Tennessee Valley Authority, Post Office Box 2000, Soddy-Daisy, Tennessee 37379-2000

January 16, 2002

State of Tennessee
Department of Environment and Conservation
Division of Water Pollution Control
401 Church Street, 6th Floor Annex
Nashville, Tennessee 37243-1534

Attention: Ms. Evelyn Haskin, Enforcement & Compliance

Dear Ms. Haskin:

TENNESSEE VALLEY AUTHORITY - DISCHARGE MONITORING REPORT FOR SEQUOYAH NUCLEAR PLANT (SQN)

Please find enclosed the follow-up fourth quarter biomonitoring Discharge Monitoring Report (DMR) of January 2002 for SQN. Please contact me at (423) 843-6700 if you have any questions or comments.

Sincerely,

Diedre B. Nida

Diedre B. Nida
Environmental Supervisor
Signatory Authority
for Richard T. Purcell
Vice President
Sequoyah Nuclear Plant

Enclosures: 2 (original & copy)
cc (Enclosure):
Chattanooga Environmental Assistance Center
Division of Water Pollution Control
State Office Building, Suite 550
540 McCallie Avenue
Chattanooga, Tennessee 37402-2013

U.S. Nuclear Regulatory Commission
ATTN: Document Control Desk
Washington, D.C. 20555

IE25

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*

Name TVA - SEQUOYAH NUCLEAR PLANT
Address BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Diedre B. Nida

NATIONAL POLLUTANT DISCHARGE ELIMINATION SYSTEM *(NPDES)*
DISCHARGE MONITORING REPORT *(DMR)*

TN0026450	101 T
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD	YEAR	MO	DAY		YEAR	MO	DAY
From	02	01	01	To	02	03	31

MAJOR
(SUBR 01)
F - FINAL
DIFFUSER DISCHARGE
EFFLUENT

*** NO DISCHARGE [ ] ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING AVERAGE	QUANTITY OR LOADING MAXIMUM	QUANTITY OR LOADING UNITS	QUALITY OR CONCENTRATION MINIMUM	QUALITY OR CONCENTRATION AVERAGE	QUALITY OR CONCENTRATION MAXIMUM	QUALITY OR CONCENTRATION UNITS	NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
IC25 STATRE 7DAY CHR CERIODAPHNIA	SAMPLE MEASUREMENT	********	********	**		********	********	(23)			
TRP3B 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	43.9 MINIMUM	********	********	PERCENT		QUART-ERLY	COMPOS
IC25 STATRE 7DAY CHR PIMEPHALES	SAMPLE MEASUREMENT	********	********	**	>100	********	********	(23)	0	1 / 90	COMPOS
TRP6C 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	43.9 MIMINUM	********	********	PERCENT		QUART-ERLY	COMPOS
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER			TELEPHONE		DATE		
Richard T. Purcell Site Vice President	I Certify under penalty of law that this document and all attachments were prepared under my direction or supervision in accordance with a system designed to assure that qualified personnel properly gather and evaluate the information submitted. Based on my inquiry of the person or persons who manage the system, or those persons directly responsible for gathering the information, the information submitted is, to the best of my knowledge and belief, true, accurate, and complete. I am aware that there are significant penalties for submitting false information, including the possibility of fine and imprisonment for knowing violations.	Diedre B. Nida Environmental Supervisor	423	843-6700	02	01	16
TYPED OR PRINTED		SIGNATURE OF PRINCIPAL EXECUTIVE OFFICER OR AUTHORIZED AGENT	AREA CODE	NUMBER	YEAR	MO	DAY

COMMENTS AND EXPLANATION OF ANY VIOLATIONS *(Reference all attachments here)*

Repeat Fathead Minnow Test for fourth quarter 2001 was sampled from 1/2/2002 to 1/7/2002. Report is attached.

January 15, 2002

Diedre B. Nida, SB 2A-SQN

SEQUOYAH NUCLEAR PLANT (SQN) TOXICITY BIOMONITORING, NPDES PERMIT NO. TN0026450, REPAET FATHEAD MINNOW TEST, JANUARY, 2002

Attached are two copies of the subject report for submission to the state of Tennessee. The report provides results of compliance testing using fathead minnows. UV treated Outfall 101 samples were tested during this study, in addition to routine (non-treated) samples, to help determine the cause (eliminate the effect) of the low statistical sensitivity and highly variable survival among replicates within treatments that occurred in the study conducted with Outfall 101 samples collected December 9-14, 2001. Each UV Treated concentration was placed in a 40 WATT Rainbow Lifeguard UV Sterilizer for 1 minute to remove possible pathogenic interferences. I have also included a copy of the report for your records.

Side by side comparison of untreated Outfall 101 samples and ultra violet light (UV) treated Outfall 101 samples showed no toxic effects to fathead minnows in either test. The resulting IC_{25} values, for both tests, were > 100 percent.

Fathead minnow survival and growth in untreated and UV treated intake samples were not significantly different from the control, based on Homoscedastic t-Tests.

Call me at (256) 386-2755 if you have any questions or comments following your review of the report.

Cynthia L Russell

Cynthia L. Russell
Biologist
Environmental Engineering Services- West
CEB 3A-M

Attachment
cc (Attachment):
Files, ER&TA, CEB 1B-M

TENNESSEE VALLEY AUTHORITY
TOXICITY TEST REPORT

INTRODUCTION/EXECUTIVE SUMMARY

1) Facility/Discharger: Sequoyah Nuclear Plant/TVA Report Date: January 15, 2002

2) County/State: Hamilton/Tennessee 3) NPDES Permit #: TN0026450

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

5) Design Flow (MGD): 3.266

6) Receiving Stream: Tennessee River (TRM 483.6) 7) 1Q20: 2,992.4

8) Outfall Tested: 101 9) Dates Sampled: January 2-7, 2002

10) Flow on day(s) sampled (MGD): 1600, 1606, 1598, 1602, 1604, 1600

11) Pertinent site conditions: No unusual conditions reported.*

12) Test Dates: January 4-11, 2002 13) Test Type: Short-term Chronic-definitive

14) Test Species: Fathead Minnows (*Pimephales promelas*)

15) Concentrations Tested (%): Untreated Outfall 101: 10.98, 22.0, 43.9, 72.0, 100.0
Untreated Intake: 100.0

UV Treated Outfall 101: 10.98, 22.0, 43.9, 72.0, 100.0
UV Treated Intake: 100.0

16) Permit Limit Endpoint (%): Outfall 101 IC_{25}= 43.9

17) Test Results (%): Untreated Outfall 101: *Pimephales promelas*: $IC_{25} > 100$

UV Treated Outfall 101: *Pimephales promelas*: $IC_{25} > 100$

18) Facility Contact: Diedre B. Nida 19) Phone #: (423) 843-6700

20) Testing Lab Name: Environmental Testing Solutions, LLC

21) Lab Contact: Jim Sumner 22) Phone #: (828) 862-8193
TVA Contact: Cynthia L. Russell Phone #: (256) 386-2755

*Production/operation data will be provided upon request or in the event toxicity exceeds permit limits

23) Notes:

UV treated Outfall 101 samples were tested during this study, in addition to routine (non-treated) samples, to help determine the cause (eliminate the effect) of the low statistical sensitivity and highly variable survival among replicates within treatments that occurred in the study conducted with Outfall 101 samples collected December 9-14, 2001. Each UV Treated concentration was placed in a 40 WATT Rainbow Lifeguard UV Sterilizer for 1 minute to remove possible pathogenic interferences.

Untreated and UV treated samples collected from Outfall 101 exhibited no toxicity to *P. promelas* based on the applicable permit limits, IC_{25} = 43.9 percent. Resulting IC_{25} values for both Untreated and UV treated effluent were > 100 percent. Fathead minnow survival and growth in Untreated and UV treated intake samples were not significantly different from the control, based on Homoscedastic t-Tests.

METHODS SUMMARY

Samples

1) Sampling Point: Outfall 101

2) Sample Type: Composite

3) Sample Information:

ID	Date/Time Collected (MM-DD/Time) (EST)	Date/Time Received (MM-DD/Time) (EST)	Arrival Temp. (°C)	Initial TRC* (mg/L)	Date/Time Used (MM-DD/Time) (EST)
101	01/02-0918 to 01/03 -0818	01/04 0814	0.8	< 0.10	01/04 1403 01/05 -1410
Intake	01/02- 1013 to 01/03- 0913	01/04 0814	0.6	< 0.10	01/04 1403 01/05 -1410
101	01/04- 0935 to 01/05-0939	01/05 1336	0.4	< 0.10	01/06 -1400 01/07 -1409
Intake	01/04- 1032 to 01/05- 0932	01/05 1336	0.6	< 0.10	01/06 -1400 01/07 -1409
101	01/06- 1328 to 01/07- 1234	01/08 0816	1.0	< 0.10	01/08 -1407 01/09 -1431 01/10 -1217
Intake	01/06- 1351 to 01/07- 1251	01/08 0816	0.8	< 0.10	01/08 -1407 01/09 -1431 01/10 -1217

*Total residual chlorine.

4) Sample manipulation:

Untreated samples of Outfall 101 and Intake were warmed to test temperature (25.0 ± 1.0°C) in a warm water bath.

Test Organisms

		Pimephales promelas
1)	Source:	Aquatic BioSystems, Inc.
2)	Age:	23.5-25.5 hours

Test Method Summary (See Appendix A for additional information)

		Pimephales promelas
1)	Test Conditions	Static, renewal
2)	Test Duration	7 days
3)	Dilution/Control Water	Moderately Hard Synthetic Water
4)	Number Replicates	4
5)	Animals per Replicate	10
6)	Test Initiation (Date/Time)	01/04/02/1403 EST
7)	Test Termination (Date/Time)	01/11/02/1420 EST
8)	Test Temperature	Mean = 24.6°C (24.0°C-25.5°C)

9) Physical/Chemical Measurements: Hardness, alkalinity, total residual chlorine, and conductivity were measured at the laboratory in each 100 percent sample. Daily temperatures were measured in one replication per 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 C for Bench Sheets)

1) Results of a *Pimephales* (Genus) *promelas* (Species) Chronic/ 7-d (Type/Duration) Toxicity Test

Conducted 01/04/02 (mm/dd/yy) - 01/11/02 (mm/dd/yy) Using Effluent From Untreated Outfall 101 (number)

Test Solutions	Percent Surviving (time intervals used - days)						
	1	2	3	4	5	6	7
Control	100	98	98	98	98	98	98
10.98% Effluent	100	95	95	95	95	95	85
22.0% Effluent	100	100	100	98	98	98	98
43.9% Effluent	100	100	100	100	100	100	100
72% Effluent	100	100	100	100	100	100	100
100% Effluent	100	100	100	100	100	100	100
Intake	100	100	100	100	100	100	100

Test Solutions	IC_{25} Mean Dry Weight (mg) (Replication)				
	1	2	3	4	Mean
Control	0.819	1.119	0.982	0.963	0.971
10.98% Effluent	1.067	0.959	0.329	1.066	0.855
22.0% Effluent	1.097	0.989	0.939	1.085	1.027
43.9% Effluent	0.962	1.004	0.983	1.018	0.991
72% Effluent	0.977	0.946	0.822	1.018	0.941
100% Effluent	1.005	0.963	0.978	1.132	1.020
Intake	0.989	1.036	1.086	0.901	1.003

IC_{25} Value $\geq$ 100%	Calculated TU Estimates* <1.0 TUc
95% Confidence Limits Upper Limit = NA Lower Limit = NA	Permit Limit 2.3 TUc

*NOTE: TUc = 100/ IC_{25}

2) Results of a *Pimephales* (Genus) *promelas* (Species) Chronic/ 7-d (Type/Duration) Toxicity Test

Conducted 01/04/02 (mm/dd/yy) - 01/11/02 (mm/dd/yy) Using Effluent From UV Treated Outfall 101 (number)

Test Solutions	Percent Surviving (time intervals used - days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
10.98% Effluent	100	100	100	100	100	100	100
22.0% Effluent	100	100	100	100	100	100	100
43.9% Effluent	100	100	100	100	100	100	100
72% Effluent	100	95	95	95	95	95	95
100% Effluent	100	100	100	100	100	100	100
Intake	100	100	100	100	100	100	100

Test Solutions	IC_{25} Mean Dry Weight (mg) (Replication)				
	1	2	3	4	Mean
Control	1.089	1.041	0.987	1.161	1.070
10.98% Effluent	0.914	1.146	1.042	1.157	1.065
22.0% Effluent	0.927	1.003	0.994	1.043	0.992
43.9% Effluent	1.004	0.928	1.055	1.026	1.003
72% Effluent	0.685	1.058	1.000	1.022	0.941
100% Effluent	1.031	0.981	0.891	0.796	0.925
Intake	1.009	0.838	0.956	0.980	0.946

IC_{25} Value $\geq$ 100%	Calculated TU Estimates † <1.0 TUc
95% Confidence Limits Upper Limit = NA Lower Limit = NA	Permit Limit 2.3 TUc

†NOTE: TUc = 100/ IC_{25}

3) REFERENCE TOXICANT TEST RESULTS (See Appendixes A and D)

Species	Date	Time	Duration	Toxicant	Results (IC_{25})
Pimephales promelas	01/04/02	1431	7-days	KCl	487.8 mg/L

SUMMARY/CONCLUSIONS

Untreated and UV treated samples collected from Outfall 101 exhibited no toxicity to *P. promelas* based on the applicable permit limits. Resulting IC_{25} values for both Untreated and UV treated effluent were > 100 percent. Fathead minnow survival and growth in Untreated and UV treated intake samples was not significantly different from the control, based on Homoscedastic t-Tests.

Appendix A

ADDITIONAL TOXICITY TEST INFORMATION

SUMMARY OF METHODS

Tests were conducted according to EPA/600/4-91/002 (July 1994) using four replicates, each containing ten test organisms, per treatment. Test vessels consisted of 400-mL beakers, each containing 250 mL of test solution. [2]

DEVIATIONS/MODIFICATIONS TO TEST PROTOCOL

The third renewal using the last samples collected was conducted greater than 1 hour from the test initiation time to meet the required 72-hour hold time.

DEVIATIONS/MODIFICATIONS TO PRETEST CULTURE OR HOLDING OF TEST ORGANISMS

The concurrent reference toxicant test conducted on this batch of fish was just below control chart IC_{25} limits. The resulting endpoint was 0.015 mg lower than the -2SD lower boundary. Two SD allowed a reduction of 22.6 percent from controls, while the reference toxicant results were reduced 24.9 percent from controls. Previous reference toxicant tests conducted within 30-days by both ETS and the culture lab were within bounds and this was the first outlier in 18 tests ETS has performed (one outlier in twenty is expected). If animals were more sensitive, effects would have been more likely in the effluent tests and both effluent tests clearly showed no toxicity. Both tests far surpassed both survival and growth test acceptability criteria.

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 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 using EPA Method 150.1.

6) Conductance was measured using EPA Method 120.1.

7) Alkalinity was measured using EPA Method 310.1.

8) Hardness was measured using EPA Method 130.2.

9) Total residual chlorine was determined 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/600/4-91/002. [2] Any known deviations were noted during the study and are reported herein.

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

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

2) Standard Toxicant: Potassium Chloride (KCl crystalline)

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. TN0026450.

2) Lewis. P. A.. D. J. Klemm. J. M. Lazorchak. T. J. Norberg-King. W. H. Peltier. M. A. Heber. Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms. EPA/600/4-91/002 (July 1994).

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

Sequoyah Nuclear Plant Biomonitoring
January 4-11, 2001

Appendix B

Diffuser Discharge Concentrations of Chemicals Used to Control Growth of Microbiologically Induced Bacteria and Asiatic Clams, During Toxicity Test Sampling,

And

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

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Growth of Microbiologically Induced Bacteria and Asiatic Clams, During Toxicity Test Sampling, March 12, 1998-January 7, 2002

Date	Sodium hypochlorite mg/L TRC	Towerbrom mg/L TRC	PCL-222 mg/L Phosphate	PCL-401 mg/L Copolymer	CL-363 mg/L DMAD	Cuprostat-PF mg/L Azole	H-130M mg/L Quat
03/12/1998	0.016	-	-	-	-	-	-
03/13/1998	0.015	-	-	-	-	-	-
03/14/1998	0.013	-	-	-	-	-	-
03/15/1998	0.030	-	-	-	-	-	-
03/16/1998	0.013	-	-	-	-	-	-
03/17/1998	0.020	-	-	-	-	-	-
03/18/1998	0.018	-	-	-	-	-	-
09/08/1998	0.015	-	0.014	0.005	-	-	0.021
09/09/1998	0.003	-	0.031	0.011	-	-	-
09/10/1998	0.014	-	0.060	0.021	-	-	-
09/11/1998	0.013	-	0.055	0.019	-	-	-
09/12/1998	< 0.001	-	0.044	0.015	-	-	-
09/13/1998	< 0.001	-	0.044	0.015	-	-	-
09/14/1998	0.008	-	0.044	0.015	-	-	-
02/22/1999	< 0.001	-	-	-	-	-	-
02/23/1999	0.005	-	-	-	-	-	-
02/24/1999	0.009	-	-	-	-	-	-
02/25/1999	0.012	-	-	-	-	-	-
02/26/1999	0.008	-	-	-	-	-	-
02/27/1999	< 0.001	-	-	-	-	-	-
02/28/1999	< 0.001	-	-	-	-	-	-
08/18/1999	-	0.015	0.069	0.024	0.006	-	-
08/19/1999	-	0.012	0.068	0.024	-	-	-
08/20/1999	-	0.023	0.070	0.024	-	0.120	-
08/21/1999	-	0.022	0.068	0.024	-	-	-
08/22/1999	-	0.022	0.068	0.024	-	-	-
08/23/1999	-	0.025	0.068	0.024	0.006	-	-
08/24/1999	-	0.016	0.067	0.023	0.020	-	-

Table B-1 (continued). Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Growth of Microbiologically Induced Bacteria and Asiatic Clams, During Toxicity Test Sampling, March 12, 1998-January 7, 2002

Date	Sodium hypochlorite mg/L TRC	Towerbrom mg/L TRC	PCL-222 mg/L Phosphate	PCL-401 mg/L Copolymer	CL-363 mg/L DMAD	Cuprostat-PF mg/L Azole	H-130M mg/L Quat
01/31/2000	-	< 0.002	0.026	0.009	-	-	-
02/01/2000	-	0.011	0.026	0.028	-	-	-
02/02/2000	-	0.028	0.026	0.009	0.006	-	-
02/03/2000	-	0.008	0.027	0.009	-	-	-
02/04/2000	-	0.006	0.027	0.009	0.005	0.109	-
02/05/2000	-	< 0.002	0.027	0.009	-	-	-
02/06/2000	-	< 0.002	0.027	0.009	-	-	-
07/26/2000	-	< 0.0057	0.055	0.019	-	-	0.021
07/27/2000	-	0.019	0.055	0.019	-	-	-
07/28/2000	-	0.0088	0.053	0.018	0.004	0.108	-
07/29/2000	-	< 0.0088	0.055	0.019	-	-	-
07/30/2000	-	< 0.0076	0.055	0.019	-	-	-
07/31/2000	-	< 0.0152	0.055	0.019	0.006	-	-
08/01/2000	-	< 0.0141	0.055	0.019	0.005	-	-
12/11/2000	-	0.0143	0.025	0.020	0.005	-	-
12/12/2000	-	0.0092	0.025	0.020	0.005	-	-
12/13/2000	-	< 0.0120	0.025	0.020	-	-	-
12/14/2000	-	< 0.0087	0.025	0.020	-	-	-
12/15/2000	-	0.0120	0.025	0.020	0.005	-	-
12/16/2000	-	< 0.0036	0.025	0.020	-	-	-
12/17/2000	-	< 0.0036	0.025	0.020	-	-	-
08/26/2001	-	0.017	0.06	0.021	0.006	-	-
08/27/2001	-	<0.0096	0.06	0.021	0.005	-	0.021
08/28/2001	-	<0.0085	0.06	0.021	-	-	-
08/29/2001	-	<0.0094	0.059	0.020	0.005	-	0.021
08/30/2001	-	<0.0123	0.06	0.021	0.005	-	-
08/31/2001	-	<0.005	0.059	0.020	-	-	-
11/25/2001	-	<0.0044	-	-	-	-	-
11/26/2001	-	<0.0119	0.024	0.02	0.005	-	-
11/27/2001	-	0.0137	0.023	0.019	0.007	-	-
11/28/2001	-	<0.0089	0.022	0.019	0.006	-	-
11/29/2001	-	0.0132	0.024	0.02	0.007	-	-
11/30/2001	-	< 0.0043	0.024	0.02	-	-	-

Table B-1 (continued). Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Growth of Microbiologically Induced Bacteria and Asiatic Clams, During Toxicity Test Sampling, March 12, 1998-January 7, 2002

Date	Sodium hypochlorite mg/L TRC	Towerbrom mg/L TRC	PCL-222 mg/L Phosphate	PCL-401 mg/L Copolymer	CL-363 mg/L DMAD	Cuprostat-PF mg/L Azole	H-130M mg/L Quat
12/09/2001	-	<0.0042	-	-	-	-	-
12/10/2001	-	<0.0042	-	-	-	-	-
12/11/2001	-	<0.0104	-	-	-	-	-
12/12/2001	-	0.0128	0.024	0.02	0.008	-	-
12/13/2001	-	<0.0088	0.024	0.02	-	-	-
12/14/2001	-	0.0134	0.024	0.02	0.007	-	-
01/02/02	-	< 0.0079	0.023	0.020	0.006	-	-
01/03/02	-	<0.0042	0.026	0.014	-	-	-
01/04/02	-	0.0124	0.024	0.014	0.009	-	-
01/05/02	-	<0.0042	-	-	-	-	-
01/06/02	-	<0.0042	-	-	-	-	-
01/07/02	-	<0.0089	0.024	0.014	0.006	-	-

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for Fathead Minnow Test, Sequoyah Nuclear Plant Effluent (SQN), Untreated Outfall 101, January 4-11, 2002

Test/ Sample ID	Temperature Initial (°C)	Temperature Final (°C)	Dissolved Oxygen Initial (mg/L)	Dissolved Oxygen Final (mg/L)	pH Initial S.U.	pH Final S.U.	Conductance Initial (µmhos)	Alkalinity mg/L $CaCO_3$	Hardness mg/L $CaCO_3$	Total Residual Chlorine (mg/L)
Fathead/ Control	24.8 (24.4-25.5)	24.4 (24.1-24.8)	8.0 (8.0-8.1)	7.5 (7.0-7.8)	8.06 (8.01-8.10)	7.75 (7.62-7.84)	308 (300-320)	67 (65-68)	90 (90-90)	-
Fathead/ 10.98%	24.8 (24.4-25.4)	24.4 (24.2-24.8)	8.0 (7.9-8.1)	7.4 (7.1-7.9)	8.07 (8.03-8.11)	7.73 (7.60-7.79)	300 (290-315)	-	-	-
Fathead/ 22.0 %	24.8 (24.4-25.4)	24.4 (24.1-24.6)	8.1 (7.9-8.2)	7.4 (7.1-7.9)	8.06 (8.02-8.10)	7.72 (7.57-7.86)	290 (281-304)	-	-	-
Fathead/ 43.9 %	24.8 (24.4-25.4)	24.4 (24.1-24.7)	8.2 (8.0-8.4)	7.4 (7.1-7.9)	8.02 (7.92-8.08)	7.69 (7.55-7.82)	265 (257-277)	-	-	-
Fathead/ 72.0 %	24.8 (24.4-25.4)	24.4 (24.1-24.8)	8.3 (8.0-8.5)	7.4 (7.0-7.8)	7.99 (7.93-8.06)	7.69 (7.54-7.79)	232 (225-242)	-	-	-
Fathead/ 100%	24.8 (24.4-25.4)	24.4 (24.1-24.8)	8.4 (8.1-8.6)	7.4 (7.0-7.9)	7.93 (7.85-8.05)	7.66 (7.53-7.81)	197 (192-207)	65 (61-69)	77 (74-80)	<0.10 (<0.10-<0.10)
Fathead/ Intake	24.8 (24.4-25.4)	24.4 (24.1-24.8)	8.4 (8.1-8.6)	7.4 (7.0-7.9)	7.87 (7.76-8.03)	7.74 (7.61-7.83)	196 (192-206)	64 (62-65)	78 (74-80)	<0.10 (<0.10-<0.10)

PHYSICAL/CHEMICAL SUMMARY (continued)

Water Chemistry Mean Values and Ranges for Fathead Minnow Test, Sequoyah Nuclear Plant Effluent (SQN), UV Treated Outfall 101, January 4-11, 2002

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	24.8 (24.4-25.4)	24.4 (24.2-24.7)	8.0 (7.9-8.1)	7.5 (7.2-7.8)	7.92 (7.66-8.04)	7.76 (7.68-7.88)	307 (296-321)	67 (65-68)	90 (90-90)	-
Fathead/ 10.98%	24.8 (24.4-25.4)	24.4 (24.2-24.7)	8.1 (7.9-8.2)	7.6 (7.2-7.8)	7.97 (7.81-8.03)	7.77 (7.68-7.87)	298 (289-318)	-	-	-
Fathead/ 22.0 %	24.8 (24.4-25.4)	24.4 (24.0-24.7)	8.2 (8.0-8.3)	7.6 (7.3-7.8)	8.00 (7.93-8.06)	7.74 (7.62-7.81)	287 (278-305)	-	-	-
Fathead/ 43.9 %	24.8 (24.4-25.4)	24.4 (24.1-24.7)	8.3 (8.1-8.4)	7.6 (7.2-7.9)	8.02 (7.96-8.06)	7.75 (7.64-7.83)	263 (256-277)	-	-	-
Fathead/ 72.0 %	24.8 (24.4-25.4)	24.4 (24.1-24.7)	8.3 (8.1-8.5)	7.6 (7.4-7.9)	8.01 (7.96-8.06)	7.72 (7.60-7.83)	228 (220-244)	-	-	-
Fathead/ 100.0 %	24.8 (24.4-25.4)	24.4 (24.1-24.7)	8.4 (8.1-8.6)	7.6 (7.3-7.9)	7.99 (7.95-8.07)	7.72 (7.59-7.81)	196 (189-207)	65 (61-69)	77 (74-80)	<0.10 (<0.10-<0.10)
Fathead/ Intake	24.8 (24.4-25.4)	24.4 (24.1-24.7)	8.4 (8.1-8.7)	7.6 (7.3-7.8)	7.97 (7.91-8.05)	7.71 (7.62-7.82)	194 (189-207)	64 (62-65)	78 (74-80)	<0.10 (<0.10-<0.10)

Sequoyah Nuclear Plant Biomonitoring
January 4 – 11, 2002

Appendix C

Chain of Custody Records and
Toxicity Bench Sheets

BIOMONITORING CHAIN OF CUSTODY RECORD

Client: TVA

Project Name: TVA SQN

P.O. Number: PO BOX 2000

Facility Sampled: SEQUOYAH NUCLEAR PLANT

NPDES Number: 0026450

Collected By: WANDA ALLEN

Environmental Testing Solutions, LLC
Research Building
1 Ecusta Road, Brevard, NC 28712
Phone: 828-862-8193
Fax: 828-862-8195

Delivered By (Circle One):

FedEx UPS Bus Client

Other (specify): MAX COULTER

General Comments:

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				
	—	—	Start	End	—	—	Y-	If Yes, Inches	No	Trace	EST Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	comp	4	0917 1/2/02 0918	0817 1/3/02 0818	2 (2.5g)				✓		221 020104.01	0.8°C	JS	01-03-02 0814	CUSTODY SEALS INTACT
SQN-INT-TOX	comp	4	1/2/02 1013	0913 1/3/02	1 (2.5g)				✓		221 020104.02	0.6°C	JS	01-03-02 0814	GOOD CONDITION ↓

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Wanda Allen	1/3/02 1000	J Jimmer	01-03-02 0814

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

Client: TVA

Project Name: TVA SQN

P.O. Number: PO BOX 2000

Facility Sampled: SEQUOYAH NUCLEAR PLANT

NPDES Number: 0026450

Collected By: WANDA ALLEN

Environmental Testing Solutions, LLC
Research Building
1 Ecusta Road, Brevard, NC 28712
Phone: 828-862-8193
Fax: 828-862-8195

Delivered By (Circle One):

FedEx UPS Bus Client

Other (specify): MAX COULTER

General Comments:

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				
	---	---	Start	End	---	---	Y-	If Yes, Inches	No	Trace	EST Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	comp	4	0935 1/4/02 1039	0835 1/5/02 0939	2 (2.5g)				✓		221 02.0105.01	0.4°C	JS	01-05-02 1336	CUSTODY SEALS INTACT
SQN-INT-TOX	comp	4	1/4/02 1032	0932 1/5/02	1 (2.5g)				✓		221 020105.02	0.6°C	JS	01-05-02 1336	GOOD CONDITION

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Wanda Allen	1/5/02 1015	J Humpe	01-05-02 1336

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

Client: TVA

Project Name: TVA SQN

P.O. Number: PO BOX 2000

Facility Sampled: SEQUOYAH NUCLEAR PLANT

NPDES Number: 0026450

Collected By: WANDA ALLEN

Environmental Testing Solutions, LLC
Research Building
1 Ecusta Road, Brevard, NC 28712
Phone: 828-862-8193
Fax: 828-862-8195

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

General Comments:

Field Identification / Sample Description	Grab/ Comp.	Ship. Temp. (°C)	Collection Date/Time Start	Collection Date/Time End	Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate) Y-	If Yes, Inches	No	Trace	Laboratory Use EST Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	comp	4	1334 1/6/02 1328	~~1234~~ 1228 1/7/02	2 (2.5g)						221 020108.01	1.0°C	JL	01-08-02 0816	CUSTODY SEALS INTACT
SQN-INT-TOX	comp	4	1/6/02 ~~1351~~ 1351	~~1234~~ ~~1251~~ 1/7/02 1251	1 (2.5g)						221 020108.02	0.8°C	JL	01-08-02 0816	GOOD CONDITION ↓
SQN															

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Wanda Allen	01-07-02 ~~12/7/01~~ 1330	[signature]	01-08-02 0816

nstructions: 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 ample 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. amples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

Sample Receipt Log

Date Received	Time Received	Received By	Received From	Sample Temperature (°C)	Project Number	Sample Number	Sample Name and Description	State	Comments
12-13-01	1240	KEKeenan	Fed-ex	0.7°C	219	011213.03	Alcoa Engineered Products	NC	
12-15-01	1231	KEKeenan	Fedex	1.8°C	217	011215.01	Monroe WWTP	NC	
12-15-01	1231	KEKeenan	J. Burrell ~~Fedex~~	3.0°C	214	011215.02	Tryon WWTP	NC	
12-15-01	1232	KEKeenan	M. Ladd	1.1°C	211	011215.03	Test America - Earth Env. Andrews WWTP	NC	
12-15-01	1232	KEKeenan	M. Ladd	1.1°C	212	011215.04	Test America - Earth Env. Franklin WWTP	NC	
12-15-01	1233	KEKeenan	Greyhound	1.5°C	213	011215.05	Precise Analytical Norfolk Southern Railway	NC	
12-15-01	1234	KEKeenan	Fed-ex	0.9°C	216	011215.06	Craven County Wood Energy	NC	
12-18-01	1120	Jumper	Fedex	7.2°C	220	011218.01	Georgetown Steel - DRI Plant	SC ~~NC~~	* Sample exceeds acceptable temp. upon receipt.
01-04-02	0814	Jumper	M. Coulter	0.8°C	221	020104.01	TVA Sequoyah Nuclear Plant Outfall 101	TN	*
01-04-02	0814	Jumper	M. Coulter	0.6°C	221	020104.02	TVA Sequoyah Nuclear Plant Intake	TN	*
01-05-02	1336	Jumper	M. Coulter	0.4°C	221	020105.01	TVA Sequoyah Nuclear Plant Outfall 101	TN	*
01-05-02	1336	Jumper	M. Coulter	0.6°C	221	020105.02	TVA Sequoyah Nuclear Plant Intake	TN	*
01-08-02	0816	Jumper	M. Coulter	1.0°C	221	020108.01	TVA Sequoyah Nuclear Plant Outfall 101	TN	*
01-08-02	0816	Jumper	M. Coulter	0.8°C	221	020108.02	TVA Sequoyah Nuclear Plant Intake	TN	*
01-08-02	1010	KEKeenan	UPS	1.4°C	222	020108.03	Tritest - RDU Airport Outfall 001	NC	
01-08-02	1010	KEKeenan	UPS	1.4°C	222	020108.04	Tritest - RDU Airport Outfall 002	NC	
01-08-02	1010	KEKeenan	UPS	1.4°C	222	020108.05	Tritest RDU Airport Outfall 003	NC	
01-08-02	1010	KEKeenan	UPS	1.4°C	222	020108.06	Tritest - RDU Airport Outfall 004	NC	
01-08-02	1107	KEKeenan	Fedex	0.5°C	223	020108.07	Anderson County Sewer Authority	SC	
01-09-02	0902	Jumper	L. Carreiro	0.2°C	224	020109.01	Asheville - North Fork WTP	NC	
[illegible]	[illegible]	Jumper	Fedex	0.7°C	225	020109.02	PCS Phosphate Outfall 007	NC	

1300 Blue Spruce Drive, Suite C
Fort Collins, Colorado 80524

Toll Free: 800/331-5916
Tel: 970/484-5091 Fax:970/484-2514

ORGANISM HISTORY

DATE: 1/3/02

SPECIES: Pimephales promelas

AGE: N/A

LIFE STAGE: Embryo

HATCH DATE: 1/3/02 Between 11:30 a.m.&1:30 p.m.MST

BEGAN FEEDING: N/A

FOOD: N/A

Water Chemistry Record:	Mean	Range
TEMPERATURE:	25 °C	--
SALINITY/CONDUCTIVITY:	--	--
TOTAL HARDNESS (as $CaCO_3$):	124 mg/l	--
TOTAL ALKALINITY (as $CaCO_3$):	90 mg/l	--
pH:	8.09	--

Comments:

Facility Supervisor

Received:
01-04-02 1144 JHumber
Temp = 22.8°C
DO = 8.9 mg/L

Fed at 1150 JS.
Organisms appear healthy.

Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002 Method 1000.0)
Species: *Pimephales promelas*

Client: TVA SEQUOYAH NUCLEAR PLANT
Facility: OUTFALL 101 - NONTREATED
NPDES #: TN 0026450

Dilution preparation information:						*Comments:*
MHS batch:		01-03-02 / 01-06-02				
Dilution prep (%)	10.98	22	43.9	72	100	
Effluent volume (mL)	131.76	264	526.8	864	1200	
Diluent volume (mL)	1068.24	936	673.2	336	0	

Test organism information:		*Test information:*	
Organism age:	24-HOURS OLD	Randomizing template:	BLUE
Date and times organisms were born between:	01-03-02 1130 TO 1330 MST 1330 1530 EST ~~(0930 TO 1130 EST)~~ JH	Incubator number:	2
Organism source:	ABS BATCH 01-03-02	*Artemia* lot number:	BS0207P
Transfer bowl information:	pH = 7.87 Temperature = 24.1°C	Oven temperature:	60°C
		Drying time:	24-HOURS

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Analyst
0	01-04-02	— JH	1438	1403	JH
1	01-05-02	1000	1600	1410	JH
2	01-06-02	1007	1610	1400	KEK
3	01-07-02	0955	1555	1409	JH
4	01-08-02	1000	1600	1407	JH
5	01-09-02	1016	1620	1431	KEK
6	01-10-02	0952	1550	1217	KEK *
7	01-11-02	—	JH	1420	KEK

* TEST WAS NOT RENEWED ± 1-HOUR FROM TEST INITIATION TO MEET 72-HOUR HOLD TIME

Control information:		Acceptance criteria
% Mortality:	2.5%	≤ 20%
Average weight per larvae:	0.9707	≥ 0.25 mg/larvae

Summary of test endpoints:	
7-day LC50	>100%
NOEC	100%
LOEC	>100%
ChV	>100%
IC25	>100%

Species: *Pimephales promelas*

Client: TVA SEQUOYAH NUCLEAR PLANT
OUTFALL 101 · NONTREATED

Date: 01-04-02

Survival and Growth Data

Day	CONTROL				10.98 %				22%			
	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~~ 9 1d	10	10	10	10	10	8 2d	10	10	10	10	10
3	9	10	10	10	10	10	8	10	10	10	10	10
4	9	10	10	10	10	10	8	10	10	10	9 1d	10
5	9	10	10	10	10	10	8	10	10	10	9	10
6	9	10	10	10	10	10	8	10	10	10	9	10
7	9	10	10	10	10	10	4 4d	10	10	10	9	10
A = Pan weight (mg)	14.973	15.151	14.956	15.001	15.013	14.938	14.962	14.820	14.965	15.158	14.942	14.910
B = Pan + Larvae weight (mg)	23.160	26.34	24.78	24.63	25.68	24.53	18.25	25.48	25.93	25.05	24.33	25.76
Larvae weight (mg) = A – B	8.187	11.189	9.824	9.629	10.667	9.592	3.288	10.660	10.965	9.894	9.388	10.850

Calculations and data reviewed: [illegible]

Comments:

Species: *Pimephales promelas*

Client: TVA SEQUOYAH NUCLEAR PLANT
OUTFALL 101 - NONTREATED

Date: 01-04-02

Survival and Growth Data

Day	43.9%				72%				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)	14.949	14.965	15.015	14.925	14.950	15.130	15.132	14.959	15.112	15.042	15.119	14.940
B = Pan + Larvae weight (mg)	24.57	25.00	24.84	25.10	24.72	24.59	23.35	25.14	25.16	24.67	24.90	26.27
Larvae weight (mg) = A – B	9.621	10.035	9.825	10.175	9.770	9.460	8.218	10.183	10.048	9.628	9.781	11.322

Calculations and data reviewed: JH

Comments:

Species: *Pimephales promelas*

Client: TVA SEQUOYAH NUCLEAR PLANT
OUTFALL 101- NONTREATED

Date: 01-04-02

Survival and Growth Data

Day	100% INTAKE											
	Y	Z	AA	BB	CC	DD	EE	FF	GG	HH	II	JJ
0	10	10	10	10								
1	10	10	10	10								
2	10	10	10	10								
3	10	10	10	10								
4	10	10	10	10								
5	10	10	10	10								
6	10	10	10	10								
7	10	10	10	10								
A = Pan weight (mg)	15.012	14.995	15.162	14.902								
B = Pan + Larvae weight (mg)	24.90	25.35	26.02	23.91								
Larvae weight (mg) = A – B	9.888	10.355	10.858	9.008								

Calculations and data reviewed: JH

Comments:

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002, Method 1002.0)

Species: *Pimephales promelas*

Quality Control

Verification of Data Entry, Calculations, and Statistical Analyses

Client: TVA Sequoyah Nuclear Plant - Nontreated

Test dates: January 4 - 11, 2002

Project number: 221

Revelwed by: [illegible]

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	9	14.973	23.160	8.187	0.8187	97.5	0.9707	12.7	Not applicable
	B	10	10	15.151	26.340	11.189	1.1189				
	C	10	10	14.956	24.780	9.824	0.9824				
	D	10	10	15.001	24.630	9.629	0.9629				
10.98%	E	10	10	15.013	25.680	10.667	1.0667	85.0	0.8552	41.5	11.9
	F	10	10	14.938	24.530	9.592	0.9592				
	G	10	4	14.962	18.250	3.288	0.3288				
	H	10	10	14.820	25.480	10.660	1.0660				
22%	I	10	10	14.965	25.930	10.965	1.0965	97.5	1.0274	7.4	-5.8
	J	10	10	15.156	25.050	9.894	0.9894				
	K	10	9	14.942	24.330	9.388	0.9388				
	L	10	10	14.910	25.760	10.850	1.0850				
43.9%	M	10	10	14.949	24.570	9.621	0.9621	100.0	0.9914	2.4	-2.1
	N	10	10	14.965	25.000	10.035	1.0035				
	O	10	10	15.015	24.840	9.825	0.9825				
	P	10	10	14.925	25.100	10.175	1.0175				
72%	Q	10	10	14.950	24.720	9.770	0.9770	100.0	0.9408	9.0	3.1
	R	10	10	15.130	24.590	9.460	0.9460				
	S	10	10	15.132	23.350	8.218	0.8218				
	T	10	10	14.957	25.140	10.183	1.0183				
100% Outfall 101	U	10	10	15.112	25.160	10.048	1.0048	100.0	1.0195	7.6	-5.0
	V	10	10	15.042	24.670	9.628	0.9628				
	W	10	10	15.119	24.900	9.781	0.9781				
	X	10	10	14.948	26.270	11.322	1.1322				
100% Intake	Y	10	10	15.012	24.900	9.888	0.9888	100.0	1.0027	7.8	-3.3
	Z	10	10	14.995	25.350	10.355	1.0355				
	AA	10	10	15.162	26.020	10.858	1.0858				
	BB	10	10	14.962	23.970	9.008	0.9008				

Outfall 101:

Dunnett's MSD value: 0.2785

PMSD: 28.7

Intake:

Dunnett's MSD value: 0.1417

PMSD: 14.6

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 19.1% 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 Growth

Start Date:	1/4/02	Test ID:	PpFRCR	Sample ID:	TVA SQN - Outfall 101 Nontreated
End Date:	1/11/02	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.8187	1.1189	0.9824	0.9629
10.98	1.0667	0.9592	0.3288	1.0660
22	1.0965	0.9894	0.9388	1.0850
43.9	0.9621	1.0035	0.9825	1.0175
72	0.9770	0.9460	0.8218	1.0183
100	1.0048	0.9628	0.9781	1.1322

Conc-%	Mean	N-Mean	Transform: Untransformed Mean	Min	Max	CV%	N	Rank Sum	1-Tailed Critical	Isotonic Mean	N-Mean
D-Control	0.9707	1.0000	0.9707	0.8187	1.1189	12.654	4			0.9707	1.0000
10.98	0.8552	0.8810	0.8552	0.3288	1.0667	41.457	4	17.00	10.00	0.9669	0.9960
22	1.0274	1.0584	1.0274	0.9388	1.0965	7.410	4	20.00	10.00	0.9669	0.9960
43.9	0.9914	1.0213	0.9914	0.9621	1.0175	2.447	4	20.00	10.00	0.9669	0.9960
72	0.9408	0.9691	0.9408	0.8218	1.0183	9.000	4	17.00	10.00	0.9669	0.9960
100	1.0195	1.0502	1.0195	0.9628	1.1322	7.565	4	20.00	10.00	0.9669	0.9960

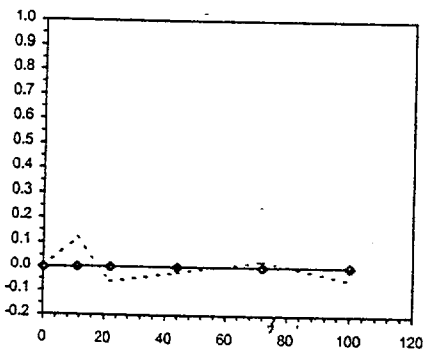
Auxiliary Tests

	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.827680886	0.884	-1.98286024	7.208732712
Bartlett's Test indicates unequal variances (p = 1.86E-03)	19.07797623	15.08631706		

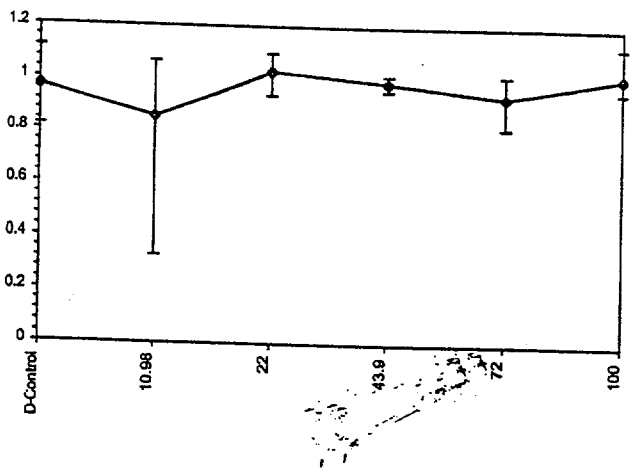
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	100	>100		1

Linear Interpolation (80 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

Statistical Analyses

Used for PMSD calculation only.

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	1/4/02	Test ID:	PpFRCR	Sample ID:	TVA SQN - Outfall 101 Nontreated
End Date:	1/11/02	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

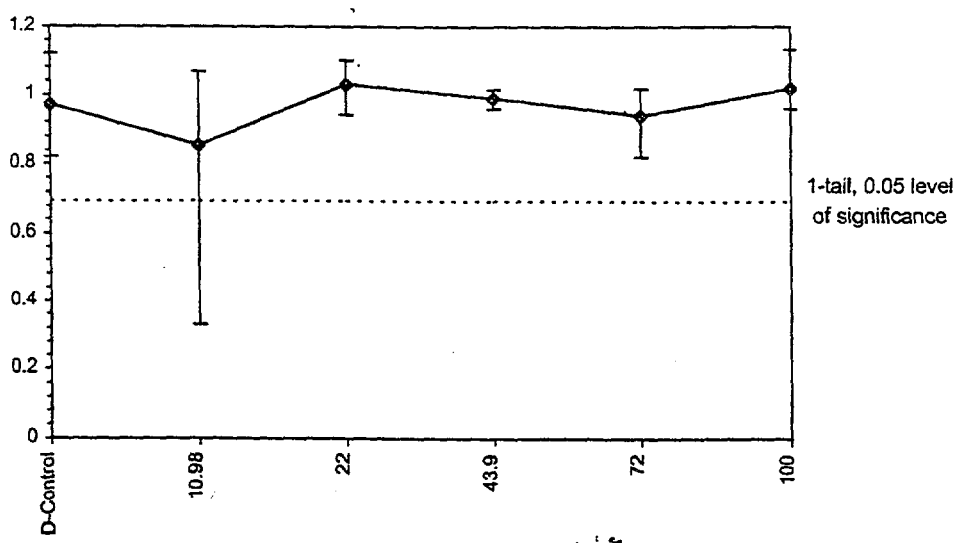
Conc-%	1	2	3	4
D-Control	0.8187	1.1189	0.9824	0.9629
10.98	1.0667	0.9592	0.3288	1.0660
22	1.0965	0.9894	0.9388	1.0850
43.9	0.9621	1.0035	0.9825	1.0175
72	0.9770	0.9460	0.8218	1.0183
100	1.0048	0.9628	0.9781	1.1322

			Transform: Untransformed						1-Tailed	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	0.9707	1.0000	0.9707	0.8187	1.1189	12.654	4			
10.98	0.8552	0.8810	0.8552	0.3288	1.0667	41.457	4	1.000	2.410	0.2785
22	1.0274	1.0584	1.0274	0.9388	1.0965	7.410	4	-0.491	2.410	0.2785
43.9	0.9914	1.0213	0.9914	0.9621	1.0175	2.447	4	-0.179	2.410	0.2785
72	0.9408	0.9691	0.9408	0.8218	1.0183	9.000	4	0.259	2.410	0.2785
100	1.0195	1.0502	1.0195	0.9628	1.1322	7.565	4	-0.422	2.410	0.2785

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.827680886	0.884	-1.98286024	7.208732712
Bartlett's Test indicates unequal variances (p = 1.86E-03)	19.07797623	15.08631706		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	100	>100		1	0.278529016	0.286928858	0.016164118	0.026713868	0.696975112	5, 18

Dose-Response Plot



Environmental Testing Solutions, LLC

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	1/4/02	Test ID:	PpPFCR	Sample ID:	TVA SQN - Intake Nontreated
End Date:	1/11/02	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.8187	1.1189	0.9824	0.9629
100	0.9888	1.0355	1.0858	0.9008

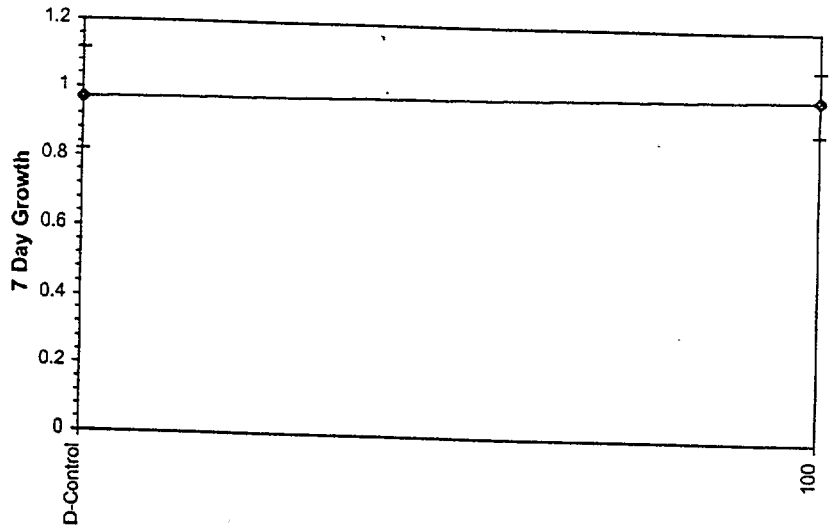
Conc-%	Mean	N-Mean	Transform: Untransformed						1-Tailed	
			Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	0.9707	1.0000	0.9707	0.8187	1.1189	12.654	4			
100	1.0027	1.0330	1.0027	0.9008	1.0858	7.844	4	-0.439	1.943	0.1417

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.974953771	0.749	-0.15580785	-0.06228885
F-Test indicates equal variances (p = 0.48)	2.439072847	47.46834564		

Hypothesis Test (1-tail, 0.05)

Homoscedastic t Test indicates no significant differences

Dose-Response Plot



Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002 Method 1000.0)
Species: *Pimephales promelas*

Client: TVA SEQUOYAH NUCLEAR PLANT
Facility: OUTFALL 101 - UNTREATED
NPDES #: TN0026450

Dilution preparation information:						*Comments:*
MHS batch:		01-03-02 / 01-06-02				EACH DILUTION WAS UV TREATED
Dilution prep (%)	10.98	22	43.9	72	100	FOR 1 MINUTE WITH A
Effluent volume (mL)	131.76	264	526.8	864	1200	40 WATT RAINBOW LIFEGUARD
Diluent volume (mL)	106&24	936	673.2	336	0	STERILIZER

Test organism information:		*Test information:*	
Organism age:	24-HOURS OLD	Randomizing template:	BLUE
Date and times organisms were born between:	01-03-02 1130 TO 1330 MST ~~(0930 TO 1130 EST)~~	Incubator number:	2
Organism source:	ABS BATCH 01-03-02	*Artemia* lot number:	BG0207P
Transfer bowl information:	pH = 7.87 Temperature = 24.1°C	Oven temperature:	60°C
		Drying time:	24-HOURS

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Analyst
0	01-04-02	—	1438	1403	
1	01-05-02	1000	1600	1351	
2	01-06-02	1007	1610	1346	KEK
3	01-07-02	0955	1555	1350	
4	01-08-02	1000	1600	1353	
5	01-09-02	1016	1620	1402	KEK
6	01-10-02	0952	1550	1200	KEK *
7	01-11-02	—		1344	

* TEST WAS NOT RENEWED ± 1-HOUR FROM TEST INITIATION TO MEET 72-HOUR HOLD TIME.

Control information:		Acceptance criteria
% Mortality:	0%	≤ 20%
Average weight per larvae:	1.0697	≥ 0.25 mg/larvae

Summary of test endpoints:	
7-day LC50	>100%
NOEC	100%
LOEC	>100%
ChV	>100%
IC25	>100%

Species: *Pimephales promelas*

Client: SEQUOYAH NUCLEAR PLANT
UV-TREATED

Date: 01-04-02

Survival and Growth Data

Day	CONTROL				10.98%				22%			
	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)	14.821	14.797	14.797	15.124	15.005	14.928	14.990	15.052	14.833	14.793	14.987	14.816
B = Pan + Larvae weight (mg)	25.71	25.21	24.67	26.74	24.14	26.40	25.41	26.625	24.10	24.82	24.93	25.24
Larvae weight (mg) = A – B	10.889	10.413	9.873	11.611	9.135	11.462	10.420	11.573	9.267	10.027	9.943	10.430

Calculations and data reviewed: [illegible]

Comments:

Species: *Pimephales promelas*

Client: SEQUOYAH NUCLEAR PLANT
UV-TREATED

Date: 01-04-02

Survival and Growth Data

Day	43.9%				72%				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	8 2d	10	10	10	10	10	10	10
3	10	10	10	10	8	10	10	10	10	10	10	10
4	10	10	10	10	8	10	10	10	10	10	10	10
5	10	10	10	10	8	10	10	10	10	10	10	10
6	10	10	10	10	8	10	10	10	10	10	10	10
7	10	10	10	10	8	10	10	10	10	10	10	10
A = Pan weight (mg)	14.813	14.794	14.794	14.817	15.121	14.896	14.892	14.912	14.807	15.090	14.865	14.770
B = Pan + Larvae weight (mg)	24.85	24.07	25.34	25.08	21.97	25.48	24.89	25.13	25.12	24.90	23.77	22.75
Larvae weight (mg) = A – B	10.037	9.276	10.546	10.263	6.849	10.584	9.998	10.218	10.313	9.810	8.905	7.960

Calculations and data reviewed: JJ

Comments:

Species: *Pimephales promelas*

Client: SEQUOYAH NUCLEAR PLANT

Date: 01-04-02

Survival and Growth Data

Day	100% INTAKE											
	Y	Z	AA	BB	CC	DD	EE	FF	GG	HH	II	JJ
0	10	10	10	10								
1	10	10	10	10								
2	10	10	10	10								
3	10	10	10	10								
4	10	10	10	10								
5	10	10	10	10								
6	10	10	10	10								
7	10	10	10	10								
A = Pan weight (mg)	15.151	14.779	14.960	14.940								
B = Pan + Larvae weight (mg)	25.24	23.16	24.52	24.74								
Larvae weight (mg) = A – B	10.089	8.381	9.560	9.800								

Calculations and data reviewed: JJ

Comments:

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002, Method 1002.0)

Species: *Pimephales promelas*

Quality Control

Verification of Data Entry, Calculations, and Statistical Analyses

Client: TVA Sequoyah Nuclear Plant - UV Treated

Test dates: January 4 - 11, 2002

Project number: 221

Reviewed by: [signature]

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	14.821	25.710	10.889	1.0889	100.0	1.0697	6.9	Not applicable
	B	10	10	14.797	25.210	10.413	1.0413				
	C	10	10	14.797	24.670	9.873	0.9873				
	D	10	10	15.129	26.740	11.611	1.1611				
10.98%	E	10	10	15.005	24.140	9.135	0.9135	100.0	1.0648	10.7	0.5
	F	10	10	14.938	26.400	11.462	1.1462				
	G	10	10	14.990	25.410	10.420	1.0420				
	H	10	10	15.057	26.630	11.573	1.1573				
22%	I	10	10	14.833	24.100	9.267	0.9267	100.0	0.9917	4.9	7.3
	J	10	10	14.793	24.820	10.027	1.0027				
	K	10	10	14.987	24.930	9.943	0.9943				
	L	10	10	14.810	25.240	10.430	1.0430				
43.9%	M	10	10	14.813	24.850	10.037	1.0037	100.0	1.0031	5.4	6.2
	N	10	10	14.794	24.070	9.276	0.9276				
	O	10	10	14.794	25.340	10.546	1.0546				
	P	10	10	14.817	25.080	10.263	1.0263				
72%	Q	10	8	15.121	21.970	6.849	0.6849	95.0	0.9412	18.3	12.0
	R	10	10	14.896	25.480	10.584	1.0584				
	S	10	10	14.892	24.890	9.998	0.9998				
	T	10	10	14.912	25.130	10.218	1.0218				
100% Outfall 101	U	10	10	14.807	25.120	10.313	1.0313	100.0	0.9247	11.2	13.6
	V	10	10	15.090	24.900	9.810	0.9810				
	W	10	10	14.865	23.770	8.905	0.8905				
	X	10	10	14.790	22.750	7.960	0.7960				
100% Intake	Y	10	10	15.151	25.240	10.089	1.0089	100.0	0.9458	7.9	11.6
	Z	10	10	14.779	23.160	8.381	0.8381				
	AA	10	10	14.960	24.520	9.560	0.9560				
	BB	10	10	14.940	24.740	9.800	0.9800				

Outfall 101:

Dunnett's MSD value: 0.1762

PMSD: 16.5

Intake:

Dunnett's MSD value: 0.1022

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 Pimephales growth by 19.1% 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 Growth

Start Date:	1/4/02	Test ID:	PpFRCR	Sample ID:	TVA SQN - Outfall 101 UV Treated
End Date:	1/11/02	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	1.0889	1.0413	0.9873	1.1611
10.98	0.9135	1.1462	1.0420	1.1573
22	0.9267	1.0027	0.9943	1.0430
43.9	1.0037	0.9276	1.0546	1.0263
72	0.6849	1.0584	0.9998	1.0218
100	1.0313	0.9810	0.8905	0.7960

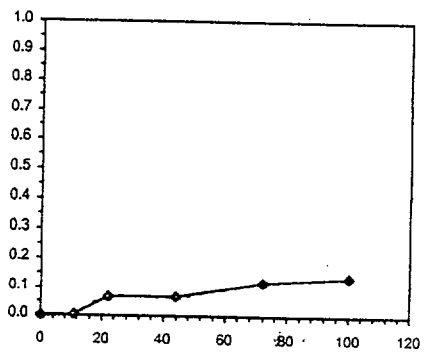
			Transform: Untransformed						1-Tailed		Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	1.0697	1.0000	1.0697	0.9873	1.1611	6.895	4				1.0697	1.0000
10.98	1.0648	0.9954	1.0648	0.9135	1.1573	10.652	4	0.067	2.410	0.1762	1.0648	0.9954
22	0.9917	0.9271	0.9917	0.9267	1.0430	4.866	4	1.067	2.410	0.1762	0.9974	0.9324
43.9	1.0031	0.9377	1.0031	0.9276	1.0546	5.427	4	0.911	2.410	0.1762	0.9974	0.9324
72	0.9412	0.8799	0.9412	0.6849	1.0584	18.336	4	1.757	2.410	0.1762	0.9412	0.8799
100	0.9247	0.8645	0.9247	0.7960	1.0313	11.215	4	1.983	2.410	0.1762	0.9247	0.8645

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.921115994	0.884	-1.09446415	1.241605879
Bartlett's Test indicates equal variances (p = 0.30)	6.01902914	15.08631706		

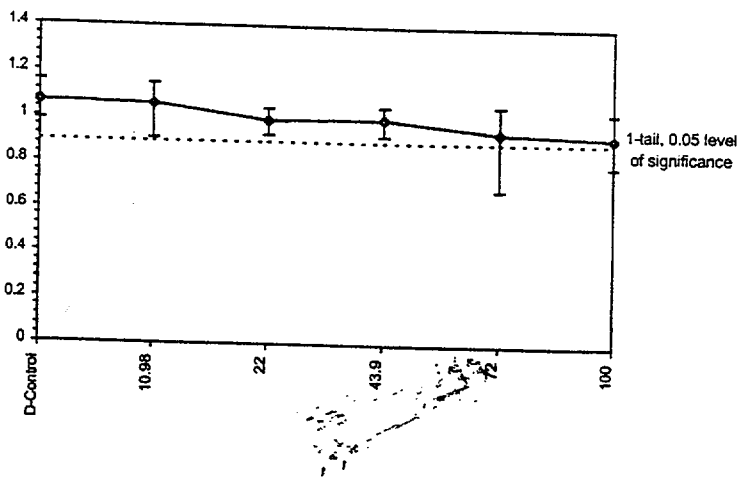
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	100	>100		1	0.176189616	0.164717072	0.01459424	0.010689478	0.283319354	5, 18

Linear Interpolation (80 Resamples)

Point	%	SD	95% CL(Exp)		Skew
IC05	18.925	17.057	0.000	107.683	2.5989
IC10	61.258				
IC15	>100				
IC20	>100				
IC25	>100				
IC40	>100				
IC50	>100				



Dose-Response Plot



Environmental Testing Solutions, LLC

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

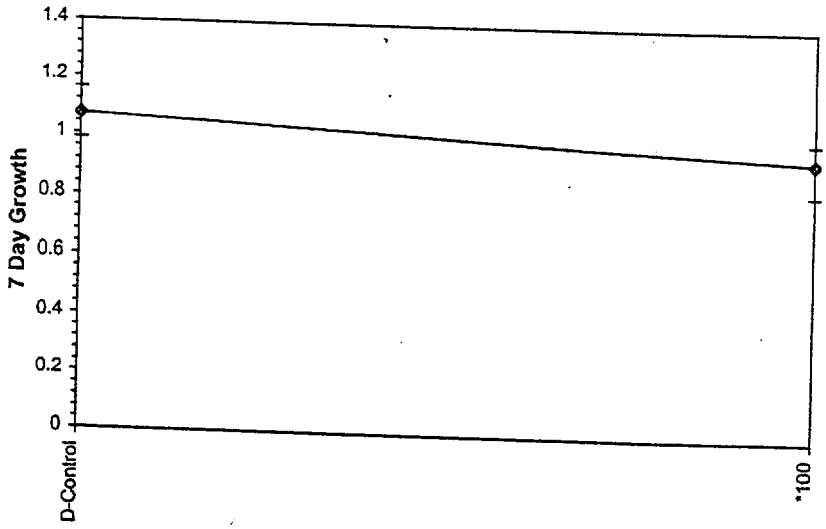
Start Date:	1/4/02	Test ID:	PpPFCR	Sample ID:	TVA SQN - Intake UV Treated
End Date:	1/11/02	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	1.0889	1.0413	0.9873	1.1611
100	1.0089	0.8381	0.9560	0.9800

Conc-%	Mean	N-Mean	Transform: Untransformed Mean	Min	Max	CV%	N	t-Stat	1-Tailed Critical	MSD
D-Control	1.0697	1.0000	1.0697	0.9873	1.1611	6.895	4			
*100	0.9458	0.8842	0.9458	0.8381	1.0089	7.925	4	2.356	1.943	0.1022

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.954538047	0.749	-0.44589651	-0.80413952
F-Test indicates equal variances (p = 0.98)	1.032824516	47.46834564		
Hypothesis Test (1-tail, 0.05)				
Homoscedastic t Test indicates significant differences				

Dose-Response Plot



Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002, Methods 1002.0)

Species: *Pimephales promelas*

Summary of Chemical Analyses

Client: TVA Sequoyah Nuclear Plant

Test dates: January 4 - 11, 2002

Project number: 221

Reviewed by: [illegible]

Concentration	Parameter	Initial Nontreated				Final Nontreated				Initial UV Treated				Final UV Treated			
		Minimum	Maximum	Mean	S	Minimum	Maximum	Mean	S	Minimum	Maximum	Mean	S	Minimum	Maximum	Mean	S
Control	pH (SU)	8.01	8.10	8.06	0.03	7.62	7.84	7.75	0.08	7.66	8.04	7.92	0.13	7.68	7.88	7.76	0.07
	DO (mg/L)	8.0	8.1	8.0	0.0	7.0	7.8	7.5	0.3	7.9	8.1	8.0	0.1	7.2	7.8	7.5	0.2
	Conductivity (µmhos/cm)	300	320	308	8					296	321	307	8				
	Alkalinity (mg/L $CaCO_3$)	65	68	67	2					65	68	67	2				
	Hardness (mg/L $CaCO_3$)	90	90	90	0					90	90	90	0				
	Initial Temperature (°C)					24.4	25.5	24.8	0.4					24.4	25.4	24.8	0.4
	Final Temperature (°C)					24.1	24.8	24.4	0.2					24.2	24.7	24.4	0.2
10.98%	pH (SU)	8.03	8.11	8.07	0.03	7.60	7.79	7.73	0.06	7.81	8.03	7.97	0.08	7.68	7.87	7.77	0.07
	DO (mg/L)	7.9	8.1	8.0	0.1	7.1	7.9	7.4	0.3	7.9	8.2	8.1	0.1	7.2	7.8	7.6	0.2
	Conductivity (µmhos/cm)	290	315	300	8					289	318	298	9				
	Initial Temperature (°C)					24.4	25.4	24.8	0.3					24.4	25.4	24.8	0.4
	Final Temperature (°C)					24.2	24.8	24.4	0.2					24.2	24.7	24.4	0.2
22%	pH (SU)	8.02	8.10	8.06	0.03	7.57	7.86	7.72	0.09	7.93	8.06	8.00	0.05	7.62	7.81	7.74	0.07
	DO (mg/L)	7.9	8.2	8.1	0.1	7.1	7.9	7.4	0.3	8.0	8.3	8.2	0.2	7.3	7.8	7.6	0.2
	Conductivity (µmhos/cm)	281	304	290	8					278	305	287	9				
	Initial Temperature (°C)					24.4	25.4	24.8	0.3					24.4	25.4	24.8	0.4
	Final Temperature (°C)					24.1	24.6	24.4	0.2					24.0	24.7	24.4	0.2
43.9%	pH (SU)	7.92	8.08	8.02	0.06	7.55	7.82	7.69	0.09	7.96	8.06	8.02	0.04	7.64	7.83	7.75	0.07
	DO (mg/L)	8.0	8.4	8.2	0.1	7.1	7.9	7.4	0.3	8.1	8.4	8.3	0.2	7.2	7.9	7.6	0.2
	Conductivity (µmhos/cm)	257	277	265	6.8					256	277	263	7				
	Initial Temperature (°C)					24.4	25.4	24.8	0.4					24.4	25.4	24.8	0.4
	Final Temperature (°C)					24.1	24.7	24.4	0.2					24.1	24.7	24.4	0.2
72%	pH (SU)	7.93	8.06	7.99	0.06	7.54	7.79	7.69	0.08	7.96	8.06	8.01	0.04	7.60	7.83	7.72	0.07
	DO (mg/L)	8.0	8.5	8.3	0.2	7.0	7.8	7.4	0.3	8.1	8.5	8.3	0.2	7.4	7.9	7.6	0.2
	Conductivity (µmhos/cm)	225	242	232	6					220	244	228	8				
	Initial Temperature (°C)					24.4	25.4	24.8	0.4					24.4	25.4	24.8	0.4
	Final Temperature (°C)					24.1	24.8	24.4	0.2					24.1	24.7	24.4	0.2
100% Outfall 101	pH (SU)	7.85	8.05	7.93	0.08	7.53	7.81	7.66	0.10	7.95	8.07	7.99	0.05	7.59	7.81	7.72	0.08
	DO (mg/L)	8.1	8.6	8.4	0.2	7.0	7.9	7.4	0.3	8.1	8.6	8.4	0.2	7.3	7.9	7.6	0.2
	Conductivity (µmhos/cm)	192	207	197	5					189	207	196	6				
	Alkalinity (mg/L $CaCO_3$)	61	69	65	4					61	69	65	4				
	Hardness (mg/L $CaCO_3$)	74	80	77	3					74	80	77	3				
	Total Residual Chlorine (mg/L)	< 0.10	< 0.10	< 0.10	0					< 0.10	< 0.10	< 0.10	0				
	Initial Temperature (°C)					24.4	25.4	24.8	0.3					24.4	25.4	24.8	0.4
	Final Temperature (°C)					24.1	24.8	24.4	0.2					24.1	24.7	24.4	0.2
100% Intake	pH (SU)	7.76	8.03	7.87	0.10	7.61	7.83	7.74	0.08	7.91	8.05	7.97	0.05	7.62	7.82	7.71	0.07
	DO (mg/L)	8.1	8.6	8.4	0.2	7.0	7.9	7.4	0.3	8.1	8.7	8.4	0.2	7.3	7.8	7.6	0.2
	Conductivity (µmhos/cm)	192	206	196	5					189	207	194	6				
	Alkalinity (mg/L $CaCO_3$)	62	66	64	2					62	66	64	2				
	Hardness (mg/L $CaCO_3$)	74	80	78	3					74	80	78	3				
	Total Residual Chlorine (mg/L)	< 0.10	< 0.10	< 0.10	0					< 0.10	< 0.10	< 0.10	0				
	Initial Temperature (°C)					24.4	25.4	24.8	0.4					24.4	25.4	24.8	0.4
	Final Temperature (°C)					24.1	24.8	24.4	0.2					24.1	24.7	24.4	0.2
Overall Temperature (°C) (including all concentrations for initial and final temperatures)						24.1	25.5	24.6	0.3					24.0	25.4	24.6	0.3

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Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002, Method 1002.0)

Species: *Pimephales promelas*

Daily Chemical Analyses

Client: TVA Sequoyah Nuclear Plant - Nontreated

Test dates: January 4 - 11, 2002

Project number: 221

Reviewed by: J Sumner

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)	8.04	7.82	8.01	7.84	8.02	7.72	8.06	7.78	8.10	7.78	8.08	7.62	8.09	7.70
	DO (mg/L)	8.1	7.8	8.0	7.6	8.0	7.0	8.0	7.7	8.0	7.4	8.0	7.3	8.0	7.5
	Conductivity (µmhos/cm)	320		304		305		302		314		314		300	
	Alkalinity (mg/L $CaCO_3$)	68								65					
	Hardness (mg/L $CaCO_3$)	90								90					
	Temperature (°C)	24.6	24.1	24.5	24.6	24.9	24.3	24.4	24.8	25.5	24.4	25.0	24.5	24.7	24.2
10.98%	pH (SU)	8.06	7.79	8.03	7.77	8.04	7.71	8.07	7.75	8.11	7.73	8.10	7.60	8.07	7.75
	DO (mg/L)	7.9	7.9	8.1	7.5	8.0	7.2	8.0	7.6	8.0	7.1	8.0	7.2	8.1	7.5
	Conductivity (µmhos/cm)	315		290		298		297		303		302		297	
	Temperature (°C)	24.7	24.2	24.5	24.6	25.0	24.3	24.4	24.8	25.4	24.4	25.0	24.5	24.7	24.2
22%	pH (SU)	8.07	7.86	8.03	7.76	8.02	7.64	8.06	7.74	8.09	7.77	8.10	7.57	8.06	7.70
	DO (mg/L)	8.1	7.9	8.1	7.5	8.1	7.1	8.0	7.4	8.1	7.1	7.9	7.1	8.2	7.6
	Conductivity (µmhos/cm)	304		281		288		283		289		295		289	
	Temperature (°C)	24.7	24.1	24.5	24.6	24.9	24.3	24.4	24.6	25.4	24.4	25.0	24.5	24.7	24.2
43.9%	pH (SU)	8.06	7.82	7.99	7.76	7.98	7.63	8.03	7.71	8.06	7.73	8.08	7.55	7.92	7.65
	DO (mg/L)	8.2	7.9	8.3	7.5	8.1	7.1	8.1	7.4	8.2	7.2	8.0	7.2	8.4	7.6
	Conductivity (µmhos/cm)	277		257		261		261		264		271		263	
	Temperature (°C)	24.6	24.1	24.4	24.6	24.9	24.3	24.4	24.7	25.4	24.4	25.1	24.5	24.7	24.2
72%	pH (SU)	8.06	7.79	7.94	7.72	7.93	7.63	7.97	7.69	8.03	7.76	8.06	7.54	7.96	7.67
	DO (mg/L)	8.3	7.8	8.5	7.5	8.2	7.1	8.3	7.4	8.3	7.0	8.0	7.1	8.4	7.7
	Conductivity (µmhos/cm)	242		225		229		226		231		236		232	
	Temperature (°C)	24.7	24.1	24.4	24.6	25.0	24.3	24.4	24.8	25.4	24.4	24.9	24.5	24.7	24.2
100% Outfall 101	pH (SU)	8.05	7.81	7.88	7.67	7.85	7.55	7.89	7.67	7.97	7.73	7.99	7.53	7.85	7.64
	DO (mg/L)	8.5	7.9	8.5	7.5	8.2	7.0	8.4	7.4	8.3	7.1	8.1	7.2	8.6	7.8
	Conductivity (µmhos/cm)	207		194		192		192		198		199		200	
	Alkalinity (mg/L $CaCO_3$)	64				61				69					
	Hardness (mg/L $CaCO_3$)	78				74				80					
	Total Residual Chlorine (mg/L)	< 0.10				< 0.10				< 0.10					
	Temperature (°C)	24.7	24.1	24.5	24.5	25.0	24.3	24.4	24.8	25.4	24.4	25.0	24.5	24.7	24.2
100% Intake	pH (SU)	8.03	7.81	7.83	7.71	7.78	7.61	7.83	7.76	7.91	7.77	7.94	7.83	7.76	7.69
	DO (mg/L)	8.5	7.9	8.6	7.5	8.1	7.0	8.4	7.5	8.3	7.0	8.2	7.2	8.6	7.7
	Conductivity (µmhos/cm)	206		193		193		192		195		198		198	
	Alkalinity (mg/L $CaCO_3$)	65 ~~66~~				65				62					
	Hardness (mg/L $CaCO_3$)	80				80				74					
	Total Residual Chlorine (mg/L)	< 0.10				< 0.10				< 0.10					
	Temperature (°C)	24.7	24.1	24.4	24.6	25.0	24.3	24.4	24.8	25.4	24.4	24.9	24.5	24.7	24.2

snp 01-04-02 vl

Species: *Pimephales promelas*

Client: TVA SEQUOYAH NUCLEAR PLANT

Date: 01-04-02

Full-strength Chemistry: OUTFALL 101 NONTREATED

Parameter	Sample Number:			Control Batch:		
	1	2	3	01-03-02	01-06-02	
pH (S.U.)	8.05	7.85	7.97	8.04	8.10	
DO (mg/L)	8.5	8.2	8.3	8.1	8.0	
Conductivity (µmhos/cm)	206	192	198	320	314	
Alkalinity (mg $CaCO_3$/L)	~~68~~ 64	61	69	68	65	
Hardness (mg $CaCO_3$/L)	78	74	80	90	90	
Chlorine (mg/L)	<0.10	<0.10	<0.10			
Collection start date	01-02-02	01-04-02	01-06-02			
Collection end time	0818	0939	1234			
Grab or Composite (duration)	23-HOUR COMPOSITE	23-HOUR COMPOSITE	23-HOUR COMPOSITE			
Temperature (°C) upon receipt	0.8°C	0.4°C	1.0°C			
Physical characteristics	LIGHT BEIGE CLEAR	CLEAR NO COLOR	CLEAR NO COLOR FLOATING BLACK PARTICLES			
Dates sample used	01-04-02 01-05-02	01-06-02 01-07-02	01-08-02 01-09-02 01-10-02			
ETS Project and Sample numbers	221 02-01-04.01	020105.01	020108.01			

Species: ***Pimephales promelas***

Client: TVA SEQUOYAH NUCLEAR PLANT

Date: 01-04-02

Full-strength Chemistry: INTAKE - NONTREATED

Parameter	Sample Number:			Control Batch:		
	1	2	3	01-03-02	01-06-02	
pH (S.U.)	8.03	7.78	7.91	8.04	8.10	
DO (mg/L)	8.5	8.1	8.3	8.1	8.0	
Conductivity (µmhos/cm)	206	193	195	320	314	
Alkalinity (mg $CaCO_3$/L)	65	65	62	68	65	
Hardness (mg $CaCO_3$/L)	80	80	74	90	90	
Chlorine (mg/L)	<0.10	<0.10	<0.10			
Collection start date	01-02-02	01-04-02	01-06-02			
Collection end time	0913	0932	1251			
Grab or Composite (duration)	~~24~~ 23-HOUR COMPOSITE	23-HOUR COMPOSITE	23-HOUR COMPOSITE			
Temperature (°C) upon receipt	0.6°C	0.6°C	0.8°C			
Physical characteristics	LIGHT BEIGE CLEAR FLOATING PARTICLES	CLEAR NO COLOR	CLEAR NO COLOR FLOATING BLACK PARTICLES			
Dates sample used	01-04-02 01-05-02	01-06-02 01-07-02	01-08-02 01-09-02 01-10-02			
ETS Project and Sample numbers	221 020104.02	020105.02	020108.02			

Species: ***Pimephales promelas***

Client: SEQUOYAH NUCLEAR PLANT NONTREATED

Date: 01-04-02

Daily Chemistry:

Concentration	Parameter	Day 0 Initial	Day 0 Final	Day 1 Initial	Day 1 Final	Day 2 Initial	Day 2 Final
CONTROL MHS	pH (S.U.)	8.04	7.82	8.01	7.84	8.02	7.72
	DO (mg/L)	8.1	7.8	8.0	7.6	8.0	7.0
	Conductivity (µmhos/cm)	320		~~296~~ 304		305	
	Temperature (°C)	24.6	24.1	24.5	24.6	24.9	24.3
10.98%	pH (S.U.)	8.06	7.79	8.03	7.77	8.04	7.71
	DO (mg/L)	7.9	7.9	8.1	7.5	8.0	7.2
	Conductivity (µmhos/cm)	315		290		298	
	Temperature (°C)	24.7	24.2	24.5	24.6	25.0	24.3
22%	pH (S.U.)	8.07	7.86	8.03	7.76	8.02	7.64
	DO (mg/L)	8.1	7.9	8.1	7.5	8.1	7.1
	Conductivity (µmhos/cm)	304		281		288	
	Temperature (°C)	24.7	24.1	24.5	24.6	24.9	24.3
43.9%	pH (S.U.)	8.06	7.82	7.99	7.76	7.98	7.63
	DO (mg/L)	8.2	7.9	8.3	7.5	8.1	7.1
	Conductivity (µmhos/cm)	277		257		261	
	Temperature (°C)	24.6	24.1	24.4	24.6	24.9	24.3
72%	pH (S.U.)	8.06	7.79	7.94	7.72	7.93	7.63
	DO (mg/L)	8.3	7.8	8.5	7.5	8.2	7.1
	Conductivity (µmhos/cm)	242		225		229	
	Temperature (°C)	24.7	24.1	24.4	24.6	25.0	24.3
100%	pH (S.U.)	8.05	7.81	7.88	7.67	7.85	7.55
	DO (mg/L)	8.5	7.9	8.5	7.5	8.2	7.0
	Conductivity (µmhos/cm)	207		194		192	
	Temperature (°C)	24.7	24.1	24.5	24.5	25.0	24.3
100% INTAKE	pH (S.U.)	8.03	7.81	7.83	7.71	7.78	7.61
	DO (mg/L)	8.5	7.9	8.6	7.5	8.1	7.0
	Conductivity (µmhos/cm)	206		193		193	
	Temperature (°C)	24.7	24.1	24.4	24.6	25.0	24.3

Species: *Pimephales promelas*

Client: SEQUOYAH NUCLEAR PLANT NONTREATED

Date: 01-04-02

Concentration	Parameter	Day 3 Initial	Day 3 Final	Day 4 Initial	Day 4 Final	Day 5 Initial	Day 5 Final	Day 6 Initial	Day 6 Final
CONTROL	pH (S.U.)	8.06	7.78	8.10	7.78	8.08	7.62	8.09	7.70
	DO (mg/L)	8.0	7.7	8.0	7.4	8.0	7.3	8.0	7.5
MHS	Conductivity (µmhos/cm)	302		314		314		300	
	Temperature (°C)	24.4	24.8	25.5	24.4	25.0	24.5	24.7	24.2
	pH (S.U.)	8.07	7.75	8.11	7.73	8.10	7.60	8.07	7.75
	DO (mg/L)	8.0	7.6	8.0	7.1	8.0	7.2	8.1	7.5
10.98%	Conductivity (µmhos/cm)	297		303		302		297	
	Temperature (°C)	24.4	24.8	25.4	24.4	25.0	24.5	24.7	24.2
	pH (S.U.)	8.06	7.74	8.09	7.77	8.10	7.57	8.06	7.70
	DO (mg/L)	8.0	7.4	8.1	7.1	7.9	7.1	8.2	7.6
22%	Conductivity (µmhos/cm)	283		289		295		289	
	Temperature (°C)	24.4	24.6	25.4	24.4	25.0	24.5	24.7	24.2
	pH (S.U.)	8.03	7.71	8.06	7.73	8.08	7.55	7.92	7.65
	DO (mg/L)	8.1	7.4	8.2	7.2	8.0	7.2	8.4	7.6
43.9%	Conductivity (µmhos/cm)	261		264		271		263	
	Temperature (°C)	24.4	24.7	25.4	24.4	25.1	24.5	24.7	24.2
	pH (S.U.)	7.97	7.69	8.03	7.76	8.06	7.54	7.96	7.67
	DO (mg/L)	8.3	7.4	8.3	7.0	8.0	7.1	8.4	7.7
72%	Conductivity (µmhos/cm)	226		231		236		232	
	Temperature (°C)	24.4	24.8	25.4	24.4	24.9	24.5	24.7	24.2
	pH (S.U.)	7.89	7.67	7.97	7.73	8 7.99	7.53	7.85	7.64
	DO (mg/L)	8.4	7.4	8.3	7.1	8.1	7.2	8.6	7.8
100%	Conductivity (µmhos/cm)	192		198		199		200	
	Temperature (°C)	24.4	24.8	25.4	24.4	25.0	24.5	24.7	24.2
	pH (S.U.)	7.83	7.76	7.91	7.77	7.94	7.83	7.76	7.69
	DO (mg/L)	8.4	7.5	8.3	7.0	8.2	7.2	8.6	7.7
100% INTAKE	Conductivity (µmhos/cm)	192		195		198		198	
	Temperature (°C)	24.4	24.8	25.4	24.4	24.9	24.5	24.7	24.2

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002, Method 1002.0)

Species: *Pimephales promelas*

Daily Chemical Analyses

Client: TVA Sequoyah Nuclear Plant - UV Treated

Test dates: January 4 - 11, 2002

Project number: 221

Reviewed by: [signature]

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)	8.00	7.77	8.03	7.74	7.66	7.69	7.86	7.83	7.93	7.88	8.04	7.68	7.90	7.76
	DO (mg/L)	8.1	7.8	8.1	7.5	8.0	7.2	8.0	7.8	7.9	7.3	8.0	7.7	8.0	7.5
	Conductivity (µmhos/cm)	321		296		305		301		309		310		304	
	Alkalinity (mg/L $CaCO_3$)	68								65					
	Hardness (mg/L $CaCO_3$)	90								90					
	Temperature (°C)	24.6	24.2	24.4	24.6	25.0	24.3	24.4	24.7	25.4	24.3	24.9	24.5	24.7	24.2
10.98%	pH (SU)	8.03	7.68	7.99	7.87	7.81	7.75	7.96	7.80	8.01	7.81	8.03	7.69	7.95	7.78
	DO (mg/L)	8.2	7.8	8.1	7.4	8.0	7.2	8.2	7.8	7.9	7.5	8.0	7.7	8.2	7.5
	Conductivity (µmhos/cm)	318		289		296		294		300		296		293	
	Temperature (°C)	24.6	24.2	24.4	24.6	25.0	24.3	24.4	24.7	25.4	24.3	24.9	24.5	24.6	24.2
22%	pH (SU)	8.06	7.76	7.99	7.81	7.93	7.70	8.01	7.80	8.05	7.78	8.05	7.62	7.94	7.70
	DO (mg/L)	8.2	7.8	8.3	7.4	8.0	7.3	8.3	7.8	8.0	7.5	8.0	7.8	8.3	7.5
	Conductivity (µmhos/cm)	305		278		286		282		289		285		285	
	Temperature (°C)	24.6	24.2	24.5	24.6	25.0	24.3	24.4	24.7	25.4	24.4	25.1	24.5	24.6	24.0
43.9%	pH (SU)	8.06	7.71	7.99	7.83	7.98	7.71	8.02	7.79	8.06	7.80	8.06	7.64	7.96	7.79
	DO (mg/L)	8.3	7.9	8.4	7.4	8.1	7.2	8.4	7.8	8.1	7.7	8.1	7.7	8.4	7.5
	Conductivity (µmhos/cm)	277		256		261		257		264		263		264	
	Temperature (°C)	24.6	24.2	24.4	24.6	25.0	24.3	24.4	24.7	25.4	24.4	25.1	24.5	24.6	24.1
72%	pH (SU)	8.06	7.74	7.99	7.83	7.98	7.68	8.00	7.76	8.05	7.75	8.06	7.60	7.96	7.70
	DO (mg/L)	8.5	7.9	8.3	7.4	8.1	7.4	8.5	7.7	8.1	7.7	8.1	7.6	8.4	7.6
	Conductivity (µmhos/cm)	244		220		228		222		231		222		226	
	Temperature (°C)	24.6	24.2	24.5	24.6	24.9	24.3	24.4	24.7	25.4	24.4	25.0	24.5	24.6	24.2
100% Outfall 101	pH (SU)	8.04	7.71	7.97	7.81	7.95	7.67	7.96	7.72	8.02	7.80	8.07	7.59	7.95	7.77
	DO (mg/L)	8.6	7.9	8.3	7.4	8.2	7.3	8.6	7.7	8.1	7.7	8.2	7.6	8.5	7.6
	Conductivity (µmhos/cm)	207		190		194		189		198		196		198	
	Alkalinity (mg/L $CaCO_3$)	64				61				69					
	Hardness (mg/L $CaCO_3$)	78				74				80					
	Total Residual Chlorine (mg/L)	< 0.10				< 0.10				< 0.10					
	Temperature (°C)	24.6	24.2	24.5	24.6	25.0	24.3	24.4	24.7	25.4	24.3	25.0	24.5	24.7	24.1
100% Intake	pH (SU)	8.05	7.68	7.97	7.82	7.92	7.62	7.94	7.71	8.01	7.76	8.01	7.64	7.91	7.74
	DO (mg/L)	8.7	7.7	8.4	7.5	8.1	7.3	8.5	7.7	8.1	7.7	8.3	7.6	8.4	7.8
	Conductivity (µmhos/cm)	207		190		189		189		194		195		195	
	Alkalinity (mg/L $CaCO_3$)	66				65				62					
	Hardness (mg/L $CaCO_3$)	80				80				74					
	Total Residual Chlorine (mg/L)	< 0.10				< 0.10				< 0.10					
	Temperature (°C)	24.6	24.2	24.5	24.6	25.0	24.3	24.4	24.7	25.4	24.3	25.0	24.5	24.6	24.1

Species: *Pimephales promelas*

Client: SEQUOYAH NUCLEAR PLANT UV TREATED

Date: 01-04-02

Daily Chemistry:

Concentration	Parameter	Day 0 Initial	Day 0 Final	Day 1 Initial	Day 1 Final	Day 2 Initial	Day 2 Final
CONTROL	pH (S.U.)	8.00	7.77	8.03	7.74	7.66	7.69
	DO (mg/L)	8.1	7.8	8.1	7.5	8.0	~~8.0~~ 7.2
NHS	Conductivity (µmhos/cm)	321		~~304~~ 296		305	
	Temperature (°C)	24.6	24.2	24.4	24.6	25.0	24.3
10.98%	pH (S.U.)	8.03	7.68	7.99	7.87	7.81	7.75
	DO (mg/L)	8.2	7.8	8.1	7.4	8.0	7.2
	Conductivity (µmhos/cm)	318		289		296	
	Temperature (°C)	24.6	24.2	24.4	24.6	25.0	24.3
22%	pH (S.U.)	8.06	7.76	7.99	7.81	7.93	7.70
	DO (mg/L)	8.2	7.8	8.3	7.4	8.0	7.3
	Conductivity (µmhos/cm)	305		278		286	
	Temperature (°C)	24.6	24.2	24.5	24.6	25.0	24.3
43.9%	pH (S.U.)	8.06	7.71	7.99	7.83	7.98	7.71
	DO (mg/L)	8.3	7.9	8.4	7.4	8.1	7.2
	Conductivity (µmhos/cm)	277		256		261	
	Temperature (°C)	24.6	24.2	24.4	24.6	25.0	24.3
72%	pH (S.U.)	8.06	7.74	7.99	7.83	7.98	7.68
	DO (mg/L)	8.5	7.9	8.3	7.4	8.1	7.4
	Conductivity (µmhos/cm)	244		220		228	
	Temperature (°C)	24.6	24.2	24.5	24.6	24.9	24.3
100%	pH (S.U.)	8.04	7.71	7.97	7.81	7.95	7.67
	DO (mg/L)	8.6	7.9	8.3	7.4	8.2	7.3
	Conductivity (µmhos/cm)	207		190		194	
	Temperature (°C)	24.6	24.2	24.5	24.6	25.0	24.3
100% INTAKE	pH (S.U.)	8.05	7.68	7.97	7.82	7.92	7.62
	DO (mg/L)	8.7	7.7	8.4	7.5	8.1	7.3
	Conductivity (µmhos/cm)	207		190		189	
	Temperature (°C)	24.6	24.2	24.5	24.6	25.0	24.3

Species: *Pimephales promelas*

Client: SEQUOYAH NUCLEAR PLANT UV TREATED

Date: 01-04-02

Concentration	Parameter	Day 3 Initial	Day 3 Final	Day 4 Initial	Day 4 Final	Day 5 Initial	Day 5 Final	Day 6 Initial	Day 6 Final
CONTROL	pH (S.U.)	7.86	7.83	7.93	7.88	~~8.05~~ 8.04	7.68	~~8.09~~ 7.90	7.76
	DO (mg/L)	8.0	7.8	7.9	7.3	8.0	7.7	8.0	7.5
MHS	Conductivity (µmhos/cm)	301		~~314~~ 309		310		304	
	Temperature (°C)	24.4	24.7	25.4	24.3	24.9	24.5	24.7	24.2
10.98%	pH (S.U.)	7.96	7.80	8.01	7.81	8.03	7.69	7.95	7.78
	DO (mg/L)	8.2	7.8	7.9	7.5	8.0	7.7	8.2	7.5
	Conductivity (µmhos/cm)	294		~~303~~ 300		296		293	
	Temperature (°C)	24.4	24.7	25.4	24.3	24.9	24.5	24.6	24.2
22%	pH (S.U.)	8.01	7.80	8.05	7.78	8.05	7.62	7.94	7.76
	DO (mg/L)	8.3	7.8	8.0	7.5	8.0	7.8	8.3	7.5
	Conductivity (µmhos/cm)	282		289		285		285	
	Temperature (°C)	24.4	24.7	25.4	24.4	25.1	24.5	24.6	24.0
43.97%	pH (S.U.)	8.02	7.79	8.06	7.80	8.06	7.64	7.96	7.79
	DO (mg/L)	8.4	7.8	8.1	7.7	8.1	7.7	8.4	7.5
	Conductivity (µmhos/cm)	257		264		263		264	
	Temperature (°C)	24.4	24.7	25.4	24.4	25.1	24.5	24.6	24.1
72%	pH (S.U.)	8.00	7.76	8.05	7.75	8.06	7.60	7.96	7.70
	DO (mg/L)	8.5	7.7	8.1	7.7	8.1	7.6	8.4	7.6
	Conductivity (µmhos/cm)	222		231		222		226	
	Temperature (°C)	24.4	24.7	25.4	24.4	25.0	24.5	24.6	24.2
100%	pH (S.U.)	7.96	7.72	8.02	7.80	~~8.04~~ 8.07	7.59	7.95	7.77
	DO (mg/L)	8.6	7.7	8.1	7.7	8.2	7.6	8.5	7.6
	Conductivity (µmhos/cm)	189		198		196		198	
	Temperature (°C)	24.4	24.7	25.4	24.3	25.0	24.5	24.1	24.1
100% INTAKE	pH (S.U.)	7.94	7.71	8.01	7.76	8.01	7.64	7.91	7.74
	DO (mg/L)	8.5	7.7	8.1	7.7	8.3	7.6	8.4	7.8
	Conductivity (µmhos/cm)	189		~~195~~ 194		195		195	
	Temperature (°C)	24.4	24.7	25.4	24.3	25.0	24.5	24.6	24.1

Alkalinity
(EPA Method 310.1)

Matrix: Water, MDL = 1.0 mg $CaCO_3$/L

Analyst	KJK
Date analyzed	01.14.02

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
5.0	INR038	INS5078	0.1	12.5	12.4	0.0202	10.1

Blk correction - 0.0 - 0.1 = 0.1 ml

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%)
INS5079	100	100	12.5	22.2	9.7	10.1	98	98%

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%)
12-22-01	MHS H_2O	100	22.2	29.1	6.9	10.1	S 70	
12-22-01	Duplicate	100	29.1	36.0	6.9	10.1	D 70	–

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)
INS5079	50	100	29.1	40.7	11.6	10.1	117

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%)
70	47	94%

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
12-17-01	MHS H_2O	100	41.0	47.9	6.9	10.1	70
01-02-02			0.2	7.1	6.9		70
01-03-02			7.1	13.8	6.7		68
01-06-02			13.8	20.2	6.4		65
01-07-02			20.5	26.9	6.4		65
01-09-02			26.9	33.2	6.3		64
01-07-02	SSW H_2O		33.2	36.5	3.3		33
01-13-02			36.5	39.5	3.0		30
020108.07	Anderson 1		39.5	50.0	10.5		110
			0.1	0.5	0.4		

Reviewed by: [signature] Date reviewed: 01-14-02

Alkalinity
(EPA Method 310.1)

Matrix: Water, MDL = 1.0 mg $CaCO_3$/L

Analyst	KEN
Date analyzed	01-14-02

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

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%)
INS5079	100	100	0.5	10.2	9.7	10.1	98	98%

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%)
020112.01	Andeeson 3	100	10.5	21.3	10.8	10.1	S 110	
020112.01	Duplicate	100	21.3	32.0	10.7	↓	D 110	—

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)
INS5079	50	100	21.3	36.9	15.6	10.1	160

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%)
110	50	100%

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
020110.02	Andeeson 2	100	36.9	47.0	10.1	10.1	100
020104.01	SON 1		0.8	7.1	6.3		64
020105.01	2		7.2	13.2	6.0		61
020108.01	3		13.2	20.0	6.8		69
020104.02	Intake 1		20.1	26.6	6.5		66
020105.02	2		26.6	33.0	6.4		65
020108.01	3		33.0	39.1	6.1		62

Reviewed by: [signature] Date reviewed: 01-14-02

Total Hardness
(EPA Method 130.2)

Matrix: Water, MDL = 1.0 mg $CaCO_3$/L

Analyst	KEW
Date analyzed	01.14.02

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
1NR011	1NR009	0.1	10.1	10.0	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%)
1NSS008	40	50	10.4	12.4	2.0	20	40	100%

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%)
12.17.01	MHS H2O	50	13.0	17.8	4.8	20	S 96	
12-17-01	Duplicate	50	17.8	22.6	4.8	20	D 96	–

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)
1NSS008	40	50	17.8	24.6	6.8	20	136

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%)
96	40	100%

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	0.0	0.0	0.0	20	ND
12.22.01	MHS H2O		24.6	29.6	4.6		92
01.02.02			29.6	34.2	4.6		92
01.03.02			34.2	38.7	4.5		90
01.06.02			38.7	43.2	4.5		90
01.07.02			43.2	47.5	4.3		86
01.09.02			0.0	4.3	4.3		86
01.07.02	SSW H2O		4.3	6.5	2.2		44
01.13.02			6.5	8.9	2.4		48
02.0108.07	Anderson 1		8.9	17.3	8.4		170

Note: If >15ml of titrant is used, sample must be diluted.

Reviewed by: [illegible]

Date reviewed 01-14-02

Total Hardness
(EPA Method 130.2)

Matrix: Water, MDL = 1.0 mg $CaCO_3$/L

Analyst	KLN
Date analyzed	01-14-02

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

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%)
INS5008	40	50	17.3	19.5	2.2	20	44	110%

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%)
020110.02	Anderson 2	50	19.5	27.1	7.6	20	S 150	
020110.02	Duplicate		27.1	34.6	7.5	20	D 150	—

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)
INS5008	40	50	27.1	36.6	9.5	20	190

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%)
150	40	100%

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Hardness (mg $CaCO_3$/L)
	Blank (should be = 0 mg $CaCO_3$/L)						
020112.01	Anderson 3	50	36.6	44.6	8.0	20	160
020104.01	SAN 1		5.4	9.3	3.9		78
020105.01	2		9.3	13.0	3.7		74
020108.01	3		13.0	17.0	4.0		80
020104.02	Intake 1		17.0	21.0	4.0		80
020105.02	2		21.0	25.0	4.0		80
020108.02	3		25.0	28.7	3.7		74

Note: If >15ml of titrant is used, sample must be diluted.

Reviewed by: [signature]

Date reviewed 01-14-02

Total Residual Chlorine
(EPA Method 330.5)

Matrix: Water, MDL = 0.10 mg/L
Meter: Accumet Model 25 pH/Ion Meter

Analyst	JI
Date analyzed	01-04-02

Iodide reagent:	INR041
Acid reagent:	INR023
Slope:	28.00

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INSS005	INSS005

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%)
INSS005	0.50	0.489	97.8%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
020104.01	TVA SEQUOYAH NUCLEAR OUTFALL 101	LIGHT BEIGE, CLEAR	S <0.00517	
↓	Duplicate		D <0.00500	0%

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00994
020104.02	TVA SEQUOYAH NUCLEAR INTAKE	LIGHT BEIGE, CLEAR FLOATING PARTICLES	<0.00729

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%)
INSS005	0.50	0.510	102.0%

Reviewed by	JI
Date reviewed	01-12-02

Total Residual Chlorine
(EPA Method 330.5)

Matrix: Water, MDL = 0.10 mg/L
Meter: Accumet Model 25 pH/Ion Meter

Analyst	KEK
Date analyzed	01.06.02

Iodide reagent:	IN R041
Acid reagent:	IN R023
Slope:	26.2

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	IN5005	IN5005

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%)
IN5005	0.50	0.507	101.4%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
020105.02	TVA-INTAKE SQN-101-INT.	NO COLOR, CLEAR	S <0.00810	
↓	Duplicate		D <0.00784	—

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00874
020105.01	TVA-OUTFALL 101, SQN-101-EH	NO COLOR, CLEAR	<0.00537

KEK

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%)
IN5005	0.50	0.522	104.4%

Reviewed by	cl
Date reviewed	01-12-02

Total Residual Chlorine
(EPA Method 330.5)

Matrix: Water, MDL = 0.10 mg/L
Meter: Accumet Model 25 pH/Ion Meter

Analyst	KH
Date analyzed	01-08-02

Iodide reagent:	INR041
Acid reagent:	INR023
Slope:	28.0

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INS500S	INS500S

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%)
INS500S	0.50	0.510	102%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
020108.07	Anderson Co. SA	slightly cloudy pale yellow, floating particles	S <0.00673	
↓	Duplicate		D <0.00610	—

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00952
020108.03	RDU-001 - Tritect, Inc	tan, slightly cloudy	<0.00369
020108.04	002	pale tan, slightly cloudy	<0.00145
020108.05	003	orange, cloudy	<0.00849
020108.06	004	pale yellow, slightly cloudy	<0.00197
020108.02	TV Intake - SQN-101-INT	no color, clear, floating bill particles	<0.00272
020108.01	TV Outfall 101 - SQN-101-EH	no color, clear, floating bill particles	<0.00530

KD

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%)
INS500S	0.50	0.528	105.6%

Reviewed by	JI
Date reviewed	01-12-02

Sequoyah Nuclear Plant Biomonitoring
January 4 – 11, 2002

Appendix D

Reference Toxicant Tests and
Control Chart Information

Environmental Testing Solutions, LLC

tassium Chloride Chronic Reference Toxicant Control Cha
for *Pimephales promelas*
using Moderately Hard Synthetic Water

Test number	Test date	7-day IC25 (g KCl/L)	CT (g/L KCl)	S	CV	CT - 2S	CT + 2S	2SD
1	03-07-00	0.5845						
2	03-17-00	0.6755	0.6300	0.0643	10.21%	0.5013	0.7587	0.1287
3	05-23-00	0.6873	0.6491	0.0563	8.67%	0.5366	0.7616	0.1125
4	06-13-00	0.7239	0.6678	0.0592	8.87%	0.5493	0.7863	0.1185
5	06-13-00	0.7014	0.6745	0.0535	7.92%	0.5676	0.7814	0.1069
6	09-19-00	0.6570	0.6716	0.0483	7.20%	0.5749	0.7683	0.0967
7	10-24-00	0.7678	0.6853	0.0572	8.34%	0.5710	0.7997	0.1144
8	11-07-00	0.6523	0.6812	0.0542	7.96%	0.5728	0.7896	0.1084
9	03-13-01	0.6874	0.6819	0.0508	7.44%	0.5804	0.7834	0.1015
10	06-26-01	0.5334	0.6671	0.0670	10.05%	0.5330	0.8011	0.1341
11	07-17-01	0.7438	0.6740	0.0677	10.04%	0.5387	0.8094	0.1354
12	08-21-01	0.6179	0.6694	0.0665	9.94%	0.5363	0.8024	0.1331
13	09-25-01	0.6008	0.6641	0.0665	10.01%	0.5311	0.7970	0.1329
14	11-01-01	0.7105	0.6674	0.0651	9.75%	0.5373	0.7975	0.1301
15	11-06-01	0.6573	0.6667	0.0627	9.41%	0.5412	0.7922	0.1255
16	11-27-01	0.5741	0.6609	0.0649	9.82%	0.5312	0.7907	0.1298
17	12-12-01	0.6324	0.6593	0.0632	9.59%	0.5328	0.7857	0.1264
18	01-04-02	0.4878	0.6497	0.0734	11.30%	0.5029	0.7966	0.1469

Note:

7-d IC25 = 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 IC25).

S = Standard deviation of the IC25 values.

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

$S_{A.75}$ = 0.38, as determined by USEPA for the method and endpoint.

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

$S_{A.90}$ = 0.45, as determined by the USEPA for the method and endpoint.

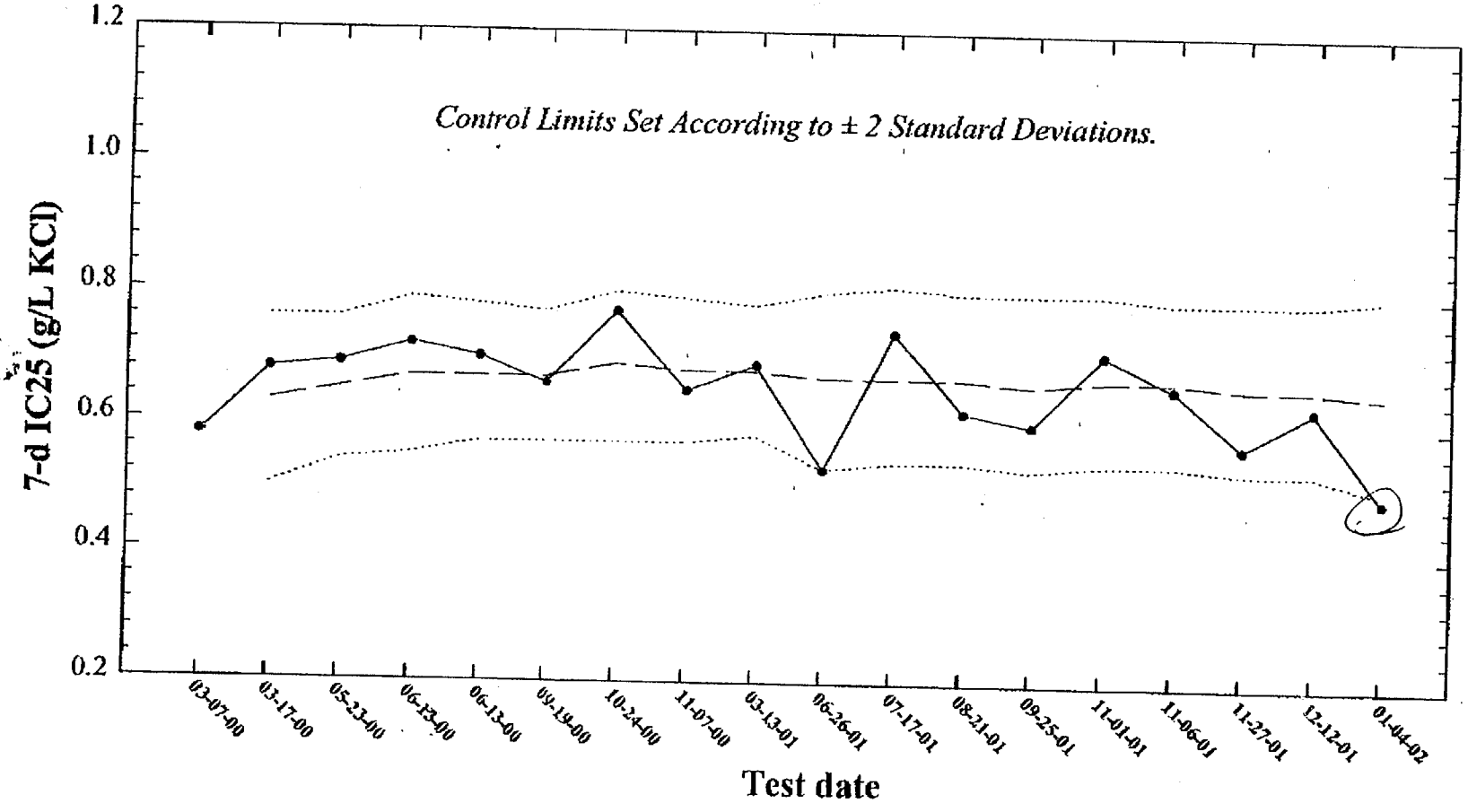
CV = Coefficient of variation of the IC25 values.

SEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity
pplications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US
ıvironmental 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 IC25 = 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 IC25)

Control Limits (mean IC25 ± 2 Standard Deviations)

Environmental Testing Solutions, LLC

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

est number	Test date	7-day IC25 (g KCl/L)	CT (g/L KCl)	S	$S_{A.75}$	$S_{A.90}$	CV	$CT - S_{A.75}$	$CT - S_{A.75}$	$CT - S_{A.90}$	$CT + S_{A.90}$	CT - 2S	CT - 2S
1	03-07-00	0.58											
2	03-17-00	0.68	0.63	0.06	0.24	0.28	0.10	0.25	1.01	0.18	1.08	0.50	0.76
3	05-23-00	0.69	0.65	0.06	0.25	0.29	0.09	0.27	1.03	0.20	1.10	0.54	0.76
4	06-13-00	0.72	0.67	0.06	0.25	0.30	0.09	0.29	1.05	0.22	1.12	0.55	0.79
5	06-13-00	0.70	0.67	0.05	0.26	0.30	0.08	0.29	1.05	0.22	1.12	0.57	0.78
6	09-19-00	0.66	0.67	0.05	0.26	0.30	0.07	0.29	1.05	0.22	1.12	0.57	0.77
7	10-24-00	0.77	0.69	0.06	0.26	0.31	0.08	0.31	1.07	0.24	1.14	0.57	0.80
8	11-07-00	0.65	0.68	0.05	0.26	0.31	0.08	0.30	1.06	0.23	1.13	0.57	0.79
9	03-13-01	0.69	0.68	0.05	0.26	0.31	0.07	0.30	1.06	0.23	1.13	0.58	0.78
10	06-26-01	0.53	0.67	0.07	0.25	0.30	0.10	0.29	1.05	0.22	1.12	0.53	0.80
11	07-17-01	0.74	0.67	0.07	0.26	0.30	0.10	0.29	1.05	0.22	1.12	0.54	0.81
12	08-21-01	0.62	0.67	0.07	0.25	0.30	0.10	0.29	1.05	0.22	1.12	0.54	0.80
13	09-25-01	0.60	0.66	0.07	0.25	0.30	0.10	0.28	1.04	0.21	1.11	0.53	0.80
14	11-01-01	0.71	0.67	0.07	0.25	0.30	0.10	0.29	1.05	0.22	1.12	0.54	0.80
15	11-06-01	0.66	0.67	0.06	0.25	0.30	0.09	0.29	1.05	0.22	1.12	0.54	0.79
16	11-27-01	0.57	0.66	0.06	0.25	0.30	0.10	0.28	1.04	0.21	1.11	0.53	0.79
17	12-12-01	0.63	0.66	0.06	0.25	0.30	0.10	0.28	1.04	0.21	1.11	0.53	0.79
18	01-04-02	0.49	0.65	0.07	0.25	0.29	0.11	0.27	1.03	0.20	1.10	0.50	0.80

Note: 7-d IC25 = 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 IC25).

S = Standard deviation of the IC25 values.

$S_{A.75}$ = Standard deviation corresponding to the the 75th percentile CV.
$S_{A.75}$ = 0.38, as determined by USEPA for the method and endpoint.

$S_{A.90}$ = Standard deviation corresponding to the the 90th percentile CV.
$S_{A.90}$ = 0.45, as determined by the USEPA for the method and endpoint.

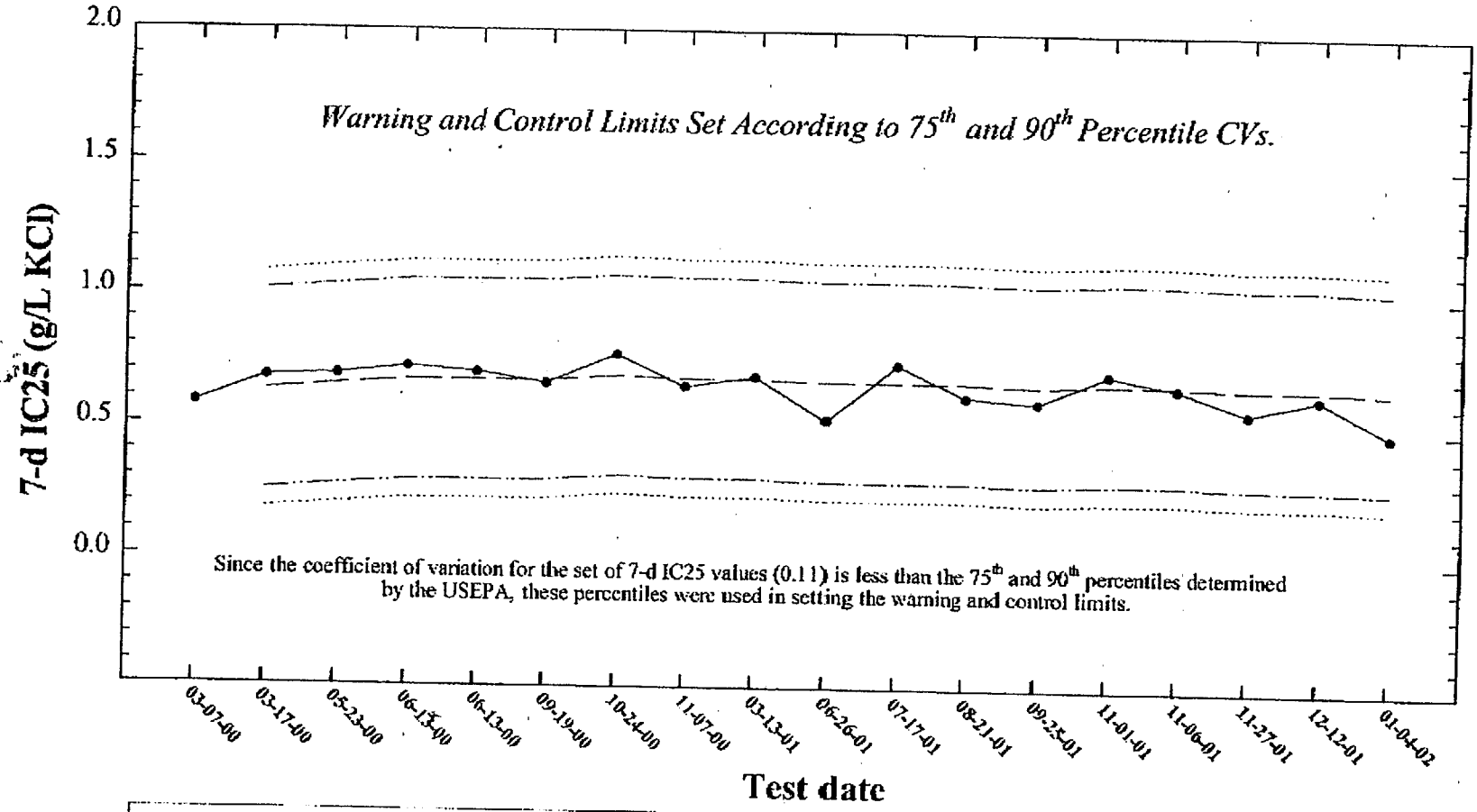
CV = Coefficient of variation of the IC25 values.

EPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National lutant 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 IC25** = 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 IC25)
- **Warning Limits** (mean IC25 ± $S_{A.75}$, $S_{A.75}$ = 0.38, as determined by USEPA for the method and endpoint)
- **Control Limits** (mean IC25 ± $S_{A.90}$, $S_{A.90}$ = 0.45, as determined by USEPA for the method and endpoint)

Potassium Chloride Chronic Reference Toxicant Test
(EPA/600/4-91/002 Method 1000.0)
Species: *Pimephales promelas*

PpKClCR Test Number: 10

Dilution preparation information:						*Comments:*
KCl CHM number:	CHM 067					OUTLIER:
Stock preparation:	50 g KCl/L: Dissolve 50 g KCl in 1-L Deionized water					IC25 < ± 2 SD AROUND
MHS batch:	01-03-02 / 01-06-02					MEAN IC25
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	

Test organism information:		*Test information:*	
Organism age:	24-HOURS OLD	Randomizing template:	YELLOW
Date and times organisms were born between:	01-03-02 1130 TO 1330 MST 1330 - 1530 EST	Incubator number:	2
Organism source:	ABS BATCH 01-03-02	*Artemia* lot number:	BG-0201P
Transfer bowl information:	pH = 7.87 Temperature = 24.3°C	Oven temperature:	60°C
		Drying time:	24-HOURS

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Analyst
0	01-04-02	——	1438	1431	[illegible]
1	01-05-02	1000	1600	1428	[illegible]
2	01-06-02	1007	1610	1422	KEK
3	01-07-02	0955	1555	1419	[illegible]
4	01-08-02	1000	1600	1426	[illegible]
5	01-09-02	1016	1620	1500	KEK
6	01-10-02	0952	1550	1436	KEK
7	01-11-02	——	——	1508	KEK

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	5%	≤ 20%	7-day LC50	669.1
Average weight per larvae:	0.80	≥ 0.25 mg/larvae	NOEC	450
			LOEC	600
			ChV	519.6
			IC25	487.8

Environmental Testing Solutions, LLC

PpKClCR Test Number: 10

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	8 2d	10	10	10	10	9 1d	10
3	10	10	10	10	10	8	10	10	10	10	9	10
4	10	10	9 1d	10	9 1d	8	10	10	10	10	9	10
5	10	9 1d	9	10	8 1d	8	10	10	10	10	9	9 1d
6	10	9	9	10	7 1d	8	10	10	9 1d	10	7 2d	9
7	10	9	9	10	7	8	10	10	8 1d	10	7	9
A = Pan weight (mg)	14.972	14.996	14.941	14.851	15.041	15.011	14.828	14.983	14.858	14.825	15.055	14.979
B = Pan + Larvae weight (mg)	23.10	21.81	23.40	23.42	20.24	20.50	22.07	22.68	22.13	22.64	19.87	21.53
Larvae weight (mg) = A – B	8.128	6.814	8.459	8.569	5.199	5.489	7.242	7.697	7.272	7.815	4.815	6.551

T: 31.97
x̄: 0.7993
CV: 10.1052

T: 25.627
x̄: 0.6407
CV: 19.4595
RC: 19.89

Calculations and data reviewed: JH
T: 26.453
x̄: 0.6613
CV: 19.7467
RC: 17.2651

Comments:

Environmental Testing Solutions, LLC

PpKClCR Test Number: 10

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	10	10	9 1d	7 3d	9 1d	7 3d	9 1d	5 5d	4 6d	4 6d	4 6d
2	6 3d	8 2d	10	7 2d	6 1d	9	6 1d	9	1 4d	4	4	3 1d
3	6	8	10	7	6	9	6	8 1d	1	4	4	3
4	6	8	9 1d	7	6	9	6	6 2d	1	4	2 2d	2 1d
5	6	7 1d	8 1d	7	6	9	1 5d	5 1d	0 1d	2 2d	1 1d	1 1d
6	6	7	8	6 1d	6	9	0 1d	2 3d	↓	0 2d	1	1
7	5 1d	7	6 2d	6	3 3d	9	↓	2	↓	↓	1	1
A = Pan weight (mg)	14.931	14.874	15.052	14.901	14.832	14.849	—	14.830	—	—	14.996	14.905
B = Pan + Larvae weight (mg)	17.47	20.78	19.71	19.66	15.86	19.60	—	16.21	—	—	15.56	15.54
Larvae weight (mg) = A – B	2.539	5.903	4.658	4.759	1.028	4.751	—	1.340	—	—	0.564	0.555

T = 17.854
x̄ = 0.4465
CV: 31.4109
RC = 44.1356

T = 1.0990
x̄ = 0.1775
CV: 115.6
RC = 77.7931

Calculations and data reviewed: JA
T = 1.1190
x̄ = 0.0280
CV: 115.4569
RC = 96.4969

Comments:

Environmental Testing Solutions, LLC
Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Survival

art Date:	1/4/02	Test ID:	PpKCICR	Sample ID:	REF-Ref Toxicant
d Date:	1/11/02	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	KCL-Potassium chloride
mple Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
mments:					

Conc-mg/L	1	2	3	4
D-Control	1.0000	0.9000	0.9000	1.0000
300	0.7000	0.8000	1.0000	1.0000
450	0.8000	1.0000	0.7000	0.9000
600	0.5000	0.7000	0.6000	0.6000
750	0.3000	0.9000	0.0000	0.2000
900	0.0000	0.0000	0.1000	0.1000

			Transform: Arcsin Square Root						1-Tailed		Number	Total
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Resp	Number
D-Control	0.9500	1.0000	1.3305	1.2490	1.4120	7.072	4				2	40
300	0.8750	0.9211	1.2306	0.9912	1.4120	17.454	4	0.617	2.410	0.3907	5	40
450	0.8500	0.8947	1.1898	0.9912	1.4120	15.281	4	0.868	2.410	0.3907	6	40
*600	0.6000	0.6316	0.8872	0.7854	0.9912	9.469	4	2.735	2.410	0.3907	16	40
*750	0.3500	0.3684	0.6128	0.1588	1.2490	75.038	4	4.427	2.410	0.3907	26	40
*900	0.0500	0.0526	0.2403	0.1588	0.3218	39.161	4	6.725	2.410	0.3907	38	40

xiliary Tests	Statistic	Critical	Skew	Kurt
apiro-Wilk's Test indicates normal distribution (p > 0.01)	0.912736952	0.884	0.908581907	3.811713373
rtlett's Test indicates equal variances (p = 0.03)	12.67539597	15.08631706		

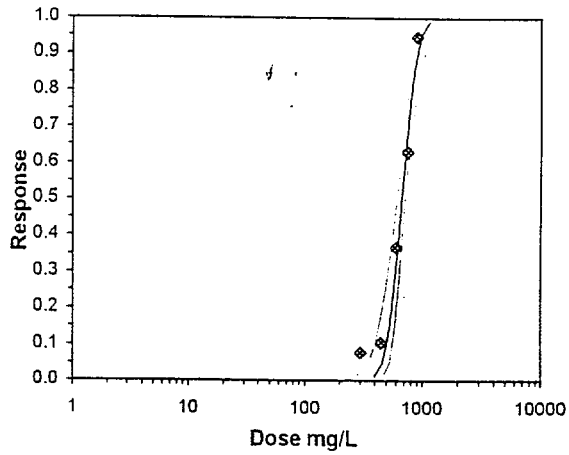
pothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
nnett's Test	450	600	519.6152423		0.291389424	0.308879788	0.716140872	0.052564329	1.4E-05	5, 18

Maximum Likelihood-Probit

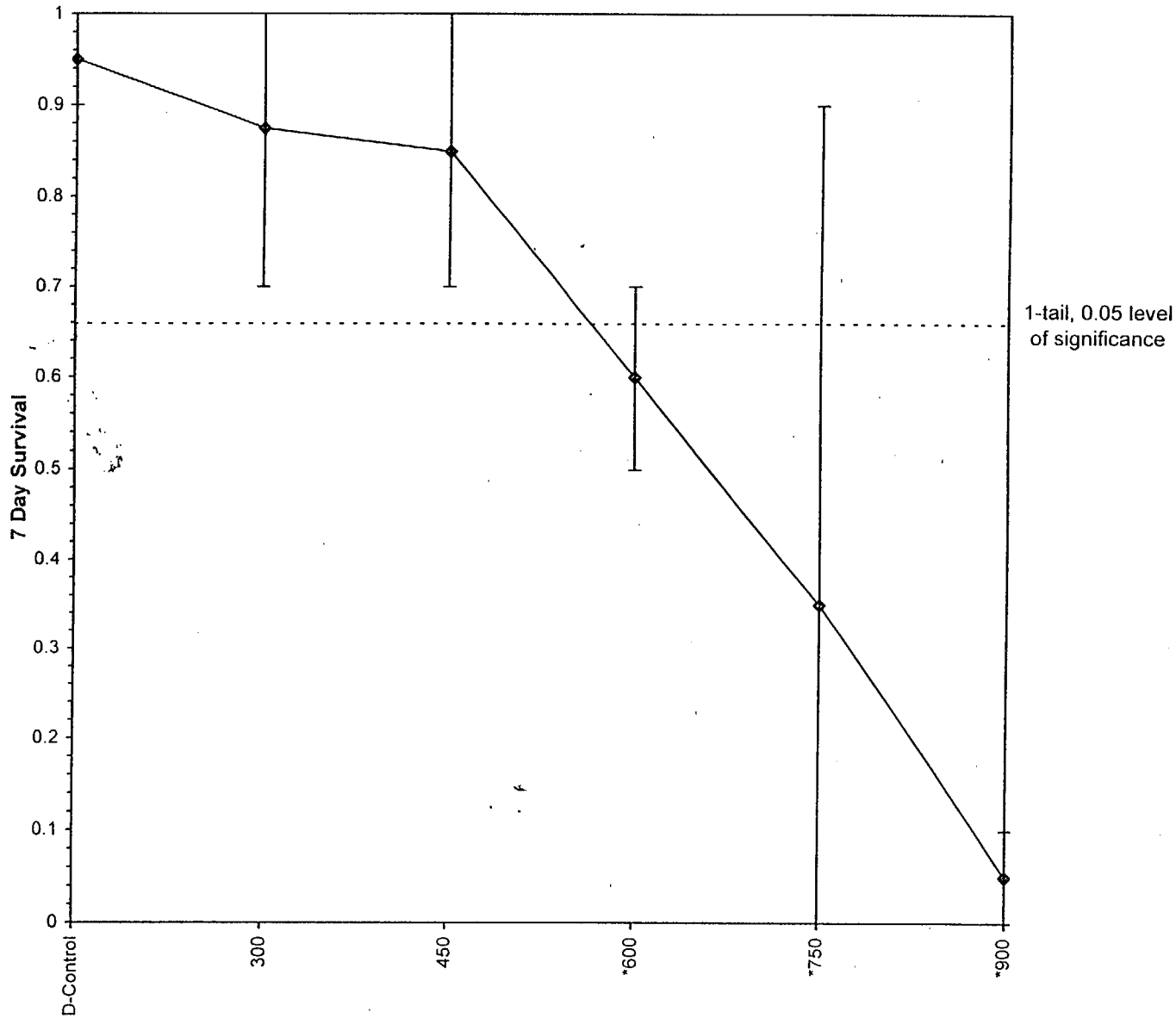
Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
0.05	2.342762563	7.814724922	0.5	2.825479568	0.098734158	7

rameter	Value	SE	95% Fiducial Limits	
pe	10.12820705	1.891366178	6.42112927	13.83528483
ercept	-23.61704209	5.3945961	-34.19045065	-13.04363353
CR	0.094023334	0.030778369	0.033697731	0.154348938

int	Probits	mg/L	95% Fiducial Limits	
01	2.674	394.2662185	274.4358598	468.8424496
05	3.355	460.3374728	349.3406584	526.7559271
10	3.718	499.9731179	396.9361633	561.0171676
15	3.964	528.6256032	432.3835662	585.7579873
20	4.158	552.5642792	462.5425809	606.5348106
25	4.326	573.9636523	489.8144023	625.2941241
40	4.747	631.6340829	563.7161328	677.7697729
50	5.000	669.0823441	610.5906447	714.7586265
60	5.253	708.7508417	657.3746155	758.3392309
75	5.674	779.964338	730.2949578	851.539691
80	5.842	810.170332	757.2813023	896.5085251
85	6.036	846.8587038	787.8892431	954.4793911
90	6.282	895.3904742	825.8761911	1035.63639
95	6.645	972.4847434	882.5994399	1172.726823
99	7.326	1135.454154	994.5752609	1488.379701



Organisms obtained from Aquatic BioSystems, Inc.


7 Day Survival
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
D-Control
300
450
*600
*750
*900
1-tail, 0.05 level
of significance

Environmental Testing Solutions, LLC
Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

art Date:	1/4/02	Test ID:	PpKClCR	Sample ID:	REF-Ref Toxicant
d Date:	1/11/02	Lab ID:	ETS-Env. Testing Solutions	Sample Type:	KCL-Potassium chloride
mple Date:		Protocol:	EPAF 91-EPA Freshwater	Test Species:	PP-Pimephales promelas
omments:					

Conc-mg/L	1	2	3	4
D-Control	0.8128	0.6814	0.8459	0.8569
300	0.5199	0.5489	0.7242	0.7697
450	0.7272	0.7815	0.4815	0.6551
600	0.2539	0.5903	0.4658	0.4759
750	0.1028	0.4731	0.0000	0.1340
900	0.0000	0.0000	0.0564	0.0555

			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	0.7993	1.0000	0.7993	0.6814	0.8569	10.106	4				0.7993	1.0000
300	0.6407	0.8016	0.6407	0.5199	0.7697	19.460	4	1.964	2.180	0.1760	0.6510	0.8145
450	0.6613	0.8274	0.6613	0.4815	0.7815	19.746	4	1.708	2.180	0.1760	0.6510	0.8145
600	0.4465	0.5586	0.4465	0.2539	0.5903	31.413	4				0.4465	0.5586
750	0.1775	0.2221	0.1775	0.0000	0.4731	115.639	4				0.1775	0.2221
900	0.0280	0.0350	0.0280	0.0000	0.0564	115.478	4				0.0280	0.0350

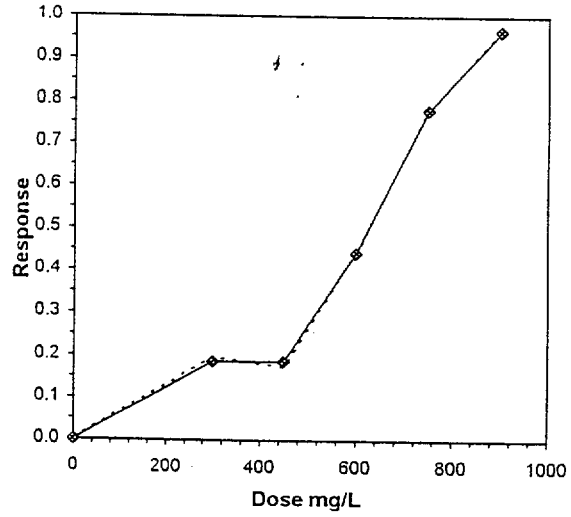
xiliary Tests	Statistic	Critical	Skew	Kurt
apiro-Wilk's Test indicates normal distribution (p > 0.01)	0.918748081	0.805	-0.488905102	-1.138415106
rtlett's Test indicates equal variances (p = 0.72)	0.649693608	9.21035099		

pothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
nnett's Test	450	450			0.176029231	0.220243017	0.029730506	0.013040269	0.158107191	2, 9

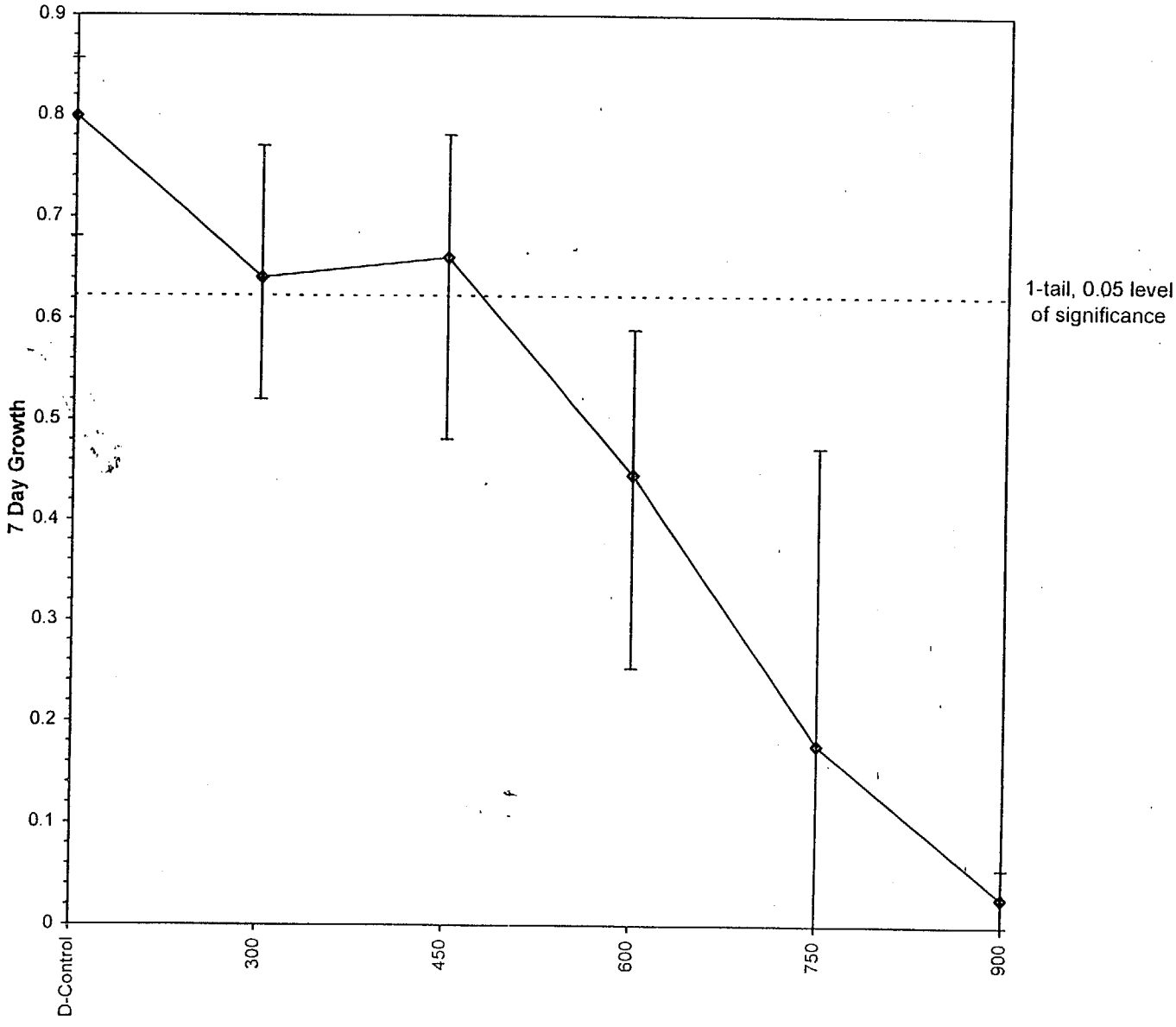
Linear Interpolation (80 Resamples)

nt	mg/L	SD	95% CL(Exp)		Skew
15*	80.87	91.08	29.19	518.68	2.1810
0*	161.74	118.92	58.39	647.06	1.0958
5*	242.61	122.21	87.58	637.39	0.3472
0	458.51	113.20	35.76	569.35	-0.3797
5	487.82	92.49	95.89	607.45	-1.2155
0	575.74	37.16	482.77	681.85	-0.0123
0	626.12	40.22	525.00	778.65	0.8685

ndicates IC estimate less than the lowest concentration



Organisms obtained from Aquatic BioSystems, Inc.


7 Day Growth
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
D-Control
300
450
600
750
900
1-tail, 0.05 level
of significance

Environmental Testing Solutions, LLC

Chronic Whole Effluent Toxicity Test (EPA/600/4-91/002, Method 1002.0)
Species: *Pimephales promelas*

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

Test number: PpKClCR # 10

Test dates: January 4-11, 2001

Reveiwed by: _____

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	14.972	23.100	8.128	0.8128	95.0	0.7993	10.1	Not applicable
	B	10	9	14.996	21.810	6.814	0.6814				
	C	10	9	14.941	23.400	8.459	0.8459				
	D	10	10	14.851	23.420	8.569	0.8569				
300	E	10	7	15.041	20.240	5.199	0.5199	87.5	0.6407	19.5	19.8
	F	10	8	15.011	20.500	5.489	0.5489				
	G	10	10	14.828	22.070	7.242	0.7242				
	H	10	10	14.983	22.680	7.697	0.7697				
450	I	10	8	14.858	22.130	7.272	0.7272	85.0	0.6613	19.7	17.3
	J	10	10	14.825	22.640	7.815	0.7815				
	K	10	7	15.055	19.870	4.815	0.4815				
	L	10	9	14.979	21.530	6.551	0.6551				
600	M	10	5	14.931	17.470	2.539	0.2539	60.0	0.4465	31.4	44.1
	N	10	7	14.877	20.780	5.903	0.5903				
	O	10	6	15.052	19.710	4.658	0.4658				
	P	10	6	14.901	19.660	4.759	0.4759				
750	Q	10	3	14.832	15.860	1.028	0.1028	35.0	0.1775	115.6	77.8
	R	10	9	14.869	19.600	4.731	0.4731				
	S	10	0	0.000	0.000	0.000	0.0000				
	T	10	2	14.870	16.210	1.340	0.1340				
900	U	10	0	0.000	0.000	0.000	0.0000	5.0	0.0280	115.5	96.5
	V	10	0	0.000	0.000	0.000	0.0000				
	W	10	1	14.996	15.560	0.564	0.0564				
	X	10	1	14.985	15.540	0.555	0.0555				

Dunnett's MSD value: 0.176

PMSD: 22.0

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 19.1% 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.

Organisms obtained from Aquatic BioSystems, Inc.

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)	CV (%)	CT for Control Growth CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	03-07-00	100	0.67	4.9		0.05	7.6	
2	03-17-00	97.5	0.40	7.5	6.2	0.06	15.7	11.7
3	05-23-00	100	0.31	3.6	5.3	0.07	21.2	14.8
4	06-13-00	100	0.45	10.1	6.5	0.10	22.2	16.7
5	06-13-00	100	0.58	13.2	7.9	0.12	20.0	17.3
6	09-19-00	100	0.67	5.3	7.4	0.07	11.2	16.3
7	10-24-00	97.5	0.83	13.3	8.3	0.22	26.9	17.8
8	11-07-00	97.5	0.67	10.5	8.6	0.13	19.3	18.0
9	03-13-01	92.5	0.34	6.5	8.3	0.08	22.8	18.5
10	06-26-01	100	0.63	9.8	8.5	0.19	30.6	19.7
11	07-17-01	100	0.52	9.1	8.5	0.07	13.8	19.2
12	08-21-01	100	0.89	8.3	8.5	0.13	15.1	18.9
13	09-25-01	100	0.85	4.9	8.2	0.09	10.4	18.2
14	11-01-01	100	0.54	2.5	7.8	0.10	18.6	18.2
15	11-06-01	100	1.00	7.4	7.8	0.17	16.9	18.2
16	11-27-01	97.5	0.70	22.8	8.7	0.18	26.1	18.7
17	12-12-01	95	0.82	16.6	9.2	0.19	23.5	18.9
18	01-04-02	95	0.80	10.1	9.2	0.18	22.0	19.1

Note:

CV = Coefficient of variation for control survival.
On average, the CV for control growth is **9.2%** in Environmental Testing Solutions, LLC *Pimephales* chronic toxicity tests.
Lower CV bound determined by USEPA (10th percentile) = 3.5%.
Upper CV bound determined by USEPA (90th 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 **19.1%** from the control.
Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.
Upper PMSD bound determined by USEPA (90th 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.

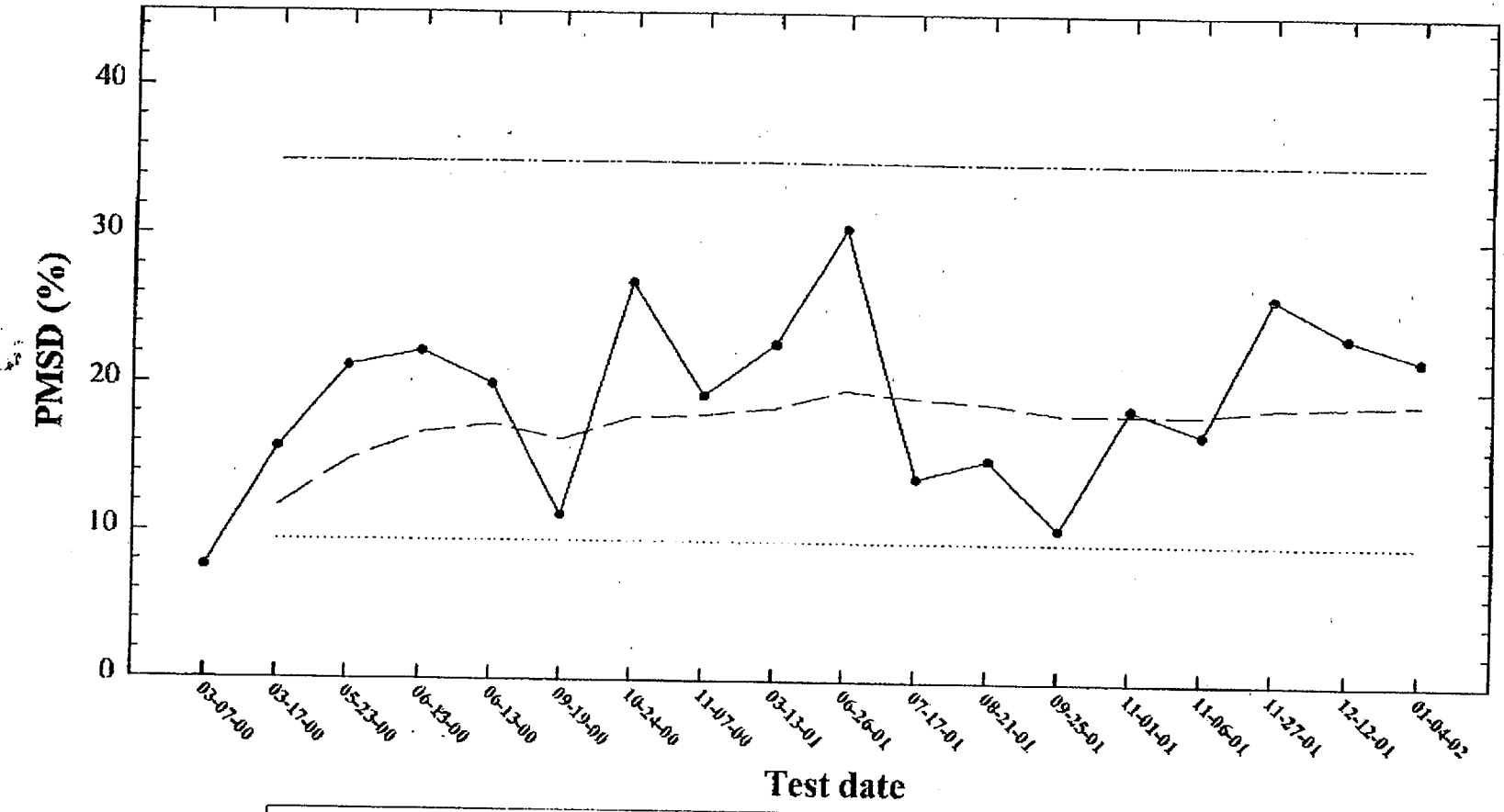
USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Organisms obtained from Aquatic BioSystems, Inc.

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 (10th percentile) = 9.4%, Upper PMSD Bound (90th percentile) = 35%

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

PpKCICR Test Number: 10

KCl Stock and Dilution water chemistry:

	Batch or preparation date	Alkalinity (mg $CaCO_3$/L)	Hardness (mg $CaCO_3$/L)
Control/Dilution Water MHS	01-0302		

01-06-02

Conductivity measurements (µmhos/cm):

Concentration	Day							Acceptance Criteria (dilutions must be remade if the conductivity falls outside the acceptable range)
	0	1	2	3	4	5	6	
CONTROL	320	304	305	302	314	314	300	280 - 360
300 mg KCl/L	857	852	860	873	845	864	859	840 - 890
450 mg KCl/L	1178	1111	1128	1143	1125	1141	1130	1100 - 1180
600 mg KCl/L	1452	1390	1392	1402	1390	1399	1409	1390 - 1460
750 mg KCl/L	1706	1665	1664	1669	1660	1681	1664	1660 - 1710
900 mg KCl/L	1937	1893	1893 ~~1888~~	1896	1890	1933	1902	1890 - 1950
KCl Stock (25 g KCl/L)	71700	68500	67800	68500	71600	69900	69400	67000 - 76000

Environmental Testing Solutions, LLC

PpKClCR Test Number: 10

Daily chemistry:

Concentration	Parameter	Day 0 Initial	Day 0 Final	Day 1 Initial	Day 1 Final	Day 2 Initial	Day 2 Final
CONTROL	pH (S.U.)	8.04	7.76	8.01	7.71	8.02	7.58
	DO (mg/L)	8.1	7.9	8.0	7.6	8.0	7.0
300 mg KCl/L	pH (S.U.)	8.10	7.84	8.08	7.85	8.10	7.69
	DO (mg/L)	8.0	7.9	8.1	7.6	8.1	7.0
450 mg KCl/L	pH (S.U.)	8.10	7.85	8.11	7.85	8.12	7.67
	DO (mg/L)	8.0	7.9	8.1	7.5	8.1	7.0
600 mg KCl/L	pH (S.U.)	8.13	7.88	8.13	7.83	8.13	7.64
	DO (mg/L)	8.1	7.9	8.2	7.6	8.1	7.1
750 mg KCl/L	pH (S.U.)	8.13	7.86	8.14	7.87	8.13	7.71
	DO (mg/L)	8.1	7.9	8.2	7.6	8.1	7.1
900 mg KCl/L	pH (S.U.)	8.14	7.86	8.16	7.93	8.13	7.83
	DO (mg/L)	8.1	7.9	8.2	7.6	8.1	7.0
Temperature (°C)		24.6	24.0	24.4	24.5	24.9	24.3

Concentration	Parameter	Day 3 Initial	Day 3 Final	Day 4 Initial	Day 4 Final	Day 5 Initial	Day 5 Final	Day 6 Initial	Day 6 Final
CONTROL	pH (S.U.)	8.06	7.74	8.10	7.67	8.08	7.72	8.09	7.60
	DO (mg/L)	8.0	7.7	8.0	7.3	8.0	7.5	8.0	7.5
300 mg KCl/L	pH (S.U.)	8.07	7.71	8.09	7.70	8.12	7.78	8.13	7.75
	DO (mg/L)	8.0	7.6	8.1	7.3	8.1	7.5	8.0	7.4
450 mg KCl/L	pH (S.U.)	8.11	7.76	8.14	7.70	8.15	7.81	8.14	7.74
	DO (mg/L)	8.0	7.4	8.2	7.1	8.3	7.5	8.0	7.5
600 mg KCl/L	pH (S.U.)	8.12	7.74	8.14	7.73	8.16	7.81	8.16	7.76
	DO (mg/L)	8.0	7.4	8.3	7.0	8.3	7.5	8.1	7.5
750 mg KCl/L	pH (S.U.)	8.13	7.80	8.15	7.71	8.18	7.86	8.17	7.80
	DO (mg/L)	8.0	7.3	8.3	7.1	8.3	7.6	8.1	7.5
900 mg KCl/L	pH (S.U.)	8.14	7.86	8.14	7.73	8.18	7.85	8.17	7.90
	DO (mg/L)	8.1	7.5	8.3	7.1	8.3	7.7	8.1	7.5
Temperature (°C)		24.4	24.7	25.4	24.3	24.9	24.4	24.6	24.2