08	5, 54
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

Point	%	SD	95% CL	Skew
IC05	>100			
IC10	>100			
IC15	>100			
IC20	>100			
IC25	>100			
IC40	>100			
IC50	>100			



Dose-Response Plot



Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013 Method 1000.0)
Species: *Pimephales promelas*

Client: TVA
Facility: Watts Bar Nuclear Plant
NPDES #: TN-0020168
Project #: 665
UV-TREATED

Dilution preparation information:						*Comments:*
Dilution prep (%)	6.25	12.5	25	50	100	Each concentration was
Effluent volume (mL)	125	250	500	1000	2000	UV-treated for 2 minutes
Diluent volume (mL)	1875	1750	1500	1000	0	using a 40-watt UV-sterilizer
Total volume (mL)	2000	2000	2000	2000	2000	

Test organism information:		*Test information:*	
Organism age:	26.5 - 29.5 - HOURS	Randomizing template:	Yellow
Date and times organisms were born between:	04-14-03 1030 TO 1330 MST	Incubator number:	3B
Organism source:	ABS BATCH 04-14-03	*Artemia* lot number:	BG18090
Transfer bowl information:	pH = 7.80 Temperature = 24.2 °C	Total drying time:	≥ 19-HOURS
		Date / Time in: 04-22-03	1500
Average transfer volume:	10.4 ml	Date / Time out: 04-23-03	0800
		Oven temperature:	100°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Control water batch used	Sample numbers used	Analyst
0	04-15-03	——	1430	1403	04-11-03	030415.01	JH
1	04-16-03	0900	1504	1411	04-11-03	030415.01	JH
2	04-17-03	0903	1510	1400	04-11-03	030417.01	JH
3	04-18-03	0900	1500	1314	04-11-03	030417.01	JH
4	04-19-03	0856	1503	1320	04-11-03	030419.01	JH
5	04-20-03	0902	1500	1333	04-11-03	030419.01	JH
6	04-21-03	0847	1500	1342	04-11-03	030419.01	JH
7	0422-03			1314	——	——	JH

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	0%	≤ 20%	7-day LC_{50}	> 100%
Average weight per initial larvae:	0.8127		NOEC	100%
Average weight per surviving larvae:	0.8127	≥ 0.25 mg/larvae	LOEC	> 100%
			ChV	> 100%
			IC_{25}	> 100%

Species: *Pimephales promelas*

Client: TVA - Watts Bar

UV- TREATED

Date: 04-15-03

Survival and Growth Data

Day	CONTROL				6.25%.				12.5%.			
	A	B	C	D	E	F	G	H	I	J	K	L
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	10	10	10	10	10	10	10	10
7	10	10	10	10	10	10	10	10	10	10	10	10
A = Pan weight (mg) Blue	14.979	14.756	15.065	14.912	14.822	14.978	15.022	14.669	15.037	15.193	14.691	15.121
B = Pan + Larvae weight (mg)	23.82	22.39	23.79	22.22	23.86	23.05	22.75	22.82	22.73	23.74	22.94	22.76
Larvae weight (mg) = A – B	8.841	7.634	8.725	7.308	9.038	8.072	7.728	8.151	7.693	8.547	8.249	7.639

Calculations and data reviewed: dl

Comments:

Species: *Pimephales promelas*

Client: TVA - Watts Bar UV-TREATED **Date:** 04-15-13

Survival and Growth Data

Day	25%.				50%.				100%.			
	M	N	O	P	Q	R	S	T	U	V	W	X
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	10	10	10	10	10	10	10	10
7	10	10	10	10	10	10	10	10	10	10	10	10
A = Pan weight (mg)	14.704	15.044	15.099	14.806	15.010	14.846	15.033	14.954	14.644	15.181	15.014	15.130
B = Pan + Larvae weight (mg)	22.04	23.10	22.84	25.92	22.18	23.64	23.05	24.27	26.01	23.48	26.20	25.76
Larvae weight (mg) = A – B	7.336	8.056	7.741	11.114	7.170	8.794	8.017	9.316	11.366	8.299	11.186	10.630

Calculations and data reviewed: JH

Comments: * Minnows appear healthy in all effluent concentrations.

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1000.0)
Species: *Pimephales promelas*

Quality Control
Verification of Data Entry, Calculations, and Statistical Analyses

Client: TVA Watts Bar Nuclear Plant, UV Treated

Test dates: April 15-22, 2003

Project number: 665

Reveiwed by: Jumsen

Concentration (%)	Replicate	Initial number of larvae	Final number of larvae	A = Pan weight (mg)	B = Pan + Larvae weight (mg)	Larvae weight (mg) = A - B	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight (mg)	Coefficient of variation (%)	Percent reduction from control (%)
Control	A	10	10	14.979	23.820	8.841	0.8841	100.0	0.8127	9.5	Not applicable
	B	10	10	14.756	22.390	7.634	0.7634				
	C	10	10	15.065	23.790	8.725	0.8725				
	D	10	10	14.912	22.220	7.308	0.7308				
6.25%	E	10	10	14.822	23.860	9.038	0.9038	100.0	0.8247	6.8	-1.5
	F	10	10	14.978	23.050	8.072	0.8072				
	G	10	10	15.022	22.750	7.728	0.7728				
	H	10	10	14.669	22.820	8.151	0.8151				
12.5%	I	10	10	15.037	22.730	7.693	0.7693	100.0	0.8032	5.5	1.2
	J	10	10	15.193	23.740	8.547	0.8547				
	K	10	10	14.691	22.940	8.249	0.8249				
	L	10	10	15.121	22.760	7.639	0.7639				
25%	M	10	10	14.704	22.040	7.336	0.7336	100.0	0.8562	20.2	-5.3
	N	10	10	15.044	23.100	8.056	0.8056				
	O	10	10	15.099	22.840	7.741	0.7741				
	P	10	10	14.806	25.920	11.114	1.1114				
50%	Q	10	10	15.010	22.180	7.170	0.7170	100.0	0.8324	11.2	-2.4
	R	10	10	14.846	23.640	8.794	0.8794				
	S	10	10	15.033	23.050	8.017	0.8017				
	T	10	10	14.954	24.270	9.316	0.9316				
100%	U	10	10	14.644	26.010	11.366	1.1366	100.0	1.0370	13.7	-27.6
	V	10	10	15.181	23.480	8.299	0.8299				
	W	10	10	15.014	26.200	11.186	1.1186				
	X	10	10	15.130	25.760	10.630	1.0630				

Outfall 112:

Dunnett's MSD value: 0.1836

PMSD: 22.6

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, LLC chronic toxicity tests when a toxicant reduces Pimephales growth by 17.2% from the control (determined through

Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.

Upper PMSD bound determined by USEPA (90th percentile) = 35%.

The lower and upper bounds were calculated by the USEPA using 205 tests conducted from 19 laboratories for Pimephales growth in chronic reference toxicant tests.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Environmental Testing Solutions, LLC

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	4/15/03	Test ID:	PpFRCR	Sample ID:	TVA, Watts Bar Nuclear Plant
End Date:	4/22/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	PP-Pimephales promelas
Comments:	UV-Treated				

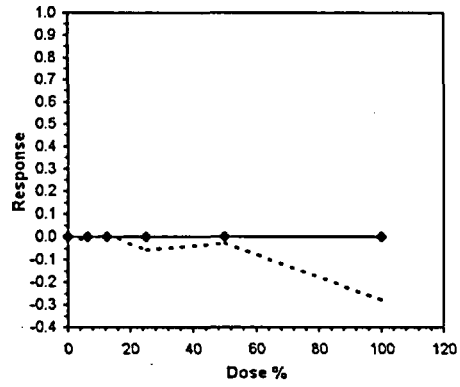
Conc-%	1	2	3	4
D-Control	0.8841	0.7634	0.8725	0.7308
6.25	0.9038	0.8072	0.7728	0.8151
12.5	0.7693	0.8547	0.8249	0.7639
25	0.7336	0.8056	0.7741	1.1114
50	0.7170	0.8794	0.8017	0.9316
100	1.1366	0.8299	1.1186	1.0630

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.8127	1.0000	0.8127	0.7308	0.8841	9.481	4				0.8610	1.0000
6.25	0.8247	1.0148	0.8247	0.7728	0.9038	6.769	4	-0.158	2.410	0.1836	0.8610	1.0000
12.5	0.8032	0.9883	0.8032	0.7639	0.8547	5.482	4	0.125	2.410	0.1836	0.8610	1.0000
25	0.8562	1.0535	0.8562	0.7336	1.1114	20.169	4	-0.571	2.410	0.1836	0.8610	1.0000
50	0.8324	1.0243	0.8324	0.7170	0.9316	11.250	4	-0.259	2.410	0.1836	0.8610	1.0000
100	1.0370	1.2760	1.0370	0.8299	1.1366	13.654	4	-2.945	2.410	0.1836	0.8610	1.0000

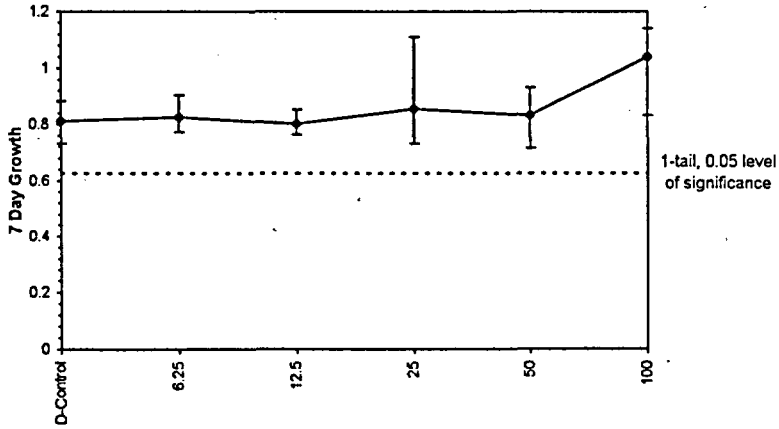
Auxiliary Tests					Statistic		Critical			Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.96759802		0.884			0.3605541	1.30981462
Bartlett's Test indicates equal variances (p = 0.24)					6.80922508		15.0863171				
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**	
Dunnett's Test	100	>100		1	0.18358002	0.22588904	0.03105137	0.01160504	0.05605605	5, 18	
Treatments vs D-Control											

Linear Interpolation (200 Resamples)

Point	%	SD	95% CL(Exp)	Skew
IC05	>100			
IC10	>100			
IC15	>100			
IC20	>100			
IC25	>100			
IC40	>100			
IC50	>100			



Dose-Response Plot



Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1000.0)
Species: *Pimephales promelas*

UV-Treated Daily Chemical Analyses

Client: Watts Bar Nuclear Plant
Test dates: April 15 - 22, 2003
Project number: 665
Reviewed by: CR

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.99	7.71	7.93	7.58	7.92	7.76	8.03	7.52	7.88	7.59	7.93	7.75	7.80	7.47
	DO (mg/L)	7.7	7.6	8.2	6.1	8.1	7.2	8.2	6.4	7.8	7.2	7.9	7.6	7.8	6.5
	Conductivity (µmhos/cm)	308		309		307		300		299		301		310	
	Alkalinity (mg/L $CaCO_3$)	64													
	Hardness (mg/L $CaCO_3$)	88													
	Temperature (°C)	25.2	25.1	25.3	25.0	25.4	24.9	25.2	25.1	25.5	24.8	25.5	24.6	25.8	24.7
6.25%	pH (SU)	7.97	7.68	7.94	7.53	7.91	7.78	8.03	7.52	7.95	7.54	7.91	7.72	7.78	7.38
	DO (mg/L)	7.8	7.5	8.2	6.6	8.2	7.5	8.3	6.5	7.8	6.8	8.1	7.6	7.9	6.1
	Conductivity (µmhos/cm)	308		306		311		303		301		305		311	
	Temperature (°C)	25.2	25.1	25.3	25.0	25.3	24.9	25.2	25.1	25.4	24.8	25.5	24.6	25.8	24.7
12.5%	pH (SU)	7.97	7.66	7.94	7.54	7.91	7.72	8.03	7.50	7.95	7.54	7.90	7.68	7.77	7.39
	DO (mg/L)	7.8	7.4	8.2	6.7	7.8	7.2	8.3	6.5	7.8	6.9	7.6	7.7	7.9	6.0
	Conductivity (µmhos/cm)	300		297		306		296		299		302		308	
	Temperature (°C)	25.2	25.1	25.3	25.0	25.3	24.9	25.3	25.1	25.4	24.8	25.6	24.6	25.8	24.7
25%	pH (SU)	7.97	7.67	7.93	7.51	7.90	7.71	8.03	7.49	7.92	7.59	7.89	7.71	7.76	7.44
	DO (mg/L)	7.8	7.5	8.2	6.5	7.8	7.2	8.2	6.1	7.6	7.0	7.7	7.6	7.8	6.1
	Conductivity (µmhos/cm)	287		285		300		289		289		292		298	
	Temperature (°C)	25.3	25.1	25.3	25.0	25.3	24.9	25.5	25.1	25.4	24.8	25.6	24.6	25.8	24.7
50%	pH (SU)	7.98	7.66	7.93	7.51	7.91	7.84	8.05	7.62	7.91	7.59	7.89	7.78	7.78	7.43
	DO (mg/L)	7.8	7.3	8.2	6.3	7.7	7.4	8.2	6.5	7.5	6.7	7.6	7.6	7.6	6.0
	Conductivity (µmhos/cm)	264		261		281		275		272		276		281	
	Temperature (°C)	25.3	25.1	25.3	25.1	25.3	24.9	25.5	25.1	25.3	24.8	25.6	24.6	25.8	24.7
100%	pH (SU)	7.99	7.77	7.95	7.66	7.90	7.94	8.11	7.75	7.91	7.88	7.88	7.85	7.79	7.65
	DO (mg/L)	7.7	7.6	8.2	6.6	7.7	7.5	8.2	6.6	7.7	7.6	7.6	7.5	7.6	6.7
	Conductivity (µmhos/cm)	216		218		248		249		241		246		250	
	Alkalinity (mg/L $CaCO_3$)	89				93				98					
	Hardness (mg/L $CaCO_3$)	110				130				130					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	25.3	25.1	25.3	25.0	25.3	24.9	25.6	25.1	25.2	24.8	25.6	24.6	25.8	24.7

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1000.0)
Species: *Pimephales promelas*

Nontreated Daily Chemical Analyses

Client: Watts Bar Nuclear Plant
Test dates: April 15 - 22, 2003
Project number: 665
Reviewed by: _____

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.89	7.72	7.93	7.60	7.98	7.51	7.96	7.49	8.00	7.56	8.01	7.73	7.72	7.50
	DO (mg/L)	7.7	7.7	8.2	6.9	7.8	6.4	8.1	6.4	7.8	7.4	7.7	7.8	7.9	6.8
	Conductivity (µmhos/cm)	305		319		306		314		288		294		313	
	Alkalinity (mg/L $CaCO_3$)	64													
	Hardness (mg/L $CaCO_3$)	88													
	Temperature (°C)	25.3	24.9	25.3	25.1	25.5	25.1	25.2	25.0	25.6	25.2	25.7	25.4	25.7	25.0
6.25%	pH (SU)	7.96	7.69	7.96	7.53	7.97	7.47	8.03	7.47	8.01	7.57	8.01	7.71	7.84	7.44
	DO (mg/L)	7.7	7.4	8.2	6.5	7.9	6.4	8.2	6.4	7.9	7.1	7.9	7.8	8.0	6.3
	Conductivity (µmhos/cm)	300		309		319		298		295		307		329	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.5	25.1	25.4	25.0	25.5	25.2	25.7	25.4	25.7	25.0
12.5%	pH (SU)	7.97	7.68	7.96	7.52	7.97	7.51	8.03	7.52	8.00	7.58	8.00	7.73	7.83	7.45
	DO (mg/L)	7.8	7.4	8.3	6.3	8.1	6.1	8.3	6.3	8.0	7.2	7.9	7.9	8.1	6.6
	Conductivity (µmhos/cm)	301		305		307		296		289		292		310	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.4	25.1	25.4	25.0	25.3	25.2	25.7	25.4	25.7	25.0
25%	pH (SU)	7.98	7.68	7.96	7.49	7.95	7.61	8.02	7.51	7.99	7.62	7.97	7.79	7.83	7.54
	DO (mg/L)	7.8	7.4	8.3	6.3	8.1	6.7	8.3	6.4	8.0	7.2	7.9	7.6	8.1	6.7
	Conductivity (µmhos/cm)	290		294		298		289		282		285		300	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.2	25.1	25.5	25.0	25.2	25.2	25.6	25.4	25.7	25.0
50%	pH (SU)	7.98	7.67	7.97	7.54	7.92	7.72	8.04	7.53	7.92	7.75	7.93	7.81	7.83	7.74
	DO (mg/L)	7.8	7.2	8.3	6.2	8.1	7.0	8.2	6.3	8.0	7.4	7.9	7.6	8.1	7.4
	Conductivity (µmhos/cm)	267		268		280		275		267		269		284	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.2	25.1	25.5	25.0	25.0	25.2	25.7	25.4	25.7	25.0
100%	pH (SU)	7.98	7.79	7.99	7.57	7.85	7.84	8.05	7.72	7.81	7.84	7.83	7.92	7.84	7.93
	DO (mg/L)	7.8	7.5	8.3	6.3	8.3	7.1	8.4	6.5	8.4	7.4	7.9	7.6	8.0	7.7
	Conductivity (µmhos/cm)	221		225		247		249		239		242		254	
	Alkalinity (mg/L $CaCO_3$)	89				93				98					
	Hardness (mg/L $CaCO_3$)	110				130				130					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	25.3	24.9	25.2	25.1	25.3	25.1	25.6	25.0	24.9	25.2	25.5	25.4	25.7	25.0

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1002.0)
Species: *Ceriodaphnia dubia*

Nontreated Daily Chemical Analyses

Client: Watts Bar Nuclear Plant
Test dates: April 15 - 22, 2003
Project number: 665
Reviewed by: CW

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.89	7.95	7.93	7.99	7.98	8.02	7.96	8.03	8.00	7.99	8.01	7.84	7.72	7.98
	DO (mg/L)	7.7	8.2	8.2	8.1	7.8	8.2	8.1	8.1	7.8	8.1	7.7	7.7	7.9	7.9
	Conductivity (µmhos/cm)	305		319		306		314		288		294		313	
	Alkalinity (mg/L $CaCO_3$)	64													
	Hardness (mg/L $CaCO_3$)	88													
	Temperature (°C)	24.7	24.8	25.3	24.5	25.2	24.4	25.1	24.4	25.7	24.4	25.4	24.3	25.5	24.6
6.25%	pH (SU)	7.96	7.95	7.96	8.02	7.97	8.01	8.03	8.07	8.01	8.00	8.01	7.84	7.84	8.01
	DO (mg/L)	7.7	8.2	8.2	8.2	7.9	8.2	8.2	8.2	7.9	8.2	7.9	7.8	8.0	7.9
	Conductivity (µmhos/cm)	300		309		319		298		295		307		329	
	Temperature (°C)	24.7	24.8	25.3	24.5	25.1	24.4	25.1	24.4	25.7	24.4	25.4	24.3	25.6	24.6
12.5%	pH (SU)	7.97	7.94	7.96	8.03	7.97	8.01	8.03	8.09	8.00	8.03	8.00	7.85	7.83	8.01
	DO (mg/L)	7.8	8.2	8.3	8.2	8.1	8.2	8.3	8.2	8.0	8.2	7.9	7.7	8.1	8.0
	Conductivity (µmhos/cm)	301		305		307		296		289		292		310	
	Temperature (°C)	24.7	24.8	25.3	24.5	25.0	24.4	25.2	24.4	25.5	24.4	25.4	24.3	25.6	24.6
25%	pH (SU)	7.98	7.95	7.96	8.09	7.95	8.02	8.02	8.15	7.99	8.08	7.97	7.89	7.83	8.06
	DO (mg/L)	7.8	8.2	8.3	8.2	8.1	8.1	8.3	8.3	8.0	8.2	7.9	7.8	8.1	8.0
	Conductivity (µmhos/cm)	290		294		298		289		282		285		300	
	Temperature (°C)	24.7	24.8	25.3	24.5	24.9	24.4	25.3	24.4	25.2	24.4	25.4	24.3	25.7	24.6
50%	pH (SU)	7.98	7.99	7.97	8.10	7.92	8.05	8.04	8.16	7.92	8.14	7.93	7.95	7.83	8.11
	DO (mg/L)	7.8	8.2	8.3	8.2	8.1	8.1	8.2	8.2	8.0	8.2	7.9	7.9	8.1	8.0
	Conductivity (µmhos/cm)	267		268		280		275		267		269		284	
	Temperature (°C)	24.7	24.8	25.3	24.5	24.7	24.4	25.3	24.4	25.1	24.4	25.4	24.3	25.8	24.6
100%	pH (SU)	7.98	8.05	7.99	8.16	7.85	8.17	8.05	8.25	7.81	8.20	7.83	8.04	7.82	8.20
	DO (mg/L)	7.8	8.2	8.3	8.2	8.3	8.1	8.4	8.2	8.4	8.3	7.9	7.8	8.0	8.0
	Conductivity (µmhos/cm)	221		225		247		249		239		242		254	
	Alkalinity (mg/L $CaCO_3$)	89				93				98					
	Hardness (mg/L $CaCO_3$)	110				130				130					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	24.7	24.8	25.3	24.5	24.6	24.4	25.4	24.4	24.8	24.4	25.4	24.3	25.8	24.6

Species: *Pimephales promelas*
Client: TVA - Watts Bar NONTREATED **Date:** 04-15-03

Daily Chemistry:

Concentration	Parameter	Day 0		Day 1		Day 2	
CONTROL	pH (S.U.)	7.89	7.72	7.93	7.60	7.98	7.51
	DO (mg/L)	7.7	7.7	8.2	6.9	7.8	6.4
	Conductivity (µmhos/cm)	305		319		306	
	Alkalinity (mg $CaCO_3$/L)	64					
	Hardness (mg $CaCO_3$/L)	88		NR		NR	
	Temperature (°C)	25.3	24.9	25.3	25.1	25.5	25.1
6.25%	pH (S.U.)	7.96	7.69	7.96	7.53	7.97	7.47
	DO (mg/L)	7.7	7.4	8.2	6.5	7.9	6.4
	Conductivity (µmhos/cm)	300		309		319	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.5	25.1
12.5%	pH (S.U.)	7.97	7.68	7.96	7.52	7.97	7.51
	DO (mg/L)	7.8	7.4	8.3	6.3	8.1	6.1
	Conductivity (µmhos/cm)	301		305		307	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.4	25.1
25%	pH (S.U.)	7.98	7.68	7.96	7.49	7.95	7.61
	DO (mg/L)	7.8	7.4	8.3	6.3	8.1	6.7
	Conductivity (µmhos/cm)	290		294		298	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.2	25.1
50%	pH (S.U.)	7.98	7.67	7.97	7.54	7.92	7.72
	DO (mg/L)	7.8	7.2	8.3	6.2	8.1	7.0
	Conductivity (µmhos/cm)	267		268		280	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.2	25.1
100%	pH (S.U.)	7.98	7.79	7.99	7.57	7.86	7.84
	DO (mg/L)	7.8	7.5	8.3	6.3	8.3	7.1
	Conductivity (µmhos/cm)	221		225		247	
	Alkalinity (mg $CaCO_3$/L)	89				93	
	Hardness (mg $CaCO_3$/L)	110				130	
	TR chlorine (mg/L)	<0.10				<0.10	
	Temperature (°C)	25.3	24.9	25.2	25.1	25.3	25.1
	pH (S.U.)						
	DO (mg/L)						
	Conductivity (µmhos/cm)						
	Alkalinity (mg $CaCO_3$/L)						
	Hardness (mg $CaCO_3$/L)						
	TR chlorine (mg/L)						
	Temperature (°C)						
		Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas*
Client: TVA - Watts Bar NONTREATED
Date: 04-15-03

Concentration	Parameter	Day 3		4		5		6	
CONTROL	pH (S.U.)	7.96	7.49	8.00	7.56	8.01	7.73	7.72	7.50
	DO (mg/L)	8.1	6.4	7.8	7.4	7.7	7.8	7.9	6.8
	Conductivity (µmhos/cm)	314		288		294		313	
	Alkalinity (mg $CaCO_3$/L)								
	Hardness (mg $CaCO_3$/L)								
	Temperature (°C)	25.2	25.0	25.6	25.2	25.7	25.4	25.7	25.0
6.25%	pH (S.U.)	8.03	7.47	8.01	7.57	8.01	7.71	7.84	7.44
	DO (mg/L)	8.2	6.4	7.9	7.1	7.9	7.8	8.0	6.3
	Conductivity (µmhos/cm)	298		295		307		329	
	Temperature (°C)	25.4	25.0	25.5	25.2	25.7	25.4	25.7	25.0
12.5%	pH (S.U.)	8.03	7.52	8.00	7.58	8.00	7.73	7.83	7.45
	DO (mg/L)	8.3	6.3	8.0	7.2	7.9	7.9	8.1	6.6
	Conductivity (µmhos/cm)	296		289		292		310	
	Temperature (°C)	25.4	25.0	25.3	25.2	25.7	25.4	25.7	25.0
25%	pH (S.U.)	8.02	7.51	7.99	7.62	7.97	7.79	7.83	7.54
	DO (mg/L)	8.3	6.4	8.0	7.2	7.9	7.6	8.1	6.7
	Conductivity (µmhos/cm)	289		282		285		300	
	Temperature (°C)	25.5	25.0	25.2	25.2	26.6	25.4	25.7	25.0
50%	pH (S.U.)	8.04	7.53	7.92	7.75	7.93	7.81	7.83	7.74
	DO (mg/L)	8.2	6.3	8.0	7.4	7.9	7.6	8.1	7.4
	Conductivity (µmhos/cm)	275		267		269		284	
	Temperature (°C)	25.5	25.0	25.0	25.2	25.7	25.4	25.7	25.0
100%	pH (S.U.)	8.05	7.72	7.81	7.84	7.83	7.92	7.84	7.93
	DO (mg/L)	8.4	6.5	8.4	7.4	7.9	7.6	8.0	7.7
	Conductivity (µmhos/cm)	249		239		242		254	
	Alkalinity (mg $CaCO_3$/L)			98					
	Hardness (mg $CaCO_3$/L)			130					
	TR Chlorine (mg/L)			<0.10					
	Temperature (°C)	25.6	25.0	24.9	25.2	25.5	25.4	25.7	25.0
	pH (S.U.)								
	~~DO (mg/L)~~								
	Conductivity (µmhos/cm)								
	Alkalinity (mg $CaCO_3$/L)								
	Hardness (mg $CaCO_3$/L)								
	TR chlorine (mg/L)								
	Temperature (°C)								
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

Species: *Ceriodaphnia dubia*
Client: TVA WATTS BAR Date: 04-15-03

Daily Chemistry:

Concentration	Parameter	Day 0		Day 1		Day 2	
CONTROL	pH (S.U.)	7.89	7.95	7.93	7.99	7.98	8.02
	DO (mg/L)	7.7	8.2	8.2	8.1	7.8	8.2
	Conductivity (µmhos/cm)	305		319		306	
	Alkalinity (mg $CaCO_3$/L)	64					
	Hardness (mg $CaCO_3$/L)	88					
	Temperature (°C)	24.7	24.8	25.3	24.5	25.2	24.4
6.25%	pH (S.U.)	7.96	7.95	7.96	8.02	7.97	8.01
	DO (mg/L)	7.7	8.2	8.2	8.2	7.9	8.2
	Conductivity (µmhos/cm)	306		309		319	
	Temperature (°C)	24.7	24.8	25.3	24.5	25.1	24.4
12.5%	pH (S.U.)	7.97	7.94	7.96	8.03	7.97	8.01
	DO (mg/L)	7.8	8.2	8.3	8.2	8.1	8.2
	Conductivity (µmhos/cm)	301		306		307	
	Temperature (°C)	24.7	24.8	25.3	24.5	25.0	24.4
25%	pH (S.U.)	7.98	7.95	7.96	8.09	7.95	8.02
	DO (mg/L)	7.8	8.2	8.3	8.2	8.1	8.1
	Conductivity (µmhos/cm)	290		294		298	
	Temperature (°C)	24.7	24.8	25.3	24.5	24.9	24.4
50%	pH (S.U.)	7.98	7.99	7.97	8.10	7.92	8.05
	DO (mg/L)	7.8	8.2	8.3	8.2	8.1	8.1
	Conductivity (µmhos/cm)	267		268		280	
	Temperature (°C)	24.7	24.8	25.3	24.5	24.7	24.4
100%	pH (S.U.)	7.98	8.05	7.99	8.16	7.86	8.17
	DO (mg/L)	7.8	8.2	8.3	8.2	8.3	8.1
	Conductivity (µmhos/cm)	221		225		247	
	Alkalinity (mg $CaCO_3$/L)	89				93	
	Hardness (mg $CaCO_3$/L)	110				130	
	TR chlorine (mg/L)	<0.10		K		<0.10	
	Temperature (°C)	24.7	24.8	25.3	24.5	24.6	24.4
	pH (S.U.)						
	DO (mg/L)						
	Conductivity (µmhos/cm)						
	Alkalinity (mg $CaCO_3$/L)						
	Hardness (mg $CaCO_3$/L)						
	TR chlorine (mg/L)					H	
	Temperature (°C)						
		Initial	Final	Initial	Final	Initial	Final

Species: *Ceriodaphnia dubia*
Client: TVA - WATTS BAR Date: 04-15-03

Concentration	Parameter	Day 3		4		5		6	
CONTROL	pH (S.U.)	7.96	8.03	8.00	7.99	8.01	7.84	7.72	7.98
	DO (mg/L)	8.1	8.1	7.8	8.1	7.7	7.7	7.9	7.9
	Conductivity (µmhos/cm)	314		288		294		313	
	Alkalinity (mg $CaCO_3$/L)	df							
	Hardness (mg $CaCO_3$/L)			df		df		df	
	Temperature (°C)	25.1	24.4	25.7	24.4	26.4	24.3	25.5	24.6
6.25%	pH (S.U.)	8.03	8.07	8.01	8.00	8.01	7.84	7.84	8.01
	DO (mg/L)	8.2	8.2	7.9	8.2	7.9	7.8	8.0	7.9
	Conductivity (µmhos/cm)	298		295		307		329	
	Temperature (°C)	25.1	24.4	25.7	24.4	25.4	24.3	25.6	24.6
12.5%	pH (S.U.)	8.03	8.09	8.00	8.03	8.00	7.85	7.83	8.01
	DO (mg/L)	8.3	8.2	8.0	8.2	7.9	7.7	8.1	8.0
	Conductivity (µmhos/cm)	296		289		292		310	
	Temperature (°C)	25.2	24.4	25.5	24.4	25.4	24.3	25.6	24.6
25%	pH (S.U.)	8.02	8.15	7.99	8.08	7.97	7.89	7.83	8.06
	DO (mg/L)	8.3	8.3	8.0	8.2	7.9	7.8	8.1	8.0
	Conductivity (µmhos/cm)	289		282		285		300	
	Temperature (°C)	25.3	24.4	25.2	24.4	25.4	24.3	25.7	24.6
50%	pH (S.U.)	8.04	8.16	7.92	8.14	7.93	7.95	7.83	8.11
	DO (mg/L)	8.2	8.2	8.0	8.2	7.9	7.9	8.1	8.0
	Conductivity (µmhos/cm)	275		267		269		284	
	Temperature (°C)	25.3	24.4	25.1	24.4	25.4	24.3	25.8	24.6
100%	pH (S.U.)	8.05	8.25	7.81	8.20	7.83	8.04	7.82	8.20
	DO (mg/L)	8.4	8.2	8.4	8.3	7.9	7.8	8.0	8.0
	Conductivity (µmhos/cm)	249		239		242		254	
	Alkalinity (mg $CaCO_3$/L)			98					
	Hardness (mg $CaCO_3$/L)			130					
	TR Chlorine (mg/L)			<0.10					
	Temperature (°C)	25.4	24.4	24.8	24.4	25.4	24.3	25.0	24.6
	pH (S.U.)								
	~~DO (mg/L)~~								
	Conductivity (µmhos/cm)								
	Alkalinity (mg $CaCO_3$/L)								
	Hardness (mg $CaCO_3$/L)					df			
	TR chlorine (mg/L)								
	Temperature (°C)								
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas*
Client: TVA - Watts Bar UV-TREATED **Date:** 04.15.03

Daily Chemistry:

Concentration	Parameter	Day 0		Day 1		Day 2	
CONTROL	pH (S.U.)	7.99	7.71	7.93	7.58	7.92	7.76
	DO (mg/L)	7.7	7.6	8.2	6.1	8.1	7.2
	Conductivity (µmhos/cm)	308		309		307	
	Alkalinity (mg $CaCO_3$/L)	64					
	Hardness (mg $CaCO_3$/L)	88					
	Temperature (°C)	25.2	25.1	25.3	25.0	25.4	24.9
6.25%.	pH (S.U.)	7.97	7.68	7.94	7.53	7.91	7.78
	DO (mg/L)	7.8	7.5	8.2	6.6	8.2	7.5
	Conductivity (µmhos/cm)	308		306		311	
	Temperature (°C)	25.2	25.1	25.3	25.0	25.3	24.9
12.5%.	pH (S.U.)	7.97	7.66	7.94	7.54	7.91	7.72
	DO (mg/L)	7.8	7.4	8.2	6.7	7.8	7.2
	Conductivity (µmhos/cm)	300		297		306	
	Temperature (°C)	25.2	25.1	25.3	25.0	25.3	24.9
25%.	pH (S.U.)	7.97	7.67	7.93	7.51	7.90	7.71
	DO (mg/L)	7.8	7.5	8.2	6.5	7.8	7.2
	Conductivity (µmhos/cm)	287		285		300	
	Temperature (°C)	25.3	25.1	25.3	25.0	25.3	24.9
50%.	pH (S.U.)	7.98	7.66	7.93	7.51	7.91	7.84
	DO (mg/L)	7.8	7.3	8.2	6.3	7.7	7.4
	Conductivity (µmhos/cm)	264		261		281	
	Temperature (°C)	25.3	25.1	25.3	25.0	25.3	24.9
100%.	pH (S.U.)	7.99	7.77	7.96	7.66	7.90	7.94
	DO (mg/L)	7.7	7.6	8.2	6.6	7.7	7.5
	Conductivity (µmhos/cm)	216		218		248	
	Alkalinity (mg $CaCO_3$/L)	89				93	
	Hardness (mg $CaCO_3$/L)	110				130	
	TR chlorine (mg/L)	<0.10				<0.10	
	Temperature (°C)	25.3	25.1	25.3	25.0	25.3	24.9
	pH (S.U.)						
	DO (mg/L)						
	Conductivity (µmhos/cm)						
	Alkalinity (mg $CaCO_3$/L)						
	Hardness (mg $CaCO_3$/L)						
	TR chlorine (mg/L)						
	Temperature (°C)						
		Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas*
Client: TVA - Watts Bar UV-TREATED Date: 04.15-03

Concentration	Parameter	Day 3		Day 4		Day 5		Day 6	
CONTROL	pH (S.U.)	8.03	7.52	7.88	7.59	7.93	7.75	7.80	7.47
	DO (mg/L)	8.2	6.4	7.8	7.2	7.9	7.6	7.8	6.5
	Conductivity (µmhos/cm)	~~314~~		299		301		310	
	Alkalinity (mg $CaCO_3$/L)	300							
	Hardness (mg $CaCO_3$/L)								
	Temperature (°C)	25.2	25.1	25.5	24.8	25.5	24.6	25.8	24.7
6.25 I.	pH (S.U.)	8.03	7.52	7.95	7.54	7.91	7.72	7.78	7.38
	DO (mg/L)	8.3	6.5	7.8	6.8	8.1	7.6	7.9	6.1
	Conductivity (µmhos/cm)	~~303~~ ~~298~~		301		306		311	
	Temperature (°C)	25.2	25.1	25.4	24.8	25.5	24.6	25.8	24.7
12.5 I.	pH (S.U.)	8.03	7.50	7.95	7.54	7.90	7.68	7.77	7.39
	DO (mg/L)	8.3	6.5	7.8	6.9	7.6	7.7	7.9	6.0
	Conductivity (µmhos/cm)	296		299		302		308	
	Temperature (°C)	25.3	25.1	25.4	24.8	25.6	24.6	25.8	24.7
25 I.	pH (S.U.)	8.03	7.49	7.92	7.59	7.89	7.71	7.76	7.44
	DO (mg/L)	8.2	6.1	7.6	7.0	7.7	7.6	7.8	6.1
	Conductivity (µmhos/cm)	289		289		292		298	
	Temperature (°C)	25.5	25.1	25.4	24.8	25.6	24.6	25.8	24.7
50 I.	pH (S.U.)	8.05	7.62	7.91	7.59	7.89	7.78	7.78	7.43
	DO (mg/L)	8.2	6.5	7.5	6.7	7.6	7.6	7.6	6.0
	Conductivity (µmhos/cm)	275		272		276		281	
	Temperature (°C)	25.5	25.1	25.3	24.8	25.6	24.6	25.8	24.7
100 I.	pH (S.U.)	8.11	7.75	7.91	7.88	7.88	7.85	7.79	7.65
	DO (mg/L)	8.7	6.6	7.7	7.6	7.6	7.5	7.6	6.7
	Conductivity (µmhos/cm)	249		241 ~~272~~		246		250	
	Alkalinity (mg $CaCO_3$/L)			98					
	Hardness (mg $CaCO_3$/L)			130					
	TR Chlorine (mg/L)			<0.10					
	Temperature (°C)	25.6	25.1	25.2	24.8	25.6	24.6	25.8	24.7
	pH (S.U.)								
	DO (mg/L)								
	Conductivity (µmhos/cm)								
	Alkalinity (mg $CaCO_3$/L)								
	Hardness (mg $CaCO_3$/L)								
	TR chlorine (mg/L)								
	Temperature (°C)								
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

Total Residual Chlorine
(EPA Method 330.5)

Matrix: Water, MDL = 0.10 mg/L
Meter: Accumet Model AR25 pH/Ion Meter

Analyst	KEW
Date analyzed	04-15-03

Iodide reagent:	1NR07976
Acid reagent:	1NR059

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	1NS5134	1NS5134

Note: For samples with a residual chlorine of > 1.0 mg/L, the calibration range must be adjusted to bracket the chlorine levels of the samples.

Laboratory control standard:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
1NS5134	0.50	0.486	97.2%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
030415.02	Prog. Energy Cape Fear 007	brown/orange	S <0.0078	
↓	Duplicate	cloudy	D <0.0038	—

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.0088
030415.03	Prog Energy Tpstream	brown/orange, cloudy	<0.00273
030415.01	TVA WBNT	no color, clear	<0.0193

Note: All samples were analyzed in excess of EPA recommended holding time (15 minutes) unless otherwise noted.

Laboratory control standard:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
1NS5134	0.50	0.464	92.8%

Reviewed by	JS
Date reviewed	04-20-03

Total Residual Chlorine
(EPA Method 330.5)

Matrix: Water, MDL = 0.10 mg/L
Meter: Accumet Model AR25 pH/Ion Meter

Analyst	KEK	Iodide reagent:	INR075
Date analyzed	04-17-03	Acid reagent:	INR059

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INSS134	INSS134

Note: For samples with a residual chlorine of > 1.0 mg/L, the calibration range must be adjusted to bracket the chlorine levels of the samples.

Laboratory control standard:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS134	0.50	0.468	93.67

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
030417.01	TVA		S <0.00065	
↓	Duplicate		D <0.00235	—

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00733

Note: All samples were analyzed in excess of EPA recommended holding time (15 minutes) unless otherwise noted.

Laboratory control standard:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS134	0.50	0.452	90.41

Reviewed by	
Date reviewed	04-20-03

Total Residual Chlorine
(EPA Method 330.5)

Matrix: Water, MDL = 0.10 mg/L

Meter: Accumet Model AR25 pH/Ion Meter

Analyst	KBL
Date analyzed	04-19-03

Iodide reagent:	INR075
Acid reagent:	INR059

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INSS134	INSS134

Note: For samples with a residual chlorine of > 1.0 mg/L, the calibration range must be adjusted to bracket the chlorine levels of the samples.

Laboratory control standard:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS134	0.50	0.511	102.2%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
030419.01	Watts Bar TVA	pale tan, slightly cloudy	S <0.000402	
↓	Duplicate		D <0.000356	–

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00992
030418.01	Prog. Energy Cape Fear	pale orange, cloudy	<0.00698
030418.02	↓ Upstream	pale orange, cloudy	<0.00491

KBL

Note: All samples were analyzed in excess of EPA recommended holding time (15 minutes) unless otherwise noted.

Laboratory control standard:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS134	0.50	0.500	100%

Reviewed by	JT
Date reviewed	04-20-03

Environmental Testing Solutions, LLC Page 1 of 1

Alkalinity
(EPA Method 310.1)
Matrix: Water, MDL = 1.0 mg $CaCO_3$/L

Analyst: KeL
Date analyzed: 04.24.03

Titrate samples to pH = 4.50 S.U.

Titrant normality and multiplier determination:

pH of Deionized water = 4.5 S.U.	Titrant reference number	Normality check standard number	Begin ml	End ml	Total ml (E)	Normality (N) of H_2SO_4 = (5 ml Na_2CO_3 x 0.05)/E = 0.25/E (acceptable range = 0.018 - 0.022)	pH Factor or Multiplier = (N x 50000)/ 100 ml sample = N x 500
7.0	INR074	INR079	0.2	12.5	12.3	0.0203	10.2

BLK 0.0 - 0.2 - 0.2ml

Laboratory control standard:

Reference standard number	True value (TV) (mg $CaCO_3$/L)	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (MV) (mg $CaCO_3$/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INR045	100	100	12.5	22.3	9.8	10.2	100	100 %

Duplicate sample precision:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
04.22.03	SSW H2O	100	22.3	25.6	3.3	10.2	S 34	
↓	Duplicate	100	25.6	28.8	3.2	↓	D 33	3.0 %

Matrix spike recovery:

Reference standard number	Spike value (SV) (mg $CaCO_3$/L)	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Spike alkalinity (A) (mg $CaCO_3$/L)
INR005	50	100	25.6	33.7	8.1	10.2	83

Sample alkalinity (B) (mg $CaCO_3$/L)	Measured spike value (MV) MV = A - B (mg $CaCO_3$/L)	% R = MV / SV x 100 (acceptable range = 75 to 125%)
33	50	100 %

Sample measurements:

Sample number	Sample ID		Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
04.17.03	MHS H2O	A	100	33.7	39.8	6.1	10.1	62
04.17.03		B		39.8	45.5	5.7		58
04.17.03		C		0.7	6.8	6.1		62
04.22.03		A		6.8	12.9	6.1		62
04.22.03		B		12.9	18.7	5.8		69
030415.01	NBN	1		18.7	27.5	8.8		89
030417.01		2		28.0	37.2	9.2		93
030419.01		3		37.2	46.9	9.7		98

Reviewed by: JJ Date reviewed: 05-01-03

Total Hardness
(EPA Method 130.2)
Matrix: Water, MDL = 1.0 mg $CaCO_3$/L

Analyst: KLK
Date analyzed: 04.24.03

Titrant normality and multiplier determination:

Titrant reference number	Normality check standard number	Begin ml	End ml	Total ml (E)	Normality (N) of EDTA = 0.2/E (acceptable range = 0.018 - 0.022)	pH Factor or Multiplier = (N x 50000)/ 50 ml sample = N x 1000
INR05D	INSS092	0.3	10.3	10	0.020	20

Laboratory control standard:

Reference standard number	True value (TV) (mg $CaCO_3$/L)	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Hardness (MV) (mg $CaCO_3$/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS101	40	50	10.3	12.4	2.1	20	42	105%

Duplicate sample precision:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Hardness (mg $CaCO_3$/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
04.22.03	SSW H2O	50	12.5	14.7	2.2	20	S 44	
↓	Duplicate	50	14.8	17.1	2.3	20	D 46	4.4%

Matrix spike recovery:

Reference standard number	Spike value (SV) (mg $CaCO_3$/L)	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Spike hardness (A) (mg $CaCO_3$/L)
INSS101	40	50	14.8	19.2	4.4	20	88

Sample hardness (B) (mg $CaCO_3$/L)	Measured spike value (MV) MV = A - B (mg $CaCO_3$/L)	% R = MV / SV x 100 (acceptable range = 75 to 125%)
44	42	105%

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Hardness (mg $CaCO_3$/L)
TV=ND	Blank (should be = 0 mg $CaCO_3$/L)	50	0.3	0.3	0.0	20	ND
04.17.03	MHS H2O A		19.2	23.6	4.4		88
04.17.03	B		23.6	28.4	4.8		96
04.17.03	C		28.5	32.7	4.2		84
04.22.03	A		32.7	37.4	4.7		94
04.22.03	B		37.4	41.6	4.2		84
030415.01	WBN 1		16.4	21.8	5.4		110
030417.01	2		21.9	28.2	6.3		130
030419.01	3		28.2	34.5	6.3		130

Note: If >15ml of titrant is used, sample must be diluted. Reviewed by: [signature] Date reviewed: 05-01-03

Watts Bar Nuclear Plant Biomonitoring
April 15 - 22, 2003

Appendix D

Reference Toxicant Test and
Control Chart

Environmental Testing Solutions, LLC

Potassium Chloride Chronic Reference Toxicant Control Chart
for *Pimephales promelas*
using Moderately Hard Synthetic Water

Test number	Test date	7-day IC_{25} (g KCl/L)	CT (g/L KCl)	S	Control Limits CT - 2S	Control Limits CT + 2S	$S_{A.10}$	Warning Limits CT - $S_{A.10}$	Warning Limits CT + $S_{A.10}$	$S_{A.25}$	Control Limits CT - $S_{A.25}$	Control Limits CT + $S_{A.25}$	CV
1	08-06-02	0.55											
2	08-13-02	0.58	0.57	0.02	0.53	0.61	0.07	0.50	0.63	0.12	0.45	0.69	0.03
3	08-20-02	0.63	0.59	0.04	0.51	0.66	0.07	0.52	0.66	0.12	0.46	0.71	0.06
4	09-10-02	0.61	0.59	0.03	0.53	0.66	0.07	0.52	0.66	0.12	0.47	0.72	0.06
5	09-17-02	0.64	0.60	0.04	0.53	0.68	0.07	0.53	0.68	0.13	0.48	0.73	0.06
6	10-01-02	0.60	0.60	0.03	0.54	0.67	0.07	0.53	0.68	0.13	0.48	0.73	0.05
7	10-01-02	0.47	0.58	0.06	0.47	0.70	0.07	0.51	0.65	0.12	0.46	0.71	0.10
8	10-08-02	0.53	0.58	0.06	0.46	0.69	0.07	0.51	0.65	0.12	0.46	0.70	0.10
9	10-15-02	0.66	0.59	0.06	0.47	0.71	0.07	0.52	0.66	0.12	0.46	0.71	0.10
10	10-22-02	0.64	0.59	0.06	0.47	0.71	0.07	0.52	0.66	0.12	0.47	0.72	0.10
11	11-05-02	0.59	0.59	0.06	0.48	0.70	0.07	0.52	0.66	0.12	0.47	0.72	0.10
12	12-03-02	0.61	0.59	0.05	0.48	0.70	0.07	0.52	0.66	0.12	0.47	0.72	0.09
13	12-03-02	0.52	0.59	0.06	0.48	0.70	0.07	0.52	0.66	0.12	0.46	0.71	0.09
14	01-07-03	0.64	0.59	0.06	0.48	0.70	0.07	0.52	0.66	0.12	0.47	0.71	0.09
15	01-14-03	0.61	0.59	0.05	0.48	0.70	0.07	0.52	0.66	0.12	0.47	0.72	0.09
16	02-04-03	0.64	0.59	0.05	0.49	0.70	0.07	0.52	0.67	0.12	0.47	0.72	0.09
17	03-18-03	0.65	0.60	0.05	0.49	0.70	0.07	0.53	0.67	0.13	0.47	0.72	0.09
18	03-18-03	0.64	0.60	0.05	0.50	0.70	0.07	0.53	0.67	0.13	0.47	0.73	0.09
19	04-08-03	0.50	0.59	0.06	0.48	0.71	0.07	0.52	0.67	0.12	0.47	0.72	0.09
20	04-15-03	0.56	0.59	0.05	0.48	0.70	0.07	0.52	0.66	0.12	0.47	0.72	0.09

Note:

7-d IC_{25} = 7-day 25% inhibition concentration. An estimation of the concentration of potassium chloride that would cause a 25% reduction in Pimephales growth for the test population.

CT = Central tendency (mean IC_{25}).

S = Standard deviation of the IC_{25} values.

$S_{A.10}$ = Standard deviation corresponding to the 10th percentile CV. $S_{A.10}$ = 0.12, as determined by USEPA for the method and endpoint.

$S_{A.25}$ = Standard deviation corresponding to the 25th percentile CV. $S_{A.25}$ = 0.21, as determined by the USEPA for the method and endpoint.

CV = Coefficient of variation of the IC25 values.

Control and warning limits were established using the standard deviation of the IC_{25} values corresponding to the 10th and 25th percentile CVs. These ranges are more stringent than the control and warning limits recommended by USEPA for the test method and endpoint.

USEPA Recommended Control and Warning Limits:

Warning Limit = Standard deviation corresponding to the 75th percentile CV. $S_{A.75}$ = 0.38.

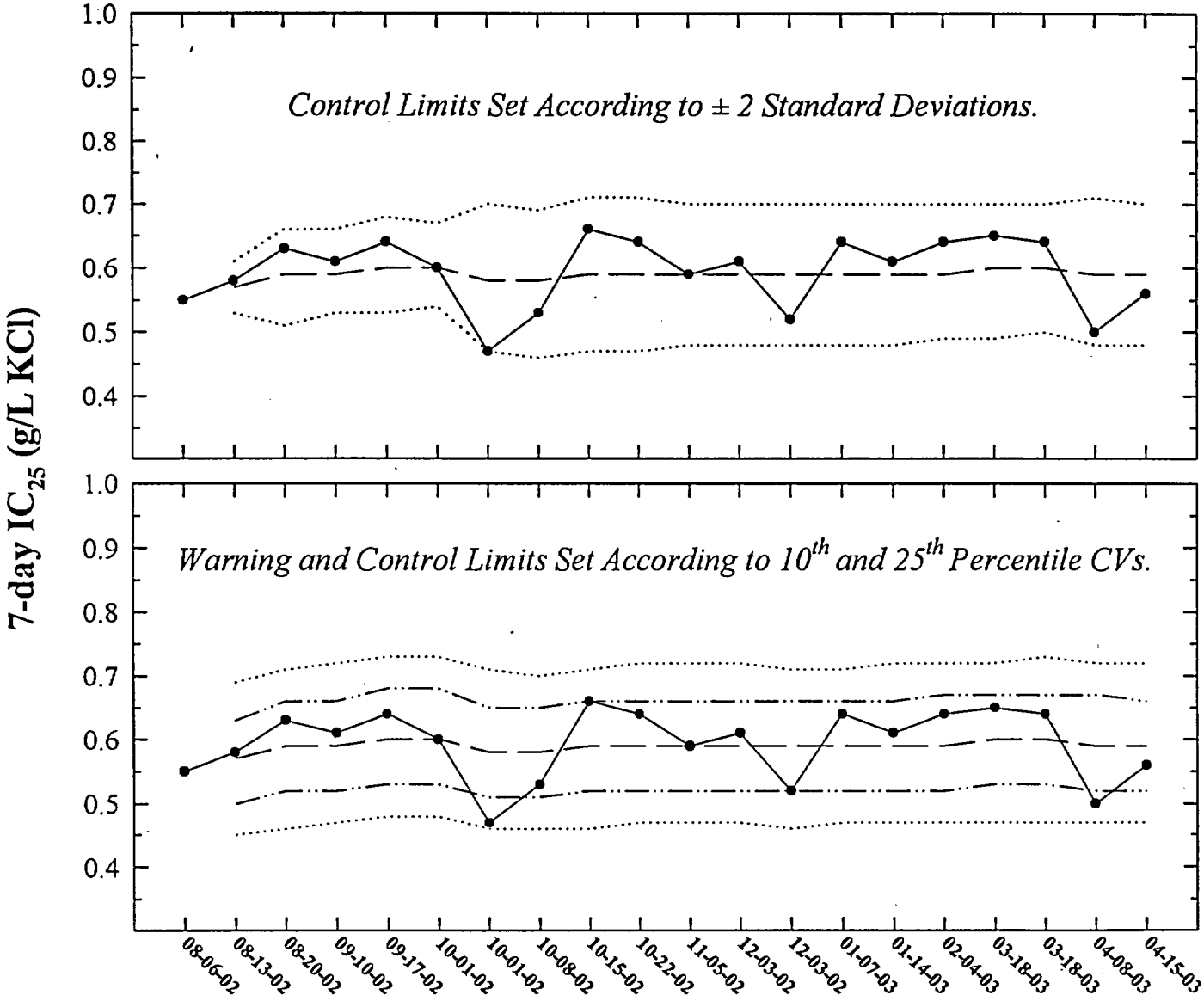
Control Limit = Standard deviation corresponding to the 90th percentile CV. $S_{A.90}$ = 0.45.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Organisms obtained from Aquatic BioSystems, Inc.

Environmental Testing Solutions, LLC

Potassium Chloride Chronic Reference Toxicant Control Chart
for *Pimephales promelas*
using Moderately Hard Synthetic Water



—●— **7-day IC_{25}** = 25% inhibition concentration. An estimation of the concentration of potassium chloride that would cause a 25% reduction in *Pimephales* growth for the test population.

— – **Central Tendency** (mean IC_{25})

—·— **Warning Limits** (mean $IC_{25} \pm S_{A.10}$)

····· **Control Limits** (mean $IC_{25} \pm S_{A.25}$ or 2 Standard Deviations)

Environmental Testing Solutions, LLC

Precision of Endpoint Measurements

Potassium Chloride Chronic Reference Toxicant Data for *Pimephales promelas* using Moderately Hard Synthetic Water

Test number	Test date	Control Survival (%)	Control Mean Growth (mg/larvae)	CT for Control Growth (mg/larvae)	CV (%)	CT for Control Growth CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	08-06-02	97.5	0.694		22.2		0.16	23.5	
2	08-13-02	100	0.659	0.677	13.6	17.9	0.12	17.5	20.5
3	08-20-02	100	0.765	0.706	8.5	14.8	0.12	16.3	19.1
4	09-10-02	100	0.854	0.743	1.2	11.4	0.11	12.5	17.5
5	09-17-02	100	0.824	0.759	13.4	11.8	0.12	14.6	16.9
6	10-01-02	97.5	0.750	0.758	18.4	12.9	0.19	25.4	18.3
7	10-01-02	100	0.975	0.789	12.7	12.8	0.13	12.8	17.5
8	10-08-02	97.5	0.929	0.806	8.0	12.2	0.18	19.5	17.8
9	10-15-02	100	1.037	0.832	16.9	12.8	0.23	21.9	18.2
10	10-22-02	100	0.822	0.831	10.6	12.5	0.13	15.6	18.0
11	11-05-02	100	0.874	0.835	2.8	11.7	0.12	13.8	17.6
12	12-03-02	100	0.852	0.836	9.1	11.4	0.12	13.7	17.3
13	12-03-02	100	0.668	0.823	10.4	11.4	0.15	22.4	17.7
14	01-07-03	100	0.886	0.828	4.1	10.8	0.14	15.7	17.5
15	01-14-03	100	0.677	0.818	3.0	10.3	0.07	11.0	17.1
16	02-04-03	97.5	0.933	0.825	14.1	10.6	0.15	16.5	17.1
17	03-18-03	100	0.838	0.826	8.0	10.4	0.15	18.5	17.1
18	03-18-03	100	0.803	0.824	21.3	11.0	0.21	26.5	17.7
19	04-08-03	100	1.083	0.838	6.1	10.8	0.09	8.0	17.1
20	04-15-03	100	0.892	0.841	17.0	11.1	0.17	18.7	17.2

Note:

CV = Coefficient of variation for control survival.
On average, the CV for control growth is **11.1%** in Environmental Testing Solutions, LLC Pimephales chronic toxicity tests.
Lower CV bound determined by USEPA (10^{th} percentile) = 3.5%.
Upper CV bound determined by USEPA (90^{th} percentile) = 20%

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference
PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, LLC chronic toxicity tests when a toxicant reduces Pimephales growth by **17.2%** from the control.
Lower PMSD bound determined by USEPA (10^{th} percentile) = 9.4%.
Upper PMSD bound determined by USEPA (90^{th} percentile) = 35%.

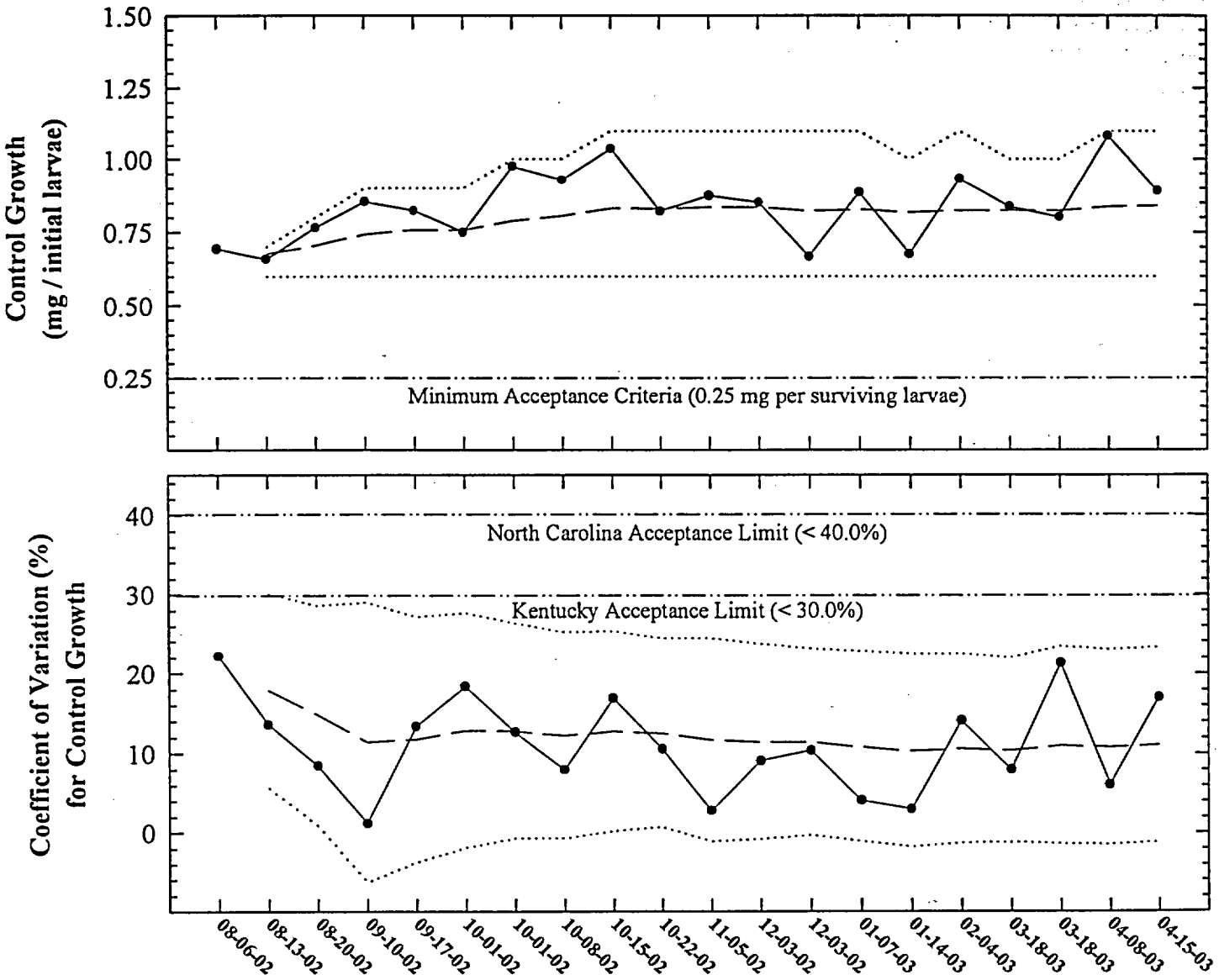
CT = Central Tendancy (mean Control Growth, CV, or mean PMSD)

The lower and upper bounds were calculated by the USEPA using 205 tests conducted from 19 laboratories for Pimephales growth in chronic reference toxicant tests.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Environmental Testing Solutions, LLC

Pimephales promelas Control Growth and Coefficient of Variation in Potassium Chloride Chronic Reference Toxicant Tests



—●— **Control Growth or Coefficient of Variation (CV)**

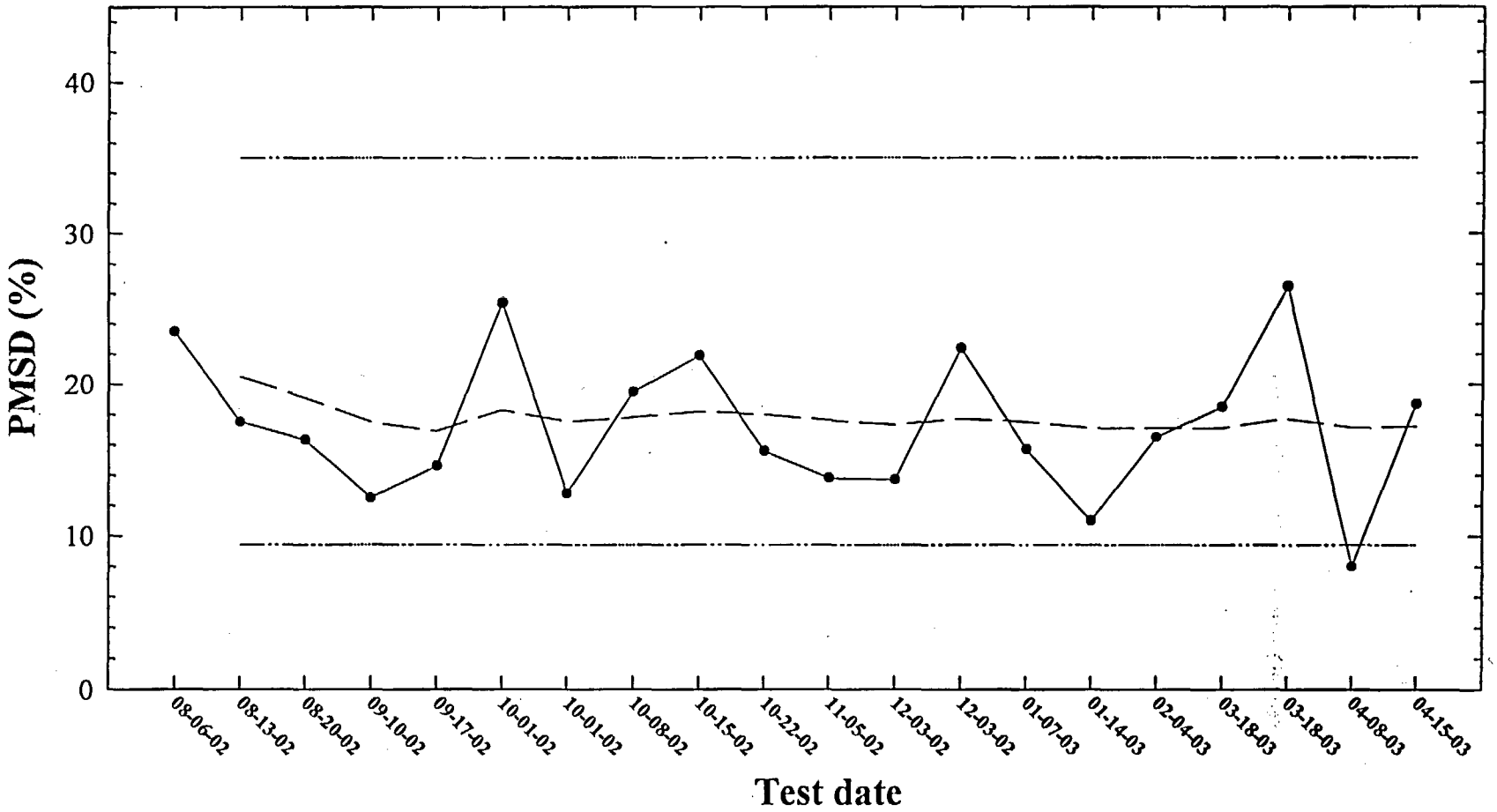
— – **Central Tendency** (mean Control Growth or CV)

····· **Control Limits** (mean Control Growth or CV ± 2 Standard Deviations)

Environmental Testing Solutions, LLC

Precision of Endpoint Measurements

Potassium Chloride Chronic Reference Toxicant Control Chart
for *Pimephales promelas*
using Moderately Hard Synthetic Water



—●— **PMSD** = percent minimum significant difference. PMSD is the minimum significant difference between the control and treatment that can be declared statistically significant.

—— - **Central Tendency** (mean PMSD)

····· **Lower and Upper PMSD Bounds**

Lower PMSD Bound (10^{th} percentile) = 9.4%, Upper PMSD Bound (90^{th} percentile) = 35%

(Lower and upper PMSD bounds were determined by USEPA for the method and endpoint.)

Potassium Chloride Chronic Reference Toxicant Test
(EPA-821-R-02-013 Method 1000.0)
Species: *Pimephales promelas*

PpKClCR Test Number: 8

Dilution preparation information:						*Comments:*
KCl CHM number:		CHM 067				
Stock preparation:		50 g KCl/L: Dissolve 50 g KCl in 1-L Deionized water				
Dilution prep (mg/L)	300	450	600	750	900	
Stock volume (mL)	6	9	12	15	18	
Diluent volume (mL)	994	991	988	985	982	
Total volume (mL)	1000	1000	1000	1000	1000	

Test organism information:		*Test information:*	
Organism age:	26 - 29 HOURS OLD	Randomizing template:	RED
Date and times organisms were born between:	04-14-03 1030 TO 1330 MST	Incubator number:	3C
Organism source:	ABS BATCH 04-14-03	*Artemia* lot number:	8618090
Transfer bowl information:	pH = 7.80 Temperature = 24.2 °C	Total drying time:	19-HOURS
		Date / Time in: 04-22-03	1500
Average transfer volume:	10.4 ml	Date / Time out: 04-23-03	0800
		Oven temperature:	100°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Control water batch used	Analyst
0	04-15-03	———	1430	1337	04-11-03	dl
1	04-16-03	0900	1504	1356	04-11-03	dl
2	04-17-03	0903	1510	1340	04-11-03	dl
3	04-18-03	0900	1500	1338	04-11-03	dl
4	04-19-03	0856	1503	1250	04-11-03	dl
5	04-20-03	0902	1500	1316	04-11-03	dl
6	04-21-03	0847	1500	1321	04-11-03	dl
7	04-22-03			1400	———	dl

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	0%	≤ 20%	7-day LC_{50}	714.2
Average weight per initial larvae:	0.8919		NOEC	450
Average weight per surviving larvae:	0.8919	≥ 0.25 mg/larvae	LOEC	600
			ChV	519.6
			IC_{25}	562.1

PpKClCR Test Number: 8

Survival and Growth Data

Day	CONTROL				300 mg KCl/L				450 mg KCl/L			
	A	B	C	D	E	F	G	H	I	J	K	L
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	10	10	10	10	10	10	10	10
7	10	10	10	10	10	10	10	10	10	10	10	10
A = Pan weight (mg) TRAY	15.084	15.000	15.042	14.857	14.896	14.952	14.860	15.067	15.167	14.638	14.649	15.195
B = Pan + Larvae weight (mg)	22.91	26.14	~~23.17~~ 23.17	23.44	23.06	22.02	22.68	24.27	23.32	23.48	22.97	22.39
Larvae weight (mg) = A – B	7.826	11.140	8.128	8.583	8.164	7.068	7.820	9.203	8.158	8.830	7.906	7.195

*Re-checked (1. LARGE LARVAE)

Calculations and data reviewed: JJ

Comments:

PpKClCR Test Number: 8

Survival and Growth Data

Day	600 mg KCl/L				750 mg KCl/L				900 mg KCl/L			
	M	N	O	P	Q	R	S	T	U	V	W	X
0	10	10	10	10	10	10	10	10	10	10	10	10
1	9 1d	7 3d	8 2d	9 1d	6 4d	5 5d	5 5d	4 6d	1 9d	1 9d	2 8d	1 9d
2	9	7	8	9	6	5	5	4	0 1d	1	2	1
3	9	7	8	9	6	5	5	4	0	1	2	1
4	9	7	8	9	6	4 1d	5	4	0	1	2	1
5	9	7	8	8 1d	6	4	4 1d	4	0	1	2	1
6	9	7	8	8	6	4	4	4	0	1	1 2d	1
7	9	7	8	8	6	4	4	4	0	1	1	1
A = Pan weight (mg)	15.116	15.097	15.129	14.954	14.947	14.725	14.765	14.714	—	14.462	14.797	14.680
B = Pan + Larvae weight (mg)	22.12	21.14	20.53	21.46	19.69	17.84	18.38	17.41	—	15.37	15.66	15.74
Larvae weight (mg) = A – B	7.004	6.043	5.401	6.506	4.743	3.115	3.615	2.696	—	0.708	0.863	1.060

Calculations and data reviewed: JH

Comments:

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1000.0)
Species: *Pimephales promelas*

Quality Control
Verification of Data Entry, Calculations, and Statistical Analyses

Test number: PpKCICR # 48

Test dates: April 15-22, 2003

Reveiwed by: JLumpkin

Concentration (mg/L KCl)	Replicate	Initial number of larvae	Final number of larvae	A = Pan weight (mg)	B = Pan + Larvae weight (mg)	Larvae weight (mg) = A - B	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight (mg)	Coefficient of variation (%)	Percent reduction from control (%)
Control	A	10	10	15.084	22.910	7.826	0.7826	100.0	0.8919	17.0	Not applicable
	B	10	10	15.000	26.140	11.140	1.1140				
	C	10	10	15.042	23.170	8.128	0.8128				
	D	10	10	14.857	23.440	8.583	0.8583				
300	E	10	10	14.896	23.060	8.164	0.8164	100.0	0.8064	11.0	9.6
	F	10	10	14.952	22.020	7.068	0.7068				
	G	10	10	14.860	22.680	7.820	0.7820				
	H	10	10	15.067	24.270	9.203	0.9203				
450	I	10	10	15.162	23.320	8.158	0.8158	100.0	0.8022	8.4	10.1
	J	10	10	14.650	23.480	8.830	0.8830				
	K	10	10	15.064	22.970	7.906	0.7906				
	L	10	10	15.195	22.390	7.195	0.7195				
600	M	10	9	15.116	22.120	7.004	0.7004	80.0	0.6239	10.9	30.1
	N	10	7	15.097	21.140	6.043	0.6043				
	O	10	8	15.129	20.530	5.401	0.5401				
	P	10	8	14.954	21.460	6.506	0.6506				
750	Q	10	6	14.947	19.690	4.743	0.4743	45.0	0.3542	25.0	60.3
	R	10	4	14.725	17.840	3.115	0.3115				
	S	10	4	14.765	18.380	3.615	0.3615				
	T	10	4	14.714	17.410	2.696	0.2696				
900	U	10	0	0.000	0.000	0.000	0.0000	7.5	0.0658	70.2	92.6
	V	10	1	14.662	15.370	0.708	0.0708				
	W	10	1	14.797	15.660	0.863	0.0863				
	X	10	1	14.680	15.740	1.060	0.1060				

Dunnett's MSD value: 0.1672

PMSD: 18.7

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, LLC chronic toxicity tests when a toxicant reduces Pimephales growth by 17.2% from the control (determined through reference toxicant testing).

Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.

Upper PMSD bound determined by USEPA (90th percentile) = 35%.

The lower and upper bounds were calculated by the USEPA using 205 tests conducted from 19 laboratories for Pimephales growth in chronic reference toxicant tests.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Environmental Testing Solutions, LLC
Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Survival

Start Date:	4/15/03	Test ID:	PpKCICR	Sample ID:	REF-Ref Toxicant
End Date:	4/22/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	KCL-Potassium chloride
Sample Date:		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	1.0000	1.0000	1.0000	1.0000
300	1.0000	1.0000	1.0000	1.0000
450	1.0000	1.0000	1.0000	1.0000
600	0.9000	0.7000	0.8000	0.8000
750	0.6000	0.4000	0.4000	0.4000
900	0.0000	0.1000	0.1000	0.1000

			Transform: Arcsin Square Root					Rank	1-Tailed	Number	Total
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	Sum	Critical	Resp	Number
D-Control	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4			0	40
300	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4	18.00	10.00	0	40
450	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4	18.00	10.00	0	40
*600	0.8000	0.8000	1.1136	0.9912	1.2490	9.478	4	10.00	10.00	8	40
*750	0.4500	0.4500	0.7351	0.6847	0.8861	13.697	4	10.00	10.00	22	40
*900	0.0750	0.0750	0.2810	0.1588	0.3218	28.997	4	10.00	10.00	37	40

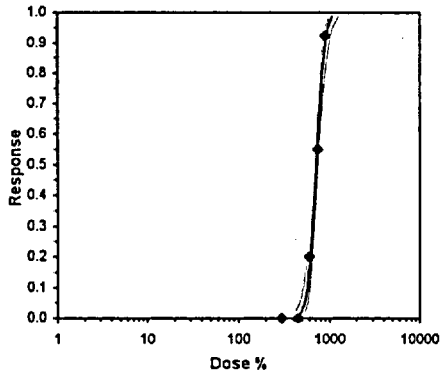
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.830562234	0.884	0.449956848	2.238040149
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	450	600	519.6152423	0.222222222
Treatments vs D-Control				

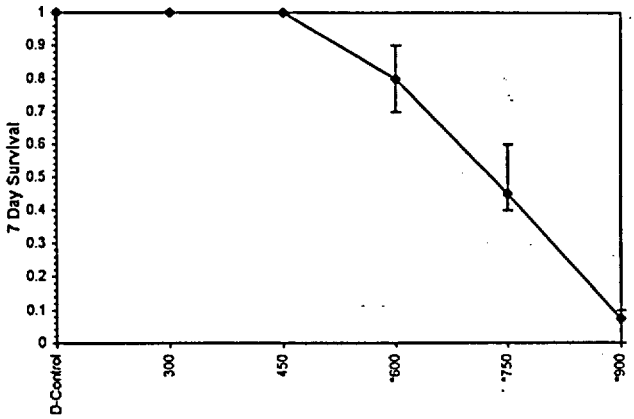
Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	12.95726761	1.79042942	9.448025883	16.46650935	0	1.365757569	7.814724922	0.71	2.853795954	0.077176765	3
Intercept	-31.9773979	5.117653608	-42.0079992	-21.9467966							
TSCR											

Point	Probits	%	95% Fiducial Limits	
EC01	2.674	472.3413312	400.2848896	521.3750248
EC05	3.355	533.1525261	471.0954071	575.3446747
EC10	3.718	568.7088027	513.2852597	607.007368
EC15	3.964	594.028635	543.459587	629.8182164
EC20	4.158	614.9536041	568.3344609	648.9760056
EC25	4.326	633.4918278	590.2082895	666.2951785
EC40	4.747	682.721288	646.5877268	714.817046
EC50	5.000	714.1607122	680.4217756	748.5909478
EC60	5.253	747.0479333	713.4874084	786.7502467
EC75	5.674	805.1019726	766.6545095	860.5455739
EC80	5.842	829.372365	787.4331737	893.3038724
EC85	6.036	858.5874515	811.6714609	933.8622463
EC90	6.282	896.8131075	842.4359998	988.4526077
EC95	6.645	956.6221399	889.0568522	1076.661109
EC99	7.326	1079.781756	981.3575575	1266.773333



Dose-Response Plot



Organisms obtained from Aquatic BioSystems, Inc.

Environmental Testing Solutions, LLC

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	4/15/03	Test ID:	PpKClCR	Sample ID:	REF-Ref Toxicant
End Date:	4/22/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	KCL-Potassium chloride
Sample Date:		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.7826	1.1140	0.8128	0.8583
300	0.8164	0.7068	0.7820	0.9203
450	0.8158	0.8830	0.7906	0.7195
600	0.7004	0.6043	0.5401	0.6506
750	0.4743	0.3115	0.3615	0.2696
900	0.0000	0.0708	0.0863	0.1060

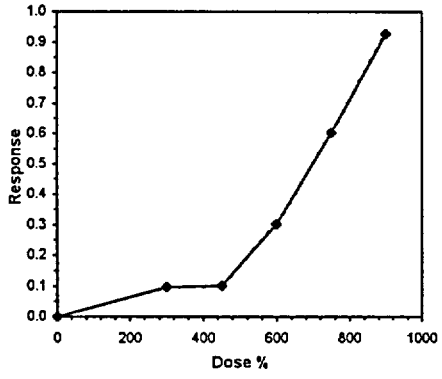
			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.8919	1.0000	0.8919	0.7826	1.1140	16.962	4				0.8919	1.0000
300	0.8064	0.9041	0.8064	0.7068	0.9203	10.997	4	1.115	2.180	0.1672	0.8064	0.9041
450	0.8022	0.8994	0.8022	0.7195	0.8830	8.420	4	1.169	2.180	0.1672	0.8022	0.8994
600	0.6239	0.6994	0.6239	0.5401	0.7004	10.939	4				0.6239	0.6994
750	0.3542	0.3971	0.3542	0.2696	0.4743	24.963	4				0.3542	0.3971
900	0.0658	0.0737	0.0658	0.0000	0.1060	70.171	4				0.0658	0.0737

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.906657279		0.805		1.102531033	1.01612156
Bartlett's Test indicates equal variances (p = 0.41)					1.793151259		9.21035099			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	450	>450		0.222222222	0.167241389	0.187506112	0.010254743	0.01177076	0.450955391	2, 9
Treatments vs D-Control										

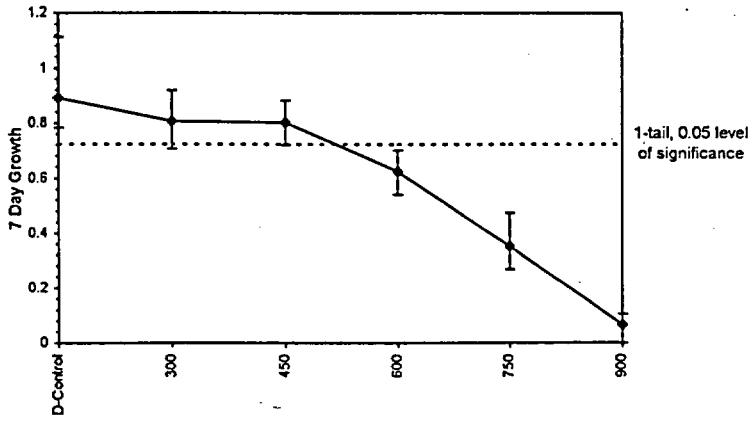
Linear Interpolation (200 Resamples)

Point	%	SD	95% CL(Exp)		Skew
IC05*	156.39	153.69	15.34	681.71	0.4635
IC10	431.66	137.87	0.00	578.46	-0.3433
IC15	487.08	111.52	35.28	615.98	-1.1116
IC20	524.58	81.35	121.96	654.88	-1.7839
IC25	562.08	55.02	377.36	664.82	-1.8367
IC40	649.34	26.44	554.05	718.39	-0.4531
IC50	698.96	24.21	625.24	776.43	0.0862

* indicates IC estimate less than the lowest concentration



Dose-Response Plot



Organisms obtained from Aquatic BioSystems, Inc.

PpKClCR Test Number: 8

Daily Chemistry:

Concentration	Parameter	Day 0		Day 1		Day 2	
CONTROL	pH (S.U.)	7.89	7.62	7.93	7.59	7.98	7.75
	DO (mg/L)	7.7	7.5	8.2	7.2	7.8	7.5
	Conductivity (µmhos/cm)	305		319		306	
	Alkalinity (mg $CaCO_3$/L)	64					
	Hardness (mg $CaCO_3$/L)	88					
	Temperature (°C)	25.3	25.1	25.7	25.1	25.6	25.0
300 mg KCl/L	pH (S.U.)	7.97	7.69	7.92	7.56	7.99	7.76
	DO (mg/L)	7.6	7.5	8.2	7.2	7.9	7.5
	Conductivity (µmhos/cm)	848		852		849	
	Temperature (°C)	25.3	25.1	25.7	25.1	25.6	25.0
450 mg KCl/L	pH (S.U.)	7.97	7.68	7.96	7.62	8.08	7.73
	DO (mg/L)	7.7	7.6	8.2	7.1	8.0	7.6
	Conductivity (µmhos/cm)	1152		1123		1130	
	Temperature (°C)	25.3	25.1	25.7	25.1	25.6	25.0
600 mg KCl/L	pH (S.U.)	7.97	7.68	7.95	7.67	8.01	7.76
	DO (mg/L)	7.7	7.6	8.2	7.3	8.0	7.4
	Conductivity (µmhos/cm)	1412		1387		1386	
	Temperature (°C)	25.3	25.1	25.7	25.1	25.6	25.0
750 mg KCl/L	pH (S.U.)	7.97	7.72	7.95	7.68	8.01	7.83
	DO (mg/L)	7.8	7.6	8.2	7.3	8.0	7.7
	Conductivity (µmhos/cm)	1693		1655		1651	
	Temperature (°C)	25.3	25.1	25.7	25.1	25.6	25.0
900 mg KCl/L	pH (S.U.)	7.96	7.73	7.95	7.75	8.01	7.82
	DO (mg/L)	7.8	7.7	8.3	7.7	8.0	7.8
	Conductivity (µmhos/cm)	1901		1870		1869	
	Temperature (°C)	25.3	25.1	25.7	25.1	25.6	25.0
		Initial	Final	Initial	Final	Initial	Final

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PpKClCR Test Number: 8

Concentration	Parameter	Day 3		4		5		6	
CONTROL	pH (S.U.)	7.96	7.51	8.00	7.54	8.01	7.62	7.72	7.56
	DO (mg/L)	8.1	6.8	7.8	7.0	7.7	7.8	7.9	6.9
	Conductivity (µmhos/cm)	314		288		294		313	
	Alkalinity (mg $CaCO_3$/L)								
	Hardness (mg $CaCO_3$/L)								
	Temperature (°C)	25.7	24.9	25.6	25.0	25.5	24.8	25.7	24.9
300 mg KCl/L	pH (S.U.)	8.02	7.49	7.99	7.54	8.01	7.62	7.80	7.50
	DO (mg/L)	8.1	6.8	7.8	6.8	7.8	7.7	8.0	6.6
	Conductivity (µmhos/cm)	858		842		850		868	
	Temperature (°C)	25.7	24.9	25.6	25.0	25.5	24.8	25.7	24.9
450 mg KCl/L	pH (S.U.)	8.02	7.46	8.00	7.52	8.02	7.61	7.83	7.49
	DO (mg/L)	8.2	6.3	7.8	6.7	7.8	7.7	8.2	6.6
	Conductivity (µmhos/cm)	1142		1120		1134		1160	
	Temperature (°C)	25.7	24.9	25.6	25.0	25.5	24.8	25.7	24.9
600 mg KCl/L	pH (S.U.)	8.04	7.55	8.03	7.53	8.03	7.64	7.85	7.46
	DO (mg/L)	8.2	6.9	7.9	6.7	7.8	7.6	8.2	6.2
	Conductivity (µmhos/cm)	1408		1392		1407		1441	
	Temperature (°C)	25.7	24.9	25.6	25.0	25.5	24.8	25.7	24.9
750 mg KCl/L	pH (S.U.)	8.04	7.58	8.03	7.53	8.03	7.69	7.87	7.49
	DO (mg/L)	8.1	6.9	7.9	6.8	7.9	7.6	8.1	6.5
	Conductivity (µmhos/cm)	1664		1652		1671		1702	
	Temperature (°C)	25.7	24.9	25.6	25.0	25.5	24.8	25.7	24.9
900 mg KCl/L	pH (S.U.)	8.05	7.61	8.02	7.54	8.03	7.69	7.80	7.61
	DO (mg/L)	8.2	7.3	7.9	7.0	7.9	7.8	8.2	7.0
	Conductivity (µmhos/cm)	1889		1876		1905		1930	
	Temperature (°C)	25.7	24.9	25.6	25.0	25.5	24.8	25.7	24.9
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

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Environmental Testing Solutions, LLC

Sodium Chloride Chronic Reference Toxicant Control Chart
for *Ceriodaphnia dubia*
using Moderately Hard Synthetic Water

Test number	Test date	7-d IC_{25} (g/L NaCl)	CT (g/L NaCl)	S	Control Limits CT - 2S	Control Limits CT + 2S	$S_{A.10}$	Warning Limits CT - $S_{A.10}$	Warning Limits CT + $S_{A.10}$	$S_{A.25}$	Control Limits CT - $S_{A.25}$	Control Limits CT + $S_{A.25}$	CV
1	02-05-02	1.07											
2	03-19-02	1.03	1.05	0.03	0.99	1.11	0.08	0.97	1.13	0.18	0.87	1.23	0.03
3	04-09-02	1.03	1.05	0.02	1.00	1.09	0.08	0.96	1.13	0.18	0.87	1.22	0.02
4	05-07-02	1.05	1.05	0.02	1.01	1.09	0.08	0.96	1.13	0.18	0.87	1.22	0.02
5	06-04-02	1.06	1.05	0.02	1.01	1.09	0.08	0.97	1.13	0.18	0.87	1.23	0.02
6	07-09-02	1.03	1.05	0.02	1.01	1.08	0.08	0.96	1.13	0.18	0.87	1.22	0.02
7	08-06-02	1.05	1.05	0.02	1.01	1.08	0.08	0.96	1.13	0.18	0.87	1.23	0.02
8	09-04-02	1.05	1.05	0.02	1.02	1.08	0.08	0.96	1.13	0.18	0.87	1.23	0.01
9	10-08-02	1.03	1.05	0.02	1.01	1.08	0.08	0.96	1.13	0.18	0.87	1.22	0.02
10	11-05-02	1.03	1.04	0.02	1.01	1.08	0.08	0.96	1.13	0.18	0.87	1.22	0.01
11	12-03-02	1.02	1.04	0.02	1.01	1.08	0.08	0.96	1.13	0.18	0.86	1.22	0.02
12	12-03-02	1.03	1.04	0.02	1.01	1.07	0.08	0.96	1.12	0.18	0.86	1.22	0.02
13	12-04-02	1.02	1.04	0.02	1.00	1.07	0.08	0.96	1.12	0.18	0.86	1.22	0.02
14	12-06-02	1.03	1.04	0.02	1.01	1.07	0.08	0.96	1.12	0.18	0.86	1.22	0.02
15	12-11-02	1.04	1.04	0.02	1.01	1.07	0.08	0.96	1.12	0.18	0.86	1.21	0.02
16	12-18-02	1.04	1.04	0.02	1.01	1.07	0.08	0.96	1.12	0.18	0.86	1.22	0.02
17	01-07-03	0.96	1.03	0.02	0.99	1.08	0.08	0.95	1.12	0.18	0.86	1.21	0.02
18	02-04-03	0.99	1.03	0.03	0.98	1.08	0.08	0.95	1.11	0.18	0.86	1.21	0.02
19	03-05-03	1.05	1.03	0.03	0.98	1.08	0.08	0.95	1.12	0.18	0.86	1.21	0.02
20	04-08-03	1.03	1.03	0.02	0.98	1.08	0.08	0.95	1.11	0.18	0.86	1.21	0.02

Note:

7-d IC_{25} = 7-day 25% inhibition concentration. An estimation of the concentration of sodium chloride that would cause a 25% reduction in Ceriodaphnia reproduction for the test population.

CT = Central tendency (mean IC_{25}).

S = Standard deviation of the IC_{25} values.

$S_{A.10}$ = Standard deviation corresponding to the 10th percentile CV. $S_{A.10}$ = 0.08, as determined by USEPA for the method and endpoint.

$S_{A.25}$ = Standard deviation corresponding to the 25th percentile CV. $S_{A.25}$ = 0.17, as determined by the USEPA for the method and endpoint.

CV = Coefficient of variation of the IC_{25} values.

Control and warning limits were established using the standard deviation of the IC_{25} values corresponding to the 10th and 25th percentile CVs. These ranges are more stringent than the control and warning limits recommended by USEPA for the test method and endpoint.

USEPA Recommended Control and Warning Limits:

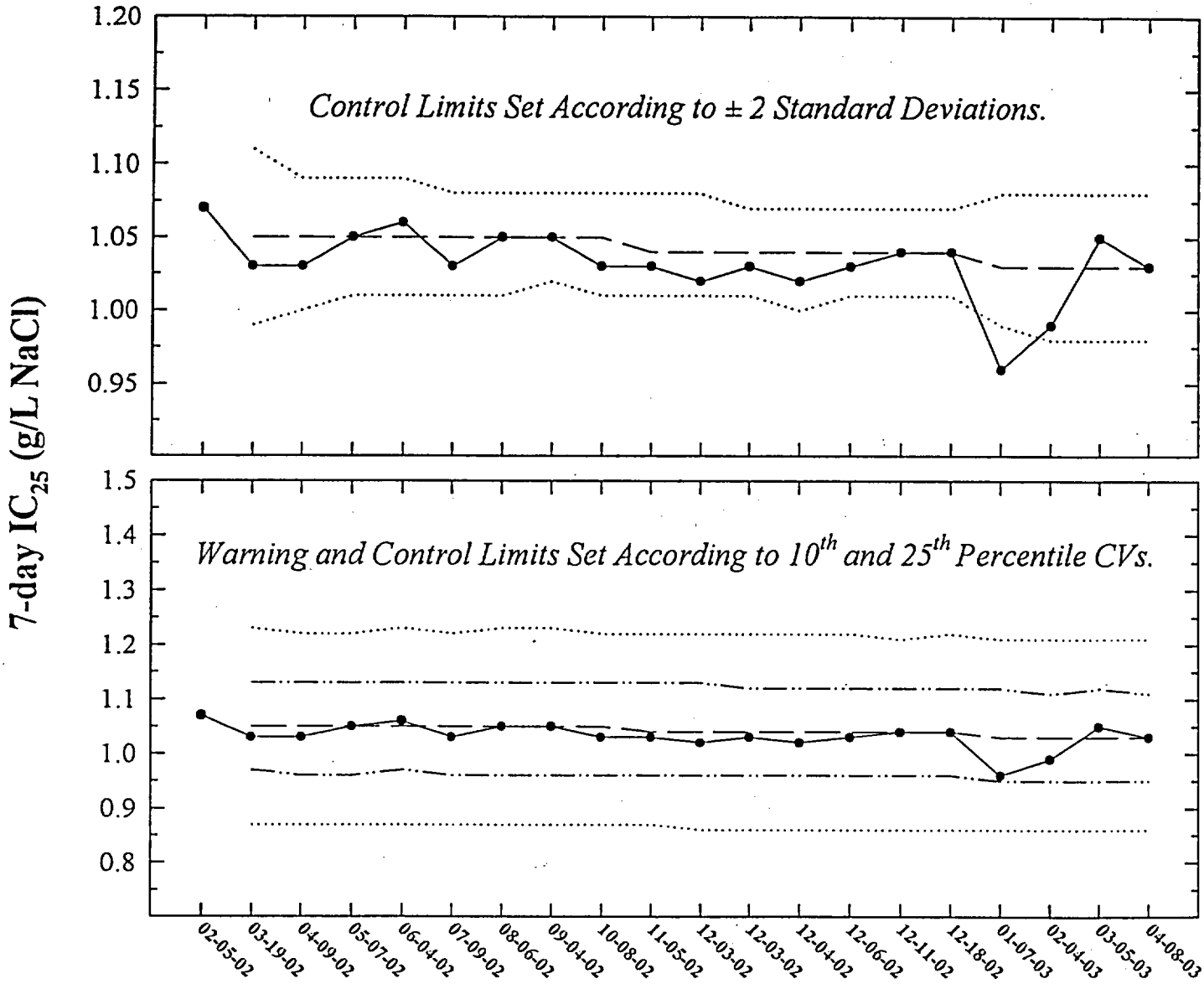
Warning Limit = Standard deviation corresponding to the 75th percentile CV. $S_{A.75}$ = 0.45.

Control Limit = Standard deviation corresponding to the 90th percentile CV. $S_{A.90}$ = 0.62.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency. Cincinnati, OH.

Environmental Testing Solutions, LLC

Sodium Chloride Chronic Reference Toxicant Control Chart for *Ceriodaphnia dubia* using Moderately Hard Synthetic Water



—•— 7-day IC_{25} = 25% inhibition concentration. An estimation of the concentration of sodium chloride that would cause a 25% reduction in *Ceriodaphnia* reproduction for the test population.

— – **Central Tendency** (mean IC_{25})

—·— **Warning Limits** (mean $IC_{25} \pm S_{A.10}$)

····· **Control Limits** (mean $IC_{25} \pm S_{A.25}$ or 2 Standard Deviations)

Environmental Testing Solutions, LLC

Precision of Endpoint Measurements

Sodium Chloride Chronic Reference Toxicant Data for *Ceriodaphnia dubia* using Moderately Hard Synthetic Water

Test number	Test date	Control Survival (%)	Control Mean Reproduction (offspring/female)	CT for Control Mean Reproduction (offspring/female)	CV (%)	CT for Control Reproduction CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	02-05-02	100	23.1		10.1		2.6	11.1	
2	03-19-02	100	26.3	24.7	8.6	9.3	3.0	11.2	11.2
3	04-09-02	100	26.2	25.2	12.3	10.3	2.8	10.7	11.0
4	05-07-02	100	27.3	25.7	8.1	9.8	2.3	8.4	10.4
5	06-04-02	100	26.0	25.8	9.9	9.8	3.8	14.7	11.2
6	07-09-02	100	29.5	26.4	8.0	9.5	3.5	11.7	11.3
7	08-06-02	100	28.4	26.7	10.4	9.6	2.7	9.5	11.0
8	09-04-02	100	31.4	27.3	6.7	9.3	3.0	9.5	10.8
9	10-08-02	100	31.1	27.7	9.2	9.3	2.9	9.4	10.7
10	11-05-02	100	29.5	27.9	8.0	9.1	2.5	8.4	10.5
11	12-03-02	90	34.0	28.4	6.2	8.9	2.7	8.0	10.2
12	12-03-02	100	33.2	28.8	6.0	8.6	3.3	9.9	10.2
13	12-04-02	100	32.5	29.1	11.0	8.8	3.2	9.8	10.2
14	12-06-02	100	29.7	29.2	13.7	9.2	3.0	10.0	10.2
15	12-11-02	100	33.8	29.5	7.4	9.0	2.9	8.5	10.1
16	12-18-02	100	30.5	29.5	7.0	8.9	2.9	9.4	10.0
17	01-07-03	100	33.2	29.7	8.1	8.9	2.9	8.6	9.9
18	02-04-03	100	32.3	29.9	5.1	8.7	2.7	8.4	9.8
19	03-05-03	100	28.7	29.8	6.2	8.5	3.5	12.1	10.0
20	04-11-03	100	26.3	29.7	6.2	8.4	2.5	9.6	9.9

Note:

CV = Coefficient of variation for control reproduction.
On average, the CV for control reproduction is 8.4% in Environmental Testing Solutions, LLC *Ceriodaphnia* chronic
Lower CV bound determined by USEPA (10^{th} percentile) = 8.9%.
Upper CV bound determined by USEPA (90^{th} percentile) = 42%

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference
PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, LLC chronic toxicity tests when a toxicant reduces Ceriodaphnia reproduction by 9.9% from the control.
Lower PMSD bound determined by USEPA (10^{th} percentile) = 11%.
Upper PMSD bound determined by USEPA (90^{th} percentile) = 37%.

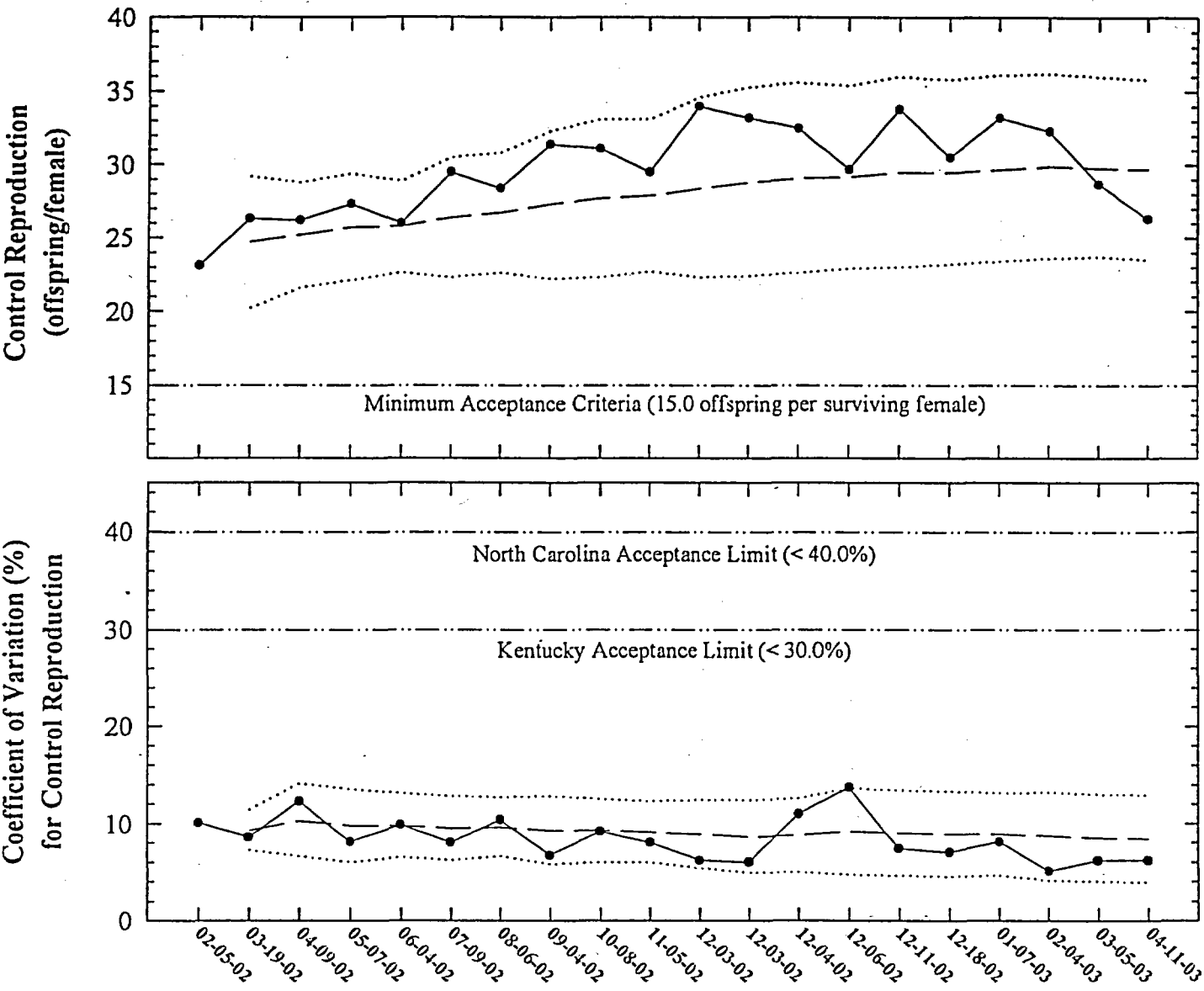
CT = Central Tendancy (Mean Control Reproduction, CV, or mean PMSD)

The lower and upper bounds were calculated by the USEPA using 393 tests conducted from 33 laboratories for Ceriodaphnia reproduction in chronic reference toxicant tests.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Environmental Testing Solutions, LLC

Ceriodaphnia dubia Control Reproduction and Coefficient of Variation in Sodium Chloride Chronic Reference Toxicant Tests



Control Reproduction or Coefficient of Variation (CV)

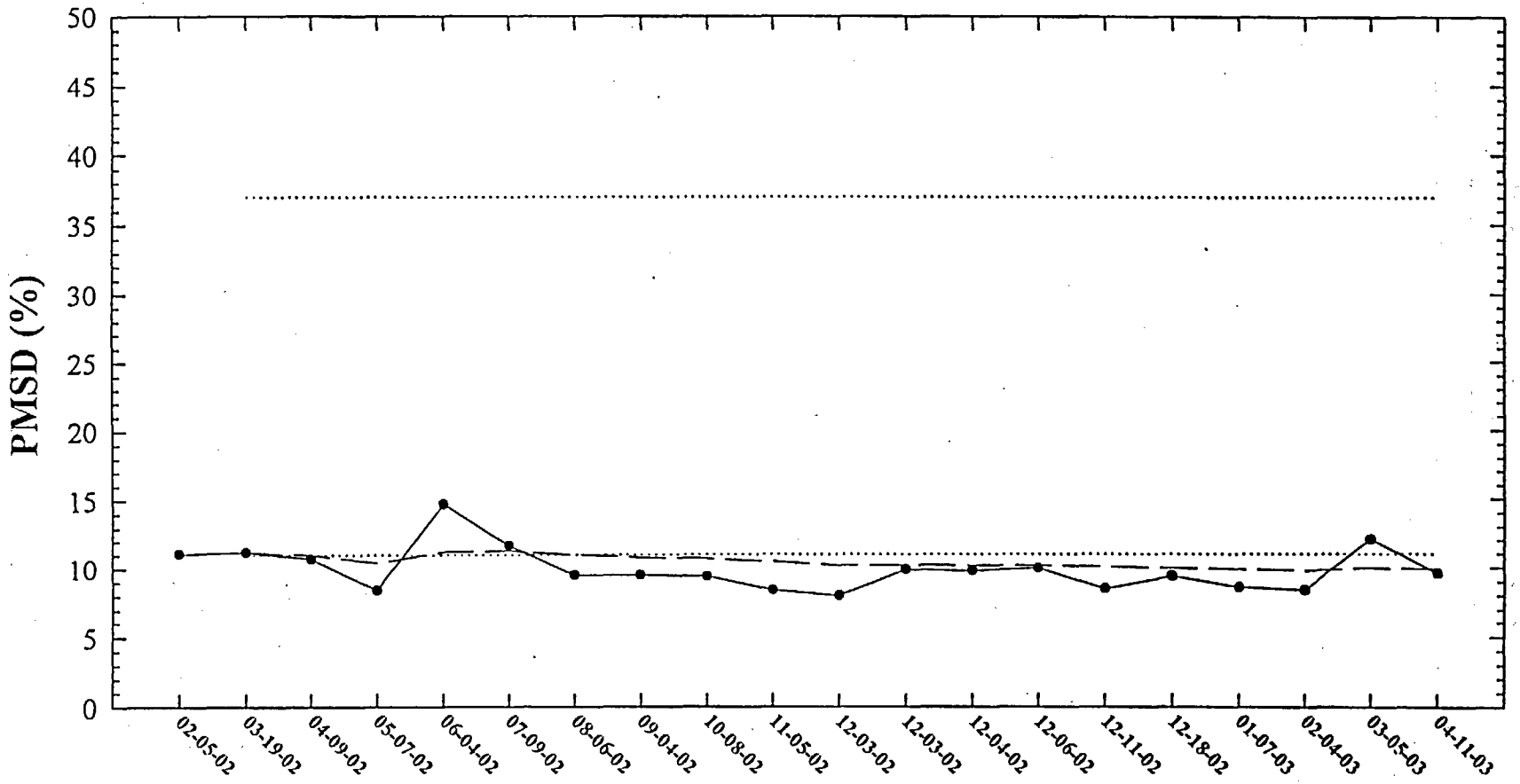
Central Tendency (mean Control Reproduction or CV)

Control Limits (mean Control Reproduction or CV ± 2 Standard Deviations)

Environmental Testing Solutions, LLC

Precision of Endpoint Measurements

Sodium Chloride Chronic Reference Toxicant Control Chart
for *Ceriodaphnia dubia*
using Moderately Hard Synthetic Water



PMSD = percent minimum significant difference. PMSD is the minimum significant difference between the control and treatment that can be declared statistically significant.

Central Tendency (mean PMSD)

Lower and Upper PMSD Bounds

Lower PMSD Bound (10^{th} percentile) = 11%, Upper PMSD Bound (90^{th} percentile) = 37%

(Lower and upper PMSD bounds were determined by USEPA for the method and endpoint.)

Sodium Chloride Chronic Reference Toxicant Test
(EPA-821-R-02-013 Method 1002.0)
Species: *Ceriodaphnia dubia*

CdNaCLCR #: 9

Dilution preparation information:						*Comments:*
NaCl CHM number:		CHM 060				
Stock preparation:		100 g NaCl/l (dissolve 50 g NaCl in 500 ml deionized water)				
Dilution prep (mg/L)	600	800	1000	1200	1400	
Stock volume (mL)	9	12	15	18	21	
Diluent volume (mL)	1491	1488	1485	1482	1479	
Total volume (mL)	1500	1500	1500	1500	1500	

Test organism information:		*Test information:*	
Organism age:	< 24-HOURS OLD	Randomizing template:	~~RED~~ GREEN
Date and times organisms were born between:	~~04-07-03~~ 04-08-03 0836 TO 1217	Incubator number and shelf location:	2 D2 / E1
Organism source:	04-01-03 A-F	YCT batch: ABS	03-25-03
Transfer bowl information:	pH = 8.11 Temperature = 24.9	*Selenastrum* batch: ABS	03-25-03

Daily renewal information:

Day	Date	Test initiation, renewal, or termination time	Control water batch used MHS	Analyst
0	04-08-03	1526	04-01-03	JH
1	04-09-03	1456	04-07-03	JH
2	04-10-03	1458	04-07-03	JH
3	04-11-03	1521	04-07-03	JH
4	04-12-03	1530	04-11-03	JH
5	04-13-03	1524	04-11-03	JH
6	04-14-03	1916	04-11-03	JH
7	04-15-03	1541	——— JH	JH

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	≤ 20%	7-day LC50	>1400
% Adults having 3rd Broods:	100%	≥ 80%	NOEC	800
% Mortality:	0%	≤ 20%	LOEC	1000
Mean Offspring/Female:	26.3	≥ 15.0 offspring/female	ChV	894.4
% CV:	6.2%	< 40.0 %	IC25	1025.5

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 9

CONTROL

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	3	4	4	3	3	3	3	5	3	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	9	10	10	7	9	11	8	10	8	8
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	2*	0	4*	0	2*	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	13	13	11	14	12	14	12	13	16	12
Total young produced		25	27	27	24	28	28	25	28	27	24
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L
X for 3rd Broods		X	X	X	X	X	X	X	X	X	X

* split brood

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	26.3

600 mg NaCl/L

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	3	5	3	3	3	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	10	13	8	10	10	10	10	8	8	9
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	2*	0	1*	0	4*	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	15	15	12	13	14	14	12	15	11	10
Total young produced		29	32	23	28	29	27	26	27	27	23
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	27.1
% Reduction from Control:	-3.0%

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 9

800 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	4	4	3	5	3	3	3	4	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	10	9	10	8	12	8	8	10	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	10	14	10	14	14	12	15	14	16	13
Total young produced		27	28	23	27	27	27	26	25	30	26
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	26.6
% Reduction from Control:	-1.17%

1000 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	4	4	3	5	3	3	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	8	8	13	10	6	10	10	7	11	8
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	2*	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	7	8	11	3	8	8	9	6	8	8
Total young produced		19	20	28	17	17	25	22	16	23	20
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

*CARRY OVER

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	20.7
% Reduction from Control:	21.3%

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 9

1200 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	3	2	2	0	4	3	3	3	2	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	7	4	9	2	6	5	10	1*	8	8
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	6	0	0	4	0	0	0	5	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	5	3	4	10	6	8	7	6	5	6
Total young produced		15	9	15	16	16	16	20	15	15	17
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

* carry over

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	15.4
% Reduction from Control:	41.4%

1400 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	0	1	0	0	2	0	0	2	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	0	0	0	0	0	2	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	1	0	0	0	1	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	0	0	0	0	0	0	0	0	0	0
Total young produced		0	1	0	1	2	0	2	3	0	0
Final Adult Mortality		L	L	D	L	L	L	L	L	L	L

Concentration:	
% Mortality:	10%
Mean Offspring/Female:	0.9
% Reduction from Control:	96.6%

Environmental Testing Solutions, LLC

Verification of *Ceriodaphnia* Reproduction Totals

Control

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	3	4	4	3	3	3	3	5	3	4	35
5	9	10	10	7	9	11	8	10	8	8	90
6	0	0	2	0	4	0	2	0	0	0	8
7	13	13	11	14	12	14	12	13	16	12	130
Total	25	27	27	24	28	28	25	28	27	24	263

1000 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	4	4	3	5	3	3	4	4	38
5	8	8	13	10	6	10	10	7	11	8	91
6	0	0	0	0	0	2	0	0	0	0	2
7	7	8	11	3	8	8	9	6	8	8	76
Total	19	20	28	17	17	25	22	16	23	20	207

600 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	3	5	3	3	3	4	4	4	37
5	10	13	8	10	10	10	10	8	8	9	96
6	0	0	0	0	2	0	1	0	4	0	7
7	15	15	12	13	14	14	12	15	11	10	131
Total	29	32	23	28	29	27	26	27	27	23	271

1200 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	3	2	2	0	4	3	3	3	2	3	25
5	7	4	9	2	6	5	10	1	8	8	60
6	0	0	0	4	0	0	0	5	0	0	9
7	5	3	4	10	6	8	7	6	5	6	60
Total	15	9	15	16	16	16	20	15	15	17	154

800 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	4	4	3	5	3	3	3	4	3	37
5	12	10	9	10	8	12	8	8	10	10	97
6	0	0	0	0	0	0	0	0	0	0	0
7	10	14	10	14	14	12	15	14	16	13	132
Total	27	28	23	27	27	27	26	25	30	26	266

1400 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	0	1	0	0	2	0	0	2	0	0	5
5	0	0	0	0	0	0	2	0	0	0	2
6	0	0	0	1	0	0	0	1	0	0	2
7	0	0	0	0	0	0	0	0	0	0	0
Total	0	1	0	1	2	0	2	3	0	0	9

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1002.0)
Species: Ceriodaphnia dubia

Quality Control
Verification of Data Entry, Calculations, and Statistical Analyses

Test number: CdNaClCR #38

Test dates: April 8-15, 2003

Reveiwed by:

Concentration (mg/L NaCl)	Replicate number										Survival (%)	Average reproduction (offspring/female)	Coefficient of variation (%)	Percent reduction from control (%)
	1	2	3	4	5	6	7	8	9	10				
Control	25	27	27	24	28	28	25	28	27	24	100	26.3	6.2	Not applicable
600	29	32	23	28	29	27	26	27	27	23	100	27.1	10.1	-3.0
800	27	28	23	27	27	27	26	25	30	26	100	26.6	6.9	-1.1
1000	19	20	28	17	17	25	22	16	23	20	100	20.7	18.5	21.3
1200	15	9	15	16	16	16	20	15	15	17	100	15.4	17.6	41.4
1400	0	1	0	1	2	0	2	3	0	0	90	0.9	122.3	96.6

Dunnett's MSD value: 2.531

PMSD: 9.6

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, LLC chronic toxicity tests when a toxicant reduces Ceriodaphnia reproduction by 9.9% from the control.

Lower PMSD bound determined by USEPA (10^{th} percentile) = 11%.

Upper PMSD bound determined by USEPA (90^{th} percentile) = 37%.

The lower and upper bounds were calculated by the USEPA using 393 tests conducted from 33 laboratories for *Ceriodaphnia* reproduction in chronic reference toxicant tests.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Environmental Testing Solutions, LLC

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	4/8/03	Test ID:	CdNaClCR	Sample ID:	REF-Ref Toxicant
End Date:	4/15/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	NACL-Sodium chloride
Sample Date		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	CD-Ceriodaphnia dubia
Comments:					

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	25.000	27.000	27.000	24.000	28.000	28.000	25.000	28.000	27.000	24.000
600	29.000	32.000	23.000	28.000	29.000	27.000	26.000	27.000	27.000	23.000
800	27.000	28.000	23.000	27.000	27.000	27.000	26.000	25.000	30.000	26.000
1000	19.000	20.000	28.000	17.000	17.000	25.000	22.000	16.000	23.000	20.000
1200	15.000	9.000	15.000	16.000	16.000	16.000	20.000	15.000	15.000	17.000
1400	0.000	1.000	0.000	1.000	2.000	0.000	2.000	3.000	0.000	0.000

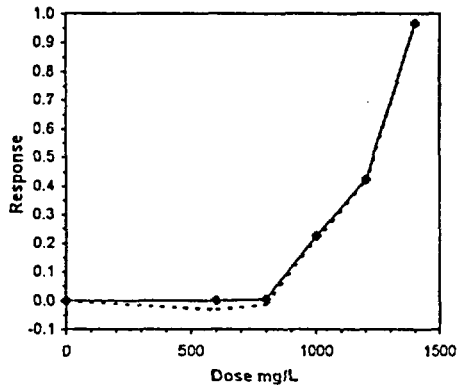
			Transform: Untransformed					Rank	1-Tailed	Isotonic	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	Sum	Critical	Mean	N-Mean
D-Control	26.300	1.0000	26.300	24.000	28.000	6.222	10			26.700	1.0000
600	27.100	1.0304	27.100	23.000	32.000	10.061	10	114.00	75.00	26.700	1.0000
800	26.600	1.0114	26.600	23.000	30.000	6.909	10	106.50	75.00	26.600	0.9963
*1000	20.700	0.7871	20.700	16.000	28.000	18.508	10	66.50	75.00	20.700	0.7753
*1200	15.400	0.5856	15.400	9.000	20.000	17.638	10	55.00	75.00	15.400	0.5768
*1400	0.900	0.0342	0.900	0.000	3.000	122.278	10	55.00	75.00	0.900	0.0337

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.93054914	1.035	0.12044696	1.50413886
Bartlett's Test indicates unequal variances (p = 8.61E-03)	15.4483681	15.0863171		

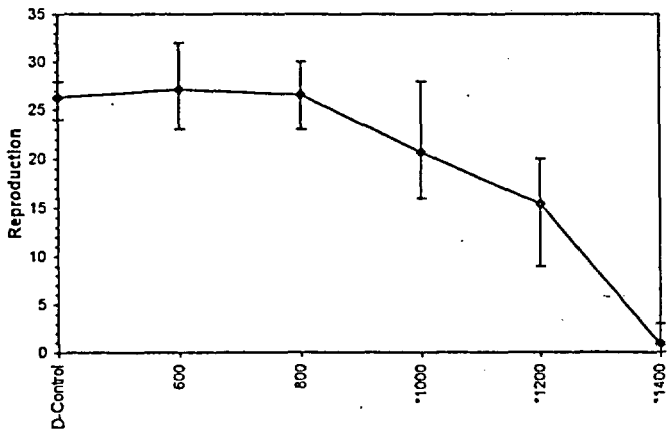
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	800	1000	894.427191	
Treatments vs D-Control				

Linear Interpolation (200 Resamples)

Point	mg/L	SD	95% CL		Skew
IC05	841.864407	20.8144632	785.890323	866.119912	-1.6817
IC10	887.118644	23.514141	841.856278	932.239823	0.4223
IC15	932.372881	30.6481604	882.569481	999.079893	0.5599
IC20	977.627119	33.5004029	917.15615	1039.27631	0.1628
IC25	1025.4717	34.9445136	952.861201	1078.60714	-0.1248
IC40	1176.60377	27.994347	1110.10498	1209.16801	-0.5026
IC50	1228.27586	11.9688245	1198.26565	1242.40026	-0.9747



Dose-Response Plot



Environmental Testing Solutions, LLC

Statistical Analyses

Used for PMSD calculation only.			Ceriodaphnia Survival and Reproduction Test-Reproduction	
Start Date:	4/8/03	Test ID:	CdNaClCR	Sample ID:	REF-Ref Toxicant
End Date:	4/15/03	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	NACL-Sodium chloride
Sample Date:		Protocol:	CHRONIC-(EPA-821-R-02-013)	Test Species:	CD-Ceriodaphnia dubia
Comments:					

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	25.000	27.000	27.000	24.000	28.000	28.000	25.000	28.000	27.000	24.000
600	29.000	32.000	23.000	28.000	29.000	27.000	26.000	27.000	27.000	23.000
800	27.000	28.000	23.000	27.000	27.000	27.000	26.000	25.000	30.000	26.000
1000	19.000	20.000	28.000	17.000	17.000	25.000	22.000	16.000	23.000	20.000
1200	15.000	9.000	15.000	16.000	16.000	16.000	20.000	15.000	15.000	17.000
1400	0.000	1.000	0.000	1.000	2.000	0.000	2.000	3.000	0.000	0.000

			Transform: Untransformed						1-Tailed	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	26.300	1.0000	26.300	24.000	28.000	6.222	10			
600	27.100	1.0304	27.100	23.000	32.000	10.061	10	-0.723	2.287	2.531
800	26.600	1.0114	26.600	23.000	30.000	6.909	10	-0.271	2.287	2.531
*1000	20.700	0.7871	20.700	16.000	28.000	18.508	10	5.059	2.287	2.531
*1200	15.400	0.5856	15.400	9.000	20.000	17.638	10	9.847	2.287	2.531
*1400	0.900	0.0342	0.900	0.000	3.000	122.278	10	22.947	2.287	2.531

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.93054914	1.035	0.12044696	1.50413886
Bartlett's Test indicates unequal variances (p = 8.61E-03)	15.4483681	15.0863171		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test Treatments vs D-Control	800	1000	894.427191		2.53106747	0.09623831	1037.24	6.12592593	1.0E-31	5, 54

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 9

Daily Chemistry:

Concentration	Parameter	Day 0		Day 1		Day 2	
CONTROL	pH (S.U.)	8.01	8.12	8.01	8.14	7.98	8.08
	DO (mg/L)	7.7	7.6	7.5	8.1	7.8	8.1
	Conductivity (µmhos/cm)	303		288		296	
	Alkalinity (mg $CaCO_3$/L)	62					
	Hardness (mg $CaCO_3$/L)	82					
	Temperature (°C)	25.3	24.5	25.5	24.4	25.4	24.2
600 mg NaCl/L	pH (S.U.)	8.09	8.04	8.08	8.12	8.06	8.03
	DO (mg/L)	7.8	7.5	7.6	8.0	8.0	8.1
	Conductivity (µmhos/cm)	1477		1462		1476	
	Temperature (°C)	25.3	24.5	25.5	24.4	25.4	24.2
800 mg NaCl/L	pH (S.U.)	8.07	8.02	8.06	8.12	8.04	8.00
	DO (mg/L)	7.8	7.5	7.6	8.1	8.0	8.0
	Conductivity (µmhos/cm)	1784		1775		1790	
	Temperature (°C)	25.3	24.5	25.5	24.4	25.4	24.2
1000 mg NaCl/L	pH (S.U.)	8.05	8.02	8.06	8.10	8.04	8.01
	DO (mg/L)	7.8	7.5	7.6	8.2	8.2	8.0
	Conductivity (µmhos/cm)	2057		2051		2059	
	Temperature (°C)	25.3	24.5	25.5	24.4	25.4	24.2
1200 mg NaCl/L	pH (S.U.)	8.04	8.02	8.04	8.11	8.04	8.00
	DO (mg/L)	7.9	7.5	7.7	8.2	8.2	8.1
	Conductivity (µmhos/cm)	2482		2472		2492	
	Temperature (°C)	25.3	24.5	25.5	24.4	25.4	24.2
1400 mg NaCl/L	pH (S.U.)	8.03	8.00	8.04	8.13	8.04	7.98
	DO (mg/L)	7.8	7.6	7.7	8.3	8.2	8.2
	Conductivity (µmhos/cm)	2925		2894		2915	
	Temperature (°C)	25.3	24.5	25.5	24.4	25.4	24.2
		Initial	Final	Initial	Final	Initial	Final

Stock 07600

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 9

Concentration	Parameter	Day 3		4		5		6	
CONTROL	pH (S.U.)	8.12	8.03	8.05	8.28	7.99	7.98	7.95	7.95
	DO (mg/L)	7.7	7.9	8.0	8.4	8.1	8.4	8.1	7.6
	Conductivity (µmhos/cm)	304		307		298		293	
	Alkalinity (mg $CaCO_3$/L)			64					
	Hardness (mg $CaCO_3$/L)			88					
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.6	25.4	24.6
600 mg NaCl/L	pH (S.U.)	8.06	8.02	8.09	8.23	8.04	7.95	8.02	7.98
	DO (mg/L)	8.0	7.9	8.2	8.4	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	1509		1481		1491		1494	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.6	25.4	24.6
300 mg NaCl/L	pH (S.U.)	8.05	8.03	8.11	8.24	8.02	7.94	7.99	7.96
	DO (mg/L)	8.3	8.0	8.2	8.3	8.3	8.4	8.4	7.7
	Conductivity (µmhos/cm)	1812		1828		1820		1841	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
000 mg NaCl/L	pH (S.U.)	8.04	8.02	8.10	8.21	8.02	7.93	7.98	7.98
	DO (mg/L)	8.3	8.1	8.2	8.3	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	2194		2205		2194		2234	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
200 mg NaCl/L	pH (S.U.)	8.03	8.02	8.10	8.25	8.02	7.93	7.98	7.96
	DO (mg/L)	8.3	8.1	8.2	8.3	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	2524		2517		2513		2560	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
400 mg NaCl/L	pH (S.U.)	8.03	8.02	8.08	8.24	8.03	7.97	7.97	7.95
	DO (mg/L)	8.3	8.0	8.2	8.3	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	2987		2912		2905		2968	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

87500

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 9

Concentration	Parameter	Day 3		4		5		6	
CONTROL	pH (S.U.)	8.12	8.03	8.05	8.28	7.99	7.98	7.95	7.95
	DO (mg/L)	7.7	7.9	8.0	8.4	8.1	8.4	8.1	7.6
	Conductivity (µmhos/cm)	304		307		298		293	
	Alkalinity (mg $CaCO_3$/L)			64					
	Hardness (mg $CaCO_3$/L)			88					
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.6	25.4	24.6
600 mg NaCl/L	pH (S.U.)	8.06	8.02	8.09	8.23	8.04	7.95	8.02	7.98
	DO (mg/L)	8.0	7.9	8.2	8.4	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	1509		1481		1491		1494	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.6	25.4	24.6
800 mg NaCl/L	pH (S.U.)	8.05	8.03	8.11	8.24	8.02	7.94	7.99	7.96
	DO (mg/L)	8.3	8.0	8.2	8.3	8.3	8.4	8.4	7.7
	Conductivity (µmhos/cm)	1812		1828		1820		1841	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
1000 mg NaCl/L	pH (S.U.)	8.04	8.02	8.10	8.21	8.02	7.93	7.98	7.98
	DO (mg/L)	8.3	8.1	8.2	8.3	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	2194		2205		2194		2234	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
200 mg NaCl/L	pH (S.U.)	8.03	8.02	8.10	8.25	8.02	7.93	7.98	7.96
	DO (mg/L)	8.3	8.1	8.2	8.3	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	2524		2517		2513		2560	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
400 mg NaCl/L	pH (S.U.)	8.03	8.02	8.08	8.24	8.03	7.97	7.97	7.95
	DO (mg/L)	8.3	8.0	8.2	8.3	8.3	8.4	8.4	7.8
	Conductivity (µmhos/cm)	2987		2912		2905		2968	
	Temperature (°C)	25.4	24.4	25.7	24.8	25.6	24.5	25.4	24.6
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

87500

TENNESSEE VALLEY AUTHORITY
TOXICITY TEST REPORT

INTRODUCTION/EXECUTIVE SUMMARY

1) Facility/Discharger: Watts Bar Nuclear Plant/TVA Report Date: May 13, 2003

2) County/State: Rhea/Tennessee 3) NPDES Permit #: TN0020168

4) Type of Facility: Nuclear-fueled electric generating plant

5) Average Flow (MGD): Flow will be determined at a later date.

6) Receiving Stream: TRM 527.9

7) 1Q20 (MGD): 2062

8) Outfall(s) Tested: 113 9) Dates Sampled: April 14-19, 2003

10) Flow on day(s) sampled (MGD): 183.02 182.25 182.91 182.16 182.98 184.04

Pertinent site conditions:

Watts Bar Hydro and WBN DSN 113 operations during this study are summarized in Appendix B. Dilution factors ranged from 59 to 60. There were no periods of zero release.

During the toxicity testing, the daily periodic bromine/chlorine treatment of WBN cooling water was in service.

12) Test Dates: April 15-22, 2003 13) Test Type: Short-term Chronic-definitive

14) Test Species: #1 Fathead Minnows (*Pimephales promelas*)
#2 Daphnids (*Ceriodaphnia dubia*)

15) Concentrations Tested (%): Outfall 113 2.58 5.20 10.3 20.6 41.2
Raw Water Intake 100

16) Permit Limit Endpoint (%): IC_{25} = 10.3%

17) Test Results (%): *Pimephales promelas*: IC_{25} >41.2%
Ceriodaphnia dubia : IC_{25} >41.2%

18) Facility Contact: Martha I. Ervin 19) Phone #: (423) 365-3585

20) Testing Lab Name: S&ME, Inc.

21) Lab Contact: Leira Douthat 22) Phone #: (865) 970-0003
TVA Contact: Cynthia L. Russell Phone #: (256) 386-2755

23) Notes: Exposure of test organisms to samples from Outfall 113 resulted in no toxicity to *Pimephales promelas* or *Ceriodaphnia dubia*. Fathead minnows exposed to intake samples were not significantly different from control for survival or growth based on Homoscedastic t-Test. There was no significant difference between control and intake samples for daphnid survival or reproduction based on Homoscedastic t-Test.

METHODS SUMMARY

Samples

1) Sampling Point: Outfall 113; Intake

2) Sample Type: Outfall 113- Composite; Intake-Grab

3) Sample Information:

ID	Date/Time Collected (MM-DD/Time)	Date/Time Received (MM-DD/Time)	Arrival Temp. (°C)	Initial TRC* (mg/L)	Date/Time Used By (MM-DD/Time)
113	04-14/1047 to 04-15/0947	04-15/1230	5.7	0.07	04-15/1345 04-16/1310
Intake	04-15/0750	04-15/1230	1.2	0.08	04-15/1345 04-16/1310
113	04-16/0825 to 04-17/0725	04-17/1050	1.9	0.09	04-17/1405 04-18/1430
Intake	04-17/0800	04-17/1050	3.2	0.06	04-17/1405 04-18/1430
113	04-18/1425 to 04-19/1325	04-19/1455	4.9	0.05	04-20/1340 04-21/1250 04-22/1225
Intake	04-19/1240	04-19/1455	5.7	0.04	04-20/1340 04-21/1250 04-22/1225

*Total residual chlorine measurements likely reflect interference with method.
NOTE: All times listed are Eastern Daylight Savings Time.

4) Sample manipulation: Samples were warmed to test temperature (25°C) in a warm water bath

Test Organisms

	Pimephales promelas	*Ceriodaphnia dubia*
1) Source:	In-house culture	In-house culture
2) Age:	< 24 hours	< 24 hours

Test Method Summary (See Appendix A for additional information)

	Pimephales promelas	*Ceriodaphnia dubia*
1) Test Conditions	Static, renewal	Static, renewal
2) Test Duration	7 days	Until at least 60% of control females have 3 broods
3) Dilution/Control Water	Moderately Hard Reconstituted Water	20% Dilute Mineral Water with trace minerals
4) Number Replicates	4	10
5) Animals per Replicate	10	1
6) Test Initiation (101)	4-15-03/1300 EDT	4-15-03/1345 EDT
7) Test Termination (101)	4-22-03/1300 EDT	4-21-03/1300 EDT
8) Test Temperature (101)	Mean = 25.4°C (24.2°C-26.0°C)	Mean = 25.2°C (24.3°C-25.9°C)

9) Physical/Chemical Measurements: Hardness, alkalinity, total residual chlorine, and conductivity were measured at the laboratory in each 100 percent sample or highest concentration tested. Daily temperatures were measured in one replicate in each test concentration. Pre- and post-exposure test solutions were analyzed daily for pH and dissolved oxygen.

10) Statistics: Statistics were performed according to methods prescribed by EPA using ToxCalc version 5.0 statistical software (Tidepool Scientific Software, McKinneyville, CA).

Results of a *Pimephales promelas* Larval Survival and Growth Toxicity Test
(Genus) (Species) (Type/Duration)

Conducted 4/15/03 - 4/22/03 Using Effluent From: Outfall 113
(mm/dd/yy) (mm/dd/yy) (number)

Test Solutions	Percent Surviving (time intervals used - days)						
	1	2	3	4*	5	6	7
Control	100	98	98	95	95	95	95
2.58% Effluent	100	95	95	90	90	90	90
5.2% Effluent	100	100	100	98	98	95	95
10.3% Effluent	98	98	98	93	90	88	88
20.6% Effluent	100	100	100	98	95	95	95
41.2% Effluent	100	100	100	93	93	93	93
Intake	100	98	98	90	88	83	78

*96-Hour LC_{50} Value: ≥ 41.2%	Calculated TU Estimate: ≤2.43 TUa Permit Limit: N/A

Test Solutions	IC_{25} Mean Dry Weight (mg) (Replicate)				
	1	2	3	4	Mean
Control	0.452	0.346	0.386	0.373	0.389
2.58% Effluent	0.451	0.286	0.437	0.325	0.375
5.2% Effluent	0.347	0.327	0.532	0.464	0.418
10.3% Effluent	0.277	0.290	0.231	0.460	0.315
20.6% Effluent	0.295	0.389	0.259	0.364	0.327
41.2% Effluent	0.308	0.333	0.376	0.191	0.302
Intake	0.135	0.299	0.360	0.351	0.286

IC_{25} Value: ≥41.2% Permit Limit: 10.3%	Calculated TU Estimates† :≤2.4 TUc Permit Limit: 9.7 TUc

†NOTE: TUa = $100/LC_{50}$; TUc = $100/IC_{25}$

2) Results of a *Ceriodaphnia* *dubia* Chronic/ 7-day (3-brood) Toxicity Test
(Genus) (Species) (Type/Duration)

Conducted 4/15/03 - 4/22/03 Using Effluent From: Outfall 113
(mm/dd/yy) (mm/dd/yy) (number)

Test Solutions	Percent Surviving (time intervals used – days)						
	1	2	3	4*	5	6	7
Control	100	100	100	100	100	100	-
2.58% Effluent	100	100	100	100	100	100	-
5.20% Effluent	100	100	100	100	100	100	-
10.3% Effluent	100	100	100	100	100	100	-
20.6% Effluent	100	100	100	100	100	100	-
41.2% Effluent	100	100	100	100	100	100	-
Intake	100	100	100	100	100	100	-

*96-Hour LC_{50} Value: > 41.2%	Calculated TU Estimate: <2.43 TUa Permit Limit: N/A

Test Solutions	Reproduction (#young/female/6 days) Data Replicates										
	1	2	3	4	5	6	7	8	9	10	Mean
Control	31	25	29	31	28	32	25	29	27	23	28.0
2.58% Effluent	35	28	32	28	24	29	28	29	27	29	28.9
5.20% Effluent	28	25	31	20	25	25	27	29	26	28	26.4
10.3% Effluent	29	23	29	25	27	30	26	29	20	25	26.3
20.6% Effluent	26	28	28	28	30	27	27	22	23	29	26.8
41.2% Effluent	30	31	22	20	23	24	26	21	26	28	25.1
Intake	18	22	19	34	26	24	27	34	26	32	26.2

IC_{25} Value: >41.2% Permit Limit: 10.3%	Calculated TU Estimates[†] :<2.4 TUc Permit Limit: 9.7 TUc

[†]NOTE: TUa = $100/LC_{50}$; TUc = $100/IC_{25}$

REFERENCE TOXICANT TEST RESULTS (See Appendixes A and D)

Species	Date	Time	Duration	Toxicant	Results (LC_{50}/IC_{25})
Pimephales promelas	4-08-03	1300	7 days	KCl	801.69 mg/L (IC_{25})
Ceriodaphnia dubia	4-08-03	1345	7 days	NaCl	1106.64 mg/L (IC_{25})

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for Fathead Minnow and Daphnid Tests, Watts Bar Nuclear Plant (WBN), Outfall 113, April 15-22, 2003

Test/ Sample ID	Temperature		Dissolved Oxygen		pH		Conductance	Alkalinity	Hardness	Total Residual Chlorine
	Initial	Final	Initial	Final	Initial	Final	Initial			
	(°C)	(°C)	(mg/L)	(mg/L)	S.U.	S.U.	(µmhos)	mg/L $CaCO_3$	mg/L $CaCO_3$	(mg/L)
Fathead/ Control	25.2 (24.3-25.8)	25.3 (24.2-25.7)	6.8 (6.7-7.0)	5.8 (5.1-6.5)	8.0 (7.9-8.2)	7.6 (7.4-7.7)	395 (388-403)	62.8 (60-64)	93.2 (80-98)	-
Fathead/ 2.58%	25.3 (24.5-25.8)	25.4 (24.9-25.8)	6.7 (6.6-6.9)	5.7 (4.9-6.3)	8.1 (8.0-8.2)	7.6 (7.3-7.7)	388 (378-398)	-	-	-
Fathead/ 5.2%	25.3 (24.5-25.8)	25.5 (25.0-25.9)	6.8 (6.6-7.0)	5.5 (5.1-6.2)	8.1 (8.0-8.2)	7.6 (7.4-7.8)	382 (372-392)	-	-	-
Fathead/ 10.3%	25.3 (24.7-25.8)	25.5 (24.9-25.9)	6.7 (6.6-6.9)	5.6 (5.4-6.2)	8.1 (8.0-8.1)	7.7 (7.4-7.9)	374 (360-392)	-	-	-
Fathead/ 20.6%	25.3 (24.7-25.8)	25.5 (25.1-25.9)	6.7 (6.6-6.9)	5.7 (5.3-6.4)	8.1 (8.0-8.2)	7.7 (7.6-7.9)	356 (342-383)	-	-	-
Fathead/ 41.2%	25.4 (24.8-25.9)	25.6 (25.3-25.9)	6.8 (6.7-6.9)	5.6 (5.1-6.5)	8.1 (8.0-8.1)	7.7 (7.5-7.9)	318 (295-360)	-	-	-
Fathead/ 100%	25.1 (24.8-25.7)	-	7.3 (6.8-7.8)	-	8.1 (8.0-8.1)	-	165 (160-173)	62.7 (60-67)	88 (80-108)	0.07 (0.05-0.09)
Fathead/ Intake	25.3 (24.8-25.8)	25.7 (25.3-26.0)	8.1 (7.2-9.0)	5.8 (5.1-6.6)	8.3 (8.0-8.5)	7.8 (7.5-8.1)	154 (144-161)	60 (58-61)	82.7 (76-90)	0.06 (0.04-0.08)
Daphnid/ Control	25.0 (24.4-25.6)	25.0 (24.3-25.6)	6.7 (6.2-6.9)	6.2 (5.7-6.4)	8.1 (8.0-8.1)	8.2 (8.0-8.4)	168 (165-172)	59.7 (58-62)	80.7 (80-82)	-
Daphnid/ 2.58%	25.4 (24.5-25.7)	25.0 (24.3-25.4)	6.6 (6.2-6.8)	6.1 (5.6-6.5)	8.1 (8.0-8.2)	8.2 (8.1-8.4)	168 (162-171)	-	-	-
Daphnid/ 5.2%	25.4 (24.7-25.9)	25.0 (24.3-25.2)	6.6 (6.2-6.9)	6.1 (5.6-6.6)	8.1 (8.0-8.1)	8.3 (8.1-8.4)	168 (165-169)	-	-	-
Daphnid/ 10.3%	25.5 (24.8-25.9)	25.0 (24.4-25.4)	6.6 (6.2-6.9)	6.2 (5.6-6.6)	8.1 (8.0-8.2)	8.4 (8.1-8.6)	168 (165-169)	-	-	-
Daphnid/ 20.6%	25.4 (24.8-25.9)	25.0 (24.4-25.5)	6.6 (6.2-7.0)	6.1 (5.3-6.6)	8.1 (8.0-8.2)	8.4 (8.0-8.6)	167 (165-168)	-	-	-
Daphnid/ 41.2%	25.3 (24.4-25.8)	25.0 (24.4-25.4)	6.7 (6.3-7.0)	6.2 (5.6-6.6)	8.1 (8.0-8.2)	8.4 (8.1-8.5)	166 (164-169)	-	-	-
Daphnid/ 100%	25.3 (24.9-25.7)	-	7.4 (7.0-7.9)	-	8.1 (8.0-8.1)	-	164 (160-168)	62.7 (60-67)	88 (80-108)	0.07 (0.05-0.09)
Daphnid/ Intake	25.4 (24.8-25.8)	25.1 (24.4-25.8)	8.2 (7.4-9.0)	6.2 (5.7-6.6)	8.2 (8.0-8.5)	8.3 (8.1-8.4)	157 (154-162)	60 (58-61)	82.7 (76-90)	0.06 (0.04-0.08)

SUMMARY/CONCLUSIONS

Exposure of test organisms to samples from Outfall 113 resulted in no toxicity to *Pimephales promelas* or *Ceriodaphnia dubia*. Fathead minnows exposed to intake samples were not significantly different from control for survival or growth based on Homoscedastic t-Test. There was no significant difference between control and intake samples for daphnid survival or reproduction based on Homoscedastic t-Test.

Appendix A

ADDITIONAL TOXICITY TEST INFORMATION

SUMMARY OF METHODS

Fathead minnow tests were conducted according to EPA/821/R/02/013 using four replicates, each containing ten test organisms, per treatment. Test vessels consisted of 500-mL polystyrene tumblers, each containing 250 mL of test solution. [2]

C. dubia tests were conducted according to EPA/821/R/02/013 using ten replicates, each containing one test organism. Test vessels consisted of 30-mL polystyrene containers, each containing 15 mL of test solution.

DEVIATIONS/MODIFICATIONS TO TEST PROTOCOL

1) *Pimephales promelas*

None

2) *Ceriodaphnia dubia*

None

DEVIATIONS/MODIFICATIONS TO PRETEST CULTURE OR HOLDING OF TEST ORGANISMS

1) *Pimephales promelas*

None

2) *Ceriodaphnia dubia*

None

PHYSICAL AND CHEMICAL METHODS

1) Reagents, Titrants, Buffers, etc.: All chemicals were certified products used before expiration dates (where applicable).

2) Instruments: All identification, service, and calibration information pertaining to S&ME laboratory instruments is recorded in calibration and maintenance log books.

3) Temperature was measured using a VWR Scientific digital NIST-traceable thermometer according to S&ME SOP. [3]

4) Dissolved oxygen was measured using a VWR Scientific Dissolved oxygen meter. The instrument was calibrated and readings were made according to EPA Method 120.1.

5) The pH was measured using a VWR Scientific Symphony pH meter equipped with an Orion combination electrode. The instrument was calibrated and readings were made according to EPA Method 150.1.

6) Conductance was measured using an Orion conductivity meter. The instrument was calibrated and readings were taken according to manufacturers instructions.

7) Alkalinity was measured using Standard Methods Titration Method 2320 B using 0.01 N HCl to an endpoint pH of 4.5.

8) Hardness was measured using Standard Methods EDTA Titrimetric Method 2340 C or EPA Method 130.2.

9) Total residual chlorine was determined using the DPD Ferrous Titrimetric Method with a Hach Colorimetric kit.

QUALITY ASSURANCE

Toxicity Test Methods: All phases of the study including, but not limited to, sample collection, handling and storage; glassware preparation; test organism culturing/acquisition and acclimation; test organism handling during test; and maintaining appropriate test conditions were conducted according to the protocol as described in this report and EPA/821/R/02/013. [2] Any known deviations were noted during the study and are reported herein.

REFERENCE TOXICANT TESTS (See Appendix D for control chart information)

Test Type: 7-day chronic tests with results expressed as IC_{25} values in mg NaCl/L for *Ceriodaphnia dubia* or KCL/L for *Pimephales promelas*.

2) Standard Toxicant: Sodium Chloride (NaCl crystalline) and Potassium Chloride (KCl crystalline)

3) Dilution Water Used: 20% Dilute Mineral Water with trace metals for *Ceriodaphnia dubia* and Moderately Hard Water for *Pimephales promelas*.

4) Statistics: IC_{25} – calculated by ToxCalc version 5.0 statistical software using EPA-recommended methods.

REFERENCES

1) NPDES Permit No. TN0020168.

2) US. Environmental Protection Agency, Office of Water. Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, EPA/821/R/02/013 (October 2002).

3) S&ME Standard Operating Procedures. May 2002

Watts Bar Nuclear Plant Biomonitoring
April 15 - 22, 2003

Appendix B

Pertinent Site Data

Calculated Dilution Based Watts Bar Nuclear Plant (WBN) and
Watts Bar (WB) Hydro Plant Releases, April 14-19, 2003

Date	Zero Release (Hours)	WB Hydro Hourly Average Flow (MGD)	WBN DSN113 Daily Flow (MGD)	Dilution (WB Hydro/WBN)	TRO (mg/L) Measured at the time of samples collection
April 14, 2003	0	10923	183.02	60	<0.02
April 15, 2003	0	10923	182.25	60	
April 16, 2003	0	10923	182.91	60	<0.02
April 17, 2003	0	10923	185.16	59	
April 18, 2003	0	10923	182.98	60	0.02
April 19, 2003	0	10923	184.04	59	

Watts Bar Nuclear Plant Biomonitoring
April 15 - 22, 2003

Appendix C

Chain of Custody Records
Toxicity Test Bench Sheets
Statistical Analyses

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: WBNP BIOTOXICITY

P.O. Number: N/A

Facility Sampled: Watts Bar Nuclear Plant

NPDES Number: TN0020168

Collected By: Martha Ervin

S & ME, Inc.
1413 Topside Road
Louisville, TN 37777

Phone: 865-970-0003

Fax: 865-970-0004

Delivered By (Circle One):
FedEx UPS Bus (Client)
Other (specify):

General Comments: Contact WBNP personnel at (423) 365-3575 if any problems are encountered. Pager number 1-800-323-4853 then number to be dialed.

Samples arrived on ice

Field Identification / Sample Description	Grab/ Comp.	Ship. Temp. (°C)	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Laboratory Use				
			Date	Time			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
~~WBN Intake 101~~	~~Grab~~				~~101- , 1gal~~										
WBN Intake 113	Grab		4/15/03	07:50	113- , 1gal				X		03-090	1.2°C	SAB	1230	clear
~~WBN OSN 101~~	~~Comp~~				~~101- , 2.5 gal~~										
WBN OSN 113	Comp	Start	4/14/03	10:47	113- , 2.5 gal				X		03-091	5.7°C	SAB	1230	clear
		end	4/15/03	09:47											
TRC = 0.00 ppm															

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Martha Ervin	4/15/03 @ 10:00	Calvin Nichols ~~FedEx~~ MTE 4/15/03	4/15/03 @ 1000
Calvin Nichols ~~FedEx~~ MTE 4/15/03	4-15-03 @ 1230	Sarah Smith	4/15/03 1230

Instructions: *Clients* should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 4°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: WBNP BIOTOXICITY

P.O. Number: N/A

Facility Sampled: Watts Bar Nuclear Plant

NPDES Number: TN0020168

Collected By: Lanny Brown
Calvin Nichols

S & ME, Inc.
1413 Topside Road
Louisville, TN 37777

Phone: 865-970-0003
Fax: 865-970-0004

Delivered By (Circle One):
FedEx UPS Bus (Client)
Other (specify):_____

General Comments: Contact WBNP personnel at (423) 365-3575 if any problems are encountered. Pager number 1-800-323-4853 then number to be dialed.

-samples arrived packed on ice.

Field Identification / Sample Description	Grab/ Comp.	Ship. Temp. (°C)	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Laboratory Use				
			Date	Time			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
~~WBN Intake 101-~~	~~Grab~~				101- , 1gal										
WBN Intake 113- 2	Grab		4/17/03	0800	113- , 1gal		X	0.10			03-095	3.2°C	lm	1058	clear
~~WBN OSN 101-~~	~~Comp~~				101- , 25 gal										
WBN OSN 113- 2	Comp	start	4/17/03	08:25	113- , 25 gal		X	0.10			03-096	1.4°C	lm	~~1058~~ 1056	clear
		end	4/17/03	07:25											

TRC=<0.02 ppm for composite sample.

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
		~~FedEx~~ MJE 4/18/03	
Calvin Nichol ~~FedEx~~ MJE 4/18/03	4-17-03 1030	[signature]	4/17/03 1050

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 4°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: WBNP BIOTOXICITY

P.O. Number: N/A

Facility Sampled: Watts Bar Nuclear Plant

NPDES Number: TN0020168

Collected By: Lanny Braun, Calvin Nichols

S & ME, Inc.
1413 Topside Road
Louisville, TN 37777

Phone: 865-970-0003

Fax: 865-970-0004

Delivered By (Circle One):
FedEx UPS Bus (Client)
Other (specify):_____

General Comments: Contact WBNP personnel at (423) 365-3575 if any problems are encountered. Pager number 1-800-323-4853 then number to be dialed.

-Samples arrived on ice.

Field Identification / Sample Description	Grab/ Comp.	Ship. Temp. (°C)	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Laboratory Use				
			Date	Time			Yes	If Yes, Inches	No	Trace	ITTS Log Number	Arrival Temp. (°C)	By	Time	Appearance
~~WBN Intake 101-~~	~~Grab~~				101- ,2.5gal										
WBN Intake 113- 3	Grab		4/19/03	12:40	113- ,2.5gal						03-100	5.7°C	RO	1320	clear
~~WBN OSN 101-~~	~~Comp~~				101- ,5 gal										
WBN OSN 113- 3	Comp	Start	4/18/03 1425	~~12:30~~ ~~8:25~~ 4/19/03 MJE 4/19/03	113- ,5 gal		0.01				03-101	4.7°C	RO	1500	clear
		end	4/19/03	13:25											
TRC = 0.02 ppm for composite															

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
		~~FedEx~~ MJE 4/17/03	
Calvin Nichols ~~FedEx~~ MJE 4/17/03	4/10/03 1455	Brooke Davis	4/19/03 14:55

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 4°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within this time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

FATHEAD MINNOW TEST INFORMATION

Test Name: TVA – Watts Bar Nuclear Plant (Outfall 113)

Test Conducted From: 4/15/03 (Day 0) To 4/22/03 (Day 7)

Sites/Concentrations: 1. Control 4. 10.3%
2. 2.58% 5. 20.6%
3. 5.2% 6. 41.2%

Stock (if applicable): _____

Control Water Type (✓):

Moderately Hard Water + Trace Minerals ☑

Other (describe): _____ ☐

Dilution Water Type (✓):

Moderately Hard Water + Trace Minerals ☑

Other (describe): _____ ☐

Source of Test Larvae (✓): Lot# 03-024

S&ME ☑

Other (describe): _____ ☐

Date Larvae Hatched: 4/14 - 4/15/03 Hatch Time: 4/14 1630 - 4/15 1130 Initials: BM

Record of Minor Test Non-Conformity

Date: _____

Description of Non-Conformity: _____

Initials: _____

Date: _____

Description of Non-Conformity: _____

Initials: _____

Test Log sheets QA Reviewed By: [signature] (Reviewer Initials) on 4/23/03 (Date)

Fathead Minnow Daily Test Information Logsheet

Test Name: TVA - Watts Bar (Outfall 113) Test Dates: 4/15 - 4/22/03

Daily Test information	Temperature Information (25±1°C)			Feeding Information (feeding interval 6± 1h)		Test Initiation, Water Change, or Test Termination		Control Water Carboy No. and Additional comments	
Date and Initials	Incubator Temp. (°C)		Therm. No.	Fed 100 uL Brine Shrimp	Feeding time	Start Time	End Time		
	a.m.	p.m.						Carboy #	Date Prep.
Day 0 RO 4/15/03	25.0	25.2	1	SHS	am: — pm: 1545	1300	1325	2	4/14/03
Day 1 RO 4/16/03	25.0	25.1	1	RO RO	am: 0900 pm: 1530	1235	1302	2	4/14/03
Day 2 RO 4/17/03	25.0	25.1	1	SHS RO	am: 0900 pm: 1530	1300	1323	5	4/15/03
Day 3 RO 4/18/03	25.0	25.2	1	SHS JM	am: 0900 pm: 1545	1325	1345	2	4/17/03
Day 4 SHS 4/19/03	25.2	25.1	1	JM RO	am: 0930 pm: 1500	1230	1300	5	4/18/03
Day 5 SHS 4/20/03	25.1	25.0	1	JM JM	am: 1000 pm: 1530	1200	1220	5	4/18/03
Day 6 RO 4/21/03	25.0	24.9	1	RO RO	am: 0930 pm: 1600	1225	1250	2	4/A/03
Day 7 JM 4/22/03	25.0	25.0	1			1300 to JM		—	—

Client: TVA Watts Bar - Outfall 113 **Analysts:** BO, SHS, JM

Location: Rhea County, Tennessee **Dates:** 4/15 - 4/22/03

CUMULATIVE SURVIVAL OF P. PROMELAS IN CHRONIC TEST

CONC.	Initials	DAY NO.	REPLICATE A	B	C	D
Control Mod. Hard	BO	1	10	10	10	10
	BO/SHS	2	10	10	10	9
	BO/SHS	3	10	10	10	9
	BO/JM	4	9	10	10	9
	SHS/JM	5	9	10	10	9
	SHS/BO	6	9	10	10	9
	JM	7	9	10	10	9
2.58% Outfall 113	BO	1	10	10	10	10
	BO/SHS	2	9	10	9	10
	BO/SHS	3	9	10	9	10
	BO/JM	4	8	9	9	10
	SHS/JM	5	8	9	9	10
	SHS/BO	6	8	9	9	10
	JM	7	8	9	9	10
5.2% Outfall 113	BO	1	10	10	10	10
	BO/SHS	2	10	10	10	10
	BO/SHS	3	10	10	10	10
	BO/JM	4	10	10	9	10
	SHS/JM	5	10	10	9	10
	SHS/BO	6	10	9	9	10
	JM	7	10	9	9	10
10.3% Outfall 113	BO	1	9*	10	10	10
	BO/SHS	2	9	10	10	10
	BO/SHS	3	9	10	10	10
	BO/JM	4	9	10	8*	10
	SHS/JM	5	9	10	7	10
	BO/SHS	6	9	9*	7	10
	JM	7	9	9	7	10
20.6% Outfall 113	BO	1	10	10	10	10
	BO/SHS	2	10	10	10	10
	BO/SHS	3	10	10	10	10
	BO/JM	4	10	9	10	10
	SHS/JM	5	10	9	9*	10
	BO/SHS	6	10	9	9	10
	JM	7	10	9	9	10

CONC.	Initials	DAY NO.	REPLICATE A	B	C	D
41.2% Outfall 113	BO	1*	10	10	10	10
	BO/SHS	2	10	10	10	10
	BO/SHS	3	10	10	10	10
	BO/JM	4	10	10	10	7*
	SHS/JM	5	10	10	10	7
	BO/SHS	6	10	10	10	7
	JM	7	10	10	10	7
Intake	BO	1	10	10	10	10
	BO/SHS	2	10	10	10	9
	BO/SHS	3	10	10	10	9
	BO/JM	4	8*	10	10	8*
	SHS/JM	5	7*	10	10	8
	BO/SHS	6	6	9*	10	8
	JM	7	5	8*	10	8
		1				
		2				
		3				
		4				
		5				
		6				
		7				
		1				
		2				
		3				
		4				
		5				
		6				
		7				
		1				
		2				
		3				
		4				
		5				
		6				
		7				

* 4/16/03 - Day 1 - BO fuzzy dead fish - 10.3%. A
* 4/16/03 - Day 1 - BO 41.2%. A, B, C, & D. Water is cloudy. Fish aren't eating yet.
4/17/03 Day 2 - Fish eating better throughout
* 4/19/03 - Day 4 - BO - 10.3%. Rep C - fuzzy dead fish & 41.2%. rep D & Intake A & C. Fish eating better. Some fish are small throughout test
* 4/20/03 - day 5 ~~- -~~ All dead fish fuzzy
* 4/21/03 - day 6 - really ~~2~~ fuzzy dead fish 10.3%. B
* 4/21/03 - BO - day 7 - Intake BO - fuzzy skeleton. Still small fish throughout test concentrations

FATHEAD MINNOW SEVEN DAY TEST
RAW DATA
SURVIVAL AND GROWTH

Client: TVA Watts Bar - Outfall 113

Test Conducted By: RO, SHS, JM

Date Weighed 4/23/03

Weights Conducted by: RO, SHS

Test Dates: 4/15-4/22/03

Comments:

Began drying (date/time) 4/22/03 1440

End of drying (date/time) 4/23/03 0910

Oven Temp (°C) 102.1°C

Exposure & Repl.	Rep.	# Live Fish	# Original Fish	Pan Tare Weight (mg)	Dry Weight Pan + Fish (mg)	Groups Dry Weight (Pan + Fish minus Tare) (mg)	Mean Dry Weight (mg)
	A	9	10	1278.19	1282.71	4.52	0.452
Control	B	10	10	1277.08	1280.54	3.46	0.346
	C	10	10	1286.42	1290.28	3.86	0.386
	D	9	10	1257.36	1261.09	3.73	0.373
	A	8	10	1278.77	1283.28	4.51	0.451
2.58%	B	9	10	1281.38	1284.24	2.86	0.286
Outfall 113	C	9	10	1284.32	1288.69	4.37	0.437
	D	10	10	1286.40	1289.65	3.25	0.325
	A	10	10	1289.95	1293.42	3.47	0.347
5.2%	B	9	10	1285.00	1288.27	3.27	0.327
Outfall 113	C	9	10	1283.75	1289.07	5.32	0.532
	D	10	10	1278.50	1283.14	4.64	0.464
	A	9	10	1278.48	1281.25	2.77	0.277
10.3%	B	9	10	1280.61	1283.51	2.90	0.290
Outfall 113	C	7	10	1287.02	1289.33	2.31	0.231
	D	10	10	1280.76	1285.36	4.60	0.460
	A	10	10	1288.17	1291.12	2.95	0.295
20.6%	B	9	10	1293.58	1297.47	3.89	0.389
Outfall 113	C	9	10	1288.60	1291.19	2.59	0.259
	D	10	10	1292.41	1296.05	3.64	0.364
	A	10	10	1253.05	1256.13	3.08	0.308
41.2%	B	10	10	1255.20	1258.53	3.33	0.333
Outfall 113	C	10	10	1261.20	1264.96	3.76	0.376
	D	7	10	1254.98	1256.89	1.91	0.191

Weights of all fish were small - may be due to late hatch or initiation date + size

LSO Q

FATHEAD MINNOW SEVEN DAY TEST
RAW DATA
SURVIVAL AND GROWTH

Client: TVA Watts Bar - Outfall 113
Test Conducted By: RD, SH, JM
Date Weighed 4/23/03
Weights Conducted by: RD, SH

Test Dates: 4/15-4/22/03
Comments:
Began drying (date/time) 4/22/03 1440
End of drying (date/time) 4/23/03 0910
Oven Temp (°C) 102.1°C

Exposure & Repl.	Rep.	# Live Fish	# Original Fish	Pan Tare Weight (mg)	Dry Weight Pan + Fish (mg)	Groups Dry Weight (Pan + Fish minus Tare) (mg)	Mean Dry Weight (mg)
	A	5	10	1257.52	1258.87	1.35	0.135
Intake	B	8	10	1254.92	1257.91	2.99	0.299
100%	C	10	10	1257.43	1261.03	3.60	0.360
	D	8	10	1258.09	1261.60	3.51	0.351
	A						
	B						
	C						
	D						
	A						
	B						
	C						
	D						
	A						
	B						
	C						
	D						
	A						
	B						
	C						
	D						

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	4/15/03 13:00	Test ID:	113wb0403f	Sample ID:	TN0020168-NPDES Permit #
End Date:	4/22/03 13:00	Lab ID:	S&ME	Sample Type:	EFF3-Power Plant
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.4520	0.3460	0.3860	0.3730
2.58	0.4510	0.2860	0.4370	0.3250
5.2	0.3470	0.3270	0.5320	0.4640
10.3	0.2770	0.2900	0.2310	0.4600
20.6	0.2950	0.3890	0.2590	0.3640
41.2	0.3080	0.3330	0.3760	0.1910

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.3893	1.0000	0.3893	0.3460	0.4520	11.568	4				0.3938	1.0000
2.58	0.3748	0.9627	0.3748	0.2860	0.4510	21.810	4	0.257	2.410	0.1358	0.3938	1.0000
5.2	0.4175	1.0726	0.4175	0.3270	0.5320	23.318	4	-0.501	2.410	0.1358	0.3938	1.0000
10.3	0.3145	0.8080	0.3145	0.2310	0.4600	31.875	4	1.326	2.410	0.1358	0.3206	0.8141
20.6	0.3268	0.8394	0.3268	0.2590	0.3890	18.414	4	1.109	2.410	0.1358	0.3206	0.8141
41.2	0.3020	0.7759	0.3020	0.1910	0.3760	26.208	4	1.548	2.410	0.1358	0.3020	0.7668

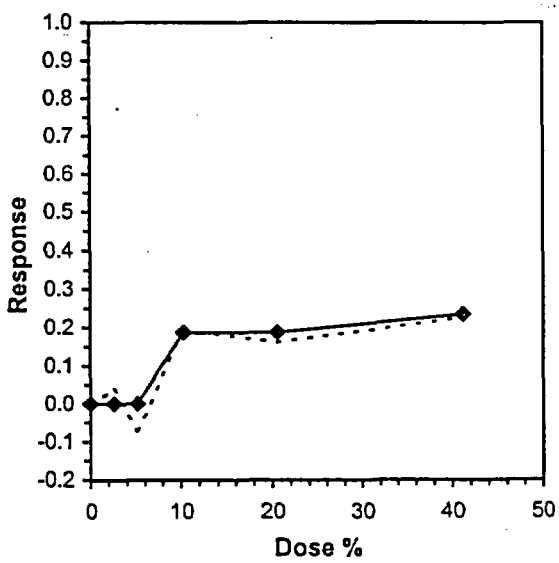
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.96103	0.884	0.26819	-0.865
Bartlett's Test indicates equal variances (p = 0.83)	2.151	15.0863		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	41.2	>41.2		2.42718	0.13583	0.34896	0.00857	0.00635	0.28925	5, 18

Linear Interpolation (80 Resamples)

Point	%	SD	95% CL(Exp)		Skew
IC05	6.572	3.206	0.000	17.585	0.9868
IC10	7.944				
IC15	9.315				
IC20	26.748				
IC25	>41.2				
IC40	>41.2				
IC50	>41.2				

pmsd = 34.9%



Larval Fish Growth and Survival Test-7 Day Survival

Start Date:	4/15/03 13:00	Test ID:	113wb0403f	Sample ID:	TN0020168
End Date:	4/22/03 13:00	Lab ID:	S&ME	Sample Type:	EFF3-Power Plant
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.9000	1.0000	1.0000	0.9000
100	0.5000	0.8000	1.0000	0.8000

			Transform: Arcsin Square Root					1-Tailed		
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	0.9500	1.0000	1.3305	1.2490	1.4120	7.072	4			
100	0.7750	0.8158	1.1029	0.7854	1.4120	23.198	4	1.670	1.943	0.2649

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.94576	0.749	-0.0833	1.67988
F-Test indicates equal variances (p = 0.13)	7.39462	47.4683		
Hypothesis Test (1-tail, 0.05)				
Homoscedastic t Test indicates no significant differences				

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	4/15/03 13:00	Test ID:	113wb0403f	Sample ID:	TN0020168
End Date:	4/22/03 13:00	Lab ID:	S&ME	Sample Type:	EFF3-Power Plant
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.4520	0.3460	0.3860	0.3730
100	0.1350	0.2990	0.3600	0.3510

			Transform: Untransformed					1-Tailed		
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	0.3893	1.0000	0.3893	0.3460	0.4520	11.568	4			
100	0.2863	0.7354	0.2863	0.1350	0.3600	36.456	4	1.812	1.943	0.1104

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.88323	0.749	-1.2131	1.69543
F-Test indicates equal variances (p = 0.20)	5.37105	47.4683		
Hypothesis Test (1-tail, 0.05)				
Homoscedastic t Test indicates no significant differences				

CERIODAPHNIA TEST INFORMATION SHEET

Test Name: TVA – Watts Bar Nuclear Plant (Outfall 113)

Test Conducted From: 4/15/03 (Day 0) To 4/21/03 LSOQA (Day 7)

Sites/Concentrations: 1. Control 4. 10.3% 7. Effluent 100%
2. 2.58% 5. 20.6%
3. 5.2% 6. 41.2%

Stock (if applicable): _____

Control Water Type (✓):

20% Dilute Mineral Water + Trace Minerals ☑

Other (describe): _____ ☐

Dilution Water Type (✓):

20% Dilute Mineral Water + Trace Minerals ☑

Other (describe): _____ ☐

Source of Test Organisms: S&ME brood board nos. 242

Age of Test Organisms:

Isolated neonates for test on 4/15/03 (date) from 0830 to 1300 (time) Initials mo

Record of Minor Test Non-Conformity

Date: _____

Description of Non-Conformity: _____

Initials: _____

Date: _____

Description of Non-Conformity: _____

Initials: _____

Test Log sheets QA Reviewed By: [signature] (Reviewer Initials) on 4/23/03 (Date)

Ceriodaphnia Daily Test Information Logsheet

Test Name: TVA - Watts Bar (Outfall 113) Test Dates: 4/15 - 4/22/03

Daily Test information	Temperature Information (cup -25±1°C)			Feeding Information				Test Initiation, Water Change, or Test Termination		Control Water Carboy No. and Additional comments	
Date and Initials	Incubator Temp. (°C)		Therm. No.	Fed 100 uL YCT	YCT Date	Fed 100 uL Selenastrum	Selanastrum Date	Start Time	End Time	Carboy #	Date Prep.
	a.m.	p.m.									
Day 0 BO 4/15/03	25.0	25.2	1	1410	2/5/03	1410	3/24/03	1345	1410	3	4/13/03
Day 1 BO 4/16/03	25.0	25.1	1	1330	2/5/03	1330	3/24/03	1310	1330	3	4/18/03
Day 2 BO 4/17/03	25.0	25.0	1	1430	2/5/03	1420	3/24/03	1405	1420	3	4/18/03
Day 3 BO 4/18/03	25.0	25.2	1	1445	2/5/03	1445	3/24/03	1430	1445	3	4/18/03
Day 4 BO 4/19/03	25.2	25.1	1	1405	2/5/03	1405	3/24/03	1340	1405	3	4/18/03
Day 5 BO 4/20/03	25.2	25.1	1	1315	2/5/03	1315	3/24/03	1250	1315	3	4/18/03
Day 6 BO 4/21/03	25.0	24.9	1	1320	2/5/03	1320	3/24/03	1300	1320	3	4/18/03
Day 7 BO 4/22/03	25.0	24.9	1					1400		—	—

Checked for split broods BAO

Ceriodaphnia 3-Brood Survival and Reproduction
Raw Data Sheet

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Analysts: BD, SHS, JH
Test Dates: 4/15 - 4/21/03

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
			--	--	--	--	--	--	--	--	--	--	---	---	---	---
	BD	1	0	→									0	10	10	0
	BD	2	0	→									0	10	10	0
Control	SHS	3	0	→									0	10	10	0
20%	BD	4	7	2	7	7	6	7	4	5	5	7	57	10	10	5.7
DMW	SHS	5	11	12	10	12	9	11	11	12	10	9	107	10	10	10.7
	SHS	6	13	11	12	12	13	14	10	12	12	7	116	10	10	11.6
		7														
	BD	Total	31	25	29	31	28	32	25	29	27	23	280	10	10	28.0

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8.	9	10				
			--	--	--	--	--	--	--	--	--	--	---	---	---	---
	BD	1	0	→									0	10	10	0
	BD	2	0	→									0	10	10	0
2.58%	SHS	3	0	→									0	10	10	0
Outfall	BD	4	7	4	7	6	6	4	6	7	8	5	60	10	10	6.0
113	SHS	5	11	10	14	9	7	10	8	10	8	12	99	10	10	9.9
	SHS	6	17	14	11	13	11	15	14	12	11	12	130	10	10	13.0
		7														
	BD	Total	35	~~58~~ 28	32	28	24	29	28	29	27	29	289	10	10	28.9

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
			--	--	--	--	--	--	--	--	--	--	---	---	---	---
	BD	1	0	→									0	10	10	0
	BD	2	0	→									0	10	10	0
5.2%	SHS	3	0	→									0	10	10	0
Outfall	BD	4	0	2	6	5	4	5	6	5	6	6	45	10	10	4.5
113	SHS	5	13	11	13	6	9	9	10	11	10	7	99	10	10	9.9
	SHS	6	15	12	12	9	12	11	11	13	10	15	120	10	10	12.0
		7														
	BD	Total	28	25	31	20	25	~~29~~ 25	~~29~~ 27	29	26	28	264	10	10	26.4

Ceriodaphnia 3-Brood Survival and Reproduction
Raw Data Sheet

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Analysts: BD, SLS, JM
Test Dates: 4/15 - 4/23/03 (PO)

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
			--	--	--	--	--	--	--	--	--	--	---	---	---	---
	PO	1	0	→									0	10	10	0
		2	0	→									0	10	10	0
10.3%	PO	3	0	→									0	10	10	0
Outfall	JM	4	3	0	5	2	5	4	2	2	2	5	30	10	10	3.0
113	JM	5	12	7	11	11	8	12	9	15	6	6	97	10	10	9.7
	PO	6	14	16	13	12	14	14	15	12	12	14	136	10	10	13.6
		7														
	PO	Total	29	23	29	25	27	30	26	29	20	25	263	10	10	26.3

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
			--	--	--	--	--	--	--	--	--	--				
	PO	1	0	→									0	10	10	0
		2	0	→									0	10	10	0
20.6%	PO	3	0	→									0	10	10	0
Outfall	JM	4	4	7	5	4	3	4	4	2	4	4	41	10	10	4.1
113	JM	5	7	9	11	13	14	7	12	6	8	10	97	10	10	9.7
	PO	6	15	12	12	11	13	16	11	14	11	15	130	10	10	13.0
		7														
	PO	Total	26	28	28	28	30	27	27	22	23	29	268	10	10	26.8

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
			--	--	--	--	--	--	--	--	--	--	---	---	---	---
	PO	1	0	→									0	10	10	0
		2	0	→									0	10	10	0
41.2%	PO	3	0	→									0	10	10	0
Outfall	JM	4	4	5	6	2	2	6	5	3	3	4	40	10	10	4.0
113	JM	5	10	9	6	5	9	8	8	5	9	11	80	10	10	8.0
	PO	6	16	17	10	13	12	10	13	13	14	13	131	10	10	13.1
		7														
	PO	Total	30	31	22	20	23	24	26	21	26	28	251	10	10	25.1

Ceriodaphnia 3-Brood Survival and Reproduction
Raw Data Sheet

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Analysts: BSD, SAS, JM
Test Dates: 4/15 - 4/21/03

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
Intake		1	0	→								→	0	10	10	0
		2	0	→								→	0	10	10	0
		3	0	→								→	0	10	10	0
		4	8	8	5	10	7	6	5	8	10	7	74	10	10	7.4
		5	7	0	4	6	5	6	8	8	0	10	54	10	10	5.4
		6	3	14	10	18	14	12	14	18	16	15	134	10	10	13.4
		7														
		Total	18	22	19	34	26	24	27	34	26	32	262	10	10	26.2

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	4/15/2003 13:45	Test ID: WB113cr	Sample ID: TN0020168-NPDES Permit #
End Date:	4/21/2003 14:00	Lab ID: S&ME Inc.	Sample Type: EFF3-Power Plant
Sample Date:		Protocol: EPAF 91-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia
Comments:			

Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	31.000	25.000	29.000	31.000	28.000	32.000	25.000	29.000	27.000	23.000
2.58	35.000	28.000	32.000	28.000	24.000	29.000	28.000	29.000	27.000	29.000
5.2	28.000	25.000	31.000	20.000	25.000	25.000	27.000	29.000	26.000	28.000
10.3	29.000	23.000	29.000	25.000	27.000	30.000	26.000	29.000	20.000	25.000
20.6	26.000	28.000	28.000	28.000	30.000	27.000	27.000	22.000	23.000	29.000
41.2	30.000	31.000	22.000	20.000	23.000	24.000	26.000	21.000	26.000	28.000

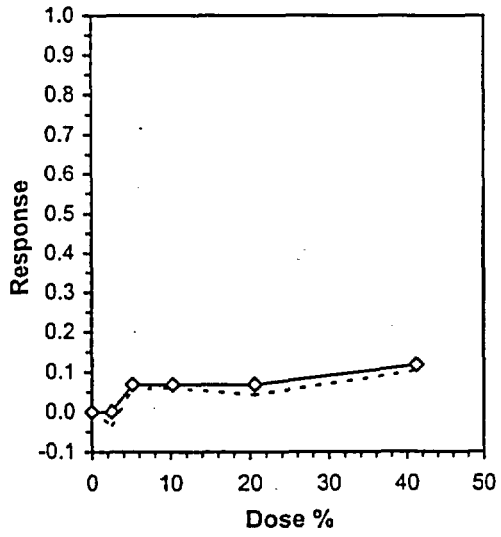
			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	28.000	1.0000	28.000	23.000	32.000	10.648	10				28.450	1.0000
2.58	28.900	1.0321	28.900	24.000	35.000	10.114	10	-0.654	2.287	3.149	28.450	1.0000
5.2	26.400	0.9429	26.400	20.000	31.000	11.321	10	1.162	2.287	3.149	26.500	0.9315
10.3	26.300	0.9393	26.300	20.000	30.000	12.031	10	1.235	2.287	3.149	26.500	0.9315
20.6	26.800	0.9571	26.800	22.000	30.000	9.440	10	0.872	2.287	3.149	26.500	0.9315
41.2	25.100	0.8964	25.100	20.000	31.000	14.960	10	2.106	2.287	3.149	25.100	0.8822

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)					0.65869		1.035		-0.1838	-0.3356
Bartlett's Test indicates equal variances (p = 0.92)					1.45653		15.0863			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	41.2	>41.2		2.42718	3.14857	0.11245	18.1367	9.47963	0.10724	5, 54

Linear Interpolation (80 Resamples)

Point	%	SD	95% CL	Skew
IC05	4.491			
IC10	33.769			
IC15	>41.2			
IC20	>41.2			
IC25	>41.2			
IC40	>41.2			
IC50	>41.2			

PMSD = 11.2%



Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	4/15/2003 13:45	Test ID: WB113Incr	Sample ID: TN00020168
End Date:	4/21/2003 14:00	Lab ID: S&ME INC.	Sample Type: EFF3-Power Plant
Sample Date:		Protocol: EPAF 91-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia
Comments:			

Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	31.000	25.000	29.000	31.000	28.000	32.000	25.000	29.000	27.000	23.000
Intake	18.000	22.000	19.000	34.000	26.000	24.000	27.000	34.000	26.000	32.000

			Transform: Untransformed						1-Tailed	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	28.000	1.0000	28.000	23.000	32.000	10.648	10			
Intake	26.200	0.9357	26.200	18.000	34.000	21.948	10	0.879	1.734	3.552

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.97568	0.868	0.04974	-0.3775
F-Test indicates equal variances (p = 0.06)	3.72	6.54109		
Hypothesis Test (1-tail, 0.05)				
Homoscedastic t Test indicates no significant differences				

$p_{mSD} = 12.7\%$

WB113- April 15-22, 2003 INITIAL AND FINAL WATER CHEMISTRY (FATHEAD)

Initial Chemistry

Fish - Control	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.4	25.6	25.2	25.1	25.8	25.1	24.3	24.3	25.8		25.2
DO	6.9	6.8	7.0	6.9	6.8	6.8	6.7	6.7	7.0		6.8
pH	7.9	8.0	8.1	7.9	8.2	8.1	8.0	7.9	8.2		8.0
Cond	396	395	394	388	393	398	403	388	403		395
Hard	98.0		96.0	96.0	96.0		80.0	80	98		93.2
Alk	63.0		60.0	63.0	64.0		64.0	60	64		62.8

Concentration 2.58%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.6	25.7	24.5	25.1	25.8	25.5	24.7	24.5	25.8		25.3
DO	6.9	6.6	6.6	6.6	6.6	6.8	6.8	6.6	6.9		6.7
pH	8.0	8.1	8.0	8.0	8.2	8.1	8.0	8.0	8.2		8.1
Cond	388	390	386	378	386	391	398	378	398		388

Concentration 5.2%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.6	25.7	24.5	24.9	25.8	25.6	24.8	24.5	25.8		25.3
DO	6.9	6.6	6.7	6.7	6.8	6.8	7.0	6.6	7.0		6.8
pH	8.0	8.1	8.0	8.0	8.2	8.1	8.0	8.0	8.2		8.1
Cond	382	384	372	376	382	384	392	372	392		382

Concentration 10.3%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.6	25.8	24.7	24.9	25.6	25.7	24.8	24.7	25.8		25.3
DO	6.8	6.6	6.7	6.7	6.7	6.8	6.9	6.6	6.9		6.7
pH	8.0	8.1	8.0	8.1	8.1	8.1	8.0	8.0	8.1		8.1
Cond	369	374	360	364	383	375	392	360	392		374

Concentration 20.6%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.7	25.8	24.8	24.7	25.6	25.8	24.9	24.7	25.8		25.3
DO	6.7	6.6	6.7	6.7	6.7	6.8	6.9	6.6	6.9		6.7
pH	8.0	8.1	8.0	8.1	8.2	8.1	8.1	8.0	8.2		8.1
Cond	349	354	342	342	373	349	383	342	383		356

Concentration 41.2%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.6	25.9	24.8	24.8	25.6	25.8	25.1	24.8	25.9		25.4
DO	6.8	6.7	6.8	6.9	6.8	6.8	6.8	6.7	6.9		6.8
pH	8.0	8.1	8.0	8.1	8.1	8.1	8.1	8.0	8.1		8.1
Cond	303	307	300	295	351	311	360	295	360		318

Concentration 100%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.3	25.7	24.8	24.8	25.0	25.4	25.0	24.8	25.7		25.1
DO	7.6	7.0	7.1	7.6	7.8	7.0	6.8	6.8	7.8		7.3
pH	8.0	8.1	8.1	8.1	8.1	8.1	8.1	8.0	8.1		8.1
Cond	168	160	164	161	163	167	173	160	173		165
Hard	80.0		108.0		76.0			76	108		88
Alk	61.0		67.0		60.0			60	67		62.7
TRC	0.07		0.09		0.05			0.05	0.09		0.07

Intake	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	24.9	25.2	24.8	25.8	25.8	25.1	25.3	24.8	25.8		25.3
DO	7.8	8.1	8.6	8.7	9.0	7.4	7.2	7.2	9.0		8.1
pH	8.3	8.2	8.2	8.1	8.0	8.5	8.5	8.0	8.5		8.3
Cond	153	161	157	158	154	154	144	144	161		154
Hard	82.0		90.0		76.0			76	90		82.7
Alk	58.0		61.0		61.0			58	61		60.0
TRC	0.08		0.06		0.04			0.04	0.08		0.06

Final Chemistry

FISH

Fish - Medium	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.7	25.6	25.6	25.6	25.3	24.2	25.1	24.2	25.7		25.3
DO	6.0	5.8	5.4	5.1	5.5	6.5	6.3	5.1	6.5		5.8
pH	7.7	7.7	7.5	7.4	7.7	7.6	7.4	7.4	7.7		7.6

Concentration 2.58%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.5	25.3	25.6	25.6	25.3	24.9	25.8	24.9	25.8		25.4
DO	5.8	5.5	4.9	5.6	5.4	6.3	6.2	4.9	6.3		5.7
pH	7.7	7.6	7.3	7.6	7.7	7.6	7.6	7.3	7.7		7.6

Concentration 5.2%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.5	25.3	25.8	25.6	25.4	25.0	25.9	25.0	25.9		25.5
DO	5.4	5.2	5.1	5.2	5.4	6.2	6.2	5.1	6.2		5.5
pH	7.8	7.6	7.4	7.6	7.7	7.7	7.6	7.4	7.8		7.6

Concentration 10.3%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.5	25.8	24.9	25.8	25.5	25.4	25.9	24.9	25.9		25.5
DO	5.4	5.4	5.4	5.4	5.4	6.2	6.2	5.4	6.2		5.6
pH	7.9	7.4	7.8	7.7	7.7	7.7	7.8	7.4	7.9		7.7

Fish Water Chemistry

Concentration 20.6%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.3	25.7	25.1	25.8	25.6	25.4	25.9	25.1	25.9		25.5
DO	5.3	5.3	5.4	5.5	5.4	6.4	6.3	5.3	6.4		5.7
pH	7.8	7.7	7.6	7.7	7.7	7.8	7.9	7.6	7.9		7.7

Concentration 41.2%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.3	25.6	25.9	25.9	25.4	25.4	25.9	25.3	25.9		25.6
DO	5.6	5.2	5.1	5.3	5.3	6.3	6.5	5.1	6.5		5.6
pH	7.8	7.5	7.5	7.6	7.7	7.8	7.9	7.5	7.9		7.7

Intake	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.5	25.8	25.6	25.6	25.3	25.8	26.0	25.3	26.0		25.7
DO	5.6	5.1	5.2	5.6	5.6	6.6	6.6	5.1	6.6		5.8
pH	7.8	7.6	7.5	7.7	7.9	8.0	8.1	7.5	8.1		7.8

Test Temperature	Mean	25.4
	Minimum	24.2
	Maximum	26.0

Word Review by: Cm 05-04-2003

Excel Reviewed by: Cm 05-04-2003

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Template No.: 1
Test Type: Fathead Minnow 7-day Chronic

Analysts: PO, SH, JM
Dates/times: 4/15/03 1300 / 4/22/03 1300
Age of Larvae: ~24hrs

Control: Mod. Hard	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.4	25.6	25.2	25.1	25.8	25.1	24.3		
Final		25.7	25.6	25.6	25.6	25.3	24.2	25.1	
D.O. Initial	6.9	6.8	7.0	6.9	6.8	6.8	6.7		
Final		6.0	5.8	5.4	5.1	5.5	6.5	6.3	
pH Initial	7.9	8.0	8.1	7.9	8.2	8.1	8.0		
Final		7.7	7.7	7.5	7.4	7.7	7.6	7.4	
Alkalinity	63	/	60	63	64	/	64		
Hardness	98	/	96	96	96	/	80		
Conductivity	396	395	394	388	393	398	MD3	~~MD3~~	
Analyst Initials	PO	SH	SH	PO	JM	SH	PO	SH	
QA Review Initials	SH	PO	PO	SH	PO	PO	SH	JM	

2.58% Outall 113	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.7	24.5	25.1	25.8	25.5	24.7		
Final		25.5	25.3	25.6	25.6	25.3	24.9	25.8	
D.O. Initial	6.9	6.6	6.6	6.6	6.6	6.8	6.8		
Final		5.8	5.5	4.9	5.6	5.4	6.3	6.2	
pH Initial	8.0	8.1	8.0	8.0	8.2	8.1	8.0		
Final		7.7	7.6	7.3	7.6	7.7	7.6	7.6	
Conductivity	388	390	386	378	386	391	398		
Analyst Initials	PO	SH	SH	PO	JM	SH	PO	SH	
QA Review Initials	SH	PO	PO	SH	PO	PO	SH	JM	

5.2% Outfall 113	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.7	24.5	24.9	25.8	25.6	24.8		
Final		25.5	25.3	25.8	25.6	25.4	25.0	25.9	
D.O. Initial	6.9	6.6	6.7	6.7	6.8	6.8	7.0		
Final		5.1	5.2	5.1	5.2	6.4	6.2	6.2	
pH Initial	8.0	8.1	8.0	8.0	8.2	8.1	8.0		
Final		7.8	7.6	7.4	7.6	7.7	7.7	7.6	
Conductivity	382	384	372	376	382	384	392		
Analyst Initials	PO	SH	SH	PO	JM	SH	PO	SH	
QA Review Initials	SH	PO	PO	SH	PO	PO	SH	JM	

* Day 0 - 4/14/03 - PO - The room temperature of the lab was 26.1°C - opened all of the doors to cool the room down - Checked the temps of the control waters: #2 Mod. Hard = 25.8°C; #5 Mod. Hard = 25.6°C; Grab #3 = 25.8°C - After 15 minutes of the doors being open the temp decreased to 25.4°C - the control water temp was approximately the same temp after 15 minutes. - Left the doors ajar. - Temp remained @ 25.4°C

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Template No.: 1
Test Type: Fathead Minnow 7-day Chronic

Analysts: BO, SHB, JM
Dates/times: 4/15/13 1350 4/22/13 1300
Age of Larvae: ~24hrs

	Day								
10.3% Outfall 113	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.8	24.7	24.9	~~24.7~~	25.7	24.8		25.6 = init. temp day 4
Final		25.5	25.8	24.9	25.8	25.5	25.4	25.9	
D.O. Initial	6.8	6.6	6.7	6.7	6.7	6.8	6.9		
Final		5.4	5.4	5.4	5.4	5.4	6.2	6.2	
pH Initial	8.0	8.1	8.0	8.1	8.1	8.1	8.0		
Final		7.9	7.4	7.8	7.7	7.7	7.7	7.8	
Conductivity	369	374	360	364	383	375	392		
Analyst Initials	BO	SHB	SHB	BO	BO	SHB	BO	SHB	
QA Review Initials	SHB	BO	BO	SHB	BO	BO	SHB	JM	

	Day								
20.6% Outfall 113	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.7	25.8	24.8	24.7	25.6	25.8	24.9		
Final		25.3	25.7	25.1	25.8	25.6	25.4	25.9	
D.O. Initial	6.7	6.6	6.7	6.7	6.7	6.8	6.9		
Final		5.3	5.3	5.4	5.5	5.4	6.4	6.3	
pH Initial	8.0	8.1	8.0	8.1	8.2	8.1	8.1		
Final		7.8	7.7	7.6	7.7	7.7	7.8	7.9	
Conductivity	349	354	342	342	373	349	383		
Analyst Initials	BO	SHB	SHB	BO	JM	SHB	BO	SHB	
QA Review Initials	SHB	BO	BO	SHB	BO	BO	SHB	JM	

	Day								
41.2% Outfall 113	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.9	24.8	24.8	25.6	25.8	25.1		
Final		25.3	25.6	25.9	25.9	25.4	25.4	25.9	
D.O. Initial	6.8	6.7	6.8	6.9	6.8	6.8	6.8		
Final		5.6	5.2	5.1	5.3	5.3	6.3	6.5	
pH Initial	8.0	8.1	8.0	8.1	8.1	8.1	8.1		
Final		7.8	7.5	7.5	7.6	7.7	7.8	7.9	
Conductivity	303	307	300	295	351	311	360		
Analyst Initials	BO	SHB	SHB	BO	JM	SHB	BO	SHB	
QA Review Initials	SHB	BO	BO	SHB	BO	BO	SHB	JM	

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Template No.: 1
Test Type: Fathead Minnow 7-day Chronic

Analysts: AO, SHB, CM
Dates/times: 4/15/1300 4/22/03/1300
Age of Larvae: ~24hrs

100% Outfall 113	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.3	25.7	24.8	24.8	25.0	25.4	25.0		
Final									
D.O. Initial	8.0	7.0	7.1	7.6	7.8	7.0	6.8		init. DO day 0 = 7.6
Final									
pH Initial	8.0	8.1	8.1	8.1	8.1	8.1	8.1		
Final									
Alkalinity	41		67		60				
Hardness	80		108		96				Day 4 - hardness 76.0
Conductivity	168	160	164	161	163	167	173		
Residual Chlorine	0.07		0.09		0.05				
Sample Date	4/15	4/15	4/17	4/17	4/17	4/19	4/19		
Analyst Initials	BO	SHB	SHB	CM	JM	SHB	AO		
QA Review Initials	SHB	AO	LSO	SHB	SHB	LSO	SHB		

Intake	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	24.9	25.2	24.8	25.8	25.8	25.1	25.3		
Final		25.5	25.8	25.6	25.6	25.3	25.8	26.0	
D.O. Initial	9.2*	8.1	8.6	8.7	9.0	7.4	7.2		
Final		5.6	5.1	5.2	5.6	5.6	6.6	6.6	
pH Initial	8.3	8.2	8.2	8.1	8.0	8.5	8.5		
Final		7.8	7.6	7.5	7.7	7.9	8.0	8.1	
Alkalinity	58		61		61				
Hardness	82		90		76				
Conductivity	153	161	157	158	154	154	144		
Residual Chlorine	0.08		0.06		0.04				
Sample Date	4/15	4/15	4/17	4/17	4/17	4/19	4/19		
Analyst Initials	CM	SHB	SHB	CM	JM	SHB	CM	SHB	
QA Review Initials	SHB	AO	CM	SHB	AO	CM	SHB	JM	

4/15/03 *aerated sample for 10 minutes to reduce D.O. - final (initial) D.O. level = 7.8

Watts Bar 113 April 15-April 22, 2003, INITIAL AND FINAL WATER CHEMISTRY (CERIO)

Initial Chemistry

CERIO

Control	0	1	2	3	4	5	6	MIN	MAX	MEAN
Temp	25.6	25.5	24.4	24.9	24.8	25.0		24.4	25.6	25.0
DO	6.9	6.4	6.8	6.9	6.7	6.2		6.2	6.9	6.7
pH	8.0	8.1	8.0	8.0	8.1	8.1		8.0	8.1	8.1
Cond	168	169	167	172	168	165		165	172	168
Hard	80		82		80			80.0	82.0	80.7
Alk	58		59		62			58.0	62.0	59.7

Concentration 2.58%	0	1	2	3	4	5	6	MIN	MAX	MEAN
Temp	25.6	25.7	24.5	25.5	25.7	25.1		24.5	25.7	25.4
DO	6.8	6.4	6.7	6.8	6.8	6.2		6.2	6.8	6.6
pH	8.0	8.1	8.0	8.1	8.2	8.1		8.0	8.2	8.1
Cond	171	169	170	167	167	162		162	171	168

Concentration 5.2%	0	1	2	3	4	5	6	MIN	MAX	MEAN
Temp	25.6	25.9	24.7	25.4	25.6	25.2		24.7	25.9	25.4
DO	6.9	6.4	6.7	6.7	6.8	6.2		6.2	6.9	6.6
pH	8.0	8.1	8.0	8.1	8.1	8.1		8.0	8.1	8.1
Cond	169	169	168	168	169	165		165	169	168

Concentration 10.3%	0	1	2	3	4	5	6	MIN	MAX	MEAN
Temp	25.7	25.9	24.8	25.3	25.8	25.4		24.8	25.9	25.5
DO	6.9	6.4	6.7	6.7	6.7	6.2		6.2	6.9	6.6
pH	8.0	8.1	8.0	8.1	8.2	8.1		8.0	8.2	8.1
Cond	168	168	168	168	169	165		165	169	168

Concentration 20.6%	0	1	2	3	4	5	6	MIN	MAX	MEAN
Temp	25.7	25.9	24.8	24.9	25.8	25.5		24.8	25.9	25.4
DO	7.0	6.4	6.7	6.8	6.7	6.2		6.2	7.0	6.6
pH	8.0	8.1	8.1	8.1	8.1	8.2		8.0	8.2	8.1
Cond	168	168	168	166	168	165		165	168	167

Concentration 41.2%	0	1	2	3	4	5	6	MIN	MAX	MEAN
Temp	25.6	25.8	24.8	24.4	25.8	25.6		24.4	25.8	25.3
DO	7.0	6.5	6.8	6.9	6.9	6.3		6.3	7.0	6.7
pH	8.0	8.1	8.1	8.1	8.1	8.2		8.0	8.2	8.1
Cond	167	169	167	164	165	166		164	169	166

Concentration 100%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.4	25.7	24.9	24.9	25.2	25.4		24.9	25.7		25.3
DO	7.5	7.3	7.2	7.4	7.9	7.0		7.0	7.9		7.4
pH	8.0	8.1	8.1	8.1	8.1	8.1		8.0	8.1		8.1
Cond	168	160	164	161	163	167		160	168		164
Hard	80		108		76			76	108		88.0
Alk	61		67		60			60	67		62.7
TRC	0.07		0.09		0.05			0.05	0.09		0.07

Intake 100%	0	1	2	3	4	5	6	MIN	MAX		MEAN
Temp	25.1	25.8	24.8	25.8	25.8	25.1		24.8	25.8		25.4
DO	7.8	8.1	8.3	8.6	9.0	7.4		7.4	9.0		8.2
pH	8.3	8.3	8.2	8.1	8.0	8.5		8.0	8.5		8.2
Cond	156	162	157	158	154	154		154	162		157
Hard	82.0		90.0		76.0			76	90		82.7
Alk	58.0		61.0		61.0			58	61		60.0
TRC	0.08		0.06		0.04			0.04	0.08		0.06

Final Chemistry

CERIO

Cerio - Medium	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.6	24.9	24.7	25.5	25.0	24.3		24.3	25.6		25.0
DO	5.9	6.4	6.4	6.3	5.7	6.4		5.7	6.4		6.2
pH	8.0	8.1	8.2	8.1	8.4	8.3		8.0	8.4		8.2

Concentration 2.58%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.4	25.0	25.1	25.2	25.0	24.3		24.3	25.4		25.0
DO	5.8	6.3	6.5	6.1	5.6	6.5		5.6	6.5		6.1
pH	8.1	8.2	8.1	8.2	8.4	8.2		8.1	8.4		8.2

Concentration 5.2%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.2	25.1	25.0	25.2	25.0	24.3		24.3	25.2		25.0
DO	5.6	6.2	6.5	6.1	5.6	6.6		5.6	6.6		6.1
pH	8.1	8.3	8.3	8.4	8.4	8.4		8.1	8.4		8.3

Concentration 10.3%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.2	25.0	25.0	25.4	25.0	24.4		24.4	25.4		25.0
DO	5.6	6.3	6.6	6.2	5.6	6.6		5.6	6.6		6.2
pH	8.2	8.4	8.5	8.6	8.6	8.1		8.1	8.6		8.4

Concentration 20.6%	1	2	3	4	5	6	7	MIN	MAX		MEAN
Temp	25.2	25.0	25.0	25.5	25.0	24.4		24.4	25.5		25.0
DO	5.3	6.4	6.6	6.2	5.6	6.5		5.3	6.6		6.1
pH	8.2	8.4	8.4	8.5	8.6	8.0		8.0	8.6		8.4

Concentration 41.2%	1	2	3	4	5	6	7	MIN	MAX	MEAN
Temp	25.3	24.9	25.0	25.4	25.0	24.4		24.4	25.4	25.0
DO	5.6	6.5	6.6	6.2	5.7	6.5		5.6	6.6	6.2
pH	8.2	8.3	8.5	8.5	8.5	8.1		8.1	8.5	8.4

Intake 100%	1	2	3	4	5	6	7	MIN	MAX	MEAN
Temp	25.3	24.9	25.1	25.8	25.0	24.4		24.4	25.8	25.1
DO	5.8	6.4	6.6	6.4	5.7	6.5		5.7	6.6	6.2
pH	8.1	8.3	8.2	8.4	8.4	8.3		8.1	8.4	8.3

Test temperature		
	Mean:	25.2
	Min:	24.3
	Max:	25.9

Word Review by: Cm 05-04-2003

Excel Reviewed by: Cm 05-04-2003

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Template No.: 1
Test Type: *Ceriodaphnia dubia* 7-day Chronic

Analysts: RO, SHS, JN
Dates/times: 4/15/03 1345 / 4/22/03 1400
Age of Neonates: 24hrs

Control: 20% DMW	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.5	24.4	24.9	24.8	25.0	24.3		
Final		25.6	24.9	24.7	25.5	25.0	24.3		
D.O. Initial	6.9	6.4	6.8	6.9	6.7	6.2	6.9		
Final		5.9	6.4	6.4	6.3	5.7	6.4		
pH Initial	8.0	8.1	8.0	8.0	8.1	8.1	8.0		
Final		8.0	8.1	8.2	8.1	8.4	8.3		
Alkalinity	58		59		62				
Hardness	80		82		80				
Conductivity	168	169	167	172	168	165	169		
Analyst Initials	RO	SHS	SHS	RO	RO	SHS	RO		
QA Initials	SHS	RO	RO	SHS	SHS	RO	SHS		

2.58% Outall 113	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.7	24.5	25.5	25.7	25.1	24.3		
Final		25.4	25.0	25.1	25.2	25.0	24.3		
D.O. Initial	6.8	6.4	6.7	6.8	6.8	6.2	7.3		
Final		5.8	6.3	6.5	6.1	5.6	6.5		
pH Initial	8.0	8.1	8.0	8.1	8.2	8.1	8.0		
Final		8.1	8.2	8.1	8.2	8.4	8.2		
Conductivity	171	169	170	167	167	162	161		
Analyst Initials	RO	SHS	SHS	RO	RO	SHS	RO		
QA Initials	SHS	RO	RO	SHS	SHS	RO	SHS		

5.2% Outfall 113	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.9	24.7	25.4	25.6	25.2	24.3		
Final		25.2	25.1	25.0	25.2	25.0	24.3		
D.O. Initial	6.9	6.4	6.7	6.7	6.8	6.2	6.9		
Final		5.6	6.2	6.5	6.1	5.6	6.6		
pH Initial	8.0	8.1	8.0	8.1	8.1	8.1	8.1		
Final		8.1	8.3	8.3	8.4	8.4	8.4		
Conductivity	169	169	168	168	169	165	164		
Analyst Initials	RO	SHS	SHS	RO	RO	SHS	RO		
QA Initials	SHS	RO	RO	SHS	SHS	RO	SHS		

* Day 0 - 4/14/03 - refer to the fathead data sheets for room temperature adjustment.

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Template No.: 1
Test Type: *Ceriodaphnia dubia* 7-day Chronic

Analysts: MO. SHS. JN
Dates/times: 4/15/1345 | 4/22/03/1400
Age of Neonates: ~8hrs

Day									
10.3% Outfall 113	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.7	25.9	24.8	25.3	25.8	25.4	24.8		
Final		25.2	25.0	25.0	25.4	25.0	24.4		
D.O. Initial	6.9	6.4	6.7	6.7	6.7	6.2	6.9		
Final		5.6	6.3	6.6	6.2	5.6	6.6		
pH Initial	8.0	8.1	9.0	8.1	8.2	8.1	8.1		
Final		8.2	8.4	8.5	8.6	8.6	8.1		
Conductivity	168	168	168	168	169	165	165		
Analyst Initials	MO	SHS	SHS	MO	MO	SHS	MO		
QA Initials	SHS	MO	MO	SHS	SHS	MO	SHS		

Day									
20.6% Outfall 113	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.7	25.9	24.8	24.9	25.8	25.5	24.8		
Final		25.2	25.0	25.0	25.5	25.0	24.4		
D.O. Initial	7.0	6.4	6.7	6.8	6.7	6.2	6.8		
Final		5.3	6.4	6.6	6.2	5.6	6.5		
pH Initial	8.0	8.1	8.1	8.1	8.1	8.2	8.1		
Final		8.2	8.4	8.4	8.5	8.6	8.0		
Conductivity	168	168	168	166	168	165	166		
Analyst Initials	MO	SHS	SHS	MO	MO	SHS	MO		
QA Initials	SHS	MO	MO	SHS	SHS	MO	SHS		

Day									
41.2% Outfall 113	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	25.8	24.8	24.4	25.8	25.6	25.4		
Final		25.3	24.9	25.0	25.4	25.0	24.4		
D.O. Initial	7.0	6.5	6.8	6.9	6.9	6.3	6.8		
Final		5.6	6.5	6.6	6.2	5.7	6.5		
pH Initial	8.0	8.1	8.1	8.1	8.1	8.2	8.1		
Final		8.2	8.3	8.5	8.5	8.5	8.1		
Conductivity	167	169	167	164	165	166	166		
Analyst Initials	MO	SHS	SHS	MO	MO	SHS	MO		
QA Initials	SHS	MO	MO	SHS	SHS	SHS MO	SHS		

Client: TVA Watts Bar - Outfall 113
Location: Rhea County, Tennessee
Template No.: 1
Test Type: *Ceriodaphnia dubia* 7-day Chronic

Analysts: RO, SHS, JN
Dates/times: 4/15 [1347] 22/03/1450
Age of Neonates: <8hrs

	Day								
100% Outfall 113	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.4	25.7	24.9	24.9	25.2	25.4	25.2		
Final									
D.O. Initial	7.5	7.3	7.2	7.4	7.9	7.0	6.9		
Final									
pH Initial	8.0	8.1	8.1	8.1	8.1	8.1	8.1		
Final		~~8.1~~							
Alkalinity	61		67		60				
Hardness	80		108		76				
Conductivity	168	160	164	161	163	167	172		
Residual Chlorine	0.07		0.09		0.05				
Sample Date	4/15	4/15	4/17	4/17	4/19	4/19	4/19		
Analyst Initials	RO	RO	SHS	RO	RO	SHS	RO		
QA Initials	SHS	SHS	RO	SHS	SHS	RO	SHS		

	Day								
Intake	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.1	25.8	24.9	25.8	25.8	25.1	25.6		
Final		25.3	24.9	25.1	25.8	25.0	24.4		
D.O. Initial	9.2*	8.1	9.3	8.6	9.1	7.4	7.2		
Final		5.8	6.4	6.6	6.4	5.7	6.5		
pH Initial	8.3	8.3	8.2	8.1	8.0	8.5	8.5		
Final		8.1	8.3	8.2	8.4	8.4	8.3		
Alkalinity	58		61		61				
Hardness	82		90		76				
Conductivity	156	162	157	158	154	154	149		
Residual Chlorine	0.08		0.06		0.04				
Sample Date	4/15	4/15	4/17	4/17	~~4/17~~	4/18	4/19		
Analyst Initials	RO	SHS	SHS	RO	RO	SHS	RO		
QA Initials	SHS	RO	RO	SHS	SHS	RO	SHS		

* day 0 - 4/14/03 - aerated sample for 10 minutes to reduce D.O.
- final D.O. after aeration = 7.8

TOTAL RESIDUAL CHLORINE ADJUSTMENT FOR MANGANESE INTERFERENCE

Facility Name	Sample Collection Date	Date Sample Received in Laboratory	Total Residual Chlorine Test Date/Initials	Total Residual Chlorine (mg/L)	Total Residual Chlorine after Manganese Adjustment
TVA Watts Bar Outfall 113	4/15/03	4/15/03	4/15/03 SAB	0.16	0.07
	4/17/03	4/17/03	4/17/03 SAB	0.13	0.09

*Used 30 g/L potassium iodide solution and 5 g/L sodium arsenite solution to adjust for manganese interference in accordance with the HACH Company Pocket Colorimeter procedures manual.

Watts Bar Nuclear Plant Biomonitoring
April 15 - 22, 2003.

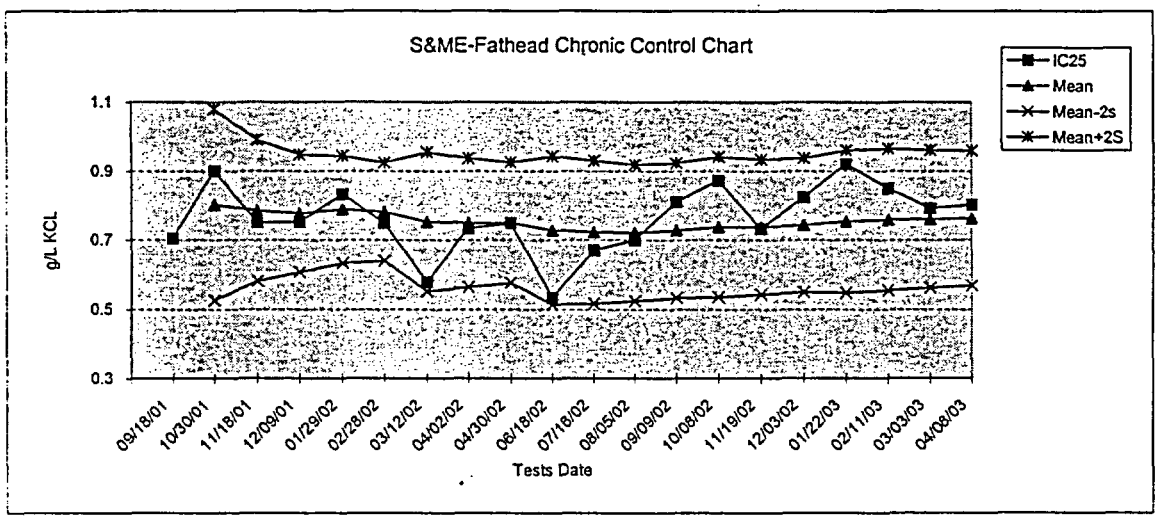
Appendix D

Reference Toxicant Tests and
Control Chart Information

SE - Fathead Chronic Control Chart

Updated April 21, 2003
Control Growth KCL Toxicity Test
Control Chart (PP)

Source	Date	IC25	Mean	S	2S	Mean-2s	Mean+2S	CV%
S&ME	09/18/01	0.7039						
S&ME	10/30/01	0.8997	0.8018	0.1385	0.2769	0.5249	1.0787	17.3
S&ME	11/18/01	0.7502	0.7846	0.1023	0.2047	0.5799	0.9893	13.0
S&ME	12/09/01	0.7514	0.7763	0.0852	0.1704	0.6059	0.9467	11.0
S&ME	01/29/02	0.8314	0.7873	0.0778	0.1556	0.6318	0.9429	9.9
S&ME	02/28/02	0.7485	0.7808	0.0714	0.1427	0.6381	0.9236	9.1
S&ME	03/12/02	0.5773	0.7518	0.1008	0.2016	0.5502	0.9534	13.4
S&ME	04/02/02	0.7327	0.7494	0.0936	0.1871	0.5623	0.9365	12.5
S&ME	04/30/02	0.7482	0.7493	0.0875	0.1750	0.5742	0.9243	11.7
S&ME	06/18/02	0.5319	0.7275	0.1074	0.2148	0.5127	0.9423	14.8
S&ME	07/16/02	0.6690	0.7222	0.1034	0.2068	0.5154	0.9290	14.3
S&ME	08/05/02	0.6990	0.7203	0.0988	0.1976	0.5226	0.9179	13.7
S&ME	09/09/02	0.8100	0.7272	0.0978	0.1956	0.5315	0.9228	13.5
S&ME	10/08/02	0.8720	0.7375	0.1016	0.2033	0.5342	0.9408	13.8
S&ME	11/19/02	0.7320	0.7371	0.0980	0.1959	0.5412	0.9331	13.3
S&ME	12/03/02	0.8230	0.7425	0.0970	0.1941	0.5484	0.9366	13.1
S&ME	01/22/03	0.9200	0.7530	0.1033	0.2067	0.5463	0.9597	13.7
S&ME	02/11/03	0.8480	0.7582	0.1027	0.2055	0.5528	0.9637	13.5
S&ME	03/03/03	0.7914	0.7600	0.1001	0.2003	0.5597	0.9602	13.2
S&ME	04/08/03	0.8020	0.7621	0.0979	0.1958	0.5663	0.9579	12.8



Potassium Chloride Chronic Reference Toxicant Control Chart
for Fathead Minnow
using Moderately Hard Synthetic Water

Test Number	Test date	7-d IC_{25} (g/L KCl)	CT (g/L KCl)	S	Control Limits CT - 2S	Control Limits CT + 2S	$S_{A.75}$	Warning Limits CT - $S_{A.75}$	Warning Limits CT + $S_{A.75}$	$S_{A.90}$	Control Limits CT - $S_{A.90}$	Control Limits CT + $S_{A.90}$	CV
1	09/18/01	0.7039											
2	10/30/01	0.8997	0.80	0.14	0.52	1.08	0.36	0.44	1.16	0.50	0.30	1.30	0.17
3	11/18/01	0.7502	0.78	0.10	0.58	0.99	0.35	0.43	1.14	0.49	0.30	1.27	0.13
4	12/09/01	0.7514	0.78	0.09	0.61	0.95	0.35	0.43	1.13	0.48	0.29	1.26	0.11
5	01/29/02	0.8314	0.79	0.08	0.63	0.94	0.35	0.43	1.14	0.49	0.30	1.28	0.10
6	02/28/02	0.7485	0.78	0.07	0.64	0.92	0.35	0.43	1.13	0.48	0.30	1.26	0.09
7	03/12/02	0.5773	0.75	0.10	0.55	0.95	0.34	0.41	1.09	0.47	0.29	1.22	0.13
8	04/02/02	0.7327	0.75	0.09	0.56	0.94	0.34	0.41	1.09	0.46	0.28	1.21	0.12
9	04/30/02	0.7482	0.75	0.09	0.57	0.92	0.34	0.41	1.09	0.46	0.28	1.21	0.12
10	06/18/02	0.5319	0.73	0.11	0.51	0.94	0.33	0.40	1.05	0.45	0.28	1.18	0.15
11	07/16/02	0.6690	0.72	0.10	0.52	0.93	0.32	0.40	1.05	0.45	0.27	1.17	0.14
12	08/05/02	0.6990	0.72	0.10	0.52	0.92	0.32	0.40	1.04	0.45	0.27	1.17	0.14
13	09/09/02	0.8100	0.73	0.10	0.53	0.92	0.33	0.40	1.05	0.45	0.28	1.18	0.13
14	10/08/02	0.8720	0.74	0.10	0.53	0.94	0.33	0.41	1.07	0.46	0.28	1.19	0.14
15	11/19/02	0.7320	0.74	0.10	0.54	0.93	0.33	0.41	1.07	0.46	0.28	1.19	0.13
16	12/03/02	0.8230	0.74	0.10	0.55	0.94	0.33	0.41	1.08	0.46	0.28	1.20	0.13
17	01/22/03	0.9200	0.75	0.10	0.55	0.96	0.34	0.41	1.09	0.47	0.29	1.22	0.14
18	02/11/03	0.8480	0.76	0.10	0.55	0.96	0.34	0.42	1.10	0.47	0.29	1.23	0.14
19	03/03/03	0.7914	0.76	0.10	0.56	0.96	0.34	0.42	1.10	0.47	0.29	1.23	0.13
20	04/08/03	0.8020	0.76	0.10	0.57	0.96	0.34	0.42	1.11	0.47	0.29	1.23	0.13

Note: 7-d IC_{25} = 7-day 25% inhibition concentration. An estimation of the concentration of sodium chloride that would cause a 25% reduction in Ceriodaphnia reproduction for the test population.

CT = Central tendency (mean IC_{25}).

S = Standard deviation of the IC_{25} values.

$S_{A.75}$ = Standard deviation corresponding to the 75th percentile CV. $S_{A.75}$ = 0.45, as determined by USEPA for the method and endpoint.

$S_{A.90}$ = Standard deviation corresponding to the 90th percentile CV. $S_{A.90}$ = 0.62, as determined by the USEPA for the method and endpoint.

CV = Coefficient of variation of the IC_{25} values.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Precision of Endpoint Measurements
Sodium Chloride Chronic Reference Toxicant Data for Fathead Minnows using Moderately Hard Synthetic Water

Test number	Test date	Control Survival (%)	Control Mean Growth (dry weight/larvae) (mg)	CV (%)	CT for Control Growth CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	09/18/01	95	0.839	7.5		0.1769	21.1	
2	10/30/01	97.5	0.705	10.1	8.8	0.147	20.9	21.0
3	11/18/01	100	0.850	8.6	8.7	0.1188	14.0	18.6
4	12/09/01	100	0.493	12.3	9.6	0.08	16.2	18.0
5	01/29/02	100	0.669	8.1	9.3	0.1115	16.7	17.8
6	02/28/02	97.5	0.723	9.9	9.4	0.2536	35.1	20.6
7	03/12/02	92.5	0.662	8.0	9.2	0.1316	19.9	20.5
8	04/02/02	95	0.813	10.4	9.4	0.1757	21.6	20.7
9	04/30/02	100	0.460	6.7	9.1	0.0789	17.1	20.3
10	06/18/02	97.5	0.696	9.2	9.1	0.1965	28.2	21.1
11	07/16/02	100	0.338	8.0	9.0	0.1407	41.6	22.9
12	08/05/02	90	0.538	6.2	8.8	0.1531	28.5	23.4
13	09/09/02	100	0.508	6.0	8.5	0.1542	30.4	23.9
14	10/08/02	97.5	0.715	11.0	8.7	0.1988	27.8	24.2
15	11/19/02	92.5	0.674	13.7	9.0	0.087	12.9	23.5
16	12/03/02	95	0.504	7.4	8.9	0.153	30.4	23.9
17	01/22/03	97.5	0.403	7.0	8.8	0.1422	35.3	24.6
18	02/11/03	92.5	0.555	8.1	8.8	0.1567	28.2	24.8
19	03/03/03	97.5	0.509	5.1	8.6	0.0673	13.2	24.2
20	04-08-03	90	0.434	10.9	8.7	0.1357	31.3	24.5

FATHEAD MINNOW TEST INFORMATION

Test Name: S&ME Chronic Reference Test

Test Conducted From: 4/8/03 (Day 0) To 4/15/03 (Day 7)

Sites/Concentrations:
1. Control
2. 250 mg/L
3. 500 mg/L
4. 750 mg/L
5. 1000 mg/L
6. 1500 mg/L

Stock (if applicable): _____

Control Water Type (✓):

Moderately Hard Water + Trace Minerals ☑

Other (describe): _____ ☐

Dilution Water Type (✓):

Moderately Hard Water + Trace Minerals ☑

Other (describe): _____ ☐

Source of Test Larvae (✓):

S&ME Lot # 03-220 ☑

Other (describe): _____ ☐

Date Larvae Hatched: 4/7 - 4/8/03 (4/7 - 4/8) Hatch Time: 1500 - 0310 Initials: BO

Record of Minor Test Non-Conformity

Date: _____

Description of Non-Conformity: _____

Initials: _____

Date: _____

Description of Non-Conformity: _____

Initials: _____

Test Log sheets QA Reviewed By: Ken S. Dotthet (Reviewer Initials) on 4/16/03 (Date)

Fathead Minnow Daily Test Information Logsheet

Test Name: Chronic Reference Test Test Dates: 4/8 - 4/15/03

Daily Test information	Temperature Information (25±1°C)			Feeding Information (feeding interval 6± 1h)		Test Initiation, Water Change, or Test Termination		Control Water Carboy No. and Additional comments	
Date and Initials	Incubator Temp. (°C)		Therm. No.	Fed 100 uL Brine Shrimp	Feeding time	Start Time	End Time		
	a.m.	p.m.						Carboy #	Date Prep.
Day 0 PO 4/8/03	25.0	25.0	1	✓	am: — pm: 1515	1300	1330	2	4/4/03
Day 1 PO 4/9/03	25.1	25.0	1	✓	am: ~~1315~~ 0915 pm: 1530	1400	1425	5	4/8/03
Day 2 PO 4/10/03	25.1	25.0	1	✓	am: ~~0900~~ 0915 pm: 1615	1400	1415	0	4/8/03
Day 3 SHS 4/11/03	25.1	25.2	1	✓	am: 0915 pm: 1600	1305	1325	5	4/9/03
Day 4 JM 4/12/03	25.0	25.0	1	✓	am: 1115 pm: 1645	1300	1330	5	4/11/03
Day 5 JM 4/13/03	25.0	25.0	1	✓	am: 1130 pm: 1700	1300	1325	5	4/11/03
Day 6 SHS 4/14/03	25.1	25.0	1	✓	am: 1000 pm: 1615	1300	1320	2	4/11/03
Day 7 SHS 4/15/03	25.0	25.0	1			1300		—	—

Client: S&ME Chronic Reference

Location: S&ME Laboratory

Analyst: BD, SHS, JM

Dates: 4/8 - 4/15/03

CUMULATIVE SURVIVAL OF P. PROMELAS IN CHRONIC TEST

			REPLICATE			
CONC.	Initials	DAY NO.	A	B	C	D
Control Mod. Hard	BD/SHS	1	10	10	10	10
	BD/SHS	2	10	10	10	10
	SHS	3	10	10	10	10
	JM	4	10	10	10	10
	JM	5	10	10	9	9
	BD/SHS	6	10	10	9	9
	SHS	7	9	10	9	8
250 mg/L (KCl)	BD/SHS	1	10	10	10	10
	BD/SHS	2	9	10	10	10
	SHS	3	9	10	10	10
	JM	4	9	10	10	10
	JM	5	9	10	10	10
	BD/SHS	6	8	9	10	9
	SHS	7	8	8	9	9
500 mg/L (KCl)	BD/SHS	1	10	10	10	10
	BD/SHS	2	10	10	10	10
	SHS	3	10	10	10	10
	JM	4	10	10	10	10
	JM	5	9	10	10	10
	BD/SHS	6	8	10	9	9
	SHS	7	7	10	8	7
750 mg/L (KCl)	BD/SHS	1	10	10	10	BD ~~9~~ 10
	BD/SHS	2	9	10	10	10
	SHS	3	8	10	9	9
	JM	4	8	10	8	9
	JM	5	8	9	8	8
	BD/SHS	6	8	9	7	7
	SHS	7	8	9	6	7
1000 mg/L (KCl)	BD/SHS	1	9	10	10	9
	BD/SHS	2	9	10	10	5
	SHS	3	SHS ~~8~~ 9	10	10	5
	JM	4	9	10	10	5
	JM	5	8	7	7	3
	BD/SHS	6	8	7	7	2
	SHS	7	6	7	7	1

			REPLICATE			
CONC.	Initials	DAY NO.	A	B	C	D
1500 mg/L (KCl)	BD/SHS	1	5	5	10	6
	BD/SHS	2	1	0	4	0
	SHS	3	0	0	0	0
	JM	4	0	0	0	0
	JM	5	0	0	0	0
	SHS	6	0	0	0	0
	SHS	7	0	0	0	0
		1				
		2				
		3				
		4				
		5				
		6				
		7				
		1				
		2				
		3				
		4				
		5				
		6				
		7				
		1				
		2				
		3				
		4				
		5				
		6				
		7				
		1				
		2				
		3				
		4				
		5				
		6				
		7				

FATHEAD MINNOW SEVEN DAY TEST
RAW DATA
SURVIVAL AND GROWTH

Client: S&ME Reference Test
Test Conducted By: BG, BH, JM
Date Weighed 4/16/03

Test Dates: 4/8-4/15/03
Comments:
Began drying (date/time) 4/15/03 1400
End drying (date/time) 4/16/03 1000

Oven Temp (°C) 107.5°C

Exposure & Repl.	Rep.	# Live Fish	# Original Fish	Pan Tare Weight (mg)	Dry Weight Pan + Fish (mg)	Groups Dry Weight (Pan + Fish minus Tare) (mg)	Mean Dry Weight (mg)
Control	A	9	10	1294.30	1298.05	3.75	0.375
	B	10	10	1291.22	1295.60	4.38	0.438
	C	9	10	1297.39	1302.30	4.91	0.491
	D	8	10	1297.00	1301.32	4.32	0.432
250 mg/L	A	8	10	1288.79	1293.31	4.52	0.452
	B	8	10	1290.03	1293.30	3.27	0.327
	C	9	10	1291.58	1294.68	3.10	0.310
	D	9	10	~~1286.43~~ 1288.42	1288.79	2.37	0.237
500 mg/L	A	7	10	1286.18	1289.21	3.03	0.303
	B	10	10	1295.35	1300.07	4.72	0.472
	C	8	10	1288.58	1292.17	~~3.59~~ 3.59	0.359
	D	7	10	1293.80	1296.28	2.48	0.248
750 mg/L	A	8	10	1294.10	1298.83	4.73	0.473
	B	9	10	1286.88	1289.36	2.48	0.248
	C	6	10	1291.29	1294.86	3.57	0.357
	D	7	10	1279.76	1283.04	3.28	0.328
1000 mg/L	A	6	10	1290.94	1294.35	3.41	0.341
	B	7	10	1292.41	1295.63	3.22	0.322
	C	7	10	1298.36	1301.87	3.51	0.351
	D	1	10	1293.36	1293.58	0.22	0.022

Larval Fish Growth and Survival Test-7 Day Survival

Start Date:	4/8/03 13:00	Test ID:	ref0403fs
End Date:	4/15/03 13:00	Lab ID:	S&ME
Sample Date:		Protocol:	EPAF 91-EPA Freshwater
Comments:			
Sample ID:		REF-Ref Toxicant	
Sample Type:		KCL-Potassium chloride	
Test Species:		PP-Pimephales promelas	

Conc-mg/L	1	2	3	4
D-Control	0.9000	1.0000	0.9000	0.8000
250	0.8000	0.8000	0.9000	0.9000
500	0.7000	1.0000	0.8000	0.7000
750	0.8000	0.9000	0.6000	0.7000
1000	0.6000	0.7000	0.7000	0.1000
1500	0.0000	0.0000	0.0000	0.0000

			Transform: Arcsin Square Root					1-Tailed		
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	0.9000	1.0000	1.2543	1.1071	1.4120	9.935	4			
250	0.8500	0.9444	1.1781	1.1071	1.2490	6.954	4	0.554	2.360	0.3246
500	0.8000	0.8889	1.1254	0.9912	1.4120	17.662	4	0.938	2.360	0.3246
750	0.7500	0.8333	1.0584	0.8861	1.2490	14.733	4	1.425	2.360	0.3246
*1000	0.5250	0.5833	0.7975	0.3218	0.9912	40.253	4	3.321	2.360	0.3246
1500	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4			

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.94207	0.868	-0.8093	1.72125
Bartlett's Test indicates equal variances (p = 0.25)	5.33512	13.2767		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	750	1000	866.025		0.26077	0.28874	0.12228	0.03782	0.04222	4, 15

Larval Fish Growth and Survival Test-7 Day Growth

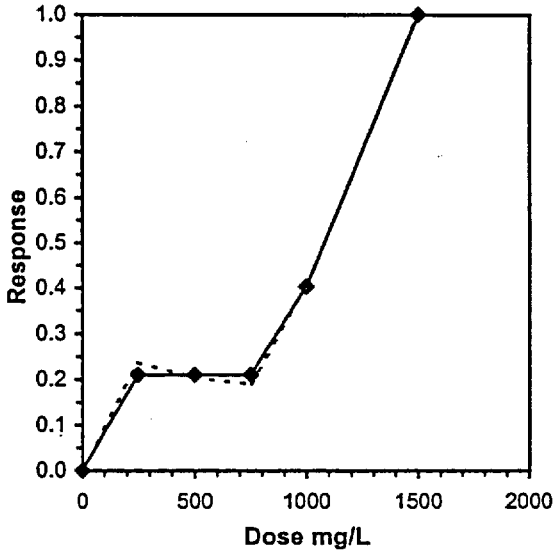
Start Date:	4/8/03 13:00	Test ID:	ref0403fg	Sample ID:	REF-Ref Toxicant
End Date:	4/15/03 13:00	Lab ID:	S&ME	Sample Type:	KCL-Potassium chloride
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-mg/L	1	2	3	4
D-Control	0.3750	0.4380	0.4910	0.4320
250	0.4520	0.3270	0.3100	0.2370
500	0.3030	0.4720	0.3590	0.2480
750	0.4730	0.2480	0.3570	0.3280
1000	0.3410	0.3220	0.3510	0.0220
1500	0.0000	0.0000	0.0000	0.0000

Linear Interpolation (80 Resamples)

Point	mg/L	SD	95% CL(Exp)		Skew
IC05*	59.5064	108.319	23.9033	806.72	2.8051
IC10*	119.013	181.187	47.8067	1137.14	2.0208
IC15*	178.519	245.365	71.71	1172.32	1.1021
IC20*	238.026	293.221	95.6134	1479.06	0.1239
IC25	801.69	301.38	0	1189.49	-0.3674
IC40	995.825	122.377	540.058	1218.91	-1.3704
IC50	1081.08	92.9263	770.357	1265.02	-0.7196

* indicates IC estimate less than the lowest concentration



Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	4/8/03 13:00	Test ID:	ref0403fg	Sample ID:	REF-Ref Toxicant
End Date:	4/15/03 13:00	Lab ID:	S&ME	Sample Type:	KCL-Potassium chloride
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-mg/L	1	2	3	4
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250	0.4520	0.3270	0.3100	0.2370
500	0.3030	0.4720	0.3590	0.2480
750	0.4730	0.2480	0.3570	0.3280

			Transform: Untransformed					1-Tailed		
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	0.4340	1.0000	0.4340	0.3750	0.4910	10.930	4			
250	0.3315	0.7638	0.3315	0.2370	0.4520	26.944	4	1.730	2.290	0.1357
500	0.3455	0.7961	0.3455	0.2480	0.4720	27.710	4	1.494	2.290	0.1357
750	0.3515	0.8099	0.3515	0.2480	0.4730	26.514	4	1.393	2.290	0.1357

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.9149		0.844		0.46543	-0.5616
Bartlett's Test indicates equal variances (p = 0.71)					1.39661		11.3449			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	750	>750			0.13567	0.3126	0.00859	0.00702	0.34353	3, 12

pmsd = 31.3

Daily Chemistry Data - Fathead Minnow 7-day Chronic

Client: S&ME Reference Test Analysts: BD, SHS, JM

Location: S&ME Inc. Laboratory Dates/times: 4/8 1305 4/15/03/1300

Template No.: 1 Age of larvae: ~~24hrs~~ 24hrs

	Day								
Control: (Mod. Hard)	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.6	24.3	25.9	24.4	25.4	25.8	25.8		
Final		24.3	24.6	24.3	25.0	25.2	25.8	25.2	
D.O. Initial	6.9	6.8	6.9	6.8	7.1	7.0	6.6		
Final		6.0	6.8	6.2	6.0	6.5	5.6	5.4	
pH Initial	7.9	7.9	7.9	8.0	8.1	8.1	8.0		
Final		7.6	7.5	7.4	7.6	7.9	7.6	7.5	
Alkalinity	62	62	59	57	62	/	63		
Hardness	98	96	96	98	98	/	100		
Conductivity	393	369	374	399	386	389	395		
Analyst Initials	BD	BD	BD	SHS	JM	JM	SHS	BD	
QA Review Initials	SHS	SHS	SHS	BD	BD	BD	BD	SHS	

	Day								
250 mg/L (KCl)	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.9	24.3	~~25.8~~	24.4	25.5	25.8	25.6		init. temp day 2 = 25.8
Final		24.1	24.2	24.3	25.0	25.3	25.4	25.2	
D.O. Initial	6.5	6.6	6.9	6.8	7.0	6.5	6.6		
Final		5.9	6.0	6.0	5.8	6.0	5.5	5.3	
pH Initial	8.0	7.9	8.0	8.1	8.1	8.2	8.0		
Final		7.6	7.7	7.5	7.6	7.8	7.6	7.5	
Conductivity	811	813	806	855	831	831	850		
Analyst Initials	BD	BD	BD	SHS	JM	JM	SHS	BD	
QA Review Initials	SHS	SHS	SHS	BD	BD	BD	BD	SHS	

	Day								
500 mg/L (KCl)	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.9	24.3	25.8	24.5	25.5	25.7	25.6		
Final		24.1	24.5	24.5	25.2	25.2	25.4	25.2	
D.O. Initial	6.4	6.4	6.8	6.7	6.8	6.3	6.6		
Final		5.9	6.4	6.0	5.4	5.8	5.6	5.5	
pH Initial	8.0	8.0	8.0	8.0	8.1	8.2	8.0		
Final		7.7	7.7	7.4	7.6	7.8	7.6	7.6	
Conductivity	1258	1266	1257	1243	1246	1256	1260		
Analyst Initials	BD	BD	BD	SHS	JM	JM	SHS	BD	
QA Review Initials	SHS	SHS	SHS	BD	BD	BD	BD	SHS	

NOTES:

Daily Chemistry Data - Fathead Minnow 7-day Chronic

Client: S&ME Reference Test — Analysts: SHS, BD

Location: S&ME Inc. Laboratory — Dates/times: 4/8/03 1300 - 4/15/03 1300

Template No.: 1 — Age of larvae: <24 hrs

	Day								
750 mg/L (KCl)	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.8	24.3	25.8	24.6	25.6	25.8	25.6		
Final		24.4	24.4	24.6	25.1	25.3	25.3	25.2	
D.O. Initial	6.5	6.6	6.7	6.6	6.8	6.1	6.3		
Final		5.8	6.2	5.9	5.8	5.6	5.5	5.6	
pH Initial	8.0	8.0	8.0	8.0	8.1	8.2	8.0		
Final		7.8	7.7	7.5	7.7	7.7	7.6	7.5	
Conductivity	1581	1563	1549	1652	1667	1648	1684		
Analyst Initials	SHS	BD	BD	SHS	JF	JF	SHS	BD	
QA Review Initials	BD	SHS	SHS	BD	BD	BD	BD	SHS	

	Day								
1000 mg/L (KCl)	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.8	24.3	25.8	24.6	25.6	25.8	25.6		
Final		24.5	24.5	24.6	25.0	25.3	25.3	24.9	
D.O. Initial	6.5	6.5	6.7	6.6	6.9	5.9	6.3		
Final		6.0	6.1	5.7	6.0	5.7	5.4	5.4	
pH Initial	8.0	8.0	8.0	8.0	8.1	8.2	8.0		
Final		7.8	7.7	7.4	7.7	7.8	7.6	7.5	
Conductivity	1975	1967	1925	2050	2050	2030	2090		
Analyst Initials	BD	BD	BD	SHS	JF	JF	SHS	BD	
QA Review Initials	SHS	SHS	SHS	BD	BD	BD	BD	SHS	

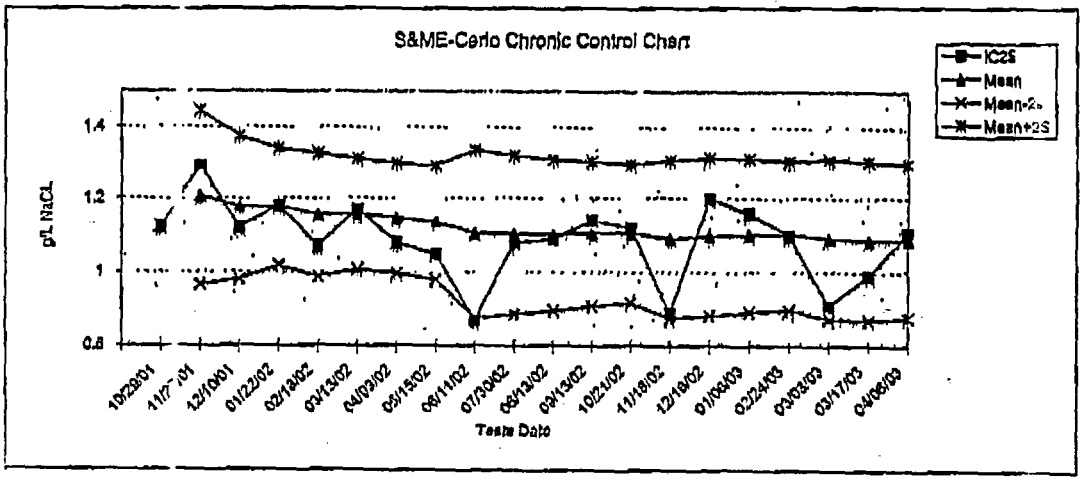
	Day								
1500 mg/L (KCl)	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.9	24.4	25.8	24.7	/	/	/		
Final		24.4	24.6	24.8	/	/	/	/	
D.O. Initial	6.5	6.5	6.5	6.6	/	/	/		
Final		6.0	6.2	5.7	/	/	/	/	
pH Initial	8.0	8.0	8.0	8.0	/	/	/		
Final		7.8	7.6	7.6	/	/	/	/	
Conductivity	2710	2780	2710	2940	/	/	/		
Analyst Initials	BD	BD	BD	SHS	JF	/	/	/	
QA Review Initials	SHS	SHS	SHS	BD		/	/	/	
Stock Solution Date	4/8	4/8	4/8	4/8	4/8	4/8		/	

NOTES:

S&ME-CD Chronic Control Chart

Updated April 15, 2003
IC25 NaCL Toxicity Test
Control Chart (CD)

Source	Date	IC25	Mean	S	2S	Mean-2s	Mean+2S	CV%
S&ME	10/29/01	1.1200						
S&ME	11/27/01	1.2900	1.2050	0.1202	0.2404	0.9646	1.4454	10.0
S&ME	12/10/01	1.1200	1.1767	0.0981	0.1963	0.9804	1.3730	8.3
S&ME	01/22/02	1.1800	1.1775	0.0802	0.1603	1.0172	1.3378	6.8
S&ME	02/18/02	1.0700	1.1560	0.0844	0.1689	0.9871	1.3249	7.3
S&ME	03/13/02	1.1700	1.1583	0.0757	0.1515	1.0069	1.3098	6.5
S&ME	04/03/02	1.0800	1.1471	0.0752	0.1504	0.9967	1.2976	6.6
S&ME	05/15/02	1.0500	1.1350	0.0776	0.1553	0.9797	1.2903	6.8
S&ME	06/11/02	0.8700	1.1056	0.1144	0.2287	0.8768	1.3343	10.3
S&ME	07/30/02	1.0800	1.1030	0.1081	0.2162	0.8868	1.3192	9.8
S&ME	08/13/02	1.0900	1.1018	0.1026	0.2053	0.8965	1.3071	9.3
S&ME	09/13/02	1.1400	1.1050	0.0985	0.1970	0.9080	1.3020	8.9
S&ME	10/21/02	1.1200	1.1062	0.0944	0.1888	0.9174	1.2949	8.5
S&ME	11/18/02	0.8900	1.0907	0.1075	0.2150	0.8757	1.3058	9.9
S&ME	12/16/02	1.2000	1.0980	0.1074	0.2148	0.8832	1.3128	9.8
S&ME	01/08/03	1.1600	1.1019	0.1049	0.2098	0.8921	1.3117	9.5
S&ME	02/24/03	1.1000	1.1013	0.1016	0.2031	0.8986	1.3049	9.2
S&ME	03/03/03	0.9100	1.0911	0.1084	0.2168	0.8743	1.3079	9.9
S&ME	03/17/03	0.9900	1.0858	0.1079	0.2157	0.8700	1.3015	9.9
S&ME	04/08/03	1.1066	1.0868	0.1051	0.2102	0.8766	1.2970	9.7



Sodium Chloride Chronic Reference Toxicant Data for *Ceriodaphnia dubia* using Moderately Hard Synthetic Water

Test number	Test date	Control Mean Reproduction (offspring/female)	CV (%)	CT for Control Reproduction CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	10-29-01	27.7	10.1		3.0	10.8	
2	11-27-01	23.2	8.6	8.6	4.3	18.5	18.5
3	12-10-01	24.8	12.3	10.5	2.1	8.5	13.5
4	01-22-02	23.5	8.1	9.7	4.3	18.3	15.1
5	02-18-02	17.3	9.9	9.7	6.1	35.3	20.1
6	03-13-02	23.3	8.0	9.4	4.7	20.2	20.1
7	04-03-02	24.6	10.4	9.6	3.2	13.0	19.0
8	05-15-02	28.6	6.7	9.2	6.2	21.7	19.3
9	06-11-02	28.7	9.2	9.2	6.1	21.3	19.6
10	07-30-02	20.6	8.0	9.0	5.0	24.3	20.1
11	08-13-02	29.7	6.2	8.7	6.2	20.9	20.2
12	09-13-02	28.8	6.0	8.5	5.0	17.4	19.9
13	10-21-02	25.4	11.0	8.7	5.5	21.7	20.1
14	11-18-02	29.3	13.7	9.1	9.6	32.8	21.0
15	12-16-02	24.9	7.4	9.0	3.6	14.5	20.6
16	01-08-03	20.8	7.0	8.8	4.0	19.2	20.5
17	02-24-03	26.8	8.1	8.8	4.0	14.9	20.1
18	03-03-03	31.4	5.1	8.6	6.2	19.7	20.1
19	03-17-03	29.0	6.2	8.4	5.6	19.3	20.1
20	04-08-03	24.9	21.5	9.1	4.5	18.1	20.0

Sodium Chloride Chronic Reference Toxicant Control Chart
for *Ceriodaphnia dubia*
using Moderately Hard Synthetic Water

Test Number	Test date	7-d IC_{25} (g/L NaCl)	CT (g/L NaCl)	S	Control Limits CT-2S	Control Limits CT+2S	$S_{A.75}$	Warning Limits CT-$S_{A.75}$	Warning Limits CT+$S_{A.75}$	$S_{A.90}$	Control Limits CT-$S_{A.90}$	Control Limits CT+$S_{A.90}$	CV
1	10-29-01	1.120											
2	11-27-01	1.290	1.205	0.12	0.96	1.45	0.54	0.66	1.75	0.75	0.46	1.95	0.10
3	12-10-01	1.120	1.177	0.10	0.98	1.37	0.53	0.65	1.71	0.73	0.45	1.91	0.08
4	01-22-02	1.180	1.178	0.08	1.02	1.34	0.53	0.65	1.71	0.73	0.45	1.91	0.07
5	02-18-02	1.070	1.156	0.08	0.99	1.32	0.52	0.64	1.68	0.72	0.44	1.87	0.07
6	03-13-02	1.170	1.158	0.08	1.01	1.31	0.52	0.64	1.68	0.72	0.44	1.88	0.07
7	04-03-02	1.080	1.147	0.08	1.00	1.30	0.52	0.63	1.66	0.71	0.44	1.86	0.07
8	05-15-02	1.050	1.135	0.08	0.98	1.29	0.51	0.62	1.65	0.70	0.43	1.84	0.07
9	06-11-02	0.870	1.106	0.11	0.88	1.33	0.50	0.61	1.60	0.69	0.42	1.79	0.10
10	07-30-02	1.080	1.103	0.11	0.89	1.32	0.50	0.61	1.60	0.68	0.42	1.79	0.10
11	08-13-02	1.090	1.102	0.10	0.90	1.31	0.50	0.61	1.60	0.68	0.42	1.78	0.09
12	09-13-02	1.140	1.105	0.10	0.91	1.30	0.50	0.61	1.60	0.69	0.42	1.79	0.09
13	10-21-02	1.120	1.106	0.09	0.92	1.29	0.50	0.61	1.60	0.69	0.42	1.79	0.09
14	11-18-02	0.890	1.091	0.11	0.88	1.31	0.49	0.60	1.58	0.68	0.41	1.77	0.10
15	12-16-02	1.200	1.098	0.11	0.88	1.31	0.49	0.60	1.59	0.68	0.42	1.78	0.10
16	01-08-03	1.160	1.102	0.10	0.89	1.31	0.50	0.61	1.60	0.68	0.42	1.79	0.10
17	02-24-03	1.100	1.102	0.10	0.90	1.30	0.50	0.61	1.60	0.68	0.42	1.78	0.09
18	03-03-03	0.910	1.091	0.11	0.87	1.31	0.49	0.60	1.58	0.68	0.41	1.77	0.10
19	03-17-03	0.990	1.086	0.11	0.87	1.30	0.49	0.60	1.57	0.67	0.41	1.76	0.10
20	04-08-03	1.107	1.087	0.11	0.88	1.30	0.49	0.60	1.58	0.67	0.41	1.76	0.10

Note: 7-d IC_{25} = 7-day 25% inhibition concentration. An estimation of the concentration of sodium chloride that would cause a 25% reduction in Ceriodaphn reproduction for the test population.

CT = Central tendency (mean IC_{25}).

S = Standard deviation of the IC_{25} values.

$S_{A.75}$ = Standard deviation corresponding to the 75th percentile CV. $S_{A.75}$ = 0.45, as determined by USEPA for the method and endpoint.

$S_{A.90}$ = Standard deviation corresponding to the 90th percentile CV. $S_{A.90}$ = 0.62, as determined by the USEPA for the method and endpoint.

CV Coefficient of variation of the IC_{25} values.

USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

CERIODAPHNIA TEST INFORMATION SHEET

Test Name: Chronic Reference Test

Test Conducted From: 4/8/03 (Day 0) To 4/15/03 (Day 7)

Sites/Concentrations: 1. control 4. 1000mg/L
2. 500mg/L 5. 1500mg/L
3. 750mg/L 6. 8000mg/L

Stock (if applicable): _____

Control Water Type (✓):

20% Dilute Mineral Water + Trace Minerals ☑

Other (describe): _____ ☐

Dilution Water Type (✓):

20% Dilute Mineral Water + Trace Minerals ☑

Other (describe): _____ ☐

Source of Test Organisms: S&ME brood board nos. 241

Age of Test Organisms:

Isolated neonates for test on 4/8/03 (date) from 0815 to 1255 (time) Initials BO

Record of Minor Test Non-Conformity:

Date: _____

Description of Non-Conformity: _____

Initials: _____

Date: _____

Description of Non-Conformity: _____

Initials: _____

Test Log sheets QA Reviewed By: Lisa S. Dunbar (Reviewer Initials) on 4/15/03 (Date)

REV.01, January 01 (ceriodaphnia test info sheet)

Ceriodaphnia Daily Test Information Logsheet

Test Name: S&ME Chronic Reference Test Test Dates: 4/8 - 4/15/03

Daily Test Information	Temperature Information (cup -25±1°C)			Feeding Information				Test Initiation, Water Change, or Test Termination		Control Water Carboy No. and Additional comments	
Date and Initials	Incubator Temp. (°C)		Therm. No.	Fed 100 uL YCT	YCT Date	Fed 100 uL Selenastrum	Selanastrum Date	Start Time	End Time	Carboy #	Date Prep.
	a.m.	p.m.									
Day 0 RO 4/8/03	25.0	25.0	1	1405	2/5/03	1405	3/24/03	1345	1405	3	4/4/03
Day 1 MO 4/9/03	25.1	25.0	1	1420	2/5/03	1420	3/24/03	1410	1420	3	4/4/03
Day 2 RO 4/10/03	25.1	25.0	1	1445	2/5/03	1445	3/24/03	1430	1445	3	4/9/03
Day 3 SJS 4/11/03	25.1	25.2	1	1350	2/5/03	1350	3/24/03	1330	1350	3	4/9/03
Day 4 JM 4/12/03	25.0	25.0	1	1355	2/5/03	1355	3/24/03	1330	1355	3	4/9/03
Day 5 JM 4/13/03	25.0	25.0	1	1405	2/5/03	1405	3/24/03	1335	1405	3	4/9/03
Day 6 SJS 4/14/03	25.1	25.0	1	1340	2/5/03	1340	3/24/03	1315	1340	3	4/13/03
Day 7 RO 4/15/03	25.0	25.0	1					1330		—	—

Ceriodaphnia 3-Brood Survival and Reproduction
Raw Data Sheet

Client: Chronic Reference Test
Location: S&ME Inc.
Analysts: BD, SAB, JM
Test Dates: 4/8 – 4/15/03

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
			1	2	3	4	5	6	7	8	9	10				
Conc.	Init.	Day	--	--	--	--	--	--	--	--	--	--	---	---	---	---
	BD	1	0										0	10	10	0
	RB	2	0										0	10	10	0
Control	SAB	3	0										0	10	10	0
	JM	4	6	2	5	5	5	2	4	4	6	4	43	10	10	4.3
	JM	5	11	9	10	9	10	9	8	7	9	6	88	10	10	8.8
	SAB	6	0	0	0	0	11	0	0	0	9	8	28	10	10	2.8
	JM	7	20	12	10	12	0	13	16	7# JM	0	0	90	10	10	9.0
		Total	37	23	25	26	26	24	28	18	24	18	249	10	10	24.9

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
			1	2	3	4	5	6	7	8	9	10				
	Init.	Day	--	--	--	--	--	--	--	--	--	--	---	---	---	---
	BD	1	0										0	10	10	0
	RB	2	0										0	10	10	0
500	SAB	3	0										0	10	10	0
mg/L	JM	4	3	5	4	5	5	4	5	3	5	4	43	10	10	4.3
NaCl	JM	5	11	11	7	8	9	8	8	7	12	10	91	10	10	9.1
	SAB	6	14	0	0	10	0	0	[illegible]	12	14	9	59	10	10	5.9
	JM	7	12	12	10	0	14	14	7	0	0	0	57	10	10	5.7
		Total	28	28	21	23	28	26	20	22	31	23	250	10	10	25.0

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
			1	2	3	4	5	6	7	8	9	10				
	Init.	Day	--	--	--	--	--	--	--	--	--	--	---	---	---	---
	BD	1	0										0	10	10	0
	RB	2	0										0	10	10	0
750	SAB	3	0										0	10	10	0
mg/L	JM	4	5	6	5	6	6	5	4	5	5	3	50	10	10	5.0
NaCl	JM	5	10	9	11	10	10	9	10	10	8	9	96	10	10	9.6
	SAB	6	12	15	10	12	12	12	0	9#	12	14	~~98~~	10	10	~~10.8~~ 9.8 JM
	JM	7	0	3+	0	0	0	0	17	19	3+	0	42	10	10	4.2
		Total	27	33	26	28	28	26	31	34	28	26	287	10	10	28.7

+ = split brood JM

Ceriodaphnia 3-Brood Survival and Reproduction
Raw Data Sheet

Client: Chronic Reference Test
Location: S&ME Inc.
Analysts: BD, SLS, JM Test Dates: 4/8 - 4/15/03

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
		1	0	→									0	10	10	0
		2	0	→									0	10	10	0
		3	0	→									0	10	10	0
1000	JM	4	4	1+	2	6	4	2	4	1+	3	3	30	10	10	3.0
mg/L	JM	5	8	8	10	9	7	5	8	7	4	5	71	10	10	7.1
NaCl	BD	6	10	9	13	15	12	14	15	13	13	9	123	10	10	12.3
	JM	7	0	0	0	0	0	0	0	3+	0	0	3	10	10	0.3
	BD	Total	22	18	25	30	23	21	27	24	20	17	227	10	10	22.7

+ 3 prior brood JM

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
		1	0	→									0	10	10	0
		2	0	→									0	10	10	0
		3	0	→									0	10	10	0
1500	JM	4	4	1	OX	1X	8	2	3	2X	0	3	31	8	10	3.0
mg/L	JM	5	2	1	1	3	1	0	0	1	X	1	8	7	10	0.8
NaCl	BD	6	3	7	1	4	3	6	10	1	1	0	33	7	10	3.3
	JM	7	0	3	1	0	9	8	7	1	1	8	32	7	10	3.2
				9	↓					↓	↓					
	BD	Total	9	25	0	8	13	14	17	2	0	12	84	7	10	8.4

* Did not count these - all brood

			Replicate										No. of Young	No. of live Adults	No. of Original Adults	Young Per Adult
	Init.	Day	1	2	3	4	5	6	7	8	9	10				
		1	0								S		0	10	10	0
		2	X	X	X	X	0	0	X	X	X	0	0	3	10	0
		3	↓	↓	↓	↓	0	0	↓	↓	↓	0	0	0	10	0
2000		4					↓	↓				↓				
mg/L		5														
NaCl		6														
		7														
			↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓
		Total	0	0	0	0	0	0	0	0	0	0				

Ceriodaphnia Survival and Reproduction Test-7 Day Survival

Start Date:	4/8/2003 13:45	Test ID: Ref403cr	Sample ID: REF-Ref Toxicant
End Date:	4/15/2003 13:30	Lab ID: S&ME INC.	Sample Type: NACL-Sodium chloride
Sample Date:		Protocol: EPAF 91-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia
Comments:			

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
500	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
750	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
1000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
1500	1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	1.0000
2000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000

Conc-mg/L	Mean	N-Mean	Resp	Not Resp	Total	N	Fisher's Exact P	1-Tailed Critical
D-Control	1.0000	1.0000	0	10	10	10		
500	1.0000	1.0000	0	10	10	10	1.0000	0.0500
750	1.0000	1.0000	0	10	10	10	1.0000	0.0500
1000	1.0000	1.0000	0	10	10	10	1.0000	0.0500
1500	0.7000	0.7000	3	7	10	10	0.1053	0.0500
2000	0.0000	0.0000	10	0	10	10		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Fisher's Exact Test	1500	2000	1732.05	

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	4/8/2003 13:45	Test ID: Ref403cr	Sample ID: REF-Ref Toxicant
End Date:	4/15/2003 13:30	Lab ID: S&ME INC.	Sample Type: NACL-Sodium chloride
Sample Date:		Protocol: EPAF 91-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia
Comments:			

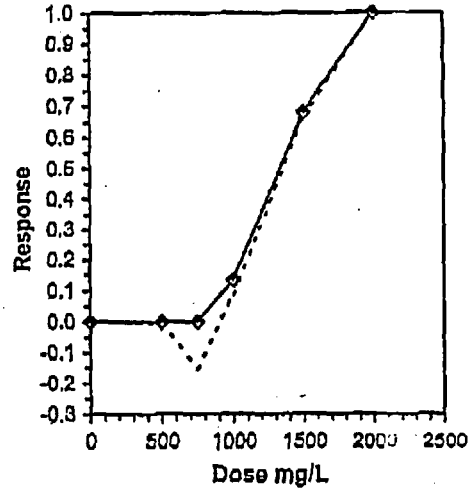
Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	37.000	23.000	25.000	26.000	26.000	24.000	26.000	16.000	24.000	18.000
500	28.000	28.000	21.000	23.000	28.000	28.000	20.000	22.000	31.000	23.000
750	27.000	33.000	26.000	28.000	28.000	26.000	31.000	34.000	28.000	26.000
1000	22.000	18.000	25.000	30.000	23.000	21.000	27.000	24.000	20.000	17.000
1500	9.000	9.000	0.000	8.000	13.000	14.000	17.000	2.000	0.000	12.000
2000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	24.900	1.0000	24.900	18.000	37.000	21.540	10				26.200	1.0000
500	25.000	1.0040	25.000	20.000	31.000	14.727	10	-0.049	2.223	4.509	26.200	1.0000
750	28.700	1.1526	28.700	26.000	34.000	10.264	10	-1.874	2.223	4.509	26.200	1.0000
1000	22.700	0.9116	22.700	17.000	30.000	17.627	10	1.085	2.223	4.509	22.700	0.8664
*1500	8.400	0.3373	8.400	0.000	17.000	71.252	10	8.136	2.223	4.509	8.400	0.3206
2000	0.000	0.0000	0.000	0.000	0.000	0.000	10				0.000	0.0000

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.98276		0.93		0.27392	0.26416
Bartlett's Test indicates equal variances (p = 0.24)					5.51255		13.2767			
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test:	1000	1500	1224.74		4.50922	0.18109	619.33	20.5667	3.3E-12	4, 45

Linear Interpolation (80 Resamples)

Point	mg/L	SD	95% CL		Skew
IC05	843.571	92.6739	775.866	1010.87	-2.5223
IC10	937.143	64.3679	837.834	1048.29	0.2867
IC15	1015.03	55.1616	899.347	1085.65	-0.1638
IC20	1080.84	48.5488	949.14	1126.69	-0.2786
IC25	1106.64	45.1459	999.048	1173.16	-0.2554
IC40	1244.06	39.1017	1166.64	1320.39	0.1802
IC50	1335.66	42.3373	1253.65	1431.73	0.2024



Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	4/8/2003 13:45	Test ID: Ref403or	Sample ID: REF-Ref Toxicant
End Date:	4/15/2003 13:30	Lab ID: S&ME INC.	Sample Type: NACL-Sodium chloride
Sample Date:		Protocol: EPAF 91-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia
Comments:			

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	37.000	23.000	25.000	26.000	26.000	24.000	28.000	18.000	24.000	18.000
500	28.000	28.000	21.000	23.000	28.000	26.000	20.000	22.000	31.000	23.000
750	27.000	33.000	26.000	28.000	28.000	26.000	31.000	34.000	28.000	26.000
1000	22.000	18.000	25.000	30.000	23.000	21.000	27.000	24.000	20.000	17.000
1500	9.000	9.000	0.000	8.000	13.000	14.000	17.000	2.000	0.000	12.000

			Transform: Untransformed						1-Tailed	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	24.900	1.0000	24.900	18.000	37.000	21.540	10			
500	25.000	1.0040	25.000	20.000	31.000	14.727	10	-0.049	2.223	4.509
750	28.700	1.1526	28.700	26.000	34.000	10.264	10	-1.874	2.223	4.509
1000	22.700	0.9116	22.700	17.000	30.000	17.627	10	1.085	2.223	4.509
*1500	8.400	0.3373	8.400	0.000	17.000	71.252	10	8.136	2.223	4.509

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.98276		0.93		0.27392	0.26416
Bartlett's Test indicates equal variances (p = 0.24)					5.51255		13.2767			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	1000	1500	1224.74		4.50922	0.18109	619.33	20.5667	3.3E-12	4, 45

Daily Chemistry Data - Ceriodaphnia 7-day Chronic

Client: Reference Test
Location: S&ME Inc.
Template No.: 1

Analysts: BO, SAS, JM
Dates/times: 4/18-24/15/03/1330
Age of Neonates: <8 hrs

Day									
Control: (20% DMW)	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.8	24.4	25.1	24.1	25.7	25.8	25.6		
Final		24.5	24.5	24.1	24.8	24.7	25.8	25.9	
D.O. Initial	6.4	6.5	6.8	6.6	7.1	6.6	6.4		
Final		6.3	6.5	6.3	6.9	6.6	5.6	6.1	
pH Initial	8.0	7.9	8.0	7.9	8.0	8.1	8.0		
Final		8.2	8.2	8.1	8.2	8.2	7.9	8.0	
Alkalinity	59	/	59	/	/	/	58		
Hardness	90	/	82	/	/	/	80		
Conductivity	174	173	170	168	167	168	174		✓checked against LCL & UCL LM QA
Analyst Initials	BO	BO	BO	SAS	JM	JM	SAS	SAS	
QA Review Initials	SAS	SAS	SAS	BO	BO	BO	BO	LO	

Day									
500 mg/L	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.8	24.2	25.1	24.1	25.9	25.8	25.6		
Final		24.7	24.3	24.1	25.0	24.7	25.9	25.9	
D.O. Initial	6.5	6.6	6.8	6.6	6.8	6.5	6.3		
Final		6.2	6.4	6.3	6.8	6.5	5.6	6.1	
pH Initial	8.0	8.0	8.0	7.9	8.1	8.1	7.9		
Final		8.1	8.2	8.0	8.1	8.1	7.9	8.0	
Conductivity	1068	1154	1127	1181	1089	1089	1135		✓checked against UCL & LCL LM QA
Analyst Initials	BO	BO	BO	SAS	JM	JM	SAS	SAS	
QA Review Initials	SAS	SAS	SAS	BO	BO	BO	BO	LO	

Day									
750 mg/L	0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.8	24.2	25.2	24.2	25.9	25.7	25.6		
Final		24.7	24.3	24.2	25.0	24.8	25.9	25.9	
D.O. Initial	6.4	6.5	6.5	6.5	6.6	6.5	6.4		
Final		6.2	6.5	6.3	6.8	6.6	5.9	6.2	
pH Initial	8.0	8.0	8.0	7.9	8.0	8.1	7.9		
Final		8.1	8.1	8.1	8.2	8.2	7.9	8.0	
Conductivity	1357	1351	1351	1471	1566	1569	1595		✓checked against UCL & LCL LM QA
Analyst Initials	BO	BO	BO	SAS	JM	JM	SAS	SAS	
QA Review Initials	SAS	SAS	SAS	BO	BO	BO	BO	LO	

Daily Chemistry Data - Ceriodaphnia 7-day Chronic

Client: Reference Test
Location: S&ME Inc.
Template No.: 1

Analysts: BO, SAS, JH
Dates/times: 4/8[1245]/4/15/03/1200
Age of Neonates: <8 hrs

1000 mg/L	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.8	24.2	25.3	24.3	24.2	25.8	25.6		day 4 ini temp 25.2
Final		24.2	24.1	24.1	25.0	24.8	25.3	25.8	
D.O. Initial	6.5	6.6	6.5	6.6	6.6	6.3	6.4		
Final		6.2	6.6	6.2	6.7	6.5	5.8	6.3	
pH Initial	8.0	8.0	8.1	8.0	8.0	8.1	7.9		
Final		8.1	8.2	8.1	8.4	8.2	8.0	8.0	
Conductivity	1878	1896	2000	2190	2070	1986	1965		✓checked against [illegible]
Analyst Initials	BO	BO	BO	SAS	JH	JH	SAS	SAS	
QA Review Initials	SAS	SAS	SAS	BO	BO	BO	BO	BO	

1500 mg/L	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.9	24.2	25.4	24.4	25.9	25.9	25.8		
Final		24.1	24.0	24.0	24.9	24.8	25.5	25.7	
D.O. Initial	6.4	6.5	6.5	6.5	6.5	6.3	6.5		
Final		6.5	6.5	6.3	6.9	6.4	5.9	6.3	
pH Initial	8.0	8.0	8.0	8.0	8.0	8.1	7.9		
Final		8.1	8.2	8.1	8.4	8.3	8.0	7.9	
Conductivity	2670	2730	2720	2750	2880	2810	2740		✓checked against [illegible]
Analyst Initials	BO	BO	BO	SAS	JH	JH	SAS	SAS	
QA Review Initials	SAS	SAS	SAS	BO	BO	BO	BO	BO	

2000 mg/L	Day 0	1	2	3	4	5	6	7	Remarks
Temperature-Initial	25.9	24.3	25.3	24.4					
Final		24.1	24.1	24.0					
D.O. Initial	6.4	6.5	6.5	6.5					
Final		6.3	6.5	6.0					
pH Initial	8.0	8.0	8.0	8.0					
Final		8.0	8.2	8.1					
Conductivity	3340	3510	3580	3720					✓checked against [illegible]
Analyst Initials	BO	BO	BO	SAS	JH				
QA Review Initials	SAS	BO	SAS	BO					
Stock Solution Date	4/8	4/8	4/8	4/8	4/8	4/8			