Tennessee Valley Authority, Post Office Box 2000, Soddy-Daisy, Tennessee 37384-2000

December 14, 2005

State of Tennessee
Department of Environment and Conservation
Division of Water Pollution Control
Enforcement & Compliance Section
6th Floor, L & C Annex
401 Church Street
Nashville, Tennessee 37243-1534

Attention: Mr. Chip Hannah

Dear Mr. Hannah:

SEQUOYAH NUCLEAR PLANT - DISCHARGE MONITORING REPORT FOR NOVEMBER 2005

Enclosed is the November 2005 Discharge Monitoring Report for Sequoyah Nuclear Plant. Please contact me at (423) 843-6700 if you have any questions or comments.

Sincerely,

Stephanie A. Howard

Stephanie A. Howard
Principal Environmental Engineer
Signatory Authority for
J. Randy Douet
Site Vice President
Sequoyah Nuclear Plant

Enclosure
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 P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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

TN0026450	101 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

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			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
TEMPERATURE, WATER DEG. CENTIGRADE	SAMPLE MEASUREMENT	********	********	**	********	********	20.4	04	0	30 / 30	MODELD
00010 Z 0 0 INSTREAM MONITORING	PERMIT REQUIREMENT	********	********	****	********	********	30.5 DAILY MX	DEG. C.		SEE PERMIT	CK REQ
TEMPERATURE, WATER DEG. CENTIGRADE	SAMPLE MEASUREMENT	********	********	**	********	********	33.2	04	0	30 / 30	RCORDR
00010 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	REPORT DAILY MX	DEG. C.		SEE PERMIT	CK REQ
TEMP. DIFF. BETWEEN SAMP. & UPSTRM DEG.C	SAMPLE MEASUREMENT	********	********	**	********	********	2.8	04	0	30 / 30	CALCTD
00016 1 W 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	5.0 DAILY MX	DEG. C.		CONTIN UOUS	CALCTD
PH	SAMPLE MEASUREMENT	********	********	**	7.4	********	7.6	12	0	6 / 30	GRAB
00400 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	6.0 MINIMUM	********	9.0 MAXIMUM	SU		WEEKLY	GRAB
SOLIDS, TOTAL SUSPENDED	SAMPLE MEASUREMENT	********	********	**	********	9	9	19	0	1 / 30	GRAB
00530 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	30 MO AVG	100 DAILY MX	MG/L		MONTHLY	GRAB
OIL AND GREASE	SAMPLE MEASUREMENT	********	********	**	********	<5	<5	19	0	1 / 30	GRAB
00556 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	15 MO AVG	20 DAILY MX	MG/L		MONTHLY	GRAB
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT	********	1563	03	********	********	********	**	0	30 / 30	RCORDR
50050 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	REPORT DAILY MX	MGD	********	********	********	****		CONTIN UOUS	RCORDR

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER			TELEPHONE		DATE		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*

No closed mode operation. CCW data for November 2005 is attached. Veliger monitoring information is attached.

CCW TRENCH

Date/Time Collected	Extractable Petroleum Hydrocarbons	Analysis Date/Time	Analyst	Method
11/02/2005 @ 0745	<0.5	11/05/2005 @ 0050	KRR	EPA 8015B

CCW CHANNEL

Date/Time Collected	Extractable Petroleum Hydrocarbons	Analysis Date/Time	Analyst	Method
11/02/2005 @ 0740	< 0.5 mg/L	11/05/2005 @ 0012	KRR	EPA 8015B

Sample Date	Mean # of ZM/m^3	% Settlers	Water Temp. (°C)	Sample Date	Mean# of Asiatic Clams/m^3	Water Temp. (°C)	LOCATION	SUB LOCATION	SAMPLE TYPE	COLLECTED BY
07/06/2005	0	0	26	07/06/2005	677	26	INPLANT	lant Raw We	QUANT	Dick Adcock
07/08/2005	122	29	26	07/08/2005	1132	26	INPLANT	lant Raw We	QUANT	Dick Adcock
07/13/2005	51	33	26	07/13/2005	998	26	INPLANT	lant Raw We	QUANT	Dick Adcock
07/15/2005	0	0	26	07/15/2005	186	26	INPLANT	lant Raw We	QUANT	Dick Adcock
07/20/2005	38	50	26	07/20/2005	303	26	INPLANT	lant Raw We	QUANT	Dick Adcock
07/22/2005	0	0	27	07/22/2005	100	27	INPLANT	lant Raw We	QUANT	Dick Adcock
07/27/2005	143	100	27	07/27/2005	341	27	INPLANT	lant Raw We	QUANT	Dick Adcock
07/28/2005	488	100	27	07/28/2005	627	27	INPLANT	lant Raw We	QUANT	Dick Adcock
07/29/2005	90	20	27	07/29/2005	663	27	INPLANT	lant Raw We	QUANT	Dick Adcock
08/03/2005	0	0	27	08/03/2005	500	27	INPLANT	lant Raw We	QUANT	Dick Adcock
08/05/2005	0	0	27	08/05/2005	250	27	INPLANT	lant Raw We	QUANT	Dick Adcock
08/09/2005	383	35	27.5	08/09/2005	1067	27.5	INPLANT	lant Raw We	QUANT	CFT
08/11/2005	0	0	27	08/11/2005	0	27	INPLANT	lant Raw We	QUANT	CFT
08/16/2005	34	0	28	08/16/2005	464	28	INPLANT	lant Raw We	QUANT	CFT
08/19/2005	100	100	27.5	08/19/2005	367	27.5	INPLANT	lant Raw We	QUANT	Dick Adcock
08/24/2005	51	100	28	08/24/2005	237	28	INPLANT	lant Raw We	QUANT	CFT
08/26/2005	133	25	27	08/26/2005	467	27	INPLANT	lant Raw We	QUANT	CFC
08/30/2005	150	100	27.5	08/30/2005	267	27.5	INPLANT	lant Raw We	QUANT	CFT
09/02/2005	17	100	27.5	09/02/2005	85	27.5	INPLANT	lant Raw We	QUANT	CFT
09/06/2005	66	100	27.5	09/06/2005	115	27.5	INPLANT	lant Raw We	QUANT	CFT
09/08/2005	100	100	27	09/08/2005	483	27	INPLANT	lant Raw We	QUANT	CFT
09/12/2005	87	100	27	09/12/2005	105	27	INPLANT	lant Raw We	QUANT	CFT
09/16/2005	100	100	27	09/20/2005	117	27	INPLANT	lant Raw We	QUANT	CKC
09/22/2005	67	100	27	09/22/2005	250	27	INPLANT	lant Raw We	QUANT	CKC
09/23/2005	83	80	27	09/23/2005	317	27	INPLANT	lant Raw We	QUANT	CFT
09/26/2005	67	75	26.5	09/26/2005	917	26.5	INPLANT	lant Raw We	QUANT	CFT
09/31/2005	239	89	25	09/31/2005	80	25	INPLANT	lant Raw We	QUANT	Dick Adcock
10/04/2005	294	100	26	10/04/2005	214	26	INPLANT	lant Raw We	QUANT	Dick Adcock
10/07/2005	100	100	25.5	10/07/2005	433	25.5	INPLANT	lant Raw We	QUANT	CFT
10/12/2005	542	100	24.5	10/12/2005	131	24.5	INPLANT	lant Raw We	QUANT	CFT
10/14/2005	67	100	24.5	10/14/2005	33	24.5	INPLANT	lant Raw We	QUANT	CFT
10/18/2005	283	100	24	10/18/2005	52	24	INPLANT	lant Raw We	QUANT	CFT
10/21/2005	33	100	24	10/21/2005	0	24	INPLANT	lant Raw We	QUANT	CFT
10/24/2005	200	100	22	10/24/2005	817	22	INPLANT	lant Raw We	QUANT	CFT
10/28/2005	0	0	17	10/28/2005	1795	17	INPLANT	lant Raw We	QUANT	Dick Adcock
11/01/2005	131	100	17	11/01/2005	105	17	INPLANT	lant Raw We	QUANT	Dick Adcock
11/03/2005	0	0	17	11/03/2005	26	17	INPLANT	lant Raw We	QUANT	Dick Adcock
11/09/2005	0	0	19.5	11/09/2005	64	19.5	INPLANT	lant Raw We	QUANT	CKC
11/10/2005	0	0	17	11/10/2005	95	17	INPLANT	lant Raw We	QUANT	DLA
11/15/2005	0	0	17	11/15/2005	125	17	INPLANT	lant Raw We	QUANT	Dick Adcock
11/18/2005	0	0	16	11/18/2005	16	16	INPLANT	lant Raw We	QUANT	Dick Adcock
11/21/2005	0	0	15	11/21/2005	80	15	INPLANT	lant Raw We	QUANT	Dick Adcock
11/23/2005	0	0	19.3	11/23/2005	0	19.3	INPLANT	lant Raw We	QUANT	CKC
11/28/2005	0	0	13	11/28/2005	0	13	INPLANT	lant Raw We	QUANT	Dick Adcock
12/01/2005	0	0	13	12/01/2005	0	13	INPLANT	lant Raw We	QUANT	Dick Adcock

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*
Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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

TN0026450	101 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

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
CHLORINE, TOTAL RESIDUAL	SAMPLE MEASUREMENT	********	********	**	********	0.020	0.030	19	0	30 / 30	GRAB
50060 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	0.10 MO AVG	0.10 INST MAX	MG/L		WEEK-DAYS	CALCTD
TEMPERATURE - C, RATE OF CHANGE	SAMPLE MEASUREMENT	********	0.3	62	********	********		**	0	30 / 30	CALCTD
82234 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	2.0 DAILY MX	DEG C/HR	********	********	********	****		CONTINUOUS	CALCTD
	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		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*

The following injections occurred: 1. H-150M (max. calc. conc. was 0.036mg/L–limit 0.050mg/L) 2. H-150M (low detection level analytical method was <0.020mg/L–limit 0.050mg/L)

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*
Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
BIOMONITORING FOR OUTFALL 101
EFFLUENT
*** NO DISCHARGE ☐ ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
IC25 STATRE 7DAY CHR CERIODAPHNIA	SAMPLE MEASUREMENT	********	********	**	>100.0	********	********	23	0	2 / 180	COMPOS
TRP3B 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	45.2 MINIMUM	********	********	PERCENT		SEE PERMIT	COMPOS
IC25 STATRE 7DAY CHR PIMEPHALES	SAMPLE MEASUREMENT	********	********	**	>100.0	********	********	23	0	2 / 180	COMPOS
TRP6C 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	45.2 MIMINUM	********	********	PERCENT		SEE PERMIT	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		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*

Toxicity was sampled 10/30-11/4 and 11/14-11/19. These sampling events cover the requirement stated in the B/CTP, "Whole effluent toxicity testing (biomonitoring) of Outfall 101 shall be undertaken once per year when oxidizing biocides (Towerbrom 960) are being used and once per year when non-oxidizing biocides (H-150M) are being used." Reports are attached.

December 13, 2005

Ruth Ann Hurt, SB 2A-SQN

SEQUOYAH NUCLEAR PLANT (SQN) TOXICITY BIOMONITORING, NPDES PERMIT NO. TN0026450, OUTFALL 101, NOVEMBER, 2005

Attached are two copies of the subject report for submission to the state of Tennessee and a copy of the report for your records. The report provides the results of compliance testing using fathead minnows and daphnids.

Outfall 101 samples collected October 30- November 4 and November 14-19, 2005, showed no toxic effects to fathead minnows or daphnids. The resulting IC_{25} values for both species were > 100 percent. Exposure of fathead minnows and daphnids to intake samples resulted in no significant differences from controls during this study period.

Environmental Testing Solutions, Inc. performed chronic toxicity tests from November 1-8, and November 16-23, 2005. The November 1-8 toxicity tests were conducted with samples collected from SQN Outfall 101 during non-oxidizing (November 1-4) and oxidizing (October 30-November 4) biocide applications. The November 16-23 toxicity tests were conducted with samples collected from SQN Outfall 101 during oxidizing (November 14-19) biocide applications. An internal peer review of the reports was performed by TVA biologists and the Senior Toxicologist and it was determined that incorrect serial dilutions were prepared. Previous permit dilutions of 10.98, 22.0, 43.9, 72.0, and 100% were used instead of the dilutions of 11.3, 22.6, 45.2, 72.6, and 100% specified in the recently renewed permit, effective September 1, 2005.

This incident has been assessed as a laboratory deviation, and it will be identified as such in the final reports. The EPA chronic test manual (EPA-821-R-02-013) states that deviations must be evaluated on a case-by-case basis and deviations may or may not invalidate a test result "depending on the degree of the departure and the objective of the test." TVA biologists and the Senior Toxicologist have considered the degree of the deviation in this particular case and find no potential or observed impact on the test results. Therefore the test results for the subject tests are considered valid.

In addition to the routine compliance test, fathead minnows were also tested in Outfall 101 and intake samples which were treated using UV exposure for pathogen removal prior to introduction of test organisms. Fish pathogens present in intake water have been the suspected cause of anomalous dose responses and high variability among replicates in previous toxicity testing at Sequoyah. At the time this study was conducted, mortality which occurred in minnows exposed to routine compliance samples was not sufficient to jeopardize statistical validity, although survival was improved in UV treated samples.

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

Cynthia L. Russell
Biologist
Environmental Engineering Services- West
CTR 2L-M

Attachment
cc (Attachment):
Files, R&TA, CTR 1B-M

SQN November 2005M

TENNESSEE VALLEY AUTHORITY
TOXICITY TEST REPORT

INTRODUCTION / EXECUTIVE SUMMARY

Report Date: December 8, 2005

1. Facility / Discharger: Sequoyah Nuclear Plant / TVA
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. 1Q10: 2,992.4
8. Outfall Tested: 101
9. Dates Sampled: October 30-November 4, 2005
10. Average Flow on Days Sampled (MGD): 1,554.4, 1,551.2, 1,550.7
11. Pertinent Site Conditions: H-150M (non-oxidizing biocide) was injected from November 1-4, 2005. The dates and times for the H-150M injection are in the following table. See Appendix B for complete additional chemical application information during the sample collection period.

Injection Location	Date/Start-Time (ET)	Date/Ending-Time (ET)
Raw Cooling Water (RCW)	11/01/2005-0940	11/04/2005-1200

Towerbrom 960 (oxidizing biocide) was injected from October 30-November 4, 2005. The dates and times for the Towerbrom 960 injections are in the following table. See Appendix B for complete additional chemical application information during the sample collection period.

Injection Location	Date/Start-Time (ET)	Date/Ending Time-(ET)
Essential Raw Cooling Water (ERCW) Train A and B	10/30/2005-0000	11/04/2005-2400
Raw Cooling Water (RCW)	11/04/2005-1345	11/04/2005-2400

12. Test Dates: November 1-8, 2005

13. Test Type: Short-term Chronic Definitive

14. Test Species: Fathead Minnows (*Pimephales promelas*)
Daphnids (*Ceriodaphnia dubia*)

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

Pimephales promelas: 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: $IC_{25} = 45.2\%$

17. Test Results: Outfall 101: *Pimephales promelas:* $IC_{25} > 100\%$
Ceriodaphnia dubia: $IC_{25} > 100\%$

UV treated Outfall 101: *Pimephales promelas:* $IC_{25} > 100\%$

18. Facility Contact: Stephanie Howard Phone #: (423) 843-6713

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

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

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

22. Notes: Outfall 101 samples collected October 30 - November 4, 2005, showed no toxic effects to fathead minnows or daphnids. The resulting IC_{25} values, for both species, were > 100 percent. Exposure of fathead minnows and daphnids to intake samples resulted in no significant differences from controls during this study period.

Fathead minnows were also exposed to UV treated Outfall 101 and intake samples since fish pathogens present in intake water have been the suspected cause of interference (anomalous dose response and high variability among replicates) in previous toxicity testing at Sequoyah. At the time this study was conducted, insignificant mortality occurred in minnows exposed to non-treated and UV treated samples.

METHODS SUMMARY

Samples:

1. Sampling Point: Outfall 101, Intake

2. Sample Type: Composite

3. Sample Information:

Sample ID	Date (MM/DD/YY)/ Time (ET) Collected	Date (MM/DD/YY)/ Time (ET) Received	Arrival Temp. (°C)	Initial TRC* (mg/L)	Date (MM/DD/YY)/ Time (ET) Used By
101	10/30/05-0500 to 10/31/05-0400	10/31/05 1823	1.1, 1.6†	<0.10	11/01/05 1202 11/02/05 1249
Intake	10/30/05-0515 to 10/31/05-0415	10/31/05 1823	0.8	<0.10	11/01/05 1202 11/02/05 1249
101	11/01/05-0700 to 11/02/05-0600	11/02/05 1500	2.5, 1.3†	<0.10	11/03/05 1217 11/04/05 1121
Intake	11/01/05-0715 to 11/02/05-0615	11/02/05 1500	1.5	<0.10	11/03/05 1217 11/04/05 1121
101	11/03/05-0600 to 11/04/05-0500	11/04/05 1424	1.3, 1.7†	<0.10	11/05/05 1134 11/06/05 1106 11/07/05 1110
Intake	11/03/05-0615 to 11/04/05-0515	11/04/05 1424	1.2	<0.10	11/05/05 1134 11/06/05 1106 11/07/05 1110

*TRC = Total Residual Chlorine

†Samples were collected in two 2.5 gallon cubitainers. Temperature was measured in each cubitainer upon arrival.

4. Sample Manipulation: Samples from Outfall 101 and intake were warmed to test temperature (25.0 ± 1.0°C) in a warm water bath.

   Aliquots of Outfall 101 and Intake samples were UV-treated through a 40-watt Smart® UV Sterilizer (manufactured by Emperor Aquatics, Inc.) for 2 minutes.

Test Organisms:	*Pimephales promelas*	*Ceriodaphnia dubia*
1. Source:	Aquatic BioSystems, Inc.	In-house Cultures
2. Age:	21-22.5 hours old	<24-hours old
Test Method Summary:		
1. Test Conditions:	Static, Renewal	Static, Renewal
2. Test Duration:	7 days	Until at least 60% of control females have 3 broods
3. Control / Dilution Water:	Moderately Hard Synthetic	Moderately Hard Synthetic
4. Number of Replicates:	4	10
5. Organisms per Replicate:	10	1
6. Test Initiation: (Date/Time)		
Outfall 101	11/01/05 - 1202 ET	11/01/05 - 1110 ET
UV Treated Outfall 101	11/01/05 - 1221 ET	
7. Test Termination: (Date/Time)		
Outfall 101	11/08/05 - 1209 ET	11/08/05 - 1119 ET
UV Treated Outfall 101	11/08/05 - 1229 ET	
8. Test Temperature: Outfall 101:	Mean = 24.7°C (24.1 - 25.2°C)	Mean = 24.9°C (24.4 - 25.3°C)
Test Temperature: UV-Treated Outfall 101:	Mean = 24.8°C (24.1 - 25.3°C)	

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

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

TOXICITY TEST RESULTS (see Appendix C for Bench Sheets)

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

Conducted November 1-8, 2005 using effluent from Outfall 101.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
10.98%	100	100	100	100	100	100	100
22.0%	100	100	100	100	98	98	98
43.9%	100	100	100	100	100	100	100
72.0%	100	100	100	100	100	100	100
100.0%	-100	100	100	100	100	100	100
Intake	100	100	100	100	100	100	98

Test Solutions (% Effluent)	Mean Dry Weight (mg) (replicate number)				
	1	2	3	4	Mean
Control	0.582	0.669	0.718	0.683	0.663
10.98%	0.676	0.621	0.737	0.671	0.676
22.0%	0.591	0.641	0.646	0.650	0.632
43.9%	0.679	0.629	0.711	0.634	0.663
72.0%	0.582	0.718	0.581	0.682	0.641
100.0%	0.712	0.691	0.609	0.621	0.658
Intake	0.592	0.656	0.606	0.697	0.638

IC_{25} Value: ≥ 100% Permit Limit: 45.2% 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: < 1.0 TUc* Permit Limit: 2.2 TUc

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

TOXICITY TEST RESULTS (see Appendix C for Bench Sheets)

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

Conducted November 1-8, 2005 using effluent from Outfall 101.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
10.98%	100	100	100	100	100	100	100
22.0%	100	100	100	100	100	100	100
43.9%	100	100	100	100	100	100	100
72.0%	100	100	100	100	100	100	100
100.0%	100	100	100	100	100	100	100

Test Solutions (% Effluent)	Reproduction (#young/female/7 days) Data (replicate number)										
	1	2	3	4	5	6	7	8	9	10	Mean
Control	32	28	26	25	31	24	30	26	26	29	27.7
10.98%	27	30	27	29	28	30	30	27	30	26	28.4
22.0%	29	30	31	31	28	30	31	33	29	28	30.0
43.9%	33	30	31	28	32	32	34	30	32	29	31.1
72.0%	36	32	30	33	31	30	31	36	33	34	32.6
100.0%	35	32	33	27	33	32	32	34	32	36	32.6

IC_{25} Value: > 100% Permit Limit: 45.2% 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: < 1.0 TUc* Permit Limit: 2.2 TUc

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

TOXICITY TEST RESULTS (see Appendix C for Bench Sheets)

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

Conducted November 1-8, 2005 using water from Intake

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

Test Solutions (% Effluent)	Reproduction (#young/female/7 days) Data (replicate number)										
	1	2	3	4	5	6	7	8	9	10	Mean
Control	26	34	27	31	29	29	29	27	29	30	29.1
Intake	36	32	31	35	29	35	33	34	34	37	33.6

IC_{25} Value: ≥ 100% Permit Limit: N/A 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: ≤ 1.0 TUc* Permit Limit: N/A

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

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

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

Conducted November 1-8, 2005 using effluent from UV Treated Outfall 101.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
10.98%	100	100	100	100	100	100	100
22.0%	100	100	100	100	100	100	100
43.9%	100	100	100	100	100	100	98
72.0%	100	100	100	100	100	100	100
100.0%	100	100	100	100	100	100	100
Intake	100	100	100	100	100	100	100

Test Solutions (% Effluent)	Mean Dry Weight (mg) (replicate number)				
	1	2	3	4	Mean
Control	0.625	0.680	0.637	0.657	0.650
10.98%	0.690	0.719	0.676	0.692	0.694
22.0%	0.592	0.649	0.640	0.667	0.637
43.9%	0.658	0.594	0.677	0.671	0.650
72.0%	0.573	0.666	0.669	0.652	0.640
100.0%	0.645	0.662	0.640	0.650	0.649
Intake	0.664	0.686	0.674	0.685	0.677

IC_{25} Value: > 100% 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: < 1.0 TUc*

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

REFERENCE TOXICANT TEST RESULTS (see Appendix A and D)

Species	Date	Time	Duration	Toxicant	Results (IC_{25})
Pimephales promelas	November 1-8, 2005	1238	7-days	KCl	0.60 g/L
Ceriodaphnia dubia	November 1-8, 2005	1101	7-days	NaCl	1.08 g/L

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for *Pimephales promelas* and *Ceriodaphnia dubia* Tests, Sequoyah Nuclear Plant Effluent (SQN), Outfall 101, November 1-8, 2005.

Test	Sample ID	Temperature (°C)		Dissolved Oxygen (mg/L)		pH (S.U.)		Conductance (µmhos/cm)	Alkalinity (mg/L $CaCO_3$)	Hardness (mg/L $CaCO_3$)	Total Residual Chlorine (mg/L)
		Initial	Final	Initial	Final	Initial	Final				
Pimephales promelas	Control	24.8 24.6 - 24.9	24.5 24.2 - 24.8	7.6 7.4 - 7.9	7.3 7.0 - 7.8	7.77 7.53 - 8.10	7.59 7.31 - 8.03	312 306 - 315	58 58 - 59	92 85 - 97	- - -
	10.98%	24.8 24.6 - 25.0	24.5 24.2 - 24.7	7.8 7.4 - 8.1	7.2 6.9 - 7.8	7.83 7.56 - 8.14	7.53 7.28 - 7.98	295 288 - 298	- - -	- - -	- - -
	22.0%	24.8 24.6 - 25.0	24.4 24.2 - 24.6	7.8 7.4 - 8.1	7.2 6.8 - 7.8	7.80 7.56 - 8.14	7.54 7.29 - 8.02	283 279 - 292	- - -	- - -	- - -
	43.9%	24.8 24.6 - 25.1	24.5 24.1 - 24.7	7.9 7.5 - 8.3	7.3 6.8 - 7.8	7.78 7.54 - 8.14	7.55 7.30 - 8.04	255 247 - 264	- - -	- - -	- - -
	72.0%	24.9 24.6 - 25.1	24.4 24.3 - 24.7	7.9 7.6 - 8.3	7.3 6.8 - 7.9	7.76 7.50 - 8.14	7.56 7.28 - 8.05	218 212 - 225	- - -	- - -	- - -
	100.0%	24.9 24.7 - 25.2	24.5 24.2 - 24.7	7.8 7.6 - 8.3	7.3 6.8 - 8.0	7.74 7.47 - 8.16	7.57 7.29 - 8.06	183 177 - 191	64 62 - 67	74 63 - 85	< 0.10 < 0.10 - < 0.10
	Intake	25.0 24.8 - 25.1	24.5 24.1 - 24.7	7.8 7.5 - 8.3	7.3 7.0 - 7.8	7.72 7.44 - 8.16	7.57 7.31 - 8.10	181 177 - 186	63 61 - 66	74 73 - 75	< 0.10 < 0.10 - < 0.10
Ceriodaphnia dubia	Control	24.7 24.6 - 24.9	25.0 24.7 - 25.3	7.6 7.4 - 7.9	7.6 7.4 - 7.9	7.77 7.53 - 8.10	7.72 7.45 - 8.07	312 306 - 315	58 58 - 59	92 85 - 97	- - -
	10.98%	24.8 24.4 - 25.1	24.9 24.6 - 25.3	7.8 7.4 - 8.1	7.6 7.4 - 7.9	7.83 7.56 - 8.14	7.73 7.49 - 8.09	295 288 - 298	- - -	- - -	- - -
	22.0%	24.8 24.5 - 25.1	25.0 24.7 - 25.3	7.8 7.4 - 8.1	7.7 7.4 - 8.0	7.80 7.56 - 8.14	7.72 7.50 - 8.10	283 279 - 292	- - -	- - -	- - -
	43.9%	24.9 24.5 - 25.1	24.9 24.7 - 25.2	7.9 7.5 - 8.3	7.7 7.5 - 8.0	7.78 7.54 - 8.14	7.73 7.51 - 8.11	255 247 - 264	- - -	- - -	- - -
	72.0%	24.9 24.6 - 25.1	25.0 24.7 - 25.2	7.9 7.6 - 8.3	7.7 7.5 - 7.9	7.76 7.50 - 8.14	7.71 7.50 - 8.11	218 212 - 225	- - -	- - -	- - -
	100.0%	24.9 24.6 - 25.2	24.9 24.6 - 25.3	7.8 7.6 - 8.3	7.6 7.4 - 7.9	7.74 7.47 - 8.16	7.70 7.47 - 8.11	183 177 - 191	64 62 - 67	74 63 - 85	< 0.10 < 0.10 - < 0.10
	Intake	24.8 24.5 - 25.0	24.9 24.8 - 25.2	7.8 7.5 - 8.3	7.7 7.4 - 7.9	7.72 7.44 - 8.16	7.69 7.48 - 8.14	181 177 - 186	63 61 - 66	74 73 - 75	< 0.10 < 0.10 - < 0.10

Overall temperature (°C)	Average	Minimum	Maximum
Pimephales promelas	24.7	24.1	25.2
Ceriodaphnia dubia	24.9	24.4	25.3

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for the *Pimephales promelas* Test, Sequoyah Nuclear Plant Effluent (SQN), UV Treated Outfall 101, November 1-8, 2005.

Test	Sample ID	Temperature (°C)		Dissolved Oxygen (mg/L)		pH (S.U.)		Conductance (µmhos/cm)
		Initial	Final	Initial	Final	Initial	Final	
Pimephales promelas	Control	24.9 24.7 - 25.0	24.5 24.1 - 24.8	7.8 7.2 - 8.3	7.4 6.8 - 8.0	7.71 7.57 - 8.11	7.56 7.31 - 8.03	300 290 - 312
	10.98%	25.0 24.8 - 25.1	24.5 24.2 - 24.8	7.8 7.3 - 8.3	7.3 6.9 - 7.9	7.70 7.50 - 8.13	7.56 7.30 - 8.04	295 287 - 307
	22.0%	25.0 24.8 - 25.1	24.6 24.4 - 24.7	7.8 7.4 - 8.3	7.3 6.9 - 7.7	7.70 7.50 - 8.12	7.55 7.30 - 7.99	284 275 - 292
	43.9%	25.0 24.8 - 25.2	24.4 24.1 - 24.7	7.9 7.4 - 8.3	7.3 6.9 - 7.8	7.69 7.49 - 8.13	7.56 7.30 - 8.04	256 252 - 261
	72.0%	25.1 24.8 - 25.2	24.5 24.2 - 24.6	7.8 7.4 - 8.2	7.4 7.0 - 7.8	7.68 7.49 - 8.14	7.60 7.29 - 8.06	221 216 - 226
	100.0%	25.1 24.9 - 25.3	24.5 24.1 - 24.6	7.9 7.5 - 8.3	7.3 7.0 - 7.8	7.67 7.48 - 8.14	7.58 7.27 - 8.09	185 181 - 193
	Intake	25.1 24.9 - 25.2	24.4 24.1 - 24.6	7.9 7.6 - 8.3	7.3 7.0 - 7.7	7.66 7.43 - 8.18	7.57 7.27 - 8.06	184 173 - 198

Overall temperature (°C)	Average	Minimum	Maximum
Pimephales promelas	24.7	24.1	25.3

SUMMARY / CONCLUSIONS

Outfall 101 samples collected October 30-November 4, 2005, showed no toxic effects to fathead minnows or daphnids. The resulting IC_{25} values, for both species, were > 100 percent. Exposure of fathead minnows and daphnids to intake samples resulted in no significant differences from controls during this study period.

Fathead minnows were also exposed to UV treated Outfall 101 and intake samples since fish pathogens present in intake water have been the suspected cause of interference (anomalous dose response and high variability among replicates) in previous toxicity testing at Sequoyah. At the time this study was conducted, insignificant mortality occurred in minnows exposed to non-treated and UV treated samples.

Appendix A

ADDITIONAL TOXICITY TEST INFORMATION

SUMMARY OF METHODS

1. *Pimephales promelas*

Tests were conducted according to EPA-821-R-02-013 (October 2002) using four replicates, each containing ten test organisms, per treatment. Test vessels consisted of 500-mL plastic disposable cups, each containing 250-mL of test solution.

2. *Ceriodaphnia dubia*

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

DEVIATIONS / MODIFICATIONS TO TEST PROTOCOL

1. *Pimephales promelas* & 2. *Ceriodaphnia dubia*

Environmental Testing Solutions, Inc. performed chronic toxicity tests from November 1-8, 2005. The toxicity tests were conducted with samples collected from SQN Outfall 101 during non-oxidizing (November 1-4) and oxidizing (October 30-November 4) biocide applications. An internal peer review of the reports was performed by TVA biologists and the Senior Toxicologist and it was determined that incorrect serial dilutions were prepared. Previous permit dilutions of 10.98, 22.0, 43.9, 72.0, and 100% were used instead of the dilutions of 11.3, 22.6, 45.2, 72.6, and 100% specified in the recently renewed permit, effective September 1, 2005

The dilutions that were used in the tests were virtually the same as what the current permit requires ($\leq$ 1.3% effluent difference) given the variability inherent in performing whole effluent toxicity tests. Additionally, the error is judged to be inconsequential relative to evaluation of the test results since no toxicity to either species (*Pimephales promelas* or *Ceriodaphnia dubia*) was observed (i.e., IC25 > 100% effluent) during either test period.

A process change to TVA's NPDES biomonitoring compliance program has been implemented whereby the contract laboratory must confirm via email to TVA the exact dates that toxicity tests will be performed and the dilution series that will be used, prior to conducting all future tests.

This incident has been assessed as a laboratory deviation, and it will be identified as such in the final reports. The EPA chronic test manual (EPA-821-R-02-013) states that deviations must be evaluated on a case-by-case basis and deviations may or may not invalidate a test result "depending on the degree of the departure and the objective of the test." TVA biologists and the Senior Toxicologist have considered the degree of the deviation in this particular case and find no potential or observed impact on the test results. Therefore the test results for the subject tests are considered valid.

DEVIATIONS / MODIFICATIONS TO PRETEST CULTURE OR HOLDING OF TEST ORGANISMS

1. *Pimephales promelas*

   None

2. *Ceriodaphnia dubia*

   None

PHYSICAL AND CHEMICAL METHODS

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

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

3. Temperature was measured by EPA Method 170.1.

4. Dissolved oxygen was measured by EPA Method 360.1.

5. The pH was measured by EPA Method 150.1.

6. Conductance was measured by EPA Method 120.1.

7. Alkalinity was measured by EPA Method 310.1.

8. Total Hardness was measured by EPA Method 130.2.

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

QUALITY ASSURANCE

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

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

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

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

3. Dilution Water Used: Moderately hard synthetic water.

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

REFERENCES

1. NPDES Permit No. TN0026450.

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

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

4. Quality Assurance Program: Standard Operating Procedures, Environmental Testing Solutions, Inc (most current version).

Sequoyah Nuclear Plant Biomonitoring
November 1-8, 2005

Appendix B

Diffuser Discharge Concentrations of Total Residual Chlorine,

Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 4, 2005

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. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 4, 2005

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	-	-	-
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	-	-	-
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	-	-

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 4, 2005

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
01/02/2002	-	< 0.0079	0.023	0.02	0.006	-	-
01/03/2002	-	< 0.0042	0.023	0.014	-	-	-
01/04/2002	-	0.0124	0.024	0.014	0.009	-	-
01/05/2002	-	< 0.0042	-	-	-	-	-
01/06/2002	-	< 0.0042	-	-	-	-	-
01/07/2002	-	< 0.0089	0.024	0.014	0.006	-	-
02/24/2002	-	< 0.004	-	-	-	-	-
02/25/2002	-	< 0.004	0.023	0.023	-	-	-
02/26/2002	-	0.0143	0.023	0.023	0.007	-	-
02/27/2002	-	< 0.0041	0.023	0.023	-	-	-
02/28/2002	-	< 0.0041	0.024	0.008	-	-	-
03/01/2002	-	< 0.0041	0.024	0.008	-	-	-
05/05/2002	-	-	-	-	-	-	-
05/06/2002	-	-	0.058	0.02	0.014	-	-
05/07/2002	-	-	0.058	0.02	0.015	-	-
05/08/2002	-	-	0.056	0.019	-	-	-
05/09/2002	-	-	0.057	0.02	0.014	-	-
05/10/2002	-	-	0.056	0.019	-	-	-
08/04/2002	-	<0.0058	-	-	-	-	-
08/05/2002	-	<0.0058	0.053	0.018	-	-	0.025
08/06/2002	-	0.0092	0.053	0.018	-	-	-
08/07/2002	-	<0.0107	0.055	0.019	0.007	-	-
08/08/2002	-	<0.0061	0.055	0.019	-	-	-
08/09/2002	-	0.0152	0.054	0.018	0.008	-	-
10/06/2002	-	<0.00497	-	-	-	-	-
10/07/2002	-	0.0153	0.054	0.018	0.009	-	-
10/08/2002	-	<0.0092	0.054	0.018	0.007	-	-
10/09/2002	-	0.0124	0.053	0.018	0.009	-	-
10/10/2002	-	0.0134	0.054	0.018	0.009	-	-
10/11/2002	-	<0.0042	0.054	0.018	-	-	-
01/12/2003	-	<0.0035	-	-	-	-	-
01/13/2003	-	<0.006	0.025	0.019	0.009	-	-
01/14/2003	-	<0.0118	0.026	0.020		-	-
01/15/2003	-	<0.0063	0.026	0.020	0.009	-	-
01/16/2003	-	<0.0034	0.026	0.020		-	-
01/17/2003	-	<0.0034	0.026	0.009	-	-	-
04/06/2003	-	<0.0073	-	-	-	-	-
04/07/2003	-	<0.0189	-	0.021	-	-	-
04/08/2003	-	<0.0117	-	0.021	-	-	-
04/09/2003	-	<0.0139	-	0.021	0.016	-	-
04/10/2003	-	<0.0113	-	0.021	0.018	-	-
04/11/2003	-	<0.0073	-	0.022	-	-	-

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 4, 2005

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
06/15/2003	-	< 0.0045	-	-	-	-	-
06/16/2003	-	< 0.0037	0.057	0.020	-	-	0.022
06/17/2003	-	< 0.0048	0.041	0.014	-	-	0.024
06/18/2003	-	< 0.0048	0.041	0.014	-	-	0.024
06/19/2003	-	< 0.0085	0.058	0.020	-	-	0.025
06/20/2003	-	< 0.0048	0.058	0.020	-	-	0.025
08/03/2003	-	<0.0050	-	-	-	-	-
08/04/2003	-	<0.0050	0.058	0.020	-	-	-
08/05/2003	-	<0.0051	0.057	0.020	-	-	0.025
08/06/2003	-	<0.0084	0.057	0.020	-	-	0.025
08/07/2003	-	0.0129	0.057	0.020	-	-	0.024
08/08/2003	-	0.0153	0.057	0.020	0.009	-	-
10/05/2003	-	<0.0043	0.057	0.020	-	-	-
10/06/2003	-	<0.0043	0.057	0.020	-	-	0.025
10/07/2003	-	<0.0090	0.057	0.020	-	-	0.025
10/08/2003	-	<0.0106	0.057	0.020	-	-	0.025
10/09/2003	-	0.0181	0.026	0.022	-	-	0.025
10/10/2003	-	0.0183	0.026	0.024	0.009	-	-
02/01/2004	-	0.0093	0.027	0.009	-	-	-
02/02/2004	-	<0.0034	0.026	0.009	-	-	-
02/03/2004	-	<0.0034	0.026	0.009	-	-	-
02/04/2004	-	0.0124	0.026	0.009	0.009	-	-
02/05/2004	-	<0.0034	0.026	0.009	-	-	-
02/06/2004	-	0.0105	0.026	0.009	0.010	-	-
05/04/2004	-	<0.0123	0.026	0.019	-	-	0.025
05/05/2004	-	<0.0144	0.026	0.014	0.009	-	0.025
05/06/2004	-	<0.0146	0.037	0.013	-	-	0.025
05/07/2004	-	0.0227	0.058	0.020	0.009	-	0.025
05/08/2004	-	0.016	0.060	0.021	-	-	-
05/09/2004	-	<0.0104	0.058	0.020	-	-	-
07/04/2004	-	0.0217	0.057	0.019	-	-	-
07/05/2004	-	<0.0085	0.057	0.020	0.009	-	-
07/06/2004	-	<0.0077	0.058	0.020	-	-	0.031
07/07/2004	-	0.0252	0.056	0.019	-	-	0.031
07/08/2004	-	0.0223	0.057	0.019	0.009	-	-
07/09/2004	-	0.0182	0.057	0.020	0.009	-	-

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 4, 2005

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	Nalco 73551 mg/L EO/PO	H-150M mg/L Quat
11/07/2004	-	<0.0187	0.000	0.014	-	-	-	-	-
11/08/2004	-	<0.0192	0.047	0.030	-	-	-	-	-
11/09/2004	-	<0.0233	0.048	0.016	-	-	0.041	-	-
11/10/2004	-	<0.0149	0.047	0.016	-	-	0.041	-	-
11/11/2004	-	<0.0149	0.049	0.017	-	-	0.043	-	-
11/12/2004	-	<0.0253	0.048	0.017	-	-	0.042	-	-
02/06/2005	-	<0.0042	0.028	0.010	-	-	-	-	-
02/07/2005	-	<0.0116	0.028	-0.010	-	-	-	0.007	-
02/08/2005	-	<0.0080	0.028	0.010	-	-	-	-	-
02/09/2005	-	0.0199	0.028	0.010	-	-	-	-	-
02/10/2005	-	<0.0042	0.028	0.010	-	-	-	-	-
02/11/2005	-	0.0155	0.028	0.010	-	-	-	0.007	-
06/05/2005	-	0.0063	-	-	-	-	-	-	-
06/06/2005	-	0.0043	-	-	-	-	-	-	0.037
06/07/2005	-	0.0103	-	-	-	-	-	-	0.037
06/08/2005	-	0.0295	-	-	-	-	-	-	0.037
06/09/2005	-	0.0129	-	-	-	-	-	-	-
06/10/2005	-	0.0184	-	-	-	-	-	-	-
07/17/2005	-	0.0109	0.026	0.009	-	-	-	-	-
07/18/2005	-	0.0150	0.026	0.009	-	-	-	-	0.036
07/19/2005	-	0.0163	0.026	0.009	-	-	-	-	0.036
07/20/2005	-	0.0209	0.026	0.009	-	-	-	0.014	0.036
07/21/2005	-	0.0242	0.026	0.009	-	-	-	-	-
07/22/2005	-	0.0238	0.054	0.018	-	-	-	0.014	-
10/30/2005	-	0.0068	-	-	-	-	-	-	-
10/31/2005	-	0.0112	-	-	-	-	-	-	-
11/01/2005	-	0.0104	-	-	-	-	-	-	0.035
11/02/2005	-	0.0104	-	-	-	-	-	-	0.036
11/03/2005	-	0.0117	-	-	-	-	-	-	0.036
11/04/2005	-	0.0165	-	-	-	-	-	-	0.035

Sequoyah Nuclear Plant Biomonitoring
November 1-8, 2005

Appendix C

Chain of Custody Records and
Toxicity Test Bench Sheets

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: Sequoyah NP Toxicity

P.O. Number: N/A

Facility Sampled: Sequoyah NP

NPDES Number: TN0026450

Collected By: Duane Brigman
CHERI WILLIAMS

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

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

General Comments:

Field Identification / Sample Description	Grab/Comp.	Collection Date/Time		Container Number & Volume Collected	Flow MGD	Rain Event? (Mark as Appropriate)				Project #2170 Laboratory Use				
	—	Date	Time	—	—	Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	Comp	10/30/05-10/31/05 0500 TO 0400		2 (2.5gal)	NA					051031.01	1.1°/1.6°C	KW	1823	*
SQN-INT-TOX	Comp	10/30/05-10/31/05 0515 TO 0415		1 (2.5 gal)	NA					051031.02	0.8°C	KW	1823	*

Sample Custody – Fill In From Top Down

* Custody seals intact. Samples received in good condition Yunoen

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Duane Brigman	10/31/05 1344	Express Courier	10/31/05 1344
Express Courier	10/31/05 1823	ETS	10/31/05 1823

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 6°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

BIOMONITORING CHAIN OF CUSTODY RECORD

Page __1__ of __1__

Client: TVA

Project Name: Sequoyah NP Toxicity

P.O. Number: N/A

Facility Sampled: Sequoyah NP

NPDES Number: TN0026450

Collected By: Duane Brigman
CHEVY WILLIAMS

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

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

General Comments:

Field Identification / Sample Description	Grab/Comp	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Project # 2178 Laboratory Use				
	—	Date	Time	—	—	Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	Comp	11/1/05-11/2/05 0700 TO 0600	0915	2 (2.5gal)	NA			✓		051102.12	25°C/13°C	KEN	1500	*
SQN-INT-TOX	Comp	11/1/05-11/2/05 0715 TO 0615	1030	1 (2.5 gal)	NA			✓		051102.13	1.5°C	KEN	1500	*

Sample Custody – Fill In From Top Down

* Custody seals intact. Samples received in good condition. -Yunnan

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Duane Brigman	11/2/2005 1100	Express Courier	11/2/2005 1100
Express Courier	11/2/2005 1500	ETS	11/2/2005 1500

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 6°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

BIOMONITORING CHAIN OF CUSTODY RECORD

Page __1__ of __1__

Client: TVA

Project Name: Sequoyah NP Toxicity

P.O. Number: N/A

Facility Sampled: Sequoyah NP

NPDES Number: TN0026450

Collected By: Duane Brigman
Chevy Williams

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

Delivered By (Circle One):

FedEx UPS Bus Client

Other (specify): Express Courier

General Comments:

Field Identification / Sample Description	Grab/Comp	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Project # 2170 Laboratory Use				
	—	Date	Time	—	—	Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	Comp	11/3/05-11/4/05 0600 TO 0500	0745	2 (2.5gal)	NA			✓		051104.01	1.3/1.7	JL	1424	*
SQN-INT-TOX	Comp	11/3/05-11/4/05 0615 TO 0515	0900	1 (2.5 gal)	NA			✓		051164.02	1.2	JL	1424	*

Sample Custody – Fill In From Top Down

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

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Duane Brigman	11/4/05 10:00	Express Courier	11/4/05 10 00
Express Courier	11/4/05 1424	ETS	11/4/05 1424

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 6°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

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

Client: TVA
Facility: Sequoyah Nuclear Plant
NPDES #: TN 0026450
Project #: 2178

County: Hamilton
Treatment: Non-treated
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.98	22	43.9	72	100	
Effluent volume (mL)	274.5	550	1097.5	1800	2500	
Diluent volume (mL)	2225.5	1950	1402.5	700	0	
Total volume (mL)	2500	2500	2500	2500	2500	

Test organism information:		*Test information:*	
Organism age:	21 TO 22.5 HOURS OLD	Randomizing template:	YELLOW
Date and times organisms were born between:	10-31-05 1330 TO 1500	Incubator number:	3C
Organism source:	ABS BATCH PP 10-31-05	*Artemia* lot number:	BG1804T
Transfer bowl information:	pH = 7.73 Temperature = 24.0 °C	Total drying time:	21-HOURS
		Date / Time in: 11-08-05	1235
Average transfer volume:	10.4 mL	Date / Time out: 11-09-05	1241
		Oven temperature:	61°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Control water batch used MHSW	Sample numbers used	Analyst
0	11-01-05	—	1500	1202	10-24-05 B	051031.01 & 02	dl
1	11-02-05	0913	1520	1249	10-24-05 B	051031.01 & 02	dl
2	11-03-05	0915	1526	1217	10-31-05 A	051102.12 & 13	dl
3	11-04-05	0856	1503	1121	10-31-05 B	051102.12 & 13	dl
4	11-05-05	0845	1450	1134	11-03-05 A	051104.01 & 02	dl
5	11-06-05	0842	1448	1106	11-03-05 B	051104.01 & 02	dl
6	11-07-05	0845	1452	1110	11-05-05 A	051104.01 & 02	dl
7	11-08-05			1209			dl

Control information:		Acceptance criteria
% Mortality:	0%	≤ 20%
Average weight per initial larvae:	0.663	
Average weight per surviving larvae:	0.663	≥ 0.25 mg/larvae

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

Species: *Pimephales promelas* **Date:** 11-01-05

Client: TVA / Sequoyah Nuclear Plant - Non-treated

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	9 d	10	10	10
6	10	10	10	10	10	10	10	10	9	10	10	10
7	1SM 10	10	10	10	10	10	1LG 10	10	9	10	10	10
A = Pan weight (mg) Color identification: Magenta Analyst: KLeeman	14.77	14.55	14.96	14.75	1601	14.68	14.77	14.71	15.01	14.29	15.02	14.42
B = Pan + Larvae weight (mg) Analyst: Jumper	20.59	21.24	21.64	21.58	22.77	20.89	22.14	21.42	20.92	20.70	21.48	20.92
Larvae weight (mg) = A – B	5.82	6.69	7.18	6.83	6.76	6.21	7.37	6.71	5.91	6.41	6.46	6.50
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.582	0.669	0.718	0.683	0.676	0.621	0.737	0.671	0.591	0.641	0.646	0.650
Average weight per initial number of larvae (mg)	0.663				0.676				0.632			
Percent reduction from control (%)					-2.0%				4.7%			

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Species: *Pimephales promelas*

Client: TVA / Sequoyah Nuclear Plant - Non-treated **Date:** 11-01-05

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 LSH	10	10 LSH	10	10 LG	10	10	10
A = Pan weight (mg) Color identification: Magenta Analyst: MKeenan	15.05	14.27	14.47	14.31	14.34	14.86	14.61	14.73	14.98	14.75	15.15	15.05
B = Pan + Larvae weight (mg) Analyst: Jumna	21.84	20.56	21.58	20.65	20.16	22.04	20.42	21.55	22.10	21.66	21.24	22.06
Larvae weight (mg) = A – B	6.79	6.29	7.11	6.34	5.82	7.18	5.81	6.82	7.12	6.91	6.09	6.21
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.679	0.629	0.711	0.634	0.582	0.718	0.581	0.682	0.712	0.691	0.609	0.621
Average weight per initial number of larvae (mg)	0.663		0.0		0.641		3.4%		0.658		0.7%	
Percent reduction from control (%)												

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Species: ***Pimephales promelas***

Client: TVA / Sequoyah Nuclear Plant - Non-treated **Date:** 11-01-05

Survival and Growth Data

Day	100% Intake			
	Y	Z	AA	BB
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	9 1d	10	10 ISH	10
A = Pan weight (mg) · Color identification: Magenta · Analyst: KEKelman	14.34	14.79	13.93	14.48
B = Pan + Larvae weight (mg) · Analyst: Glumner	20.26	21.35	19.99	21.45
Larvae weight (mg) = A – B	5.92	6.56	6.06	6.97
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.592	0.656	0.606	0.697
Average weight per initial number of larvae (mg) / Percent reduction from control (%)	0.638		3.8%	

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: dh

Comments:

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 01-08, 2005

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

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

Project number: 2178

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	Not for Compliance Assessment, Internal Laboratory QC: Weight / Surviving number of larvae (mg)	Not for Compliance Assessment, Internal Laboratory QC: Mean weight / Surviving number of larvae (mg)	Not for Compliance Assessment, Internal Laboratory QC: Coefficient of variation (Mean weight per surviving number of larvae) (%)	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight / Initial number of larvae (mg)	Coefficient of variation (Mean weight per initial number of larvae) (%)	Percent reduction from control (%)
Control	A	10	10	14.77	20.59	5.82	0.582	0.663	8.7	0.582	100.0	0.663	8.7	Not applicable
	B	10	10	14.55	21.24	6.69	0.669			0.669				
	C	10	10	14.46	21.64	7.18	0.718			0.718				
	D	10	10	14.75	21.58	6.83	0.683			0.683				
10.98%	E	10	10	16.01	22.77	6.76	0.676	0.676	7.0	0.676	100.0	0.676	7.0	-2.0
	F	10	10	14.68	20.89	6.21	0.621			0.621				
	G	10	10	14.77	22.14	7.37	0.737			0.737				
	H	10	10	14.71	21.42	6.71	0.671			0.671				
22%	I	10	9	15.01	20.92	5.91	0.657	0.648	1.0	0.591	97.5	0.632	4.4	4.7
	J	10	10	14.29	20.70	6.41	0.641			0.641				
	K	10	10	15.02	21.48	6.46	0.646			0.646				
	L	10	10	14.42	20.92	6.50	0.650			0.650				
43.9%	M	10	10	15.05	21.84	6.79	0.679	0.663	5.9	0.679	100.0	0.663	5.9	0.0
	N	10	10	14.27	20.56	6.29	0.629			0.629				
	O	10	10	14.47	21.58	7.11	0.711			0.711				
	P	10	10	14.31	20.65	6.34	0.634			0.634				
72%	Q	10	10	14.34	20.16	5.82	0.582	0.641	10.9	0.582	100.0	0.641	10.9	3.4
	R	10	10	14.86	22.04	7.18	0.718			0.718				
	S	10	10	14.61	20.42	5.81	0.581			0.581				
	T	10	10	14.73	21.55	6.82	0.682			0.682				
100%	U	10	10	14.98	22.10	7.12	0.712	0.658	7.7	0.712	100.0	0.658	7.7	0.7
	V	10	10	14.75	21.66	6.91	0.691			0.691				
	W	10	10	15.15	21.24	6.09	0.609			0.609				
	X	10	10	15.85	22.06	6.21	0.621			0.621				
100% Intake	Y	10	9	14.34	20.26	5.92	0.658	0.654	5.7	0.592	97.5	0.638	7.5	3.8
	Z	10	10	14.79	21.35	6.56	0.656			0.656				
	AA	10	10	13.93	19.99	6.06	0.606			0.606				
	BB	10	10	14.48	21.45	6.97	0.697			0.697				

Outfall 101:
Dunnett's MSD value: 0.0862
PMSD: 13.0

Intake:
Dunnett's MSD value: 0.0731
PMSD: 11.0

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, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 13.2% from the control (determined through reference toxicant testing).
Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.
Upper PMSD bound determined by USEPA (90th percentile) = 35%.
The lower and upper bounds were calculated by the USEPA using 205 tests conducted from 19 laboratories for *Pimephales* growth in chronic reference toxicant tests.

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

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 01-08, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/1/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN Outfall 101, Non-treated
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

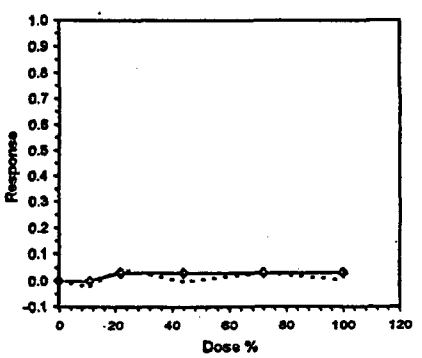
Conc-%	1	2	3	4
D-Control	0.5820	0.6690	0.7180	0.6830
10.98	0.6760	0.6210	0.7370	0.6710
22	0.5910	0.6410	0.6460	0.6500
43.9	0.6790	0.6290	0.7110	0.6340
72	0.5820	0.7180	0.5810	0.6820
100	0.7120	0.6910	0.6090	0.6210

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6630	1.0000	0.6630	0.5820	0.7180	8.718	4				0.6696	1.0000
10.98	0.6763	1.0200	0.6763	0.6210	0.7370	7.025	4	-0.370	2.410	0.0862	0.6696	1.0000
22	0.6320	0.9532	0.6320	0.5910	0.6500	4.364	4	0.866	2.410	0.0862	0.6486	0.9685
43.9	0.6633	1.0004	0.6633	0.6290	0.7110	5.876	4	-0.007	2.410	0.0862	0.6486	0.9685
72	0.6408	0.9664	0.6408	0.5810	0.7180	10.921	4	0.622	2.410	0.0862	0.6486	0.9685
100	0.6583	0.9928	0.6583	0.6090	0.7120	7.734	4	0.133	2.410	0.0862	0.6486	0.9685

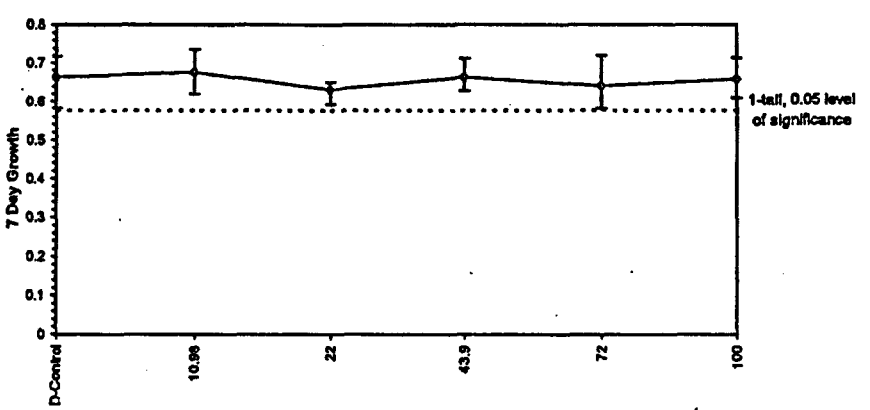
Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.958355725		0.884		-0.0946889	-1.10001503
Bartlett's Test indicates equal variances (p = 0.78)					2.487827778		15.08627224			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	100	>100		1	0.086238688	0.130073435	0.001059367	0.002560944	0.8330639	5, 18
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



TVA / Sequoyah Nuclear Plant, Intake
Non-treated
November 01-08, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/1/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN Intake, Non-treated
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

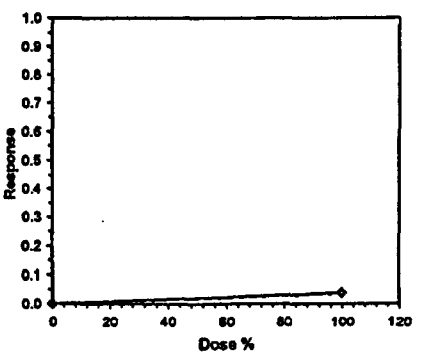
Conc-%	1	2	3	4
D-Control	0.5820	0.6690	0.7180	0.6830
100	0.5920	0.6560	0.6060	0.6970

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6630	1.0000	0.6630	0.5820	0.7180	8.718	4				0.6630	1.0000
100	0.6378	0.9619	0.6378	0.5920	0.6970	7.544	4	0.672	1.943	0.0731	0.6378	0.9619

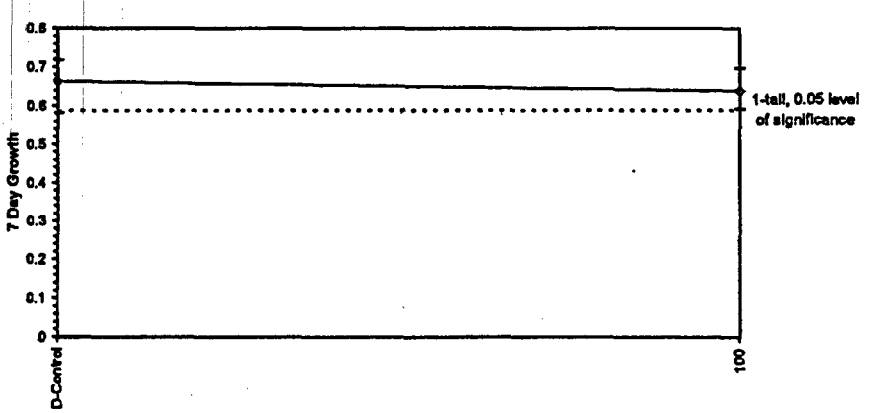
Auxiliary Tests	Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.942614555		0.749		-0.43182941	-0.80370079
F-Test indicates equal variances (p = 0.77)	1.443104506		47.46722794			
Hypothesis Test (1-tail, 0.05)	MSDu	MSDp	MSB	MSE	F-Prob	df
Homoscedastic t Test indicates no significant differences	0.07306709	0.110206772	0.001275125	0.002827792	0.526889384	1, 6
Treatments vs D-Control						

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



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

Client: TVA
Facility: Sequoyah Nuclear Plant
NPDES #: TN 0026450
Project #: 2178

County: Hamilton
Treatment: Non-treated
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.98	22	43.9	72	100	
Effluent volume (mL)	274.5	550	1097.5	1800	2500	
Diluent volume (mL)	2225.5	1950	1402.5	700	0	
Total volume (mL)	2500	2500	2500	2500	2500	

Test organism information:		*Test information:*	
Organism age:	< 24-HOURS OLD	Randomizing template:	ORANGE & WHITE
Date and times organisms were born between:	11-01-05 0731 TO 1000	Incubator number and shelf location:	~~2B2~~ 3EZ
Organism source:	10-25-05 A-F	YCT batch:	09-30-05
Transfer bowl information:	pH = 7.91 Temperature = 24.8	*Selenastrum* batch:	10-16-05

Daily renewal information:

Day	Date	Test initiation, renewal, or termination time	Control water batch used MHSW	Sample numbers used	Analyst
0	11-01-05	1110	10-24-05B	051031.01 + 02	dl
1	11-02-05	1019	10-24-05B	051031.01 & 02	dl
2	11-03-05	1014	10-31-05 A	051102. ~~13 & 14R~~ 12 & 13	dl
3	11-04-05	1020	10-31-05 B	051102. ~~13 & 14R~~ 12 & 13	dl
4	11-05-05	1023	11-03-05 A	051104.01 & 02	dl
5	11-06-05	1025	11-03-05B	051104.01 & 02	dl
6	11-07-05	1017	11-05-05 A	051104.01 & 02	dl
7	11-08-05	1119			dl

Control information:	1	2	Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	0%	≤ 20%	7-day LC50	> 100%
% Adults having 3rd Broods:	100%	100%	≥ 80%	NOEC	100%
% Mortality:	0%	0%	≤ 20%	LOEC	> 100%
Mean Offspring/Female:	27.7	29.1	≥ 15.0 offspring/female	ChV	> 100%
% CV:	9.8%	7.8%	< 40.0 %	IC25	> 100%

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated Date: 11-01-05

CONTROL - 1 *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	5	4	4	4	3	5	3	3	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	13	10	0	10	11	*1	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	9	0	0	9	10	11	11	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	14	13	13	11	16	11	15	12	12	15
Total young produced		32	28	26	25	31	24	30	26	26	29
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L
X for 3rd Broods		X	X	X	X	X	X	X	X	X	X

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	27.7

CONC: 10.98% *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	3	3	4	4	4	5	5	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	0	11	10	11	11	0	10	10	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	11	0	0	0	0	11	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	12	15	13	16	13	15	15	12	15	11
Total young produced		27	30	27	29	28	30	30	27	30	26
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	28.4
% Reduction from Control:	-2.5%

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated **Date:** 11-01-05

CONC: 22% *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	4	4	5	4	4	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	0	0	*2	0	0	0	12	0	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	12	14	10	10	10	13	0	12	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	14	14	13	15	13	16	14	17	13	13
Total young produced		29	30	31	31	28	30	31	33	29	28
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead) *SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	30.0
% Reduction from Control:	-8.37%

CONC: 43.9% *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	5	4	3	5	4	4	4	5	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	10	*2	0	0	0	0	0	12	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	+1	0	12	12	11	13	12	10	0	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	17	15	13	13	16	15	18	16	15	15
Total young produced		33	30	31	28	32	32	34	30	32	29
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead) *SPLIT BROOD

+ CARRY OVER

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	31.1
% Reduction from Control:	-12.3%

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated **Date:** 11-01-05

CONC: 72% ***Survival and Reproduction Data***

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

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	32.6
% Reduction from Control:	-17.7%

CONC: 100% ***Survival and Reproduction Data***

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	5	4	4	4	6	4	4	4	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	#1	12	10	0	0	0	0	13	13
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	14	11	0	0	14	11	12	12	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	17	15	17	13	15	15	16	18	15	18
Total young produced		35	32	33	27	33	32	32	34	32	36
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead) #SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	32.6
% Reduction from Control:	-17.7%

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated **Date:** 11-01-05

CONTROL - 2 *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	3	5	4	4	5	4	3	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	10	12	9	11	11	0	10	0	0	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	12	0	9	11	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	13	17	14	16	13	13	16	14	14	15
Total young produced		26	34	27	31	29	29	29	27	29	30
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	29.1

CONC: 100% Intake *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	5	4	4	4	5	4	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	0	11	0	0	13	0	10	12	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	11	11	0	12	10	0	14	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	20	16	16	19	15	17	15	20	18	21
Total young produced		36	32	31	35	29	35	34 (33)	34	34	37
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	33.6
% Reduction from Control:	-15.5%

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 01-08, 2005

Verification of *Ceriodaphnia* Reproduction Totals

Control-1

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	5	4	4	4	3	5	3	3	3	39
5	13	10	0	10	11	1	0	0	0	0	45
6	0	0	9	0	0	9	10	11	11	11	61
7	14	13	13	11	16	11	15	12	12	15	132
Total	32	28	26	25	31	24	30	26	26	29	277

10.98%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	3	3	4	4	4	5	5	3	39
5	11	0	11	10	11	11	0	10	10	12	86
6	0	11	0	0	0	0	11	0	0	0	22
7	12	15	13	16	13	15	15	12	15	11	137
Total	27	30	27	29	28	30	30	27	30	26	284

22%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	4	4	5	4	4	4	4	4	41
5	11	0	0	2	0	0	0	12	0	11	36
6	0	12	14	10	10	10	13	0	12	0	81
7	14	14	13	15	13	16	14	17	13	13	142
Total	29	30	31	31	28	30	31	33	29	28	300

43.9%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	5	4	3	5	4	4	4	5	4	42
5	11	10	2	0	0	0	0	0	12	0	35
6	1	0	12	12	11	13	12	10	0	10	81
7	17	15	13	13	16	15	18	16	15	15	153
Total	33	30	31	28	32	32	34	30	32	29	311

72%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	5	6	4	4	4	4	5	4	4	44
5	0	12	0	0	0	0	0	0	13	0	25
6	13	0	10	11	13	12	12	14	0	11	96
7	19	15	14	18	14	14	15	17	16	19	161
Total	36	32	30	33	31	30	31	36	33	34	326

100%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	5	4	4	4	6	4	4	4	5	44
5	0	1	12	10	0	0	0	0	13	13	49
6	14	11	0	0	14	11	12	12	0	0	74
7	17	15	17	13	15	15	16	18	15	18	159
Total	35	32	33	27	33	32	32	34	32	36	326

Control-2

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	3	5	4	4	5	4	3	4	4	4	40
5	10	12	9	11	11	0	10	0	0	11	74
6	0	0	0	0	0	12	0	9	11	0	32
7	13	17	14	16	13	13	16	14	14	15	145
Total	26	34	27	31	29	29	29	27	29	30	291

100% Intake

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	5	4	4	4	5	4	4	4	4	43
5	0	0	11	0	0	13	0	10	12	12	58
6	11	11	0	12	10	0	14	0	0	0	58
7	20	16	16	19	15	17	15	20	18	21	177
Total	36	32	31	35	29	35	33	34	34	37	336

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 01-08, 2005

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

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

Project number: 2178

Reveiwed by: Jumma

Concentration (%)	Replicate number										Survival (%)	Average reproduction (offspring/female)	Coefficient of variation (%)	Percent reduction from pooled controls (%)
	1	2	3	4	5	6	7	8	9	10				
Control - 1	32	28	26	25	31	24	30	26	26	29	100	27.7	9.8	Not applicable
10.98%	27	30	27	29	28	30	30	27	30	26	100	28.4	5.6	-2.5
22%	29	30	31	31	28	30	31	33	29	28	100	30.0	5.2	-8.3
43.9%	33	30	31	28	32	32	34	30	32	29	100	31.1	6.0	-12.3
72%	36	32	30	33	31	30	31	36	33	34	100	32.6	6.8	-17.7
100%	35	32	33	27	33	32	32	34	32	36	100	32.6	7.4	-17.7
Control - 2	26	34	27	31	29	29	29	27	29	30	100	29.1	7.8	Not applicable
100% Intake	36	32	31	35	29	35	33	34	34	37	100	33.6	7.2	-15.5

Outfall 101:
Dunnett's MSD value: 2.148
PMSD: 7.8

Intake:
Dunnett's MSD value: 1.821
PMSD: 6.3

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, Inc. chronic toxicity tests when a toxicant reduces *Ceriodaphnia* reproduction by 9.7% from the control.

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

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

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

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

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 01-08, 2005

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	11/1/2005	Test ID:	CdFRCR	Sample ID:	TVA / SQN Outfall 101, Non-treated
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia
Comments:					

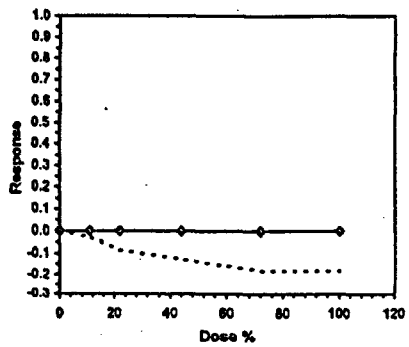
Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	32.000	28.000	26.000	25.000	31.000	24.000	30.000	26.000	26.000	29.000
10.98	27.000	30.000	27.000	29.000	28.000	30.000	30.000	27.000	30.000	26.000
22	29.000	30.000	31.000	31.000	28.000	30.000	31.000	33.000	29.000	28.000
43.9	33.000	30.000	31.000	28.000	32.000	32.000	34.000	30.000	32.000	29.000
72	36.000	32.000	30.000	33.000	31.000	30.000	31.000	36.000	33.000	34.000
100	35.000	32.000	33.000	27.000	33.000	32.000	32.000	34.000	32.000	36.000

			Transform: Untransformed						1-Tailed		Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	27.700	1.0000	27.700	24.000	32.000	9.784	10				30.400	1.0000
10.98	28.400	1.0253	28.400	26.000	30.000	5.555	10	-0.745	2.287	2.148	30.400	1.0000
22	30.000	1.0830	30.000	28.000	33.000	5.212	10	-2.449	2.287	2.148	30.400	1.0000
43.9	31.100	1.1227	31.100	28.000	34.000	5.958	10	-3.620	2.287	2.148	30.400	1.0000
72	32.600	1.1769	32.600	30.000	36.000	6.813	10	-5.217	2.287	2.148	30.400	1.0000
100	32.600	1.1769	32.600	27.000	36.000	7.402	10	-5.217	2.287	2.148	30.400	1.0000

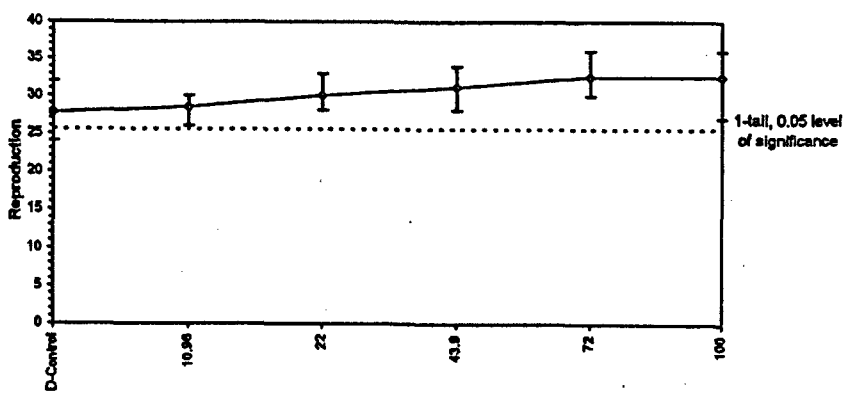
Auxiliary Tests				Statistic		Critical		Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)				0.528656304		1.035		-0.08559173	-0.07714908
Bartlett's Test indicates equal variances (p = 0.49)				4.433810234		15.08627224			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	100	>100		1	2.147790172	0.077537551	43.24	4.411111111	1.0E-06	5, 54
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



TVA / Sequoyah Nuclear Plant, Intake
Non-treated
November 01-08, 2005

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	11/1/2005	Test ID:	CdFRCR	Sample ID:	TVA / SQN Intake, Non-treated
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia
Comments:					

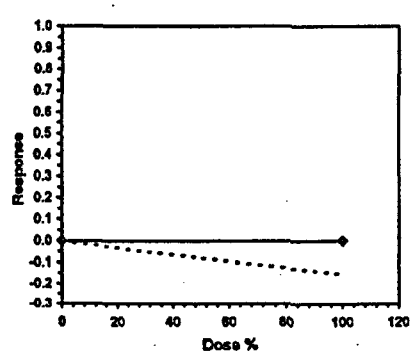
Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	26.000	34.000	27.000	31.000	29.000	29.000	29.000	27.000	29.000	30.000
100	36.000	32.000	31.000	35.000	29.000	35.000	33.000	34.000	34.000	37.000

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	29.100	1.0000	29.100	26.000	34.000	7.845	10				31.350	1.0000
100	33.600	1.1546	33.600	29.000	37.000	7.181	10	-4.284	1.734	1.821	31.350	1.0000

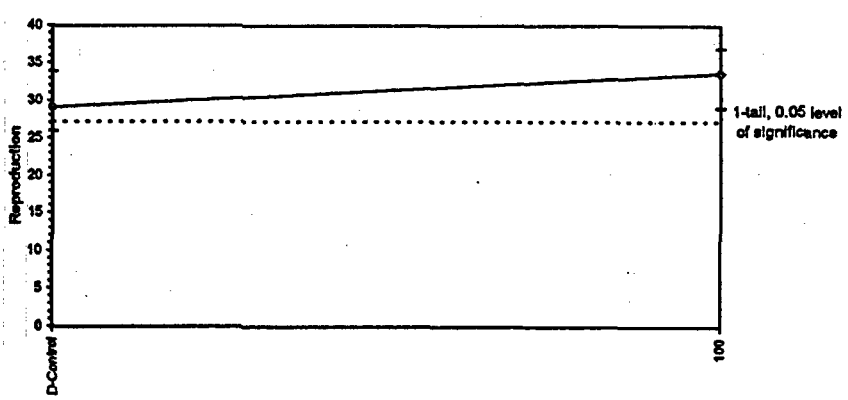
Auxiliary Tests	Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.986979127		0.868		0.080465966	0.153076601
F-Test indicates equal variances (p = 0.87)	1.117270827		6.541089535			
Hypothesis Test (1-tail, 0.05)	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Homoscedastic t Test indicates no significant differences	1.821454797	0.062592948	101.25	5.516666667	4.5E-04	1, 18
Treatments vs D-Control						

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



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

Client: TVA
Facility: Sequoyah Nuclear Plant
NPDES #: TN 0026450
Project #: 2178

County: Hamilton
Treatment: UV-treated
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.98	22	43.9	72	100	Each concentration was treated
Effluent volume (mL)	274.5	550	1097.5	1800	2500	for 2 minutes with a UV sterilizer
Diluent volume (mL)	2225.5	1950	1402.5	700	0	to remove pathogenic
Total volume (mL)	2500	2500	2500	2500	2500	interferences.

Test organism information:		*Test information:*	
Organism age:	21.5 TO 23 HOURS OLD	Randomizing template:	BLUE
Date and times organisms were born between:	10-31-05 1330 TO 1500	Incubator number:	2.8
Organism source:	ABS BATCH Pp 10-31-05	*Artemia* lot number:	BG1804T
Transfer bowl information:	pH = 7.73 Temperature = 24.0 °C	Total drying time:	24-HOURS
		Date / Time in: 11-08-05	1235
Average transfer volume:	10.4 mL	Date / Time out: 11-09-05	1241
		Oven temperature:	61°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	MHS batch used	Sample numbers used	Analyst
0	11-01-05	———	1500	1221	10-24-05 B	051031.01 & 02	
1	11-02-05	0913	1520	1237	10-24-05 B	051031.01 & 02	
2	11-03-05	0915	1526	1230	10-31-05 A	051102.12 & 13	
3	11-04-05	0856	1503	1137	10-31-05 B	051102.12 & 13	
4	11-05-05	0845	1450	1148	11-03-05 A	051104.01 & 02	
5	11-06-05	0842	1448	1130	11-03-05 B	051104.01 & 02	
6	11-07-05	0845	1452	1127	11-05-05 A	051104.01 & 02	
7	11-08-05			1229			

Control information:		Acceptance criteria
% Mortality:	0%	≤ 20%
Average weight per initial larvae:	0.650	
Average weight per surviving larvae:	0.650	≥ 0.25 mg/larvae

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

Species: *Pimephales promelas* **Date:** 11-01-05

Client: TVA / Sequoyah Nuclear Plant - UV-treated

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 (16)	10	10	10 (1SM)	10	10	10
A = Pan weight (mg) Color identification: Lt. Blue Analyst: KHeenan	16.22	14.93	15.80	15.06	14.99	16.21	14.77	15.94	14.37	14.59	14.75	14.65
B = Pan + Larvae weight (mg) Analyst: Jumar	22.47	21.73	22.23	21.63	21.89	23.40	21.53	22.86	20.29	21.08	21.15	21.32
Larvae weight (mg) = A – B = C	6.25	6.80	6.37	6.57	6.90	7.19	6.76	6.92	5.92	6.49	6.40	6.67
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.625	0.680	0.637	0.657	0.690	0.719	0.676	0.692	0.592	0.649	0.640	0.667

Average weight per initial number of larvae (mg)	Percent reduction from control (%)							
0.650		0.694		-6.8%		0.637		2.0%

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Species: ***Pimephales promelas***

Client: TVA / Sequoyah Nuclear Plant - UV-treated **Date:** 11-01-05

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	9[1d]	10	10	10[ISA]	10	10	10	10	10	10	10
A = Pan weight (mg) Color identification: Blue Analyst: KEKeenan	16.09	14.64	14.77	14.55	14.54	14.46	14.27	14.77	14.57	15.07	16.30	14.53
B = Pan + Larvae weight (mg) Analyst: JHumphrey	22.67	20.58	21.54	21.26	20.27	21.12	20.96	21.29	21.02	21.69	22.70	21.03
Larvae weight (mg) = A – B = C	6.58	5.94	6.77	6.71	5.73	6.66	6.69	6.52	6.45	6.62	6.40	6.50
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.658	0.594	0.677	0.671	0.573	0.666	0.669	0.652	0.645	0.662	0.640	0.650
Average weight per initial number of larvae (mg)	0.650				0.640				0.649			
Percent reduction from control (%)	0.0%				1.5%				0.1%			

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: JH

Comments:

Species: ***Pimephales promelas***

Client: TVA / Sequoyah Nuclear Plant - UV-treated **Date:** 11-01-05

Survival and Growth Data

Day	100% Intake			
	Y	Z	AA	BB
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) Color identification: Lt Blue Analyst: KHellman	15.88	15.30	14.23	16.19
B = Pan + Larvae weight (mg) Analyst: JVanover	22.52	22.16	20.97	23.04
Larvae weight (mg) = A – B = C	6.64	6.86	6.74	6.85
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.664	0.686	0.674	0.685
Average weight per initial number of larvae (mg)	0.677			
Percent reduction from control (%)	-4.2%			

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: JH

Comments:

TVA / Sequoyah Nuclear Plant, Outfall 101
UV-treated
July 19-26, 2005

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

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

Project number: 2178

Reviewed by:

Concentration (%)	Replicate	Initial number of larvae	Final number of larvae	A = Pan weight (mg)	B = Pan + Larvae weight (mg)	Larvae weight (mg) = A - B	Not for Compliance Assessment, Internal Laboratory QC: Weight / Surviving number of larvae (mg)	Mean weight / Surviving number of larvae (mg)	Coefficient of variation (Mean weight per surviving number of larvae) (%)	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight / Initial number of larvae (mg)	Coefficient of variation (Mean weight per initial number of larvae) (%)	Percent reduction from control (%)
Control	A	10	10	16.22	22.47	6.25	0.625	0.650	3.7	0.625	100.0	0.650	3.7	Not applicable
	B	10	10	14.93	21.73	6.80	0.680			0.680				
	C	10	10	15.86	22.23	6.37	0.637			0.637				
	D	10	10	15.06	21.63	6.57	0.657			0.657				
10.98%	E	10	10	14.99	21.89	6.90	0.690	0.694	2.6	0.690	100.0	0.694	2.6	-6.8
	F	10	10	16.21	23.40	7.19	0.719			0.719				
	G	10	10	14.77	21.53	6.76	0.676			0.676				
	H	10	10	15.94	22.86	6.92	0.692			0.692				
22%	I	10	10	14.37	20.29	5.92	0.592	0.637	5.0	0.592	100.0	0.637	5.0	2.0
	J	10	10	14.59	21.08	6.49	0.649			0.649				
	K	10	10	14.75	21.15	6.40	0.640			0.640				
	L	10	10	14.65	21.32	6.67	0.667			0.667				
43.9%	M	10	10	16.09	22.67	6.58	0.658	0.667	1.4	0.658	97.5	0.650	5.9	0.0
	N	10	9	14.64	20.58	5.94	0.660			0.594				
	O	10	10	14.77	21.54	6.77	0.677			0.677				
	P	10	10	14.55	21.26	6.71	0.671			0.671				
72%	Q	10	10	14.54	20.27	5.73	0.573	0.640	7.1	0.573	100.0	0.640	7.1	1.5
	R	10	10	14.46	21.12	6.66	0.666			0.666				
	S	10	10	14.27	20.96	6.69	0.669			0.669				
	T	10	10	14.77	21.29	6.52	0.652			0.652				
100%	U	10	10	14.57	21.02	6.45	0.645	0.649	1.5	0.645	100.0	0.649	1.5	0.1
	V	10	10	15.07	21.69	6.62	0.662			0.662				
	W	10	10	16.30	22.70	6.40	0.640			0.640				
	X	10	10	14.53	21.03	6.50	0.650			0.650				
100% Intake	Y	10	10	15.88	22.52	6.64	0.664	0.677	1.5	0.664	100.0	0.677	1.5	-4.2
	Z	10	10	15.30	22.16	6.86	0.686			0.686				
	AA	10	10	14.23	20.97	6.74	0.674			0.674				
	BB	10	10	16.19	23.04	6.85	0.685			0.685				

Outfall 101:
Dunnett's MSD value: 0.0517
PMSD: 8.0

Intake:
Dunnett's MSD value: 0.0255
PMSD: 3.9

MSD = Minimum Significant Difference
PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 13.2% from the control (determined through reference toxicant testing).
Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.
Upper PMSD bound determined by USEPA (90th percentile) = 35%.
The lower and upper bounds were calculated by the USEPA using 205 tests conducted from 19 laboratories for *Pimephales* growth in chronic reference toxicant tests.

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

TVA / Sequoyah Nuclear Plant, Outfall 101
UV-treated
November 01-08, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/1/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN Outfall 101, UV-treated
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

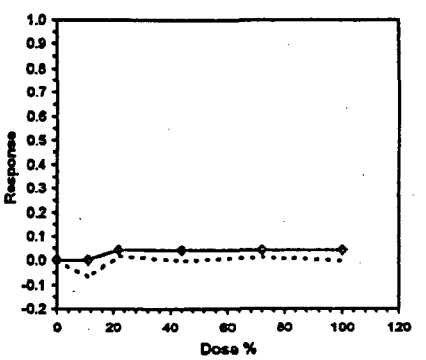
Conc-%	1	2	3	4
D-Control	0.6250	0.6800	0.6370	0.6570
10.98	0.6900	0.7190	0.6760	0.6920
22	0.5920	0.6490	0.6400	0.6670
43.9	0.6580	0.5940	0.6770	0.6710
72	0.5730	0.6660	0.6690	0.6520
100	0.6450	0.6620	0.6400	0.6500

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6498	1.0000	0.6498	0.6250	0.6800	3.709	4				0.6720	1.0000
10.98	0.6943	1.0685	0.6943	0.6760	0.7190	2.588	4	-2.074	2.410	0.0517	0.6720	1.0000
22	0.6370	0.9804	0.6370	0.5920	0.6670	5.028	4	0.594	2.410	0.0517	0.6441	0.9584
43.9	0.6500	1.0004	0.6500	0.5940	0.6770	5.872	4	-0.012	2.410	0.0517	0.6441	0.9584
72	0.6400	0.9850	0.6400	0.5730	0.6690	7.075	4	0.454	2.410	0.0517	0.6441	0.9584
100	0.6493	0.9992	0.6493	0.6400	0.6620	1.452	4	0.023	2.410	0.0517	0.6441	0.9584

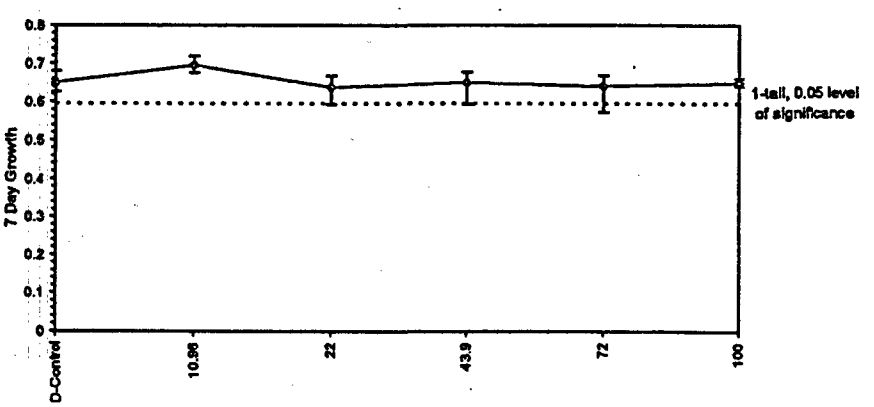
Auxiliary Tests				Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)				0.894263983		0.884		-1.08984758	0.760436643
Bartlett's Test indicates equal variances (p = 0.25)				6.584905624		15.08627224			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	100	>100		1	0.051714098	0.079590762	0.001727475	0.000920903	0.148802772	5, 18
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



TVA / Sequoyah Nuclear Plant, Intake
UV-treated
November 01-08, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/1/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN Intake, UV-treated
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

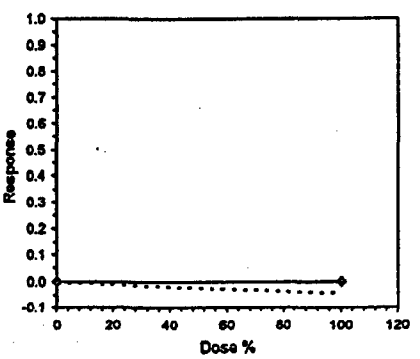
Conc-%	1	2	3	4
D-Control	0.6250	0.6800	0.6370	0.6570
100	0.6640	0.6860	0.6740	0.6850

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6498	1.0000	0.6498	0.6250	0.6800	3.709	4				0.6635	1.0000
100	0.6773	1.0423	0.6773	0.6640	0.6860	1.532	4	-2.096	1.943	0.0255	0.6635	1.0000

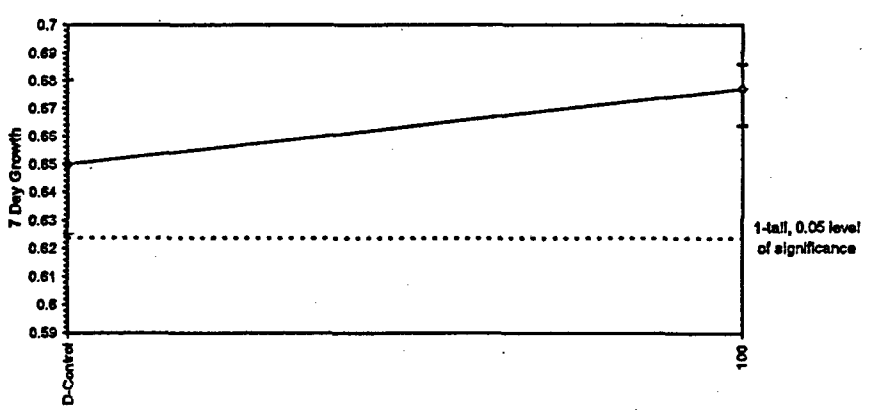
Auxiliary Tests	Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.954809606		0.749		0.360875074	0.148968767
F-Test indicates equal variances (p = 0.20)	5.399690151		47.46722794			
Hypothesis Test (1-tail, 0.05)	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Homoscedastic t Test indicates no significant differences	0.02549383	0.039236367	0.0015125	0.00084425	0.080913521	1, 6
Treatments vs D-Control						

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 01-08, 2005
Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1000.0)
Species: *Pimephales promelas*

Daily Chemical Analyses

Project number: 2178

Reviewed by:

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.73	7.63	7.79	7.47	7.67	7.48	7.53	7.38	7.87	7.31	7.70	8.03	8.10	7.82
	DO (mg/L)	7.9	7.8	7.7	7.4	7.5	7.2	7.4	7.0	7.7	7.3	7.5	7.4	7.7	7.2
	Conductivity (µmhos/cm)	314		315		306		310		314		313		310	
	Alkalinity (mg/L $CaCO_3$)	59				58		59		58		58		58	
	Hardness (mg/L $CaCO_3$)	92				87		97		95		93		85	
	Temperature (°C)	24.6	24.6	24.8	24.8	24.9	24.5	24.7	24.6	24.8	24.2	24.8	24.4	24.7	24.2
10.98%	pH (SU)	7.87	7.60	7.90	7.34	7.80	7.34	7.87	7.34	7.64	7.28	7.56	7.98	8.14	7.80
	DO (mg/L)	8.0	7.8	8.1	7.0	7.9	7.2	7.8	6.9	7.6	7.1	7.4	7.0	7.8	7.3
	Conductivity (µmhos/cm)	295		298		288		298		292		295		296	
	Temperature (°C)	24.6	24.6	24.9	24.7	25.0	24.4	24.7	24.6	24.9	24.4	24.8	24.4	24.7	24.2
22%	pH (SU)	7.86	7.61	7.84	7.43	7.73	7.34	7.84	7.33	7.64	7.29	7.56	8.02	8.14	7.77
	DO (mg/L)	8.1	7.8	8.1	7.1	7.8	7.1	7.9	6.8	7.7	7.3	7.4	7.1	7.8	7.2
	Conductivity (µmhos/cm)	284		292		279		284		280		283		281	
	Temperature (°C)	24.6	24.6	24.9	24.5	25.0	24.4	24.7	24.4	24.9	24.4	24.8	24.4	24.8	24.2
43.9%	pH (SU)	7.83	7.59	7.81	7.44	7.71	7.33	7.80	7.30	7.62	7.30	7.54	8.04	8.14	7.86
	DO (mg/L)	8.3	7.8	8.0	7.2	7.8	7.0	8.0	6.8	7.7	7.5	7.5	7.2	7.8	7.5
	Conductivity (µmhos/cm)	256		264		254		256		253		253		247	
	Temperature (°C)	24.7	24.7	24.9	24.6	25.0	24.6	24.8	24.7	25.1	24.1	24.7	24.4	24.6	24.3
72%	pH (SU)	7.81	7.63	7.79	7.46	7.69	7.32	7.76	7.30	7.61	7.28	7.50	8.05	8.14	7.86
	DO (mg/L)	8.3	7.9	8.0	7.3	7.8	6.8	8.0	7.0	7.6	7.2	7.6	7.2	7.9	7.4
	Conductivity (µmhos/cm)	219		225		217		217		218		218		212	
	Temperature (°C)	24.8	24.6	25.0	24.6	25.1	24.3	24.9	24.7	25.1	24.3	24.7	24.3	24.6	24.3
100%	pH (SU)	7.79	7.61	7.77	7.47	7.66	7.33	7.73	7.29	7.59	7.31	7.47	8.06	8.16	7.89
	DO (mg/L)	8.3	8.0	7.9	7.0	7.6	6.8	7.8	7.0	7.6	7.4	7.6	7.3	7.9	7.5
	Conductivity (µmhos/cm)	184		184		182		177		191		181		183	
	Alkalinity (mg/L $CaCO_3$)	63				62				67					
	Hardness (mg/L $CaCO_3$)	75				85				63					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	25.0	24.7	25.0	24.6	25.1	24.3	24.9	24.6	25.2	24.3	24.7	24.6	24.7	24.2
100% Intake	pH (SU)	7.76	7.61	7.77	7.45	7.64	7.31	7.67	7.31	7.57	7.34	7.44	8.10	8.16	7.86
	DO (mg/L)	8.3	7.8	8.1	7.3	7.5	7.0	7.6	7.0	7.6	7.2	7.6	7.4	7.9	7.3
	Conductivity (µmhos/cm)	182		186		179		177		180		179		183	
	Alkalinity (mg/L $CaCO_3$)	66				61				63					
	Hardness (mg/L $CaCO_3$)	73				73				75					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	25.1	24.7	25.1	24.6	25.0	24.5	24.8	24.7	25.1	24.1	24.8	24.4	24.8	24.4

Species: *Pimephales promelas* **Date:** 11-01-05
Client: TVA / Sequoyah Nuclear Plant – Non-treated

Daily Chemistry:

		Day					
		0 KEK		KEK 1		2	
	Analyst	AAB	AAB	AAB	AAB	AAB	AAB
Concentration	Parameter						
CONTROL MHSW	pH (S.U.)	7.73	7.63	7.79	7.47	7.67	7.48
	DO (mg/L)	7.9	7.8	7.7	7.4	7.5	7.2
	Conductivity (µmhos/cm)	314		315		306	
	Alkalinity (mg $CaCO_3$/L)	59				58	
	Hardness (mg $CaCO_3$/L)	92				87	
	Temperature (°C)	24.6	24.6	24.8	24.8	24.9	24.5
10.98%	pH (S.U.)	7.87	7.60	7.90	7.34	7.80	7.34
	DO (mg/L)	8.0	7.8	8.1	7.0	7.9	7.2
	Conductivity (µmhos/cm)	295		298		288	
	Temperature (°C)	24.6	24.6	24.9	24.7	25.0	24.4
22%	pH (S.U.)	7.86	7.61	7.84	7.43	7.73	7.34
	DO (mg/L)	8.1	7.8	8.1	7.1	7.8	7.1
	Conductivity (µmhos/cm)	284		292		279	
	Temperature (°C)	24.6	24.6	24.9	24.5	25.0	24.4
43.9%	pH (S.U.)	7.83	7.59	7.81	7.44	7.71	7.33
	DO (mg/L)	8.3	7.8	8.0	7.2	7.8	7.0
	Conductivity (µmhos/cm)	256		264		254	
	Temperature (°C)	24.7	24.7	24.9	24.6	25.0	24.6
72%	pH (S.U.)	7.81	7.63	7.79	7.46	7.69	7.32
	DO (mg/L)	8.3	7.9	8.0	7.3	7.8	6.8
	Conductivity (µmhos/cm)	219		225		217	
	Temperature (°C)	24.8	24.6	25.0	24.6	25.1	24.3
100%	pH (S.U.)	7.79	7.61	7.77	7.47	7.66	7.33
	DO (mg/L)	8.3	8.0	7.9	7.0	7.6	6.8
	Conductivity (µmhos/cm)	184		184		182	
	Alkalinity (mg $CaCO_3$/L)	63				62	
	Hardness (mg $CaCO_3$/L)	75				85	
	TR chlorine (mg/L)	<0.10				<0.10	
	Temperature (°C)	25.0	24.7	25.0	24.6	25.1	24.3
100% Intake	pH (S.U.)	7.76	7.61	7.77	7.45	7.64	7.31
	DO (mg/L)	8.3	7.8	8.1	7.3	7.5	7.0
	Conductivity (µmhos/cm)	182		186		179	
	Alkalinity (mg $CaCO_3$/L)	66				61	
	Hardness (mg $CaCO_3$/L)	73				73	
	TR chlorine (mg/L)	<0.10				<0.10	
	Temperature (°C)	25.1	24.7	25.1	24.6	25.0	24.5
		Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas* **Date:** 11-01-05
Client: TVA / Sequoyah Nuclear Plant – Non-treated

		Day							
		3		4 JN		JN 5		6	
	Analyst	AAB	KEN	KEN	KEN	KEN	AAB	AAB	AAB
Concentration	Parameter								
CONTROL	pH (S.U.)	7.53	7.30 (7.0)	7.67	7.31	7.70	8.03	8.10	7.82
	DO (mg/L)	7.4	7.4*	7.7	7.3	7.5	7.4	7.7	7.2
HHSW	Conductivity (µmhos/cm)	310		314		313		310	
	Alkalinity (mg $CaCO_3$/L)	59		58		58		58	
	Hardness (mg $CaCO_3$/L)	97		95		93		85	
	Temperature (°C)	24.7	24.6	24.8	24.2	24.8	24.4	24.7	24.2
10.98%	pH (S.U.)	7.87	7.34	7.64	7.28	7.56	7.98	8.14	7.80
	DO (mg/L)	7.8	6.9	7.6	7.1	WZ.2 (7.4)	7.0	7.8	7.3
	Conductivity (µmhos/cm)	298		292		295		278 (284)	
	Temperature (°C)	24.7	24.6	24.9	24.4	24.8	24.4	24.7	24.2
22%	pH (S.U.)	7.84	7.33	7.64	7.29	7.56	8.02	8.14	7.77
	DO (mg/L)	7.9	6.8	7.7	7.3	7.4	7.1	7.8	7.2
	Conductivity (µmhos/cm)	284		280		283		268 (281)	
	Temperature (°C)	24.7	24.4	24.9	24.4	24.8	24.4	24.8	24.2
43.9%	pH (S.U.)	7.80	7.30	7.62	7.30	7.54	8.04	8.14	7.86
	DO (mg/L)	8.0	6.8	7.7	7.5	7.5	7.2	7.8	7.5
	Conductivity (µmhos/cm)	256		253		253		247	
	Temperature (°C)	24.8	24.7	25.1	24.1	24.7	24.4	24.6	24.3
72%	pH (S.U.)	7.96	7.30	7.61	7.28	7.50	8.05	8.14	7.86
	DO (mg/L)	8.0	7.0	7.6	7.2	7.6	7.2	7.9	7.4
	Conductivity (µmhos/cm)	217		218		218		212	
	Temperature (°C)	24.9	24.7	25.1	24.3	24.7	24.3	24.6	24.3
100%	pH (S.U.)	7.73	7.29	7.59	7.31	7.47	8.06	8.16	7.89
	DO (mg/L)	7.8	7.0	7.6	7.4	7.6	7.3	7.9	7.5
	Conductivity (µmhos/cm)	177		191		181		183	
	Alkalinity (mg $CaCO_3$/L)			66 (7)					
	Hardness (mg $CaCO_3$/L)			63					
	TR Chlorine (mg/L)			<0.10					
	Temperature (°C)	24.9	24.6	25.2	24.3	24.7	24.6	24.7	24.2
100% Intake	pH (S.U.)	7.63	7.31	7.57	7.34	7.44	8.10	8.16	7.86
	DO (mg/L)	17 7.6 ms	7.0	7.6	7.2	7.6	7.4	NR 7.8 (7.9)	7.3
	Conductivity (µmhos/cm)	177		100		179		183	
	Alkalinity (mg $CaCO_3$/L)			63					
	Hardness (mg $CaCO_3$/L)			75					
	TR chlorine (mg/L)			<0.10					
	Temperature (°C)	24.8	24.7	25.1	24.1	24.8	24.4	24.8	24.4
		Initial	Final	Initial	Final	Initial	* Final	* Initial	* Final

* Higher overall pH due to new probe.

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 01-08, 2005
Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1002.0)
Species: *Ceriodaphnia dubia*

Daily Chemical Analyses

Project number: 2178

Reviewed by: Jumner

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.73	7.71	7.79	7.63	7.67	7.66	7.53	7.55	7.87	7.45	7.70	8.07	8.10	7.95
	DO (mg/L)	7.9	7.9	7.7	7.8	7.5	7.4	7.4	7.4	7.7	7.5	7.5	7.6	7.7	7.8
	Conductivity (µmhos/cm)	314		315		306		310		314		313		310	
	Alkalinity (mg/L $CaCO_3$)	59				58		59		58		58		58	
	Hardness (mg/L $CaCO_3$)	92				87		97		95		93		85	
	Temperature (°C)	24.7	24.9	24.6	25.1	24.7	24.9	24.7	25.2	24.9	24.7	24.7	25.3	24.9	24.8
10.98%	pH (SU)	7.87	7.74	7.90	7.62	7.80	7.65	7.87	7.54	7.64	7.49	7.56	8.09	8.14	7.97
	DO (mg/L)	8.0	7.9	8.1	7.5	7.9	7.5	7.8	7.4	7.6	7.6	7.4	7.7	7.8	7.7
	Conductivity (µmhos/cm)	295		298		288		298		292		295		296	
	Temperature (°C)	24.8	24.7	24.4	25.0	24.7	24.9	24.9	25.2	25.1	24.6	24.7	25.3	25.1	24.9
22%	pH (SU)	7.86	7.72	7.84	7.61	7.73	7.63	7.84	7.54	7.64	7.50	7.56	8.10	8.14	7.97
	DO (mg/L)	8.1	8.0	8.1	7.7	7.8	7.5	7.9	7.4	7.7	7.6	7.4	7.7	7.8	7.7
	Conductivity (µmhos/cm)	284		292		279		284		280		283		281	
	Temperature (°C)	24.8	24.7	24.5	25.3	24.8	25.0	24.9	25.2	25.1	24.9	24.7	25.2	25.1	24.9
43.9%	pH (SU)	7.83	7.74	7.81	7.62	7.71	7.61	7.80	7.53	7.62	7.51	7.54	8.11	8.14	7.96
	DO (mg/L)	8.3	8.0	8.0	7.8	7.8	7.5	8.0	7.5	7.7	7.6	7.5	7.6	7.8	7.9
	Conductivity (µmhos/cm)	256		264		254		256		253		253		247	
	Temperature (°C)	24.8	24.7	24.5	24.9	24.9	24.9	24.9	25.2	25.1	24.9	24.8	25.2	25.0	24.7
72%	pH (SU)	7.81	7.70	7.79	7.62	7.69	7.59	7.76	7.50	7.61	7.50	7.50	8.11	8.14	7.95
	DO (mg/L)	8.3	7.8	8.0	7.6	7.8	7.5	8.0	7.5	7.6	7.7	7.6	7.6	7.9	7.9
	Conductivity (µmhos/cm)	219		225		217		217		218		218		212	
	Temperature (°C)	24.8	24.8	24.6	25.0	24.9	24.9	24.9	25.2	25.0	24.9	24.7	25.2	25.1	24.7
100%	pH (SU)	7.79	7.73	7.77	7.60	7.66	7.56	7.73	7.47	7.59	7.48	7.47	8.11	8.16	7.92
	DO (mg/L)	8.3	7.9	7.9	7.7	7.6	7.5	7.8	7.4	7.6	7.4	7.6	7.7	7.9	7.9
	Conductivity (µmhos/cm)	184		184		182		177		191		181		183	
	Alkalinity (mg/L $CaCO_3$)	63				62				67					
	Hardness (mg/L $CaCO_3$)	75				85				63					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	24.8	24.9	24.6	24.9	25.1	24.8	25.0	25.3	25.1	24.7	24.8	25.2	25.2	24.6
100% Intake	pH (SU)	7.76	7.73	7.77	7.57	7.64	7.54	7.67	7.48	7.57	7.50	7.44	8.14	8.16	7.90
	DO (mg/L)	8.3	7.8	8.1	7.6	7.5	7.5	7.6	7.4	7.6	7.6	7.6	7.9	7.9	7.8
	Conductivity (µmhos/cm)	182		186		179		177		180		179		183	
	Alkalinity (mg/L $CaCO_3$)	66				61				63					
	Hardness (mg/L $CaCO_3$)	73				73				75					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	24.8	24.8	24.5	24.9	24.8	24.9	24.9	25.1	24.9	24.8	24.8	25.2	25.0	24.9

Species: *Ceriodaphnia dubia* **Date:** 11-01-05

Client: Sequoyah Nuclear Plant - Non-treated

Daily Chemistry:

		Day					
		0 MEK		MEK 1		2	
	Analyst	AAB	AAB	AAB	AAB	AAB	AAB
Concentration	Parameter						
CONTROL MHSW	pH (S.U.)	7.73	7.71	7.79	7.63	7.67	7.66
	DO (mg/L)	7.9	7.9	7.7	7.8	7.5	7.4
	Conductivity (µmhos/cm)	314		315		306	
	Alkalinity (mg $CaCO_3$/L)	59				58	
	Hardness (mg $CaCO_3$/L)	92				87	
	Temperature (°C)	24.7	24.9	24.6	25.1	24.7	24.9
10.98%	pH (S.U.)	7.87	7.74	7.90	7.62	7.80	7.65
	DO (mg/L)	8.0	7.9	8.1	7.5	7.7	7.5
	Conductivity (µmhos/cm)	295		298		288	
	Temperature (°C)	24.8	24.7	24.4	25.0	24.7	24.9
22%	pH (S.U.)	7.86	7.72	7.84	7.61	7.73	7.63
	DO (mg/L)	8.1	8.0	8.1	7.7	7.8	7.5
	Conductivity (µmhos/cm)	284		292		279	
	Temperature (°C)	24.8	24.7	24.5	25.3	24.8	25.0
43.9%	pH (S.U.)	7.83	7.74	7.81	7.62	7.71	7.61
	DO (mg/L)	8.3	8.0	8.0	7.8	7.8	7.5
	Conductivity (µmhos/cm)	256		274 AM		254	
	Temperature (°C)	24.8	24.7	24.5	24.9	24.9	24.9
72%	pH (S.U.)	7.81	7.70	7.79	7.62	7.69	7.59
	DO (mg/L)	8.3	7.8	8.0	7.6	7.8	7.5
	Conductivity (µmhos/cm)	219		264 AM		217	
	Temperature (°C)	24.8	24.8	24.6	25.0	24.9	24.9
100%	pH (S.U.)	7.79	7.73	7.77	7.60	7.66	7.56
	DO (mg/L)	8.3	7.9	7.9	7.7	7.6	7.5
	Conductivity (µmhos/cm)	184		225 AM		182	
	Alkalinity (mg $CaCO_3$/L)	63				62	
	Hardness (mg $CaCO_3$/L)	75				85	
	TR chlorine (mg/L)	<0.16				<0.10	
	Temperature (°C)	24.8	24.9	24.6	24.9	25.1	24.8
100% Intake	pH (S.U.)	7.76	7.73	7.77	7.57	7.64	7.54
	DO (mg/L)	8.3	7.8	8.1	7.6	7.5	7.5
	Conductivity (µmhos/cm)	182		186		179	
	Alkalinity (mg $CaCO_3$/L)	66				61	
	Hardness (mg $CaCO_3$/L)	73				73	
	TR chlorine (mg/L)	<0.10				<0.10	
	Temperature (°C)	24.8	24.8	24.5	24.9	24.8	24.9
		Initial	Final	Initial	Final	Initial	Final

Environmental Testing Solutions, Inc.

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated **Date:** 11-01-05

		Day							
		3		4 JN		JN 5		6	
	Analyst	AAB	KEN	KEN	KEN	KEN	AAB	AAB	
Concentration	Parameter								
CONTROL	pH (S.U.)	7.53	7.55	7.87	7.45	7.70	8.07	8.10	7.95
	DO (mg/L)	7.4	7.4	7.9	7.5	7.5	7.6	7.7	7.8
MHSW	Conductivity (µmhos/cm)	310		314		313		310	
	Alkalinity (mg $CaCO_3$/L)	59		58		58		58	
	Hardness (mg $CaCO_3$/L)	91 92		95		88 93		85	
	Temperature (°C)	24.7	25.2	24.9	24.7	24.7	25.3	24.9	24.8
10.98%	pH (S.U.)	7.87	7.54	7.64	7.49	7.56	8.09	8.14	7.97
	DO (mg/L)	7.8	7.4	7.6	7.6	7.4	7.7	7.8	7.7
	Conductivity (µmhos/cm)	298		292		295		296 278	
	Temperature (°C)	24.9	25.2	25.1	24.6	24.7	25.3	25.1	24.9
22%	pH (S.U.)	7.84	7.54	7.64	7.50	7.56	8.10	8.14	7.97
	DO (mg/L)	7.9	7.4	7.7	7.6	7.4	7.7	7.8	7.7
	Conductivity (µmhos/cm)	284		280		283		266 281	
	Temperature (°C)	24.9	25.2	25.1	24.9	24.7	25.2	25.1	24.9
43.9%	pH (S.U.)	7.80	7.53	7.62	7.51	7.54	8.11	8.14	7.96
	DO (mg/L)	8.0	7.5	7.7	7.6	7.5	7.6	7.8	7.9
	Conductivity (µmhos/cm)	256		253		253		247	
	Temperature (°C)	24.9	25.2	25.1	24.9	24.8	25.2	25.0	24.7
72%	pH (S.U.)	7.76	7.50	7.61	7.50	7.50	8.11	8.14	7.95
	DO (mg/L)	8.0	7.5	7.6	7.7	7.6	7.6	7.9	7.9
	Conductivity (µmhos/cm)	217		218		218		212	
	Temperature (°C)	24.9	25.2	25.0	24.9	24.7	25.2	25.1	24.7
100%	pH (S.U.)	7.73	7.47	7.59	7.48	7.47	8.11	8.16	7.92
	DO (mg/L)	7.8	7.4	7.6	7.4	7.6	7.7	7.9	7.9
	Conductivity (µmhos/cm)	177		191		181		183	
	Alkalinity (mg $CaCO_3$/L)			68 67					
	Hardness (mg $CaCO_3$/L)			63					
	TR Chlorine (mg/L)			<0.10					
	Temperature (°C)	25.0	25.3	25.1	24.7	24.8	25.2	25.2	24.6
100% Intake	pH (S.U.)	7.67	7.48	7.57	7.50	7.44	8.14	8.16	7.90
	DO (mg/L)	7.6	7.4	7.6	7.6	7.6	7.9	7.9	7.8
	Conductivity (µmhos/cm)	177		180		179		183	
	Alkalinity (mg $CaCO_3$/L)			63					
	Hardness (mg $CaCO_3$/L)			75					
	TR chlorine (mg/L)			<0.10					
	Temperature (°C)	24.9	25.1	24.9	24.8	24.8	* 25.2	25.0	24.9
		Initial	Final	Initial	Final	Initial	* Final	* Initial	* Final

* Higher overall pH due to new probe.

TVA / Sequoyah Nuclear Plant, Outfall 101
UV-treated
November 01-08, 2005

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

Daily Chemical Analyses

Project number: 2178

Reviewed by:

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.71	7.64	7.74	7.47	7.61	7.38	7.61	7.31	7.59	7.31	7.57	8.03	8.11	7.79
	DO (mg/L)	8.3	8.0	8.1	7.6	7.6	6.9	7.2	6.8	7.6	7.3	7.7	7.6	7.8	7.3
	Conductivity (µmhos/cm)	300		312		298		298		298		302		290	
	Temperature (°C)	24.7	24.5	24.9	24.8	25.0	24.5	24.9	24.6	24.8	24.1	24.8	24.5	25.0	24.3
10.98%	pH (SU)	7.77	7.67	7.75	7.50	7.61	7.35	7.59	7.30	7.50	7.31	7.57	8.04	8.13	7.78
	DO (mg/L)	8.3	7.9	8.3	7.5	7.5	6.9	7.3	6.9	7.7	7.2	7.5	7.5	7.7	7.2
	Conductivity (µmhos/cm)	297		307		291		295		294		297		287	
	Temperature (°C)	24.9	24.5	25.0	24.8	25.1	24.4	25.1	24.6	24.8	24.5	24.9	24.4	24.9	24.2
22%	pH (SU)	7.75	7.63	7.77	7.51	7.61	7.37	7.57	7.30	7.50	7.31	7.57	7.99	8.12	7.77
	DO (mg/L)	8.3	7.7	8.3	7.6	7.7	6.9	7.4	7.0	7.7	7.3	7.6	7.5	7.7	7.2
	Conductivity (µmhos/cm)	282		292		279		291		282		286		275	
	Temperature (°C)	24.9	24.6	25.1	24.5	25.1	24.7	25.1	24.5	24.8	24.6	24.9	24.6	24.9	24.4
43.9%	pH (SU)	7.75	7.62	7.78	7.52	7.60	7.36	7.55	7.31	7.49	7.30	7.55	8.04	8.13	7.80
	DO (mg/L)	8.3	7.8	8.3	7.6	7.9	6.9	7.4	7.0	7.7	7.1	7.7	7.3	7.7	7.2
	Conductivity (µmhos/cm)	259		261		252		260		254		257		252	
	Temperature (°C)	24.9	24.6	25.1	24.6	25.2	24.7	25.2	24.5	24.9	24.3	24.9	24.3	24.8	24.1
72%	pH (SU)	7.73	7.68	7.78	7.52	7.59	7.41	7.53	7.29	7.49	7.38	7.53	8.06	8.14	7.85
	DO (mg/L)	8.2	7.7	8.1	7.8	8.0	7.0	7.4	7.0	7.7	7.4	7.5	7.3	7.8	7.3
	Conductivity (µmhos/cm)	218		226		216		222		221		222		220	
	Temperature (°C)	25.1	24.6	25.2	24.6	25.2	24.6	25.2	24.5	25.1	24.3	25.0	24.5	24.8	24.2
100%	pH (SU)	7.72	7.65	7.78	7.52	7.59	7.38	7.50	7.27	7.48	7.33	7.51	8.09	8.14	7.85
	DO (mg/L)	8.3	7.8	8.1	7.7	7.9	7.1	7.5	7.0	7.9	7.2	7.6	7.3	7.8	7.2
	Conductivity (µmhos/cm)	186		193		183		181		185		182		185	
	Temperature (°C)	25.3	24.6	25.3	24.6	25.2	24.6	25.2	24.6	25.1	24.3	24.9	24.5	24.9	24.1
100% Intake	pH (SU)	7.71	7.64	7.79	7.52	7.56	7.34	7.43	7.27	7.48	7.34	7.50	8.06	8.18	7.83
	DO (mg/L)	8.3	7.7	8.1	7.4	7.8	7.0	7.6	7.1	7.8	7.2	7.6	7.4	8.0	7.0
	Conductivity (µmhos/cm)	191		198		178		173		182		180		183	
	Temperature (°C)	25.2	24.6	25.1	24.6	25.1	24.6	25.1	24.5	25.0	24.2	25.1	24.5	24.9	24.1

Species: *Pimephales promelas* Date: 11-01-05
Client: TVA / Sequoyah Nuclear Plant – UV-treated

Daily Chemistry:

		Day					
		0 KEY	KEY 1			2	
	Analyst	AAB	AAB	AAB	AAB	AAB	AAB
Concentration	Parameter						
CONTROL	pH (S.U.)	7.71	7.64	7.74	7.47	7.61	7.38
	DO (mg/L)	8.3	8.0	8.1	~~8.0~~ 7.6	7.6	6.9
	Conductivity (µmhos/cm)	~~314~~ 300		312		~~298~~ 298	298
	Temperature (°C)	24.7	24.5	24.9	24.8	25.0	24.5
10.98%	pH (S.U.)	7.77	7.67	7.75	7.50	7.61	7.35
	DO (mg/L)	8.3	7.9	8.3	7.5	7.5	6.9
	Conductivity (µmhos/cm)	297		307		291	
	Temperature (°C)	24.9	24.5	25.0	24.8	25.1	24.4
22%	pH (S.U.)	7.75	7.63	7.77	7.51	7.61	7.37
	DO (mg/L)	8.3	7.7	8.3	7.6	7.7	6.9
	Conductivity (µmhos/cm)	282		292		279	
	Temperature (°C)	24.9	24.6	25.1	24.5	25.1	24.7
43.9%	pH (S.U.)	7.75	7.62	7.78	7.52	7.60	7.36
	DO (mg/L)	8.3	7.8	8.3	7.6	7.9	6.9
	Conductivity (µmhos/cm)	259		261		252	
	Temperature (°C)	24.9	24.6	25.1	24.6	25.2	24.7
72%	pH (S.U.)	7.73	7.68	7.78	7.52	7.59	7.41
	DO (mg/L)	8.2	7.7	8.1	7.8	8.0	7.0
	Conductivity (µmhos/cm)	218		226		216	
	Temperature (°C)	25.1	24.6	25.2	24.6	25.2	24.6
100%	pH (S.U.)	7.72	7.65	7.78	7.52	7.59	7.38
	DO (mg/L)	8.3	7.8	8.1	7.7	7.9	7.1
	Conductivity (µmhos/cm)	186		193		183	
	Temperature (°C)	25.3	24.6	25.3	24.6	25.2	24.6
100% Intake	pH (S.U.)	7.71	7.64	7.79	7.52	7.56	7.34
	DO (mg/L)	8.3	7.7	8.1	7.4	7.8	7.0
	Conductivity (µmhos/cm)	191		198		178	
	Temperature (°C)	25.2	24.6	25.1	24.6	25.1	24.6
		Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas* **Date:** 11-01-05
Client: TVA / Sequoyah Nuclear Plant – UV-treated

		Day							
		3		4 JN		JN 5		6	
	Analyst	AAB	KEK	KEK	KEK	KEK	AAB	AAB	AAB
Concentration	Parameter								
CONTROL	pH (S.U.)	7.61	7.31	7.59	7.31	7.57	8.03	8.11	7.79
	DO (mg/L)	7.2	6.8	7.6	7.3	7.7	7.6	7.8	7.3
	Conductivity (µmhos/cm)	298		298		302		290	
	Temperature (°C)	24.9	24.6	24.8	24.1	24.8	24.5	25.0	24.3
10.98%	pH (S.U.)	7.59	7.30	7.50	7.31	7.57	8.04	8.13	7.78
	DO (mg/L)	7.3	6.9	7.7	7.2	7.5	7.5	7.7	7.2
	Conductivity (µmhos/cm)	295		294		297		287	
	Temperature (°C)	25.1	24.6	24.8	24.5	24.9	24.4	24.9	24.2
22%	pH (S.U.)	7.57	7.30	7.50	7.31	7.57	7.99	8.12	7.77
	DO (mg/L)	7.4	7.0	7.7	7.3	7.6	7.5	7.7	7.2
	Conductivity (µmhos/cm)	291		282		286		275	
	Temperature (°C)	25.1	24.5	24.8	24.6	24.9	24.6	24.9	24.4
43.9%	pH (S.U.)	7.55	7.31	7.49	7.30	7.55	8.04	8.13	7.80
	DO (mg/L)	7.4	7.0	7.7	7.1	7.7	7.3	7.7	7.2
	Conductivity (µmhos/cm)	260		254		257		252	
	Temperature (°C)	25.2	24.5	24.9	24.3	24.9	24.3	24.8	24.1
72%	pH (S.U.)	7.53	7.29	7.49	7.30	7.53	8.06	8.14	7.85
	DO (mg/L)	7.4	7.0	7.7	7.4	7.5	7.3	7.8	7.3
	Conductivity (µmhos/cm)	222		221		222		220	
	Temperature (°C)	25.2	24.5	25.1	24.3	25.0	24.5	24.8	24.2
100%	pH (S.U.)	7.50	7.27	7.48	7.33	7.51	8.09	8.14	7.85
	DO (mg/L)	7.5	7.0	7.9	7.2	7.6	7.3	7.8	7.2
	Conductivity (µmhos/cm)	181		185		182		185	
	Temperature (°C)	25.2	24.6	25.1	24.3	24.9	24.5	24.9	24.1
100% Intake	pH (S.U.)	7.43	7.27	7.48	7.34	7.50	8.06	8.18	7.83
	DO (mg/L)	7.6	7.1	7.8	7.2	7.6	7.4	8.0	7.0
	Conductivity (µmhos/cm)	173		182		180		183	
	Temperature (°C)	25.1	24.5	25.0	24.2	25.1	24.5	24.9	24.1
		Initial	Final	Initial	Final	Initial	*Final	*Initial	* Final

* Higher overall pH due to new probe.

Environmental Testing Solutions, Inc.

-49.8%

Total Residual Chlorine
(EPA Method 330.5)

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

Analyst	AAB
Date analyzed	11-01-05

Iodide reagent:	IN2178 (1646)
Acid reagent:	IN2168

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	Ins5325	Ins5325

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%)
Ins5325	0.50	0.513	102.6%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
051031.02	SQN-INT-TOR	No color, clear	S <0.0212	
↓	Duplicate		D <0.0139	-

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.0292
051031.01	SQN-101-TOR-#2	No color, clear	<0.00542
			AAB

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%)
Ins5325	0.50	0.510	102%

Reviewed by	KG
Date reviewed	11-01-05

Total Residual Chlorine
(EPA Method 330.5)

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

Analyst	AAB	Iodide reagent:	INR178 (7/7/20)
Date analyzed	11-03-05	Acid reagent:	INR168

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INSS325	INSS325

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%)
INSS325	0.50	0.458	91.6%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
051103.02	pcsphosphate 100	NO color, clear	S <0.00824	
↓	Duplicate		D <0.00936	-

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.0851
051103.01	pcsphosphate 107	light yellow, clear	<0.00526
051102.12	SAN 101 - ~~FOX#1~~ 8	light beige, clear	<0.00193
051102.13	SAN INT ~~FOX~~ 8	light beige, clear	<0.00076
			AAB

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%)
INSS325	0.50	0.519	103.8%

Reviewed by	
Date reviewed	11-03-05

Environmental Testing Solutions, Inc.

Total Residual Chlorine
(EPA Method 330.5)

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

Analyst	MER
Date analyzed	11-05-05

Iodide reagent:	INR178
Acid reagent:	INR116B

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INSS325	INSS325

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%)
INSS325	0.50	0.503	100.67.

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
051104.01	TVA		S <0.0941	
↓	SAN - 101 Duplicate	no color clear	D <0.0742	—

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00941
051104.02	TVA-SAN-INT	no color, clear	<0.00650
051105.01	Monroe WWTP	pale yellow, clear	<0.00422
051105.01	AAF McQuay	yellow, clear, particles	<0.00130
051105.02	Belews Creek SS	no color. Clear	<0.00214
051105.03	Marshall SS	no color. clear	0.0302
051105.04	Scarlett Acres MHP	Grey, cloudy, blk particles	<0.00899
051105.06	North Cary WWTP	tan, clear	<0.0221
051105.07	Schlage Lock	no color, clear	<0.0176
			↓

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%)
INSS325	0.50	0.492	98.47.

Reviewed by	MER
Date reviewed	11-10-05

Environmental Testing Solutions, Inc.

Alkalinity
(EPA Method 310.1)

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

Time started: 1006
Time ended: 1022

Analyst	KEW
Date analyzed	10.30.05

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
4.69	IN R184	IN R157	0.0	12.2	12.2	~~0.025~~	10.2

BK correction 0.0 - 0.0 - 0.0m1 → 0.0205

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%)
IN K183	100	100	12.2	21.8	9.6	10.2	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%)
10.26.05	GSW1420	100	21.8	24.9	3.1	10.2	S 32	
↓	Duplicate	↓	24.9	28.0	3.1	↓	D 32	-

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)
IN R183	50	100	24.9	32.8	7.9	10.2	81

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%)
32	49	98.

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
10.24.05A	MHS H_2O	100	32.8	38.7	5.9	10.2	60
10.24.05B	↓	↓	38.7	44.5	5.8	↓	59
051025.01	TVA JDF-001		6.8	16.2	9.4		96
051025.02	↓ INT	↓	16.5	22.8	6.3		64
25705	B.R. Park 10.24.05	25	22.9	26.5	3.6	(4) ↓	150
			KEW				

Reviewed by: KEW Date reviewed: 10.30.05

Environmental Testing Solutions, Inc.

Alkalinity
(EPA Method 310.1)

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

Time started: 1310
Time ended: 1408

Analyst	HEA
Date analyzed	11.02.05

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
4.35	INR184	INR157	0.0	12.0	12.0	0.0208	10.4

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%)
INR103	100	100	12.0	21.2	9.2	10.4	96	96%

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%)
10.31.05B	MHS H_2O	100	21.2	26.9	5.7	10.4	S 59	
↓	Duplicate	↓	26.9	32.6	5.7	↓	D 59	-

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)
INR103	50	100	26.9	37.4	10.5	10.4	110

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%)
59	51	102.02%

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
10.31.05A	MHS H_2O	100	37.4	43.0	5.6	10.4	58
11.03.05A	MHS H_2O	↓	43.0	48.6	5.6	↓	58
11.03.05B	MHS H_2O		0.0	5.6	5.6		58
11.05.05A	MHS H_2O		5.6	11.2	5.6		58
11.05.05B	MHS H_2O		11.2	16.9	5.7		59
051031.01	WA-SQN 101 1		17.4	23.5	6.1		63
051102.12	↓ 2		23.5	29.5	6.0		62
051104.01	↓ 3		29.6	36.0	6.4		66 (67)
051031.02	WA SQN 101 WT1	↓	36.0	42.3	6.3	↓	66

Reviewed by: [signature]

Date reviewed: 11-13-05

Environmental Testing Solutions, Inc.

Total Hardness
(EPA Method 130.2)

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

Time started: 0943
Time ended: 0955

Analyst	KLK
Date analyzed	10.30.05

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
INR175	INR123	0.0	9.8	9.8	0.0204	20.4

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%)
INSS309	40	50	9.8	11.9	2.1	20.4	43	107.5%

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%)
10.26.05	ESW	50	11.9	13.9	2.0	20.4	S 41	
↓	Duplicate	↓	13.9	16.0	2.1	↓	D 43	4.8%

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)
INSS309	40	50	13.9	17.9	4.0	20.4	82

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%)
43	39	97.5%

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	20.4	ND
10.24.05A	MHS H_2O		17.9	22.7	4.8		98
10.24.05B	↓		22.7	27.2	4.5		92
051025.01	TVA JOF - 001		27.2	34.2	7.0		143 = 140
051025.02	↓ INT	↓	34.2	37.9	3.7	↓	76

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

Reviewed by: KLK

Date reviewed: 10-30-05

Alkalinity
(EPA Method 310.1)

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

Analyst	MH
Date analyzed	11.01.05

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%)
INRIB3	100	100	0.0	9.4	9.4	10.4	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%)
051104.02	TVA SQN INT 3	100	9.4	15.5	6.1	10.4	S 63	
↓	Duplicate	↓	15.5	21.7	6.2	↓	D 64	1.67%

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)
INRIB3	50	100	15.5	26.5	11.0	10.4	110

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%)
63	46	92%

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
051102.13	TVA SQN INT 2	100	26.5	32.4	5.9	10.4	61
051101.01	TVA WBN 101 1		32.4	39.7	7.3		76
051103.03	2		39.7	46.5	6.8		71
051105.08	3		0.0	7.5	7.5		78
051101.02	TVA WBN 101 INT 1		7.5	14.2	6.7		70
051103.04	2		14.2	21.3	7.1		74
051105.09	3		21.3	28.3	7.0		73
051101.03	TVA WBN 113 1		28.3	35.5	7.2		75
051103.04 05	2		35.5	42.8	7.3		76

Reviewed by: [signature]

Date reviewed: 11-13-05

Alkalinity
(EPA Method 310.1)

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

Analyst	KEN
Date analyzed	11.06.05

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%)
INR103	100	100	1.5	10.8	9.3	10.4	97	97

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%)
051101.04	TVA WBN INT13 1	100	11.0	17.7	6.7	10.4	S 70	
↓	Duplicate	↓	17.8	24.6	6.8	↓	D 71	1.4%

Matrix spike recovery:

Reference standard number	Spike value (SV) (mg $CaCO_3$/L)	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Spike alkalinity (A) (mg $CaCO_3$/L)
INR103	50	100	17.8	29.5	11.7	10.4	120

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	49	98%

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
051105.10	TVA WBN 113 3	100	29.5	37.0	7.5	10.4	~~79~~ (78)
051103.06	TVA WBN 113 INT 2	↓	37.0	43.8	6.8	↓	71
~~051103.11~~	↓ 3	↓	39.4	46.3	6.9	↓	72
051105.11							

Reviewed by: [signature] Date reviewed: 11-13-05

Environmental Testing Solutions, Inc.

Total Hardness
(EPA Method 130.2)

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

Analyst: KER
Date analyzed: 11.06.05

Time started: 1104
Time ended: 1155

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
INR175	INR123	0.0	9.9	9.9	0.0202	20.2

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%)
INSS309	40	50	9.9	12.0	2.1	20.2	42	105.0%

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%)
10.31.04	MHS H_2O	50	12.0	16.3	4.3	20.2	S 87	
↓	Duplicate	↓	16.3	20.6	4.3	↓	D 87	—

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)
INSS309	40	30	20.~~0~~ 16.3	~~0~~ 22.7	6.4	20.2	130

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%)
87	43	107.5%

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	20.2	ND
10.31.05B	MHS H_2O		22.7	27.5	4.8		97
11.03.05A			27.5	32.2	4.7		95
11-03-05B			32.2	36.8	4.6		93
11.05.05A			36.8	41.0	4.2		86
11.05.05B	TVA		41.0	45.1	4.1		83
051031.01	SQN 1		0.0	3.7	3.7		75
051102.12	2		3.7	7.9	4.2		85
051104.01	3		7.9	11.0	3.1		63
051031.02	~~TVA SQN INT 1~~ TVA SQN INT 1		11.1	14.7	3.6		73

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

Reviewed by: [signature]

Date reviewed: 11-13-05

Environmental Testing Solutions, Inc.

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

Analyst	KEK
Date analyzed	11.06.05

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%)
INSS309	40	50	14.8	16.8	2.0	20.2	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%)
051102.13	TJA SQN INT 2	50	16.8	20.4	3.6	20.2	S 73	
↓	Duplicate	↓	20.4	24.0	3.6	↓	D 73	—

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)
INSS309	40	50	24.0	29.7	5.7	20.2	120

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%)
75	45	112.5%

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)						
051104.02	TJA SQN INT 3	50	24.0	27.7	3.7	20.2	75
051101.01	TJA WBN 101 1		34.2	38.6	4.4		89
051103.03	↓ 2		29.7	34.2	4.5		91
051108~051105.08	↓ 3		38.6	43.3	4.7		95
051101.02	TJA WBN 101 INT 1		43.3	47.5	4.2		85
051103.04	↓ 2		0.0	4.2	4.2		85
051105.09	↓ 3		4.2	8.8	4.6		93
051101.03 051101~	TJA WBN 113 1		8.8	13.5	4.7		95
051103.05	↓ 2	↓	13.5	18.0	4.5	↓	91

Note: If >15ml of titrant is used, sample must be diluted. Reviewed by: [signature]

Date reviewed: 11-13-05

Environmental Testing Solutions, Inc.

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

Analyst	KCK
Date analyzed	11.06-05

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%)
INSS309	40	50	18.1	20.1	2.0	20.2	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%)
051105.10	WBN 113 3	50	20.1	24.6	4.5	20.2	S 91	
↓	Duplicate	↓	24.6	29.1	4.5	↓	D 91	—

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)
INSS309	40	60	24.6	31.1	6.5	20.2	130

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%)
91	39	97.5%

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)						K
051101.04	TVA WBN 113 INT 1	50	31.1	35.1	4.0	20.2K	20.2 82K 81
051103.06	2	↓	35.1	39.3	4.2	↓	85
051105.11	3	↓	39.3	43.5	4.2	↓	85

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

Reviewed by: [signature]

Date reviewed: 11-13-05

Sequoyah Nuclear Plant Biomonitoring
November 1-8, 2005

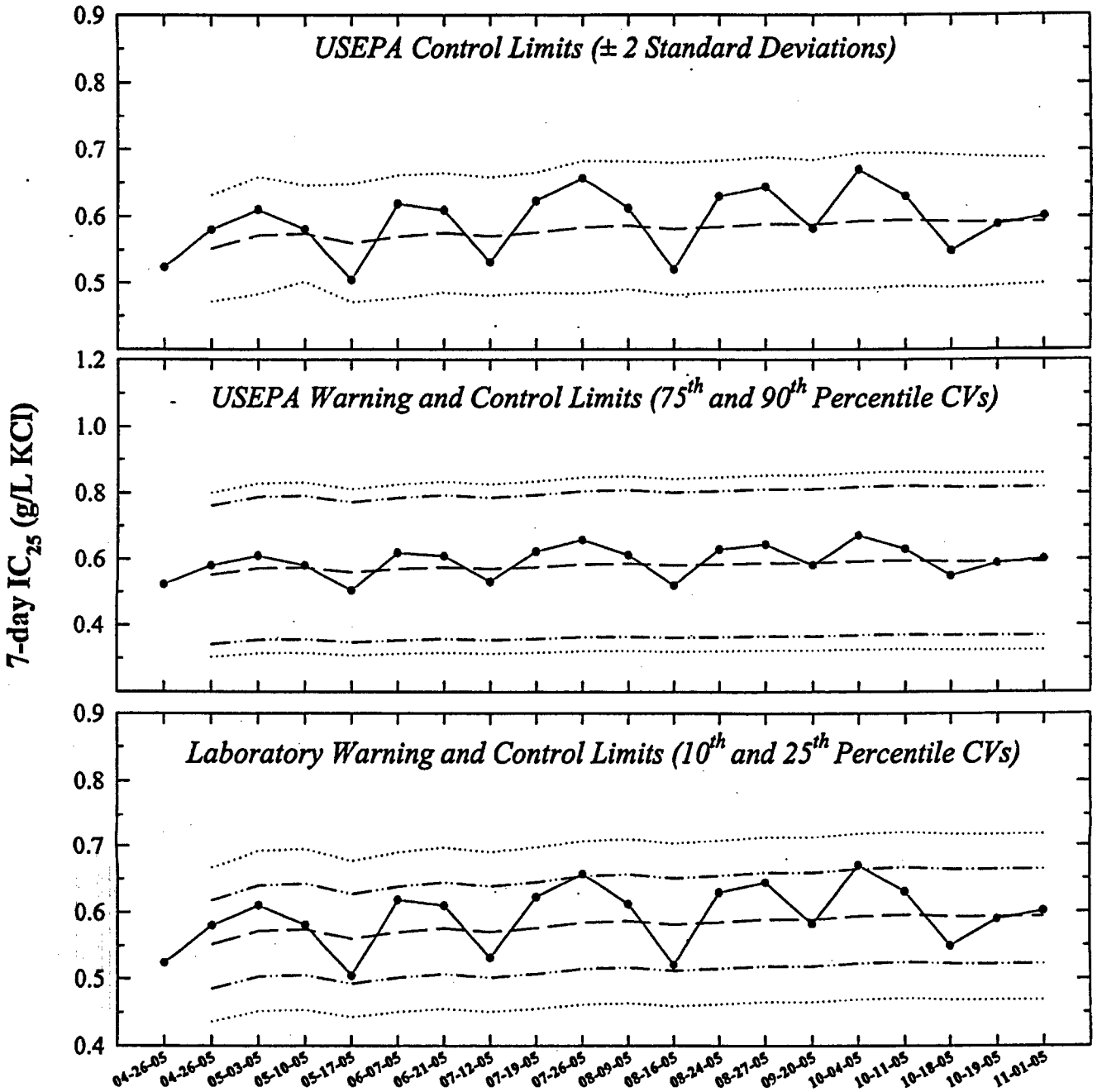
Appendix D

Reference Toxicant Test and
Control Chart

Environmental Testing Solutions, Inc.

Pimephales promelas

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



Test date

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

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

—··—·· **Warning Limits** (mean $IC_{25} \pm S_{A.10}$ or $S_{A.75}$)

····· **Control Limits** (mean $IC_{25} \pm S_{A.25}$, $S_{A.90}$, or 2 Standard Deviations)

Environmental Testing Solutions, Inc.

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

Test number	Test date	7-day IC_{25} (g/L KCl)	CT (g/L KCl)	S	State and USEPA Control Limits		$S_{A.10}$	Laboratory Warning Limits		$S_{A.25}$	Laboratory Control Limits		$S_{A.75}$	USEPA Warning Limits		$S_{A.90}$	USEPA Control Limits		CV
					CT - 2S	CT + 2S		$CT - S_{A.10}$	$CT + S_{A.10}$		$CT - S_{A.25}$	$CT + S_{A.25}$		$CT - S_{A.75}$	$CT + S_{A.75}$		$CT - S_{A.90}$	$CT + S_{A.90}$	
1	04-26-05	0.52																	
2	04-26-05	0.58	0.55	0.04	0.47	0.63	0.07	0.49	0.62	0.12	0.44	0.67	0.21	0.34	0.76	0.25	0.30	0.80	0.07
3	05-03-05	0.61	0.57	0.04	0.48	0.66	0.07	0.50	0.64	0.12	0.45	0.69	0.22	0.35	0.79	0.26	0.31	0.83	0.08
4	05-10-05	0.58	0.57	0.04	0.50	0.65	0.07	0.50	0.64	0.12	0.45	0.69	0.22	0.36	0.79	0.26	0.32	0.83	0.06
5	05-17-05	0.50	0.56	0.04	0.47	0.65	0.07	0.49	0.63	0.12	0.44	0.68	0.21	0.35	0.77	0.25	0.31	0.81	0.08
6	06-07-05	0.62	0.57	0.05	0.48	0.66	0.07	0.50	0.64	0.12	0.45	0.69	0.22	0.35	0.79	0.26	0.31	0.83	0.08
7	06-21-05	0.61	0.58	0.04	0.49	0.66	0.07	0.51	0.64	0.12	0.45	0.70	0.22	0.36	0.79	0.26	0.32	0.83	0.08
8	07-12-05	0.53	0.57	0.04	0.48	0.66	0.07	0.50	0.64	0.12	0.45	0.69	0.22	0.35	0.79	0.26	0.31	0.83	0.08
9	07-19-05	0.62	0.58	0.05	0.49	0.67	0.07	0.51	0.64	0.12	0.45	0.70	0.22	0.36	0.79	0.26	0.32	0.83	0.08
10	07-26-05	0.66	0.58	0.05	0.48	0.68	0.07	0.51	0.65	0.12	0.46	0.71	0.22	0.36	0.81	0.26	0.32	0.85	0.09
11	08-09-05	0.61	0.59	0.05	0.49	0.68	0.07	0.52	0.66	0.12	0.46	0.71	0.22	0.36	0.81	0.26	0.32	0.85	0.08
12	08-16-05	0.52	0.58	0.05	0.48	0.68	0.07	0.51	0.65	0.12	0.46	0.70	0.22	0.36	0.80	0.26	0.32	0.84	0.09
13	08-24-05	0.63	0.58	0.05	0.49	0.68	0.07	0.51	0.65	0.12	0.46	0.71	0.22	0.36	0.81	0.26	0.32	0.85	0.08
14	08-27-05	0.64	0.59	0.05	0.49	0.69	0.07	0.52	0.66	0.12	0.47	0.71	0.22	0.37	0.81	0.26	0.32	0.85	0.08
15	09-20-05	0.58	0.59	0.05	0.49	0.68	0.07	0.52	0.66	0.12	0.46	0.71	0.22	0.36	0.81	0.26	0.32	0.85	0.08
16	10-04-05	0.67	0.59	0.05	0.49	0.70	0.07	0.52	0.66	0.12	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.86	0.09
17	10-11-05	0.63	0.60	0.05	0.50	0.70	0.07	0.52	0.67	0.13	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.86	0.08
18	10-18-05	0.55	0.59	0.05	0.49	0.69	0.07	0.52	0.66	0.12	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.86	0.08
19	10-19-05	0.59	0.59	0.05	0.50	0.69	0.07	0.52	0.66	0.12	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.86	0.08
20	11-01-05	0.60	0.59	0.05	0.50	0.69	0.07	0.52	0.66	0.12	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.86	0.08

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

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

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

Laboratory Control and Warning Limits

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

$S_{A.10}$ = Standard deviation corresponding to the 10th percentile CV. ($S_{A.10}$ = 0.12)

$S_{A.25}$ = Standard deviation corresponding to the 25th percentile CV. ($S_{A.25}$ = 0.21)

USEPA Control and Warning Limits

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

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

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

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

Organisms obtained from Aquatic BioSystems, Inc.

Environmental Testing Solutions, Inc.

Precision of Endpoint Measurements

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

Test number	Test date	Control Survival (%)	Control Mean Growth (mg/larvae)	CT for Control Growth (mg/larvae)	CV (%)	CT for Control Growth CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	04-26-05	100	0.742		3.9		0.09	12.3	
2	04-26-05	97.5	0.702	0.722	7.2	5.6	0.10	13.5	12.9
3	05-03-05	100	0.760	0.734	2.5	4.5	0.10	12.9	12.9
4	05-10-05	100	0.772	0.744	10.7	6.1	0.10	13.5	13.1
5	05-17-05	100	0.858	0.767	6.0	6.1	0.06	7.4	11.9
6	06-07-05	100	0.848	0.780	3.3	5.6	0.09	10.1	11.6
7	06-21-05	97.5	0.622	0.758	7.2	5.8	0.09	14.5	12.0
8	07-12-05	100	0.765	0.759	5.6	5.8	0.10	13.4	12.2
9	07-19-05	100	0.596	0.740	2.9	5.5	0.06	9.7	11.9
10	07-26-05	100	0.807	0.747	11.5	6.1	0.11	13.2	12.0
11	08-09-05	100	0.622	0.736	5.5	6.0	0.11	17.0	12.5
12	08-16-05	100	0.599	0.724	12.7	6.6	0.11	18.9	13.0
13	08-24-05	100	0.802	0.730	14.9	7.2	0.14	17.4	13.4
14	08-27-05	100	0.678	0.727	4.0	7.0	0.11	16.5	13.6
15	09-20-05	100	0.814	0.732	8.4	7.1	0.11	13.3	13.6
16	10-04-05	100	0.785	0.736	4.5	6.9	0.06	7.3	13.2
17	10-11-05	100	0.555	0.725	4.1	6.8	0.11	19.4	13.5
18	10-18-05	100	0.741	0.726	4.9	6.7	0.04	5.9	13.1
19	10-19-05	100	0.556	0.717	5.9	6.6	0.08	14.6	13.2
20	11-01-05	100	0.694	0.716	1.2	6.3	0.09	12.4	13.2

Note:

CV = Coefficient of variation for control growth.
On average, the CV for control growth is 6.3% in Environmental Testing Solutions, Inc. *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, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 13.2% 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 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.

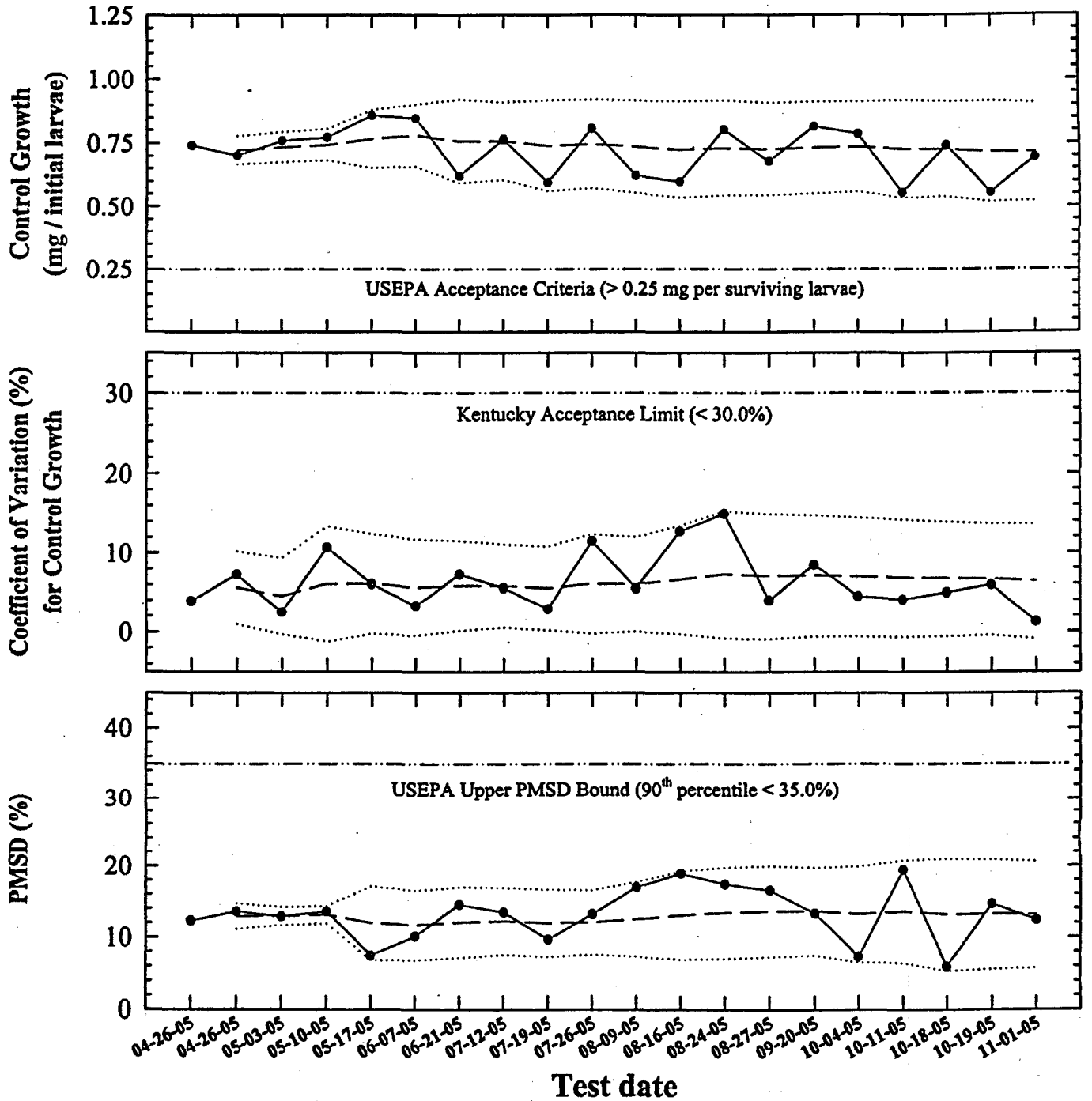
USEPA. 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination Program. EPA-833-R-00-003. US Environmental Protection Agency, Cincinnati, OH.

Organisms obtained from Aquatic BioSystems, Inc.

11-01-05

Environmental Testing Solutions, Inc.

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



Control Reproduction, Coefficient of Variation (CV), or Percent Minimum Significant Difference (PMSD) PMSD is the minimum significant difference between the control and treatment that can be declared statistically significant.

Central Tendency (mean Control Growth, CV, or PMSD)

Control Limits (mean Control Growth, CV, or PMSD ± 2 Standard Deviations)

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

PpKClCR Test Number: 81

Dilution preparation information:						*Comments:*
KCl CHM number:		CHM 188				
Stock preparation:		50 g KCl/L: Dissolve 50 g KCl in 1-L Deionized water				
Dilution prep (mg/L)	300	450	600	750	900	
Stock volume (mL)	6	9	12	15	18	
Diluent volume (mL)	994	991	988	985	982	
Total volume (mL)	1000	1000	1000	1000	1000	

Test organism information:		*Test information:*	
Organism age:	21.75 TO 23.25 HOURS OLD	Randomizing template:	GREEN
Date and times organisms were born between:	10-31-05 1330 TO 1500	Incubator number and shelf location:	3D
Organism source:	ABS BATCH Pp 10-31-05	*Artemia* lot number:	BG-1804T
Transfer bowl information:	pH = 7.73 SU Temperature = 24.0 °C	Total drying time:	24-HOURS
		Date / Time in: 11-08-05	1235
Average transfer volume:	10.4 mL	Date / Time out: 11-09-05	1241
		Oven temperature:	61°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	MHS batch used	Analyst
0	11-01-05	——	1500	1238	10-24-05 B	dl
1	11-02-05	0913	1520	1300	10-24-05 B	dl
2	11-03-05	0915	1526	1244	10-31-05 A	dl
3	11-04-05	0856	1503	1200	10-31-05 B	dl
4	11-05-05	0845	1450	1207	11-03-05 A	dl
5	11-06-05	0842	1448	1211	11-03-05 B	dl
6	11-07-05	0845	1452	1200	11-03-05 A	dl
7	11-08-05			1143		dl

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	0%	≤ 20%	7-day LC_{50}	727.7
Average weight per initial larvae:	0.694		NOEC	450
Average weight per surviving larvae:	0.694	≥ 0.25 mg/larvae	LOEC	600
			ChV	519.6
			IC_{25}	601.3

Species: ***Pimephales promelas*** PpKClCR Test Number: 81

Survival and Growth Data

Day	Control				300 mg KCl/L				450 mg KCl/L			
	A	B	C	D	E	F	G	H	I	J	K	L
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	10	10	10	10	10	10	10	9 1d
7	10	10	10	10	1SM 10	10	10	10	10	10	10	9
A = Pan weight (mg) Tray color code:: Gold Analyst: KEXUAN	15.97	14.65	15.89	14.56	15.99	14.48	14.60	16.12	14.61	14.78	14.56	16.10
B = Pan + Larvae weight (mg) Analyst: Juman	22.91	21.55	22.95	21.43	21.69	21.59	21.73	22.44	21.82	21.88	20.92	22.26
Larvae weight (mg) = A – B = C	6.94	6.90	7.06	6.87	5.70	7.11	7.13	6.32	7.21	7.10	6.36	6.16
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.694	0.690	0.706	0.687	0.570	0.711	0.713	0.632	0.721	0.710	0.636	0.616
Average weight per initial number of larvae (mg)	0.694				0.657				0.671			
Percent reduction from control (%)					5.47%				3.47%			

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Species: ***Pimephales promelas*** PpKClCR Test Number: 81

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	10	10	9^{1d}	9^{1d}	7^{3d}	7^{3d}	6^{4d}	8^{2d}	4^{6d}	2^{8d}	2^{8d}	4^{6d}
2	10	10	9	9	6^{1d}	6^{1d}	6	7^{1d}	3^{1d}	2	1^{1d}	3^{1d}
3	10	10	9	9	6	6	6	7	3	1^{1d}	1	3
4	10	10	8^{1d}	8^{1d}	6	6	6	7	~~1~~3	1	1	2^{1d}
5	10	10	8	8	5^{1d}	6	6	7	2^{1d}	1	1	1^{1d}
6	10	10	8	8	3^{2d}	5^{1d}	5^{1d}	7	2	1	1	0^{1d}
7	ISM 10	9^{1d}	8	7^{1d}	3	5 LG	5 LG	7	2	1^{LG}	1	0
A = Pan weight (mg) Tray color code:: Gold Analyst: Keeleran	14.46	15.99	16.24	14.40	15.04	15.90	14.56	14.99	14.42	14.78	14.95	~~14.70~~ —
B = Pan + Larvae weight (mg) Analyst: Jensen	20.01	21.57	21.68	18.71	17.12	19.86	18.54	19.78	15.66	15.89	15.42	—
Larvae weight (mg) = A – B = C	5.55	5.58	5.44	4.31	2.08	3.96	3.98	4.79	1.24	1.11	0.47	—
Weight per initial number of larvae (mg) = C / initial number of larvae	0.555	0.558	0.544	0.431	0.208	0.396	0.398	0.479	0.124	0.111	0.047	0
Average weight per initial number of larvae (mg)	0.522				0.370				0.071			
Percent reduction from control (%)	24.8%				46.7%				89.8%			

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: JL

Comments:

Environmental Testing Solutions, Inc.

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

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

Test number: PpKCKCR # 121 (#81 at 351 Depot St.)
Test dates: November 01-08, 2005

Reviewed 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 / Surviving number of larvae (mg)	Mean weight/ Surviving number of larvae (mg)	Coefficient of variation (Mean weight per surviving number of larvae) (%)	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight/ Initial number of larvae (mg)	Coefficient of variation (%)	Percent reduction from control (%)
Control	A	10	10	15.97	22.91	6.94	0.694	0.694	1.2	0.694	100.0	0.694	1.2	Not applicable
	B	10	10	14.65	21.55	6.90	0.690			0.690				
	C	10	10	15.89	22.95	7.06	0.706			0.706				
	D	10	10	14.56	21.43	6.87	0.687			0.687				
300	E	10	10	15.99	21.69	5.70	0.570	0.657	10.5	0.570	100.0	0.657	10.5	5.4
	F	10	10	14.48	21.59	7.11	0.711			0.711				
	G	10	10	14.60	21.73	7.13	0.713			0.713				
	H	10	10	16.12	22.44	6.32	0.632			0.632				
450	I	10	10	14.61	21.82	7.21	0.721	0.688	5.5	0.721	97.5	0.671	7.8	3.4
	J	10	10	14.78	21.88	7.10	0.710			0.710				
	K	10	10	14.56	20.92	6.36	0.636			0.636				
	L	10	9	16.10	22.26	6.16	0.684			0.616				
600	M	10	10	14.46	20.01	5.55	0.555	0.618	8.3	0.555	85.0	0.522	11.7	24.8
	N	10	9	15.99	21.57	5.58	0.620			0.558				
	O	10	8	16.24	21.68	5.44	0.680			0.544				
	P	10	7	14.40	18.71	4.31	0.616			0.431				
750	Q	10	3	15.04	17.12	2.08	0.693	0.741	8.2	0.208	50.0	0.370	31.0	46.7
	R	10	5	15.90	19.86	3.96	0.792			0.396				
	S	10	5	14.56	18.54	3.98	0.796			0.398				
	T	10	7	14.99	19.78	4.79	0.684			0.479				
900	U	10	2	14.42	15.66	1.24	0.620	0.733	45.6	0.124	10.0	0.071	92.0	89.8
	V	10	1	14.78	15.89	1.11	1.110			0.111				
	W	10	1	14.95	15.42	0.47	0.470			0.047				
	X	10	0	0.00	0.00	0.00	0.000			0.000				

Dunnett's MSD value: 0.0860
PMSD: 12.4

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, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 13.2% from the control (determined through reference toxicant testing).
Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.
Upper PMSD bound determined by USEPA (90th percentile) = 35%.
The lower and upper bounds were calculated by the USEPA using 205 tests conducted from 19 laboratories for Pimephales growth in chronic reference toxicant tests.

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

Organisms obtained from Aquatic BioSystems, Inc.

Environmental Testing Solutions, Inc.

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/1/2005	Test ID:	PpKCICR	Sample ID:	REF-Ref Toxicant
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	KCL-Potassium chloride
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas

Comments:

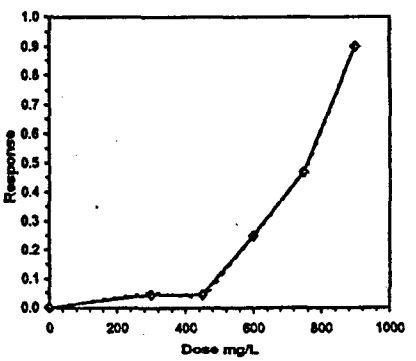
Conc-mg/L	1	2	3	4
D-Control	0.6940	0.6900	0.7060	0.6870
300	0.5700	0.7110	0.7130	0.6320
450	0.7210	0.7100	0.6360	0.6160
600	0.5550	0.5580	0.5440	0.4310
750	0.2080	0.3960	0.3980	0.4790
900	0.1240	0.1110	0.0470	0.0000

			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	0.6943	1.0000	0.6943	0.6870	0.7060	1.202	4				0.6943	1.0000
300	0.6565	0.9456	0.6565	0.5700	0.7130	10.496	4	1.005	2.290	0.0860	0.6636	0.9559
450	0.6708	0.9662	0.6708	0.6160	0.7210	7.828	4	0.626	2.290	0.0860	0.6636	0.9559
*600	0.5220	0.7519	0.5220	0.4310	0.5580	11.679	4	4.585	2.290	0.0860	0.5220	0.7519
750	0.3703	0.5333	0.3703	0.2080	0.4790	31.025	4				0.3703	0.5333
900	0.0705	0.1015	0.0705	0.0000	0.1240	81.996	4				0.0705	0.1015

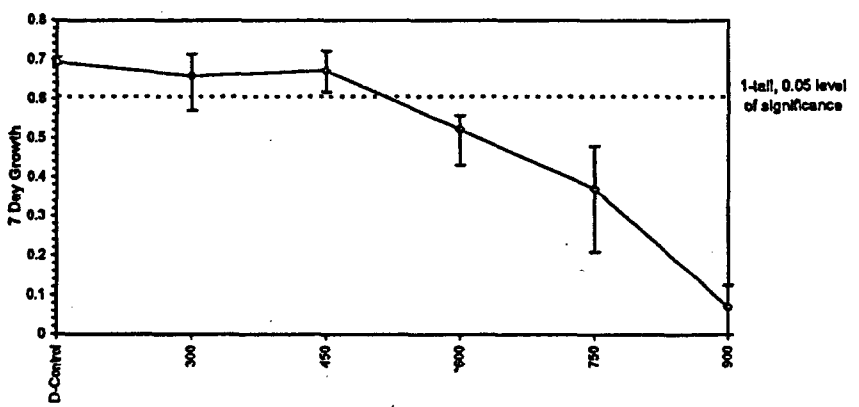
Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.918339193		0.844		-0.70822808	-0.46093538
Bartlett's Test indicates equal variances (p = 0.05)					7.722052574		11.34486675			
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	450	600	519.6152423		0.065033244	0.12392257	0.024022417	0.002822875	0.002669202	3, 12
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

Point	mg/L	SD	95% CL(Exp)		Skew
IC05	454.33	115.00	0.00	492.73	-0.4417
IC10	491.09	34.09	423.33	531.92	-4.8418
IC15	527.86	18.07	466.90	575.11	-0.2885
IC20	564.62	22.88	500.05	623.42	-0.1223
IC25	601.30	23.32	519.54	660.95	-0.3808
IC40	704.23	33.91	608.03	791.05	-0.1302
IC50	761.57	26.97	649.82	802.50	-0.8499



Dose-Response Plot



Organisms obtained from Aquatic BioSystems, Inc.

11-01-05

Species: ***Pimephales promelas*** PpKClCR Test Number: 81

Daily Chemistry:

		Day					
		0		KEX 1		2	
	Analyst	AAB	AAB	AAB	AAB	AAB	AAB
Concentration	Parameter						
CONTROL MHSW	pH (S.U.)	7.73	7.73	7.79	7.60	7.67	7.44
	DO (mg/L)	7.9	7.7	7.7	7.2	~~AB 7.6~~ (7.5)	7.0
	Conductivity (µmhos/cm)	314		315		306	
	Alkalinity (mg $CaCO_3$/L)	59				58	
	Hardness (mg $CaCO_3$/L)	92		&		87	
	Temperature (°C)	24.6	24.5	24.9	24.7	(24.5) ~~24.7~~	24.3
300 mg KCl/L	pH (S.U.)	7.77	7.68	7.83	7.55	7.74	7.41
	DO (mg/L)	8.0	7.9	7.8	7.2	~~AB 7.9~~ (7.5)	6.9
	Conductivity (µmhos/cm)	836		838		862	
	Temperature (°C)	24.5	24.5	25.0	24.6	25.1	24.2
450 mg KCl/L	pH (S.U.)	7.79	7.69	7.81	7.53	7.76	7.39
	DO (mg/L)	8.0	7.9	8.0	7.1	~~AB 8.0~~ (7.6)	7.0
	Conductivity (µmhos/cm)	1100		1130		1080	
	Temperature (°C)	24.7	24.5	25.0	24.8	25.1	24.2
600 mg KCl/L	pH (S.U.)	7.79	7.68	7.82	7.51	7.76	7.41
	DO (mg/L)	8.1	8.0	8.0	7.1	~~AB 8.0~~ (7.6)	7.1
	Conductivity (µmhos/cm)	1370		1370		1350	
	Temperature (°C)	24.6	24.7	25.1	24.8	25.1	24.5
750 mg KCl/L	pH (S.U.)	7.79	7.67	7.81	7.57	7.76	7.42
	DO (mg/L)	8.0	8.1	8.0	7.2	~~AB 8.0~~ (7.6)	7.1
	Conductivity (µmhos/cm)	(1690) 1560 &		1620		1630	
	Temperature (°C)	24.6	24.6	25.1	24.7	25.0	24.6
900 mg KCl/L	pH (S.U.)	7.80	7.90	7.86	7.54	7.77	7.45
	DO (mg/L)	8.1	8.1	8.1	7.2	~~AB 8.0~~ (7.6)	7.2
	Conductivity (µmhos/cm)	(1950) 1800 &		1920		1890	
	Temperature (°C)	24.6	24.6	25.1	24.7	25.1	24.4
STOCK	Conductivity (µmhos/cm)	75200		71500		73,300	
		Initial	Final	Initial	Final	Initial	Final

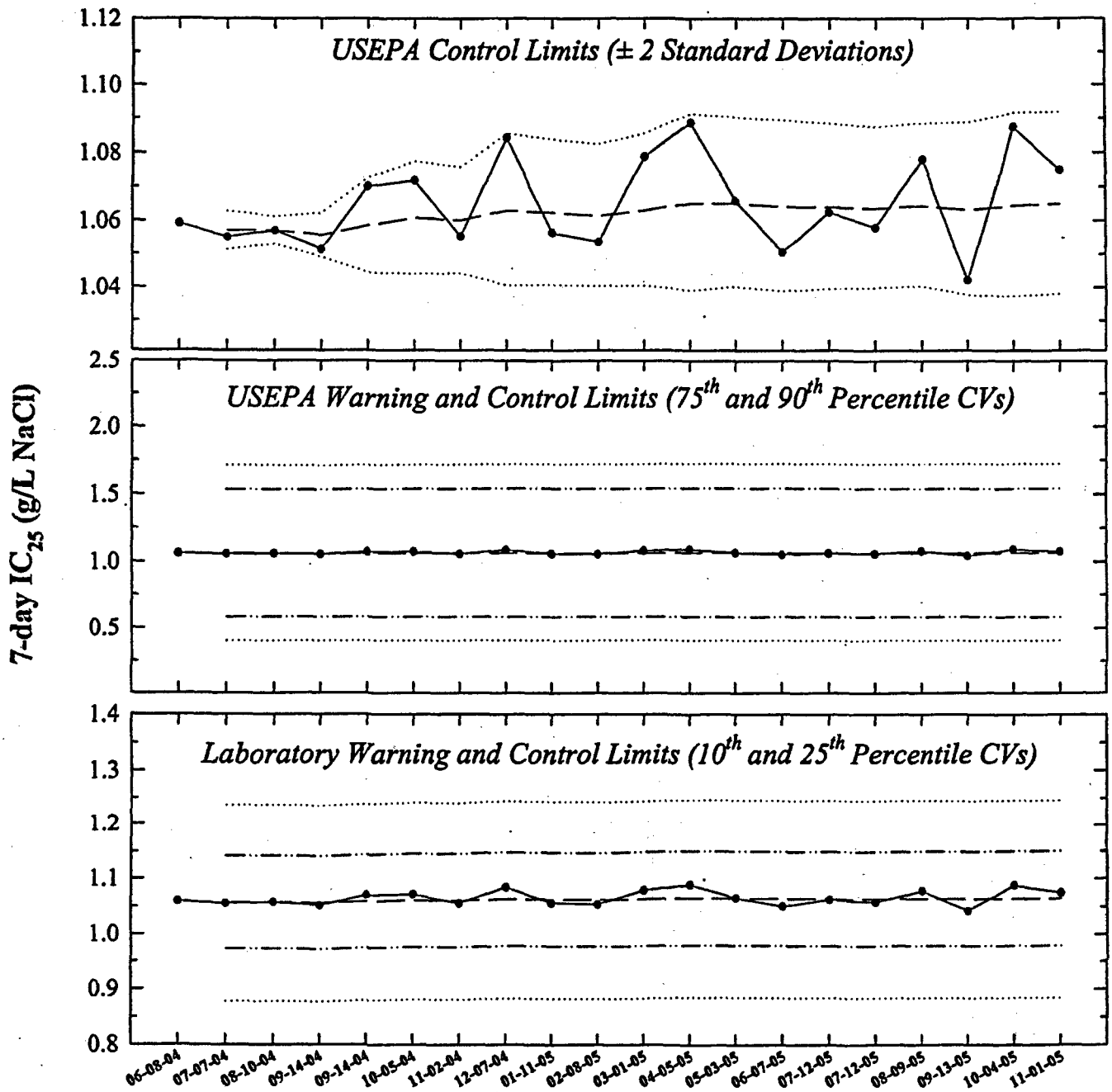
Species: *Pimephales promelas* PpKClCR Test Number: 81

		Day							
		3		4 JN		JN 5		6	
	Analyst	AAB	KEK	KEK	KEK	KEK	AAB	AAB	AAB
Concentration	Parameter								
CONTROL MHSW	pH (S.U.)	7.53	7.59	7.87	7.29	7.70	~~8.~~ (8.03)	8.10	7.78
	DO (mg/L)	7.4	7.6	7.7	7.4	7.5	7.3	7.7	7.2
	Conductivity (µmhos/cm)	310		314		313		310	
	Alkalinity (mg $CaCO_3$/L)	59		58		58		58	
	Hardness (mg $CaCO_3$/L)	97		95		93		85	
	Temperature (°C)	24.7	24.5	24.8	24.2 ~~24.8~~	24.7	24.5	24.9	24.5
300 mg KCl/L	pH (S.U.)	7.59	7.60	7.85	7.33	7.70	7.98	8.11	7.80
	DO (mg/L)	7.4	7.0	7.8	7.1	7.7	7.5	7.9	7.2
	Conductivity (µmhos/cm)	842		827		837		823	
	Temperature (°C)	24.9	24.5	24.9	24.3	24.6	24.5	24.9	24.3
450 mg KCl/L	pH (S.U.)	7.60	7.58	7.85	7.40	7.70	7.96	8.10	7.78
	DO (mg/L)	7.5	6.9	7.8	7.3	7.8	7.5	7.8	7.2
	Conductivity (µmhos/cm)	1140		1140		1120		1140	
	Temperature (°C)	24.9	24.3	24.7	24.3	24.6	24.4	24.9	24.3
600 mg KCl/L	pH (S.U.)	7.61	7.58	7.85	7.40	7.71	7.96	8.10	7.77
	DO (mg/L)	7.6	6.9	7.8	7.3	7.9	7.2	7.8	7.2
	Conductivity (µmhos/cm)	1350		1380		1380		1380	
	Temperature (°C)	24.8	24.3	24.7	24.3	24.6	24.4	24.8	24.3
750 mg KCl/L	pH (S.U.)	7.61	7.58	7.85	7.39	7.71	7.99	8.09	7.82
	DO (mg/L)	7.6	7.0	7.8	7.2	7.7	7.3	7.8	7.3
	Conductivity (µmhos/cm)	1620		1650		1660		1630	
	Temperature (°C)	24.8	24.4	24.7	24.1	24.6	24.3	24.8	24.3
900 mg KCl/L	pH (S.U.)	7.62	7.62	7.85	7.43	7.72	7.99	8.09	7.81
	DO (mg/L)	7.6	7.1	7.6	7.5	7.8	7.4	7.8	7.3
	Conductivity (µmhos/cm)	1890		1930		1910		1930	
	Temperature (°C)	24.8	24.3	24.7	24.1	24.7	24.6	24.9	24.2
STOCK	Conductivity (µmhos/cm)	73,300		70800		75200		76,300	
		Initial	Final	Initial	Final	Initial	*Final	*Initial	*Final

* Higher overall pH due to new probe.

Environmental Testing Solutions, Inc.

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



Test date

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

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

—·—·— **Warning Limits** (mean $IC_{25} \pm S_{A.10}$ or $S_{A.75}$)

····· **Control Limits** (mean $IC_{25} \pm S_{A.25}$, $S_{A.90}$, or 2 Standard Deviations)

Environmental Testing Solutions, Inc.

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

Test number	Test date	7-day IC_{25} (g/L NaCl)	CT (g/L NaCl)	S	State and USEPA Control Limits CT - 2S	CT + 2S	$S_{A.10}$	Laboratory Warning Limits CT - $S_{A.10}$	CT + $S_{A.10}$	$S_{A.25}$	Laboratory Control Limits CT - $S_{A.25}$	CT + $S_{A.25}$	$S_{A.75}$	USEPA Warning Limits CT - $S_{A.75}$	CT + $S_{A.75}$	$S_{A.90}$	USEPA Control Limits CT - $S_{A.90}$	CT + $S_{A.90}$	CV
1	06-08-04	1.06																	
2	07-07-04	1.06	1.06	0.00	1.05	1.06	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.66	0.40	1.71	0.00
3	08-10-04	1.06	1.06	0.00	1.05	1.06	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.66	0.40	1.71	0.00
4	09-14-04	1.05	1.06	0.00	1.05	1.06	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.65	0.40	1.71	0.00
5	09-14-04	1.07	1.06	0.01	1.04	1.07	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.66	0.40	1.71	0.01
6	10-05-04	1.07	1.06	0.01	1.04	1.08	0.08	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
7	11-02-04	1.06	1.06	0.01	1.04	1.08	0.08	0.98	1.14	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
8	12-07-04	1.08	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
9	01-11-05	1.06	1.06	0.01	1.04	1.08	0.08	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
10	02-08-05	1.05	1.06	0.01	1.04	1.08	0.08	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
11	03-01-05	1.08	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
12	04-05-05	1.09	1.07	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.73	0.01
13	05-03-05	1.07	1.07	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.73	0.01
14	06-07-05	1.05	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.72	0.01
15	07-12-05	1.06	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.59	1.54	0.66	0.40	1.72	0.01
16	07-12-05	1.06	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.59	1.54	0.66	0.40	1.72	0.01
17	08-09-05	1.08	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.72	0.01
18	09-13-05	1.04	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
19	10-04-05	1.09	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.72	0.01
20	11-01-05	1.08	1.07	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.73	0.01

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

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

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

Laboratory Control and Warning Limits

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

$S_{A.10}$ = Standard deviation corresponding to the 10th percentile CV. ($S_{A.10}$ = 0.08)

$S_{A.25}$ = Standard deviation corresponding to the 25th percentile CV. ($S_{A.25}$ = 0.17)

USEPA Control and Warning Limits

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

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

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

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

Environmental Testing Solutions, Inc.

Precision of Endpoint Measurements

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

Test number	Test date	Control Survival (%)	Control Mean Reproduction (offspring/female)	CT for Control Mean Reproduction (offspring/female)	CV (%)	CT for Control Reproduction CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	06-08-04	100	32.9		6.8		3.0	9.0	
2	07-07-04	100	33.3	33.1	6.3	6.6	2.6	7.8	8.4
3	08-10-04	100	27.4	31.2	5.8	6.3	2.3	8.6	8.4
4	09-14-04	100	28.7	30.6	4.9	6.0	2.4	8.2	8.4
5	09-14-04	100	28.8	30.2	7.5	6.3	3.0	10.5	8.8
6	10-05-04	100	29.6	30.1	8.2	6.6	2.8	9.6	8.9
7	11-02-04	100	30.5	30.2	8.5	6.8	2.7	8.8	8.9
8	12-07-04	100	31.8	30.4	4.9	6.6	2.5	7.9	8.8
9	01-11-05	100	31.0	30.4	9.4	6.9	3.1	10.0	8.9
10	02-08-05	100	30.0	30.4	6.8	6.9	2.4	8.0	8.8
11	03-01-05	100	29.9	30.4	6.6	6.9	2.9	9.9	8.9
12	04-05-05	100	32.4	30.5	6.9	6.9	3.1	9.6	9.0
13	05-03-05	100	29.9	30.5	7.0	6.9	2.9	9.8	9.1
14	06-07-05	100	30.4	30.5	5.0	6.7	2.6	8.5	9.0
15	07-12-05	100	30.4	30.5	7.5	6.8	2.7	8.8	9.0
16	07-12-05	100	31.1	30.5	7.2	6.8	3.2	10.2	9.1
17	08-09-05	100	28.3	30.4	7.3	6.8	2.9	10.3	9.2
18	09-13-05	100	27.9	30.2	7.3	6.9	3.9	13.9	9.4
19	10-04-05	100	27.0	30.1	5.8	6.8	3.0	11.1	9.5
20	11-01-05	100	28.4	30.0	10.3	7.0	3.8	13.2	9.7

Note:

CV = Coefficient of variation for control reproduction.
On average, the CV for control reproduction is 7.0% in Environmental Testing Solutions, Inc. *Ceriodaphnia* chronic toxicity tests.
Lower CV bound determined by USEPA (10th percentile) = 8.9%.
Upper CV bound determined by USEPA (90th percentile) = 42%

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference
PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, Inc. chronic toxicity tests when a toxicant reduces *Ceriodaphnia* reproduction by 9.7% from the control.
Lower PMSD bound determined by USEPA (10th percentile) = 11%.
Upper PMSD bound determined by USEPA (90th percentile) = 37%.

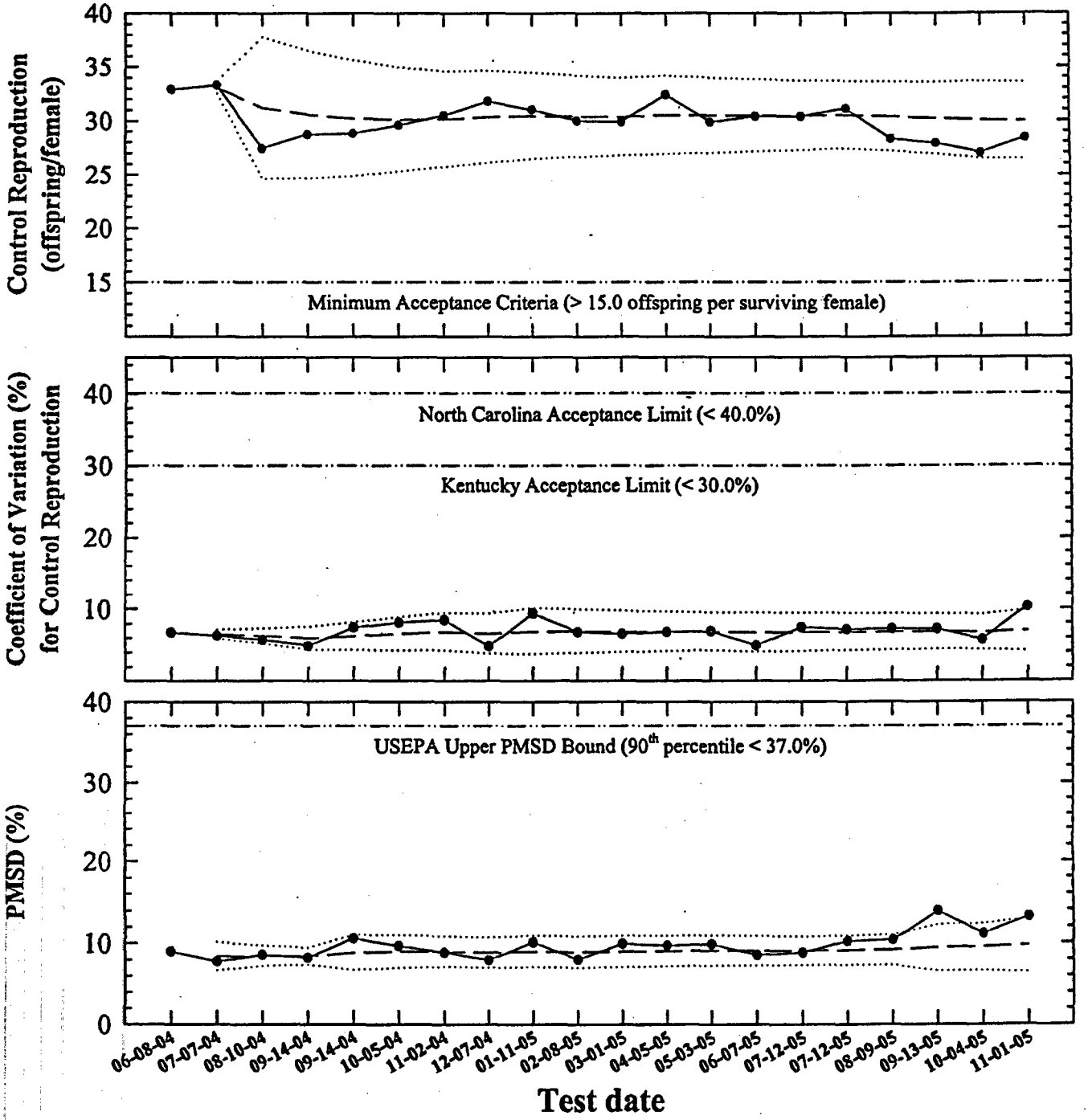
CT = Central Tendancy (Mean Control Reproduction, CV, or PMSD)

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

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

Environmental Testing Solutions, Inc.

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



—●— **Control Reproduction, Coefficient of Variation (CV), or Percent Minimum Significant Difference (PMSD)** PMSD is the minimum significant difference between the control and treatment that can be declared statistically significant.

— — · **Central Tendency** (mean Control Reproduction, CV, or PMSD)

····· **Control Limits** (mean Control Reproduction, CV, or PMSD ± 2 Standard Deviations)

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

CdNaCLCR #: 42

Dilution preparation information:						*Comments:*
NaCl CHM number:		CHM 120				
Stock preparation:		100 g NaCl/l (dissolve 50 g NaCl in 500 ml deionized water)				
Dilution prep (mg/L)	600	800	1000	1200	1400	
Stock volume (mL)	9	12	15	18	21	
Diluent volume (mL)	1491	1488	1485	1482	1479	
Total volume (mL)	1500	1500	1500	1500	1500	

Test organism information:		*Test information:*	
Organism age:	< 24-hours old	Randomizing template:	Yellow
Date and times organisms were born between:	11-01-05 0731 TO 1000	Incubator number and shelf location:	~~2B~~ 3E1
Organism source:	10-25-05 A-F	YCT batch:	09-30-05
Transfer bowl information:	pH = 7.91 SU Temperature = 24.8°C	*Selenastrum* batch:	10-16-05

Daily renewal information:

Day	Date	Test initiation, renewal, or termination time	MHS water batch used	Analyst
0	11-01-05	1101	10-24-05 B	dl
1	11-02-05	1006	10-24-05 B	dl
2	11-03-05	1004	10-31-05 A	dl
3	11-04-05	1008	10-31-05 B	dl
4	11-05-05	1010	11-03-05 A ~~10-31-05 B~~	dl
5	11-06-05	1016	11-03-05 B	dl
6	11-07-05	1003	11-05-05 A	dl
7	11-08-05	1057	[redacted]	dl

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	≤ 20%	7-day LC50	> 1400
% Adults having 3rd Broods:	100%	≥ 80%	NOEC	1000
% Mortality:	0%	≤ 20%	LOEC	1200
Mean Offspring/Female:	28.4	≥ 15.0 offspring/female	ChV	1095.4
% CV:	10.3%	< 40.0 %	IC25	1075.0

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

CONTROL

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	4	4	3	4	3	4	3	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	12	10	12	10	*2	0	11	0	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	13	0	0	0	0	12	11	0	11	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	14	18	12	14	12	12	11	14	13	13
Total young produced		31	34	26	30	25	30	25	29	27	27
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L
X for 3rd Broods		X	X	X	X	X	X	X	X	X	X

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	28.4

600 mg NaCl/L

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	5	4	4	4	3	4	4	3	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	11	*1	11	11	10	10	10	10	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	13	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	12	13	15	12	14	17	12	15	15	16
Total young produced		28	29	33	27	29	30	26	29	28	33
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead) *SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	29.2
% Reduction from Control:	-2.8%

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

800 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	5	4	4	3	3	4	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	14	10	11	10	12	9	10	0	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	1*	0	0	0	0	10	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	13	11	13	11	16	14	14	12	14	12
Total young produced		29	30	27	27	29	29	27	26	28	27
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	27.9
% Reduction from Control:	1.8%

1000 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	3	3	4	3	4	4	4	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	*1	13	0	12	0	9	9	0	10	9
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	10	0	10	0	11	0	0	9	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	17	14	13	14	14	11	10	10	13	12
Total young produced		32	31	26	29	29	23	23	23	27	24
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	26.7
% Reduction from Control:	6.0%

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

1200 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	3	1	2	5	2	2	1	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	5	5	8	6	5	8	4	0	8	6
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	4	0	0	0	0	0	3	0	0
	Adult mortality	L	L	L	~~L~~	L	L	L	L	L	L
7	Young produced	13	11	0	12	0	0	0	0	6	0
Total young produced		22	24	11	19	7	13	6	5	15	9
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	13.1
% Reduction from Control:	53.9%

1400 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	0	2	0	0	3	3	0	0	1
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	2	3	7	3	4	4	5	9	4	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	7
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	1	5	5	9	3	0	6	0	5	0
Total young produced		7	8	14	12	7	7	14	9	9	8
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	9.5
% Reduction from Control:	66.5%

Environmental Testing Solutions, Inc.

Verification of *Ceriodaphnia* Reproduction Totals

Control

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	4	4	3	4	3	4	3	4	37
5	0	12	10	12	10	2	0	11	0	10	67
6	13	0	0	0	0	12	11	0	11	0	47
7	14	18	12	14	12	12	11	14	13	13	133
Total	31	34	26	30	25	30	25	29	27	27	284

1000 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	3	3	4	3	4	4	4	3	36
5	1	13	0	12	0	9	9	0	10	9	63
6	10	0	10	0	11	0	0	9	0	0	40
7	17	14	13	14	14	11	10	10	13	12	128
Total	32	31	26	29	29	23	23	23	27	24	267

600 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	5	4	4	4	3	4	4	3	5	40
5	12	11	1	11	11	10	10	10	10	12	98
6	0	0	13	0	0	0	0	0	0	0	13
7	12	13	15	12	14	17	12	15	15	16	141
Total	28	29	33	27	29	30	26	29	28	33	292

1200 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	3	1	2	5	2	2	1	3	27
5	5	5	8	6	5	8	4	0	8	6	55
6	0	4	0	0	0	0	0	3	0	0	7
7	13	11	0	12	0	0	0	0	6	0	42
Total	22	24	11	19	7	13	6	5	15	9	131

800 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	5	4	4	3	3	4	4	4	4	39
5	12	14	10	11	10	12	9	10	0	11	99
6	0	0	0	1	0	0	0	0	10	0	11
7	13	11	13	11	16	14	14	12	14	12	130
Total	29	30	27	27	29	29	27	26	28	27	279

1400 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	0	2	0	0	3	3	0	0	1	13
5	2	3	7	3	4	4	5	9	4	0	41
6	0	0	0	0	0	0	0	0	0	7	7
7	1	5	5	9	3	0	6	0	5	0	34
Total	7	8	14	12	7	7	14	9	9	8	95

Environmental Testing Solutions, Inc.

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

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

Test number: CdNaClCR #71 (#42 at 351 Depot St.)

Test dates: November 01-08, 2005

Reveiwed by:

Concentration (mg/L NaCl)	Replicate number										Survival (%)	Average reproduction (offspring/female)	Coefficient of variation (%)	Percent reduction from control (%)
	1	2	3	4	5	6	7	8	9	10				
Control	31	34	26	30	25	30	25	29	27	27	100	28.4	10.3	Not applicable
600	28	29	33	27	29	30	26	29	28	33	100	29.2	7.9	-2.8
800	29	30	27	27	29	29	27	26	28	27	100	27.9	4.6	1.8
1000	32	31	26	29	29	23	23	23	27	24	100	26.7	12.9	6.0
1200	22	24	11	19	7	13	6	5	15	9	100	13.1	51.6	53.9
1400	7	8	14	12	7	7	14	9	9	8	100	9.5	29.5	66.5

Dunnett's MSD value: 3.751

PMSD: 13.2

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, Inc. chronic toxicity tests when a toxicant reduces *Ceriodaphnia* reproduction by 9.7% from the control.

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

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

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

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

Environmental Testing Solutions, Inc.
Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	11/1/2005	Test ID:	CdNaClCR	Sample ID:	REF-Ref Toxicant
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	NACL-Sodium chloride
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia
Comments:					

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	31.000	34.000	26.000	30.000	25.000	30.000	25.000	29.000	27.000	27.000
600	28.000	29.000	33.000	27.000	29.000	30.000	26.000	29.000	28.000	33.000
800	29.000	30.000	27.000	27.000	29.000	29.000	27.000	26.000	28.000	27.000
1000	32.000	31.000	26.000	29.000	29.000	23.000	23.000	23.000	27.000	24.000
1200	22.000	24.000	11.000	19.000	7.000	13.000	6.000	5.000	15.000	9.000
1400	7.000	8.000	14.000	12.000	7.000	7.000	14.000	9.000	9.000	8.000

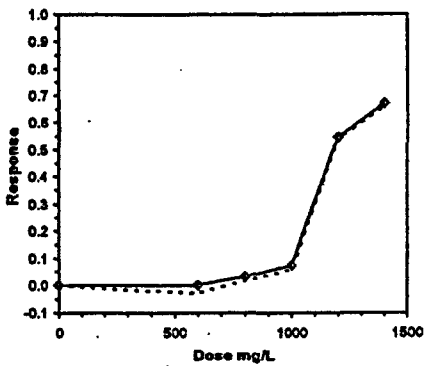
			Transform: Untransformed					Rank	1-Tailed	Isotonic	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	Sum	Critical	Mean	N-Mean
D-Control	28.400	1.0000	28.400	25.000	34.000	10.259	10			28.800	1.0000
600	29.200	1.0282	29.200	26.000	33.000	7.876	10	113.00	75.00	28.800	1.0000
800	27.900	0.9824	27.900	26.000	30.000	4.612	10	102.00	75.00	27.900	0.9688
1000	26.700	0.9401	26.700	23.000	32.000	12.860	10	90.00	75.00	26.700	0.9271
*1200	13.100	0.4613	13.100	5.000	24.000	51.579	10	55.00	75.00	13.100	0.4549
*1400	9.500	0.3345	9.500	7.000	14.000	29.461	10	55.00	75.00	9.500	0.3299

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.74299985	1.035	0.57349241	1.15846555
Bartlett's Test indicates unequal variances (p = 1.19E-04)	25.3599415	15.0862722		

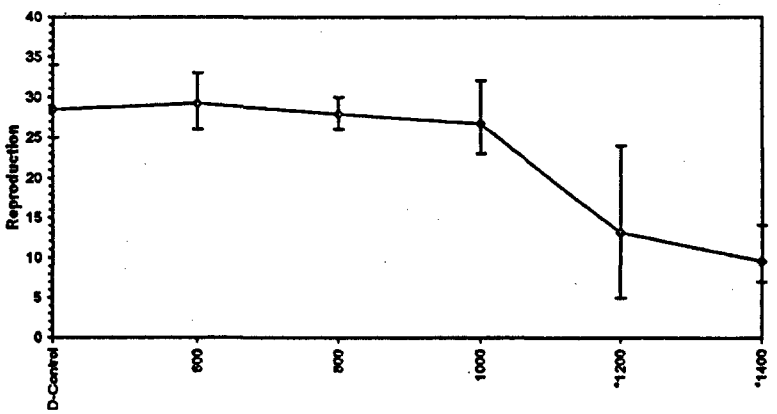
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	1000	1200	1095.44512	
Treatments vs D-Control				

Linear Interpolation (200 Resamples)

Point	mg/L	SD	95% CL		Skew
IC05	890	110.963805	675.69375	1017.77749	-0.6514
IC10	1011.47059	45.5475413	869.819444	1037.68214	-1.6247
IC15	1032.64706	20.514908	983.356136	1059.1445	-1.5647
IC20	1053.82353	17.4425533	1018.4389	1080.63192	-0.1269
IC25	1075	18.7206497	1041.48686	1116.95268	0.2515
IC40	1138.52941	26.3589432	1100.74816	1215.26767	1.1061
IC50	1180.88235	36.6868776	1138.88223	1289.27212	1.1729



Dose-Response Plot



Environmental Testing Solutions, Inc.
Statistical Analyses

Used for PMSD calculation only.			Ceriodaphnia Survival and Reproduction Test-Reproduction			
Start Date:	11/1/2005	Test ID:	CdNaClCR	Sample ID:		REF-Ref Toxicant
End Date:	11/8/2005	Lab ID:	BTS-Envir. Testing Sol.	Sample Type:		NACL-Sodium chloride
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:		CD-Ceriodaphnia dubia
Comments:						

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	31.000	34.000	26.000	30.000	25.000	30.000	25.000	29.000	27.000	27.000
600	28.000	29.000	33.000	27.000	29.000	30.000	26.000	29.000	28.000	33.000
800	29.000	30.000	27.000	27.000	29.000	29.000	27.000	26.000	28.000	27.000
1000	32.000	31.000	26.000	29.000	29.000	23.000	23.000	23.000	27.000	24.000
1200	22.000	24.000	11.000	19.000	7.000	13.000	6.000	5.000	15.000	9.000
1400	7.000	8.000	14.000	12.000	7.000	7.000	14.000	9.000	9.000	8.000

			Transform: Untransformed						1-Tailed	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	28.400	1.0000	28.400	25.000	34.000	10.259	10			
600	29.200	1.0282	29.200	26.000	33.000	7.876	10	-0.488	2.287	3.751
800	27.900	0.9824	27.900	26.000	30.000	4.612	10	0.305	2.287	3.751
1000	26.700	0.9401	26.700	23.000	32.000	12.860	10	1.036	2.287	3.751
*1200	13.100	0.4613	13.100	5.000	24.000	51.579	10	9.328	2.287	3.751
*1400	9.500	0.3345	9.500	7.000	14.000	29.461	10	11.523	2.287	3.751

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)					0.74299985		1.035		0.57349241	1.15846555
Bartlett's Test indicates unequal variances (p = 1.19E-04)					25.3599415		15.0862722			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	1000	1200	1095.44512		3.7506703	0.13206586	767.706667	13.4518519	2.5E-20	5, 54
Treatments vs D-Control										

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

Daily Chemistry:

		Day					
		0		1		2	
	Analyst	AAB	AAB	AAB	AAB	AAB	AAB
Concentration	Parameter						
CONTROL MHSW	pH (S.U.)	7.73	7.86	7.79	7.74	7.67	7.69
	DO (mg/L)	7.9	8.0	7.7	7.4	7.5	7.4
	Conductivity (µmhos/cm)	314		315		306	
	Alkalinity (mg $CaCO_3$/L)	59				58	
	Hardness (mg $CaCO_3$/L)	92				87	
	Temperature (°C)	24.7	24.9	24.6	25.2	24.5	24.8
600 mg NaCl/L	pH (S.U.)	7.77	7.77	7.77	7.63	7.59	7.61
	DO (mg/L)	7.9	8.0	8.0	7.5	7.6	7.5
	Conductivity (µmhos/cm)	1590		1620		17 AB (1470)	
	Temperature (°C)	24.8	24.9	24.6	25.0	24.7	24.9
800 mg NaCl/L	pH (S.U.)	7.76	7.75	7.75	7.61	7.66	7.60
	DO (mg/L)	8.2	7.9	8.2	7.5	7.5	7.5
	Conductivity (µmhos/cm)	1910		1930		1830	
	Temperature (°C)	24.7	24.8	24.4	25.0	24.7	25.1
1000 mg NaCl/L	pH (S.U.)	7.74	7.73	7.75	7.58	7.66	7.58
	DO (mg/L)	8.2	7.9	8.2	7.6	7.7	7.5
	Conductivity (µmhos/cm)	2160		2170		21 AB (2230)	
	Temperature (°C)	24.7	24.9	24.4	24.9	24.8	25.1
1200 mg NaCl/L	pH (S.U.)	7.73	7.74	7.74	7.58	7.64	7.59
	DO (mg/L)	8.2	7.9	8.1	7.3	7.6	7.5
	Conductivity (µmhos/cm)	2590		2570		2580	
	Temperature (°C)	24.7	25.1	24.4	25.0	24.8	25.0
1400 mg NaCl/L	pH (S.U.)	7.72	7.73	7.74	7.56	7.65	7.62
	DO (mg/L)	8.3	8.0	8.1	7.4	7.6	7.6
	Conductivity (µmhos/cm)	2930		2950		2990	
	Temperature (°C)	24.7	25.0	24.5	24.9	24.7	25.1
STOCK	Conductivity (µmhos/cm)	126,000		—K		—N	
		Initial	Final	Initial	Final	Initial	Final

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

		Day							
		3		4 JN		JN 5		6	
	Analyst	AAB	KEK	KEK	KEK	KEK	AAB	AAB	JK
Concentration	Parameter								
CONTROL MHSW	pH (S.U.)	7.53	7.64	7.87	7.57	7.70	8.12	8.10	7.95
	DO (mg/L)	7.4	7.5	7.7	7.5	7.5	7.7	7.7	7.7
	Conductivity (µmhos/cm)	310		314		313		310	
	Alkalinity (mg $CaCO_3$/L)	59		58		58		58	
	Hardness (mg $CaCO_3$/L)	97		95		93		85	
	Temperature (°C)	24.8	25.0	24.9	24.8	24.7	25.2	24.9	25.1
600 mg NaCl/L	pH (S.U.)	7.62	7.60	7.64	7.58	7.58	8.04	8.11	7.97
	DO (mg/L)	7.5	7.5	7.5	7.6	7.5	7.6	7.7	7.8
	Conductivity (µmhos/cm)	1450		1470		1450		1480	
	Temperature (°C)	24.7	25.2	25.0	24.7	24.6	25.3	24.8	25.1
800 mg NaCl/L	pH (S.U.)	7.61	7.59	7.65	7.58	7.61	8.04	8.11	7.96
	DO (mg/L)	7.6	7.6	7.6	7.6	7.5	7.6	7.6	7.9
	Conductivity (µmhos/cm)	1870		1910		1870		1830	
	Temperature (°C)	24.7	25.2	25.0	24.7	24.6	25.3	24.9	25.3
1000 mg NaCl/L	pH (S.U.)	7.60	7.58	7.64	7.59	7.61	8.02	8.10	7.94
	DO (mg/L)	7.6	7.5	7.7	7.5	7.6	7.6	7.7	7.9
	Conductivity (µmhos/cm)	2210		2320		2290		2270	
	Temperature (°C)	24.7	25.0	25.0	24.7	24.6	25.0	25.0	25.3
1200 mg NaCl/L	pH (S.U.)	7.59	7.59	7.64	7.56	7.61	8.01	8.09	7.96
	DO (mg/L)	7.6	7.5	7.7	*7.8 7.5	7.7	7.6	7.7	7.9
	Conductivity (µmhos/cm)	2550		2670		2680		2530	
	Temperature (°C)	24.7	25.2	25.0	24.9	24.6	25.0	24.8	25.0
1400 mg NaCl/L	pH (S.U.)	7.62	7.58	7.64	7.56	7.62	8.02	8.08	7.95
	DO (mg/L)	7.7	7.6	7.6	7.7	7.6	7.7	7.8	7.9
	Conductivity (µmhos/cm)	2980		3080		3010		3010	
	Temperature (°C)	24.6	25.2	25.0	24.7	24.6	25.1	24.9	25.0
STOCK	Conductivity (µmhos/cm)	—*		127000		—*		—*	
		Initial	Final	Initial	Final	Initial	* Final	* Initial	* Final

* Higher overall pH due to new probe.

TENNESSEE VALLEY AUTHORITY
TOXICITY TEST REPORT

INTRODUCTION / EXECUTIVE SUMMARY

Report Date: December 13, 2005

1. Facility / Discharger: Sequoyah Nuclear Plant / TVA

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. 1Q10: 2,992.4

8. Outfall Tested: 101

9. Dates Sampled: November 14-19, 2005

10. Average Flow on Days Sampled (MGD): 1560.9, 1529.0, 1499.9

11. Pertinent Site Conditions:

Towerbrom 960 (oxidizing biocide) was injected from November 14-19, 2005. The dates and times for the Towerbrom 960 injections are in the following table.

Injection Location	Date/Start Time (ET)	Date/Ending Time (ET)
Essential Raw Cooling Water (ERCW) Train A and B	11/14/2005 0000 11/16/2005 0920	11/15/2005 2310 11/19/2005 2400
Raw Cooling Water (RCW)	11/14/2005 0000	11/19/2005 2400

Note: Essential Raw Cooling Water (ERCW) Train A and B is located outside the plant and had to be shut down from 11/15/2005 at 2310 to 11/16/2005 at 0920 due to a NOAA issued tornado watch.

12. Test Dates: November 16-23, 2005

13. Test Type: Short-term Chronic Definitive

14. Test Species: Fathead Minnows (*Pimephales promelas*)
Daphnids (*Ceriodaphnia dubia*)

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

Pimephales promelas: 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: $IC_{25} = 45.2\%$

17. Test Results: Outfall 101: *Pimephales promelas:* $IC_{25} > 100\%$
Ceriodaphnia dubia: $IC_{25} > 100\%$

UV treated Outfall 101: *Pimephales promelas:* $IC_{25} > 100\%$

18. Facility Contact: Stephanie Howard Phone #: (423) 843-6700

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

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

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

22. Notes: Outfall 101 samples collected November 14-19, 2005, showed no toxic effects to fathead minnows or daphnids. The resulting IC_{25} values, for both species, were > 100 percent. Exposure of fathead minnows and daphnids to intake samples resulted in no significant differences from controls during this study period.

Fathead minnows were also exposed to UV treated Outfall 101 and intake samples since fish pathogens present in intake water have been the suspected cause of interference (anomalous dose response and high variability among replicates) in previous toxicity testing at Sequoyah. At the time this study was conducted, insignificant mortality occurred in minnows exposed to non-treated and UV treated samples.

METHODS SUMMARY

Samples:

1. Sampling Point: Outfall 101, Intake

2. Sample Type: Composite

3. Sample Information:

Sample ID	Date (MM/DD/YY)/ Time (ET) Collected	Date (MM/DD/YY)/ Time (ET) Received	Arrival Temp. (°C)	Initial TRC* (mg/L)	Date (MM/DD/YY)/ Time (ET) Used By
101	11/14/05 0615 to 11/15/05 0515	11/15/05 1450	1.9, 2.6†	<0.10	11/16/05 1358 11/17/05 1302
Intake	11/14/05 0630 to 11/15/05 0530	11/15/05 1450	2.0	<0.10	11/16/05 1358 11/17/05 1302
101	11/16/05 0630 to 11/17/05 0530	11/17/05 1415	1.5, 1.1†	<0.10	11/18/05 1318 11/19/05 1300
Intake	11/16/05 0630 to 11/17/05 0530	11/17/05 1415	0.8	<0.10	11/18/05 1318 11/19/05 1300
101	11/18/05 0545 to 11/19/05 0445	11/19/05 1355	1.6, 1.8†	<0.10	11/20/05 1312 11/21/05 1305 11/22/05 1310
Intake	11/18/05 0615 to 11/19/05 0515	11/19/05 1355	1.0	<0.10	11/20/05 1312 11/21/05 1305 11/22/05 1310

*TRC = Total Residual Chlorine

†Samples were collected in two 2.5 gallon cubitainers. Temperature was measured in each cubitainer upon arrival.

4. Sample Manipulation: Samples from Outfall 101 and intake were warmed to test temperature (25.0 ± 1.0°C) in a warm water bath.

   Aliquots of Outfall 101 and Intake samples were UV-treated through a 40-watt Smart® UV Sterilizer (manufactured by Emperor Aquatics, Inc.) for 2 minutes.

	Pimephales promelas	*Ceriodaphnia dubia*
Test Organisms:		
1. Source:	Aquatic BioSystems, Inc.	In-house Cultures
2. Age:	23.0-24.5 hours old	<24-hours old
Test Method Summary:		
1. Test Conditions:	Static, Renewal	Static, Renewal
2. Test Duration:	7 days	Until at least 60% of control females have 3 broods
3. Control / Dilution Water:	Moderately Hard Synthetic	Moderately Hard Synthetic
4. Number of Replicates:	4	10
5. Organisms per Replicate:	10	1
6. Test Initiation: (Date/Time)		
Outfall 101	11/16/05 - 1358 ET	11/16/05 - 1237 ET
UV Treated Outfall 101	11/16/05 - 1344 ET	
7. Test Termination: (Date/Time)		
Outfall 101	11/23/05 - 1306 ET	11/23/05 - 1151 ET
UV Treated Outfall 101	11/23/05 - 1322 ET	
8. Test Temperature: Outfall 101:	Mean = 24.9C (24.4 - 25.2°C)	Mean = 25.0°C (24.7 - 25.4°C)
Test Temperature: UV-Treated Outfall 101:		Mean = 25.0°C (24.3 - 25.4°C)

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

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

TOXICITY TEST RESULTS (see Appendix C for Bench Sheets)

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

Conducted November 16-23, 2005 using effluent from Outfall 101.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	98
10.98%	100	100	100	100	100	98	98
22.0%	100	100	100	98	98	95	95
43.9%	100	100	100	100	100	100	100
72.0%	100	100	100	100	100	100	100
100.0%	100	100	100	98	98	98	98
Intake	100	100	100	100	100	100	98

Test Solutions (% Effluent)	Mean Dry Weight (mg) (replicate number)				
	1	2	3	4	Mean
Control	0.663	0.653	0.566	0.648	0.633
10.98%	0.617	0.663	0.677	0.689	0.662
22.0%	0.629	0.699	0.704	0.671	0.676
43.9%	0.689	0.636	0.701	0.704	0.683
72.0%	0.687	0.713	0.704	0.775	0.720
100.0%	0.618	0.705	0.661	0.699	0.671
Intake	0.689	0.607	0.642	0.583	0.630

IC_{25} Value: ≥ 100% Permit Limit: 45.2% 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: ≤ 1.0 TUc* Permit Limit: 2.2 TUc

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

TOXICITY TEST RESULTS (see Appendix C for Bench Sheets)

2. Results of a *Ceriodaphnia dubia* Chronic/ 7-day Toxicity Test.
(Genus species) (Type / Duration)
Conducted November 16-23, 2005 using effluent from Outfall 101.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
10.98%	100	100	100	100	100	100	100
22.0%	100	100	100	100	100	100	100
43.9%	100	100	100	100	100	100	100
72.0%	100	100	100	100	100	100	100
100.0%	100	100	100	100	100	100	100

Test Solutions (% Effluent)	Reproduction (#young/female/7 days) Data (replicate number)										
	1	2	3	4	5	6	7	8	9	10	Mean
Control	29	31	36	27	32	32	29	28	31	27	30.2
10.98%	31	35	32	30	30	34	31	31	32	33	31.9
22.0%	34	32	32	32	33	31	33	32	34	30	32.3
43.9%	37	30	33	29	30	34	30	33	33	33	32.2
72.0%	35	31	29	32	38	34	35	33	29	36	33.2
100.0%	35	39	36	34	32	32	33	32	36	35	34.4

IC_{25} Value: ≥ 100%
Permit Limit: 45.2%

95% Confidence Limits:
Upper Limit: NA
Lower Limit: NA

Calculated TU Estimates: < 1.0 TUc*

Permit Limit: 2.2 TUc

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

TOXICITY TEST RESULTS (see Appendix C for Bench Sheets)

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

Conducted November 16-23, 2005 using water from Intake

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

Test Solutions (% Effluent)	Reproduction (#young/female/7 days) Data (replicate number)										
	1	2	3	4	5	6	7	8	9	10	Mean
Control	33	28	30	29	30	30	34	32	34	33	31.3
Intake	35	40	35	35	33	36	38	35	37	34	35.8

IC_{25} Value: ≥ 100% Permit Limit: N/A 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: < 1.0 TUc* Permit Limit: N/A

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

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

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

Conducted November 16-23, 2005 using effluent from UV Treated Outfall 101.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control	100	100	100	100	100	100	100
10.98%	100	100	100	100	100	100	98
22.0%	100	100	100	100	100	100	100
43.9%	100	100	100	100	100	100	100
72.0%	100	100	100	100	100	100	100
100.0%	100	100	100	100	100	100	100
Intake	100	100	100	100	100	100	100

Test Solutions (% Effluent)	Mean Dry Weight (mg) (replicate number)				
	1	2	3	4	Mean
Control	0.562	0.718	0.688	0.670	0.660
10.98%	0.732	0.656	0.551	0.547	0.622
22.0%	0.638	0.642	0.638	0.571	0.622
43.9%	0.525	0.602	0.776	0.563	0.617
72.0%	0.617	0.678	0.565	0.638	0.625
100.0%	0.617	0.690	0.611	0.564	0.621
Intake	0.602	0.618	0.621	0.580	0.605

IC_{25} Value: > 100% 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: < 1.0 TUc*

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

REFERENCE TOXICANT TEST RESULTS (see Appendix A and D)

Species	Date	Time	Duration	Toxicant	Results (IC_{25})
Pimephales promelas	November 16-23, 2005	1416	7-days	KCl	0.70 g/L
Ceriodaphnia dubia	November 1-8, 2005	1101	7-days	NaCl	1.08 g/L

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for *Pimephales promelas* and *Ceriodaphnia dubia* Tests, Sequoyah Nuclear Plant Effluent (SQN), Outfall 101, November 16-23, 2005.

Test	Sample ID	Temperature (°C)		Dissolved Oxygen (mg/L)		pH (S.U.)		Conductance	Alkalinity	Hardness	Total Residual
		Initial	Final	Initial	Final	Initial	Final	(µmhos/cm)	(mg/L $CaCO_3$)	(mg/L $CaCO_3$)	Chlorine (mg/L)
Pimephales promelas	Control	24.9 24.7 - 25.1	24.7 24.6 - 24.9	7.8 7.7 - 7.9	7.4 7.0 - 7.7	7.96 7.68 - 8.05	7.88 7.71 - 8.02	315 307 - 321	61 58 - 62	95 92 - 96	- - -
	10.98%	24.9 24.8 - 25.1	24.7 24.4 - 24.9	7.8 7.4 - 8.0	7.4 6.9 - 7.5	8.02 7.92 - 8.10	7.83 7.62 - 8.00	296 288 - 303	- - -	- - -	- - -
	22.0%	24.9 24.8 - 25.1	24.8 24.6 - 24.9	7.7 7.4 - 7.9	7.3 6.7 - 7.6	8.02 7.93 - 8.10	7.83 7.66 - 7.96	282 276 - 288	- - -	- - -	- - -
	43.9%	25.0 24.8 - 25.2	24.7 24.6 - 24.9	7.7 7.5 - 7.9	7.3 6.8 - 7.5	8.02 7.93 - 8.11	7.86 7.67 - 7.93	257 248 - 270	- - -	- - -	- - -
	72.0%	25.0 24.8 - 25.2	24.8 24.6 - 25.0	7.7 7.5 - 7.9	7.3 6.9 - 7.5	8.02 7.94 - 8.10	7.87 7.68 - 7.97	218 212 - 223	- - -	- - -	- - -
	100.0%	25.0 24.8 - 25.2	24.7 24.6 - 24.9	7.7 7.5 - 8.0	7.3 6.7 - 7.7	8.02 7.95 - 8.10	7.86 7.68 - 7.96	181 175 - 186	63 62 - 65	72 70 - 74	< 0.10 < 0.10 - < 0.10
	Intake	25.0 24.9 - 25.2	24.8 24.6 - 24.9	7.8 7.6 - 7.9	7.3 6.9 - 7.7	8.01 7.94 - 8.09	7.86 7.68 - 7.97	181 173 - 187	63 61 - 64	75 72 - 76	< 0.10 < 0.10 - < 0.10
Ceriodaphnia dubia	Control	24.8 24.7 - 24.9	25.1 24.9 - 25.3	7.8 7.7 - 7.9	7.8 7.5 - 8.0	7.96 7.68 - 8.05	8.00 7.93 - 8.07	315 307 - 321	61 58 - 62	95 92 - 96	- - -
	10.98%	24.8 24.7 - 25.0	25.2 24.9 - 25.3	7.8 7.4 - 8.0	7.9 7.5 - 8.2	8.02 7.92 - 8.10	8.02 7.96 - 8.09	296 288 - 303	- - -	- - -	- - -
	22.0%	24.9 24.7 - 25.0	25.2 24.9 - 25.4	7.7 7.4 - 7.9	7.9 7.6 - 8.3	8.02 7.93 - 8.10	8.02 7.99 - 8.11	282 276 - 288	- - -	- - -	- - -
	43.9%	24.9 24.8 - 25.0	25.1 24.8 - 25.2	7.7 7.5 - 7.9	7.9 7.6 - 8.3	8.02 7.93 - 8.11	8.03 7.97 - 8.11	257 248 - 270	- - -	- - -	- - -
	72.0%	24.9 24.7 - 25.1	25.1 24.9 - 25.3	7.7 7.5 - 7.9	7.9 7.5 - 8.1	8.02 7.94 - 8.10	8.06 7.99 - 8.14	218 212 - 223	- - -	- - -	- - -
	100.0%	24.9 24.7 - 25.1	25.1 25.0 - 25.2	7.7 7.5 - 8.0	7.9 7.5 - 8.3	8.02 7.95 - 8.10	8.08 8.01 - 8.16	181 175 - 186	63 62 - 65	72 70 - 74	< 0.10 < 0.10 - < 0.10
	Intake	24.9 24.8 - 24.9	25.1 24.8 - 25.3	7.8 7.6 - 7.9	7.9 7.4 - 8.1	8.01 7.94 - 8.09	8.06 7.98 - 8.13	181 173 - 187	63 61 - 64	75 72 - 76	< 0.10 < 0.10 - < 0.10

Overall temperature (°C)	Average	Minimum	Maximum
Pimephales promelas	24.9	24.4	25.2
Ceriodaphnia dubia	25.0	24.7	25.4

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for the *Pimephales promelas* Test, Sequoyah Nuclear Plant Effluent (SQN), UV-Treated Outfall 101, November 16-23, 2005.

Test	Sample ID	Temperature (°C)		Dissolved Oxygen (mg/L)		pH (S.U.)		Conductance (µmhos/cm)
		Initial	Final	Initial	Final	Initial	Final	
Pimephales promelas	Control	24.9 24.9 - 25.1	24.8 24.6 - 24.9	7.8 7.6 - 7.9	7.4 7.0 - 7.6	8.01 7.92 - 8.11	7.83 7.66 - 7.98	305 290 - 314
	10.98%	25.1 25.0 - 25.2	24.9 24.6 - 25.2	7.8 7.6 - 8.0	7.3 7.0 - 7.5	8.02 7.92 - 8.12	7.84 7.70 - 7.96	297 289 - 305
	22.0%	25.1 24.9 - 25.3	24.9 24.6 - 25.1	7.8 7.5 - 7.9	7.4 6.8 - 7.6	8.02 7.93 - 8.12	7.87 7.66 - 7.98	283 275 - 291
	43.9%	25.1 24.9 - 25.2	24.8 24.5 - 24.9	7.8 7.6 - 7.9	7.3 6.8 - 7.6	8.02 7.93 - 8.12	7.85 7.65 - 7.98	258 247 - 274
	72.0%	25.1 24.9 - 25.3	24.8 24.6 - 24.9	7.8 7.6 - 7.9	7.3 6.9 - 7.5	8.02 7.94 - 8.12	7.87 7.71 - 8.02	220 217 - 224
	100.0%	25.1 24.9 - 25.3	24.8 24.3 - 25.1	7.8 7.7 - 8.0	7.3 7.0 - 7.6	8.02 7.94 - 8.12	7.87 7.73 - 8.03	185 182 - 188
	Intake	25.1 24.9 - 25.4	24.8 24.5 - 25.1	7.8 7.5 - 8.3	7.4 7.2 - 7.8	8.02 7.90 - 8.12	7.90 7.70 - 8.02	182 176 - 191

Overall temperature (°C)	Average	Minimum	Maximum
Pimephales promelas	25.0	24.3	25.4

SUMMARY / CONCLUSIONS

Outfall 101 samples collected November 14-19, 2005, showed no toxic effects to fathead minnows or daphnids. The resulting IC_{25} values, for both species, were > 100 percent. Exposure of fathead minnows and daphnids to intake samples resulted in no significant differences from controls during this study period.

Fathead minnows were also exposed to UV treated Outfall 101 and intake samples since fish pathogens present in intake water have been the suspected cause of interference (anomalous dose response and high variability among replicates) in previous toxicity testing at Sequoyah. At the time this study was conducted, insignificant mortality occurred in minnows exposed to non-treated and UV treated samples.

Appendix A

ADDITIONAL TOXICITY TEST INFORMATION

SUMMARY OF METHODS

1. *Pimephales promelas*

Tests were conducted according to EPA-821-R-02-013 (October 2002) using four replicates, each containing ten test organisms, per treatment. Test vessels consisted of 500-mL plastic disposable cups, each containing 250-mL of test solution.

2. *Ceriodaphnia dubia*

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

DEVIATIONS / MODIFICATIONS TO TEST PROTOCOL

1. *Pimephales promelas* and 2. *Ceriodaphnia dubia*

Environmental Testing Solutions, Inc. performed chronic toxicity tests from November 16-23, 2005. The toxicity tests were conducted with samples collected from SQN Outfall 101 during oxidizing (November 14-19) biocide applications. An internal peer review of the reports was performed by TVA biologists and the Senior Toxicologist and it was determined that incorrect serial dilutions were prepared. Previous permit dilutions of 10.98, 22.0, 43.9, 72.0, and 100% were used instead of the dilutions of 11.3, 22.6, 45.2, 72.6, and 100% specified in the recently renewed permit, effective September 1, 2005.

The dilutions that were used in the tests were virtually the same as what the current permit requires ($\leq$ 1.3% effluent difference) given the variability inherent in performing whole effluent toxicity tests. Additionally, the error is judged to be inconsequential relative to evaluation of the test results since no toxicity to either species (*Pimephales promelas* or *Ceriodaphnia dubia*) was observed (i.e., IC25 > 100% effluent) during either test period.

A process change to TVA's NPDES biomonitoring compliance program has been implemented whereby the contract laboratory must confirm via email to TVA the exact dates that toxicity tests will be performed and the dilution series that will be used, prior to conducting all future tests.

This incident has been assessed as a laboratory deviation. The EPA chronic test manual (EPA-821-R-02-013) states that deviations must be evaluated on a case-by-case basis and deviations may or may not invalidate a test result "depending on the degree of the departure and the objective of the test." TVA biologists and the Senior Toxicologist have considered the degree of the deviation in this particular case and find no potential or observed impact on the test results. Therefore the test results for the subject tests are considered valid.

DEVIATIONS / MODIFICATIONS TO PRETEST CULTURE OR HOLDING OF TEST ORGANISMS

1. *Pimephales promelas*

   None

2. *Ceriodaphnia dubia*

   None

PHYSICAL AND CHEMICAL METHODS

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

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

3. Temperature was measured by EPA Method 170.1.

4. Dissolved oxygen was measured by EPA Method 360.1.

5. The pH was measured by EPA Method 150.1.

6. Conductance was measured by EPA Method 120.1.

7. Alkalinity was measured by EPA Method 310.1.

8. Total Hardness was measured by EPA Method 130.2.

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

QUALITY ASSURANCE

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

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

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

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

3. Dilution Water Used: Moderately hard synthetic water.

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

REFERENCES

1. NPDES Permit No. TN0026450.

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

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

4. Quality Assurance Program: Standard Operating Procedures, Environmental Testing Solutions, Inc (most current version).

Sequoyah Nuclear Plant Biomonitoring
November 16-23, 2005

Appendix B

Diffuser Discharge Concentrations of Total Residual Chlorine,

Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 19, 2005

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. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 19, 2005

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	-	-	-
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	-	-	-
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	-	-

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 19, 2005

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
01/02/2002	-	< 0.0079	0.023	0.02	0.006	-	-
01/03/2002	-	< 0.0042	0.023	0.014	-	-	-
01/04/2002	-	0.0124	0.024	0.014	0.009	-	-
01/05/2002	-	< 0.0042	-	-	-	-	-
01/06/2002	-	< 0.0042	-	-	-	-	-
01/07/2002	-	< 0.0089	0.024	0.014	0.006	-	-
02/24/2002	-	< 0.004	-	-	-	-	-
02/25/2002	-	< 0.004	0.023	0.023	-	-	-
02/26/2002	-	0.0143	0.023	0.023	0.007	-	-
02/27/2002	-	< 0.0041	0.023	0.023	-	-	-
02/28/2002	-	< 0.0041	0.024	0.008	-	-	-
03/01/2002	-	< 0.0041	0.024	0.008	-	-	-
05/05/2002	-	-	-	-	-	-	-
05/06/2002	-	-	0.058	0.02	0.014	-	-
05/07/2002	-	-	0.058	0.02	0.015	-	-
05/08/2002	-	-	0.056	0.019	-	-	-
05/09/2002	-	-	0.057	0.02	0.014	-	-
05/10/2002	-	-	0.056	0.019	-	-	-
08/04/2002	-	<0.0058	-	-	-	-	-
08/05/2002	-	<0.0058	0.053	0.018	-	-	0.025
08/06/2002	-	0.0092	0.053	0.018	-	-	-
08/07/2002	-	<0.0107	0.055	0.019	0.007	-	-
08/08/2002	-	<0.0061	0.055	0.019	-	-	-
08/09/2002	-	0.0152	0.054	0.018	0.008	-	-
10/06/2002	-	<0.00497	-	-	-	-	-
10/07/2002	-	0.0153	0.054	0.018	0.009	-	-
10/08/2002	-	<0.0092	0.054	0.018	0.007	-	-
10/09/2002	-	0.0124	0.053	0.018	0.009	-	-
10/10/2002	-	0.0134	0.054	0.018	0.009	-	-
10/11/2002	-	<0.0042	0.054	0.018	-	-	-
01/12/2003	-	<0.0035	-	-	-	-	-
01/13/2003	-	<0.006	0.025	0.019	0.009	-	-
01/14/2003	-	<0.0118	0.026	0.020		-	-
01/15/2003	-	<0.0063	0.026	0.020	0.009	-	-
01/16/2003	-	<0.0034	0.026	0.020		-	-
01/17/2003	-	<0.0034	0.026	0.009	-	-	-
04/06/2003	-	<0.0073	-	-	-	-	-
04/07/2003	-	<0.0189	-	0.021	-	-	-
04/08/2003	-	<0.0117	-	0.021	-	-	-
04/09/2003	-	<0.0139	-	0.021	0.016	-	-
04/10/2003	-	<0.0113	-	0.021	0.018	-	-
04/11/2003	-	<0.0073	-	0.022	-	-	-

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 19, 2005

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
06/15/2003	-	< 0.0045	-	-	-	-	-
06/16/2003	-	< 0.0037	0.057	0.020	-	-	0.022
06/17/2003	-	< 0.0048	0.041	0.014	-	-	0.024
06/18/2003	-	< 0.0048	0.041	0.014	-	-	0.024
06/19/2003	-	< 0.0085	0.058	0.020	-	-	0.025
06/20/2003	-	< 0.0048	0.058	0.020	-	-	0.025
08/03/2003	-	<0.0050	-	-	-	-	-
08/04/2003	-	<0.0050	0.058	0.020	-	-	-
08/05/2003	-	<0.0051	0.057	0.020	-	-	0.025
08/06/2003	-	<0.0084	0.057	0.020	-	-	0.025
08/07/2003	-	0.0129	0.057	0.020	-	-	0.024
08/08/2003	-	0.0153	0.057	0.020	0.009	-	-
10/05/2003	-	<0.0043	0.057	0.020	-	-	-
10/06/2003	-	<0.0043	0.057	0.020	-	-	0.025
10/07/2003	-	<0.0090	0.057	0.020	-	-	0.025
10/08/2003	-	<0.0106	0.057	0.020	-	-	0.025
10/09/2003	-	0.0181	0.026	0.022	-	-	0.025
10/10/2003	-	0.0183	0.026	0.024	0.009	-	-
02/01/2004	-	0.0093	0.027	0.009	-	-	-
02/02/2004	-	<0.0034	0.026	0.009	-	-	-
02/03/2004	-	<0.0034	0.026	0.009	-	-	-
02/04/2004	-	0.0124	0.026	0.009	0.009	-	-
02/05/2004	-	<0.0034	0.026	0.009	-	-	-
02/06/2004	-	0.0105	0.026	0.009	0.010	-	-
05/04/2004	-	<0.0123	0.026	0.019	-	-	0.025
05/05/2004	-	<0.0144	0.026	0.014	0.009	-	0.025
05/06/2004	-	<0.0146	0.037	0.013	-	-	0.025
05/07/2004	-	0.0227	0.058	0.020	0.009	-	0.025
05/08/2004	-	0.016	0.060	0.021	-	-	-
05/09/2004	-	<0.0104	0.058	0.020	-	-	-
07/04/2004	-	0.0217	0.057	0.019	-	-	-
07/05/2004	-	<0.0085	0.057	0.020	0.009	-	-
07/06/2004	-	<0.0077	0.058	0.020	-	-	0.031
07/07/2004	-	0.0252	0.056	0.019	-	-	0.031
07/08/2004	-	0.0223	0.057	0.019	0.009	-	-
07/09/2004	-	0.0182	0.057	0.020	0.009	-	-

Table B-1. Sequoyah Nuclear Plant Diffuser (Outfall 101) Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling
March 12, 1998-November 19, 2005

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	Nalco 73551 mg/L EO/PO	H-150M mg/L Quat
11/07/2004	-	<0.0187	0.000	0.014	-	-	-	-	-
11/08/2004	-	<0.0192	0.047	0.030	-	-	-	-	-
11/09/2004	-	<0.0233	0.048	0.016	-	-	0.041	-	-
11/10/2004	-	<0.0149	0.047	0.016	-	-	0.041	-	-
11/11/2004	-	<0.0149	0.049	0.017	-	-	0.043	-	-
11/12/2004	-	<0.0253	0.048	0.017	-	-	0.042	-	-
02/06/2005	-	<0.0042	0.028	0.010	-	-	-	-	-
02/07/2005	-	<0.0116	0.028	0.010	-	-	-	0.007	-
02/08/2005	-	<0.0080	0.028	0.010	-	-	-	-	-
02/09/2005	-	0.0199	0.028	0.010	-	-	-	-	-
02/10/2005	-	<0.0042	0.028	0.010	-	-	-	-	-
02/11/2005	-	0.0155	0.028	0.010	-	-	-	0.007	-
06/05/2005	-	0.0063	-	-	-	-	-	-	-
06/06/2005	-	0.0043	-	-	-	-	-	-	0.037
06/07/2005	-	0.0103	-	-	-	-	-	-	0.037
06/08/2005	-	0.0295	-	-	-	-	-	-	0.037
06/09/2005	-	0.0129	-	-	-	-	-	-	-
06/10/2005	-	0.0184	-	-	-	-	-	-	-
07/17/2005	-	0.0109	0.026	0.009	-	-	-	-	-
07/18/2005	-	0.0150	0.026	0.009	-	-	-	-	0.036
07/19/2005	-	0.0163	0.026	0.009	-	-	-	-	0.036
07/20/2005	-	0.0209	0.026	0.009	-	-	-	0.014	0.036
07/21/2005	-	0.0242	0.026	0.009	-	-	-	-	-
07/22/2005	-	0.0238	0.054	0.018	-	-	-	0.014	-
10/30/2005	-	0.0068	-	-	-	-	-	-	-
10/31/2005	-	0.0112	-	-	-	-	-	-	-
11/01/2005	-	0.0104	-	-	-	-	-	-	0.035
11/02/2005	-	0.0104	-	-	-	-	-	-	0.036
11/03/2005	-	0.0117	-	-	-	-	-	-	0.036
11/04/2005	-	0.0165	-	-	-	-	-	-	0.035
11/14/2005	-	0.0274	-	-	-	-	-	-	-
11/15/2005	-	0.0256	-	-	-	-	-	-	-
11/16/2005	-	0.0234	-	-	-	-	-	-	-
11/17/2005	-	0.0231	-	-	-	-	-	-	-
11/18/2005	-	0.0200	-	-	-	-	-	-	-
11/19/2005	-	0.0116	-	-	-	-	-	-	-

Sequoyah Nuclear Plant Biomonitoring
November 16-23, 2005

Appendix C

Chain of Custody Records and Toxicity Test Bench Sheets

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Comment: Lines in this form are ¾, 1 ½, and 2 ½ point.

Client: TVA

Project Name: Sequoyah NP Toxicity

P.O. Number: N/A

Facility Sampled: Sequoyah NP

NPDES Number: TN0026450

Collected By: Duane Brigman; Chevy Ward

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

Delivered By (Circle One):

FedEx UPS Bus Client

Other (specify): Express Courier

General Comments:

project # 2819 & 2820

Field Identification / Sample Description	Grab/Comp.	Collection Date/Time		Container Number & Volume Collected	Flow MGD	Rain Event? (Mark as Appropriate)				Laboratory Use				
		Date	Time			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	Comp	11/14/05-11/15/05	0615-0515	2 (2.5gal)	NA			X		051115.04	1.9°C/2.6°C	KBR	1450	*
SQN-INT-TOX	Comp	11/14/05-11/15/05	0630-0530	1 (2.5 gal)	NA			X		051115.05	2.0°C	KBR	1450	*

* Custody seals intact. Samples received in good condition. J Turner

Sample Custody – Fill In From Top Down

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Duane Brigman J. Duane Bry	11/15/05 9:52am	Express Courier Steve Melton	11/15/05 9:52 am
Express Courier	11/15/05 1450	ETS KKleman ETS	11/15/05 1450
Steve Melton ETS			

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 6°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

Client: TVA

Project Name: Sequoyah NP Toxicity

P.O. Number: N/A

Facility Sampled: Sequoyah NP

NPDES Number: TN0026450

Collected By: Duane Brigman and Chevy Williams

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

Delivered By (Circle One):

FedEx UPS Bus Client

Other (specify): Express Courier

General Comments:
ambient Temp @ collection: 32°C

project # 2220

Field Identification / Sample Description	Grab/Comp.	Collection Date/Time		Container Number & Volume Collected	Flow MGD	Rain Event? (Mark as Appropriate)				Laboratory Use				
		Date	Time			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	Comp	11/16/05-11/17/05	6:30a 5:30a	2 (2.5gal)	NA			X		051117.03	1.5 / 1.1	JA	1415	*
SQN-INT-TOX	Comp	11/16/05-11/17/05	6:30a 5:30a	1 (2.5 gal)	NA			X		051117.04	0.8	JA	1415	*

Sample Custody – Fill In From Top Down

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

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Duane Brigman	11/17/05 1004	Express Courier	11/17/05 1004
Express Courier	11/17/05 1115	ETS	11/17/05 1415

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 6°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

BIOMONITORING CHAIN OF CUSTODY RECORD

Page __1__ of __1__

Client: TVA

Project Name: Sequoyah NP Toxicity

P.O. Number: N/A

Facility Sampled: Sequoyah NP

NPDES Number: TN0026450

Collected By: Duane Brigman and Chevy Williams

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

Delivered By (Circle One):

FedEx UPS Bus Client

Other (specify): Express Courier

General Comments:

Field Identification / Sample Description	Grab/Comp	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				project # 2220 Laboratory Use				
		Date	Time			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time	Appearance
SQN-101-TOX	Comp	11/18/05-11/19/05	0545-0445	2 (2.5gal)	NA			X		051119.12	1.6°C/1.8C	KEM	1355	+
SQN-INT-TOX	Comp	11/18/05-11/19/05	~~0545~~DB 0615-0515	1 (2.5 gal)	NA			X		051119.13	1.0°C	KEM	1355	+

Sample Custody – Fill In From Top Down

+ Custody seals intact. Samples received in good condition. CJ

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
Duane Brigman D. Duane Brigman	11/19/2005 0930	Express Courier L. Le Jennings	11/19/2005 0930
Express Courier L. Le Jennings	11/19/2005 1355	ETS KKellman	11/19/2005 1355

Instructions: Clients should fill in all areas except those in the "Laboratory Use" block. Biomonitoring samples are preserved by storing them at 6°C and shipping them in ice. The hold time for each sample is 36 hours from the time of collection. Therefore, please collect and ship in such a way that the laboratory will receive the samples with ample time to initiate testing within that time frame. Samples shipped overnight on Friday via FedEx or UPS must be marked for Saturday delivery or they will not arrive until the following Monday.

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

Client: TVA
Facility: Sequoyah Nuclear Plant
NPDES #: TN 0026450
Project #: 2220

County: Hamilton
Treatment: Non-treated
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.98	22	43.9	72	100	
Effluent volume (mL)	274.5	550	1097.5	1800	2500	
Diluent volume (mL)	2225.5	1950	1402.5	700	0	
Total volume (mL)	2500	2500	2500	2500	2500	

Test organism information:		*Test information:*	
Organism age:	23 TO 24.5 HOURS OLD	Randomizing template:	YELLOW
Date and times organisms were born between:	11-15-05 1330 TO 1500	Incubator number:	3C
Organism source:	ABS BATCH PP 11-15-05	*Artemia* lot number:	BG18041T
Transfer bowl information:	pH = 8.10 Temperature = 24.2 °C	Total drying time:	24 HOURS
		Date / Time in: 11-23-05	1350
Average transfer volume:	10.4 mL	Date / Time out: 11-24-05	1356
		Oven temperature:	60°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	Control water batch used MHSW	Sample numbers used	Analyst
0	11-16-05	———	1500	1358	11-11-05A	051115.04 & 05	
1	11-17-05	0900	1506	1302	11-11-05B	051115.04 & 05	
2	11-18-05	0912	1520	1318	11-11-05B	051117.03 & 04	
3	11-19-05	0847	1452	1300	11-17-05A	051117.03 & 04	
4	11-20-05	0905	1506	1312	11-17-05A	051119.12 & 13	
5	11-21-05	0908	1511	1305	11-17-05B	051119.12 & 13	
6	11-22-05	0900	1500	1310	11-17-05B	051119.12 & 13	
7	11-23-05			1306			KEK

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	0%	≤ 20%	7-day LC_{50}	>100%
Average weight per initial larvae:	0.633		NOEC	100%
Average weight per surviving larvae:	~~0.653~~ 0.648	≥ 0.25 mg/larvae	LOEC	>100%
			ChV	>100%
			IC_{25}	>100%

Species: *Pimephales promelas* **Date:** 11-16-05

Client: TVA / Sequoyah Nuclear Plant - Non-treated

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	9 id	10	10	10
5	10	10	10	10	10	10	10	10	9	10	10	10
6	10	10	10	10	9 id	10	10	10	9	9 id	10	10
7	10	10	9 id	10	9	10	10	10	9	9 id	10	10
A = Pan weight (mg) Color identification: Magenta Analyst: JN	14.60	15.09	14.52	14.75 ~~14.76~~ JN	14.56 ~~14.57~~ JN	14.70	14.24	14.54	14.13	15.03	14.58	15.06
B = Pan + Larvae weight (mg) Analyst: TPK	21.23	21.62	20.18	21.23	20.73	21.33	21.01	21.43	20.42	22.02	21.62	21.77
Larvae weight (mg) = A – B	6.63	6.53	5.66	6.48	6.17	6.63	6.77	6.89	6.29	6.99	7.04	6.71
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.663	0.653	0.566	0.648	0.617	0.663	0.677	0.689	0.629	0.699	0.704	0.671
Average weight per initial number of larvae (mg)	0.633				0.662		-4.67%		0.676		-6.8%	
Percent reduction from control (%)												

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Species: *Pimephales promelas*

Client: TVA / Sequoyah Nuclear Plant - Non-treated **Date:** 11-16-05

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	9 id	10
5	10	10	10	10	10	10	10	10	10	10	9	10
6	10	10	10	10	10	10	10	10	10	10	9	10
7	10	10	10	10	10	10	10	10 LG	10 ISH	10	9	10
A = Pan weight (mg) Color identification: Magenta Analyst: JN	16.24	15.79	14.60	14.47	14.88	15.16	14.47	14.32	15.05	14.54 ~~14.53~~ JN	14.49	15.69
B = Pan + Larvae weight (mg) Analyst: KGKeenan	23.13	22.15	21.61	21.51	21.75	22.29	21.51	22.07	21.23	21.59	21.10	22.68
Larvae weight (mg) = A – B	6.89	6.36	7.01	7.04	6.87	7.13	7.04	7.75	6.18	7.05	6.61	6.99
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.689	0.636	0.701	0.704	0.687	0.713	0.704	0.775	0.618	0.705	0.661	0.699
Average weight per initial number of larvae (mg)	0.683				0.720				0.671			
Percent reduction from control (%)			-7.9%				-13.8%				-6.0%	

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: JA

Comments:

Species: *Pimephales promelas*

Client: TVA / Sequoyah Nuclear Plant - Non-treated **Date:** 11-16-05

Survival and Growth Data

Day	100% Intake			
	Y	Z	AA	BB
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	9rd	10	10 ISH
A = Pan weight (mg) Color identification: Magenta Analyst: JN	14.84	14.35	15.23	14.79
B = Pan + Larvae weight (mg) Analyst: KeKleman	21.73	20.42	21.65	20.62
Larvae weight (mg) = A – B	6.89	6.07	6.42	5.83
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.689	0.607	0.642	0.583
Average weight per initial number of larvae (mg) / Percent reduction from control (%)	0.630		0.47₀	

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: JL

Comments:

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 16-23, 2005

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

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

Project number: 2220 **Reveiwed by:**

							Not for Compliance Assessment, Internal Laboratory QC							
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 / Surviving number of larvae (mg)**	**Mean weight / Surviving number of larvae (mg)**	**Coefficient of variation (Mean weight per surviving number of larvae) (%)**	**Weight / Initial number of larvae (mg)**	**Mean survival (%)**	**Mean weight / Initial number of larvae (mg)**	**Coefficient of variation (Mean weight per initial number of larvae) (%)**	**Percent reduction from control (%)**
Control	A	10	10	14.60	21.23	6.63	0.663	0.648	2.2	0.663	97.5	0.633	7.1	Not applicable
	B	10	10	15.09	21.62	6.53	0.653			0.653				
	C	10	9	14.52	20.18	5.66	0.629			0.566				
	D	10	10	14.75	21.23	6.48	0.648			0.648				
10.98%	E	10	9	14.56	20.73	6.17	0.686	0.679	1.7	0.617	97.5	0.662	4.8	-4.6
	F	10	10	14.70	21.33	6.63	0.663			0.663				
	G	10	10	14.24	21.01	6.77	0.677			0.677				
	H	10	10	14.54	21.43	6.89	0.689			0.689				
22%	I	10	9	14.13	20.42	6.29	0.699	0.713	6.3	0.629	95.0	0.676	5.1	-6.8
	J	10	9	15.03	22.02	6.99	0.777			0.699				
	K	10	10	14.58	21.62	7.04	0.704			0.704				
	L	10	10	15.06	21.77	6.71	0.671			0.671				
43.9%	M	10	10	16.24	23.13	6.89	0.689	0.683	4.6	0.689	100.0	0.683	4.6	-7.9
	N	10	10	15.79	22.15	6.36	0.636			0.636				
	O	10	10	14.60	21.61	7.01	0.701			0.701				
	P	10	10	14.47	21.51	7.04	0.704			0.704				
72%	Q	10	10	14.88	21.75	6.87	0.687	0.720	5.3	0.687	100.0	0.720	5.3	-13.8
	R	10	10	15.16	22.29	7.13	0.713			0.713				
	S	10	10	14.47	21.51	7.04	0.704			0.704				
	T	10	10	14.32	22.07	7.75	0.775			0.775				
100%	U	10	10	15.05	21.23	6.18	0.618	0.689	7.2	0.618	97.5	0.671	6.0	-6.0
	V	10	10	14.54	21.59	7.05	0.705			0.705				
	W	10	9	14.49	21.10	6.61	0.734			0.661				
	X	10	10	15.69	22.68	6.99	0.699			0.699				
100% Intake	Y	10	10	14.84	21.73	6.89	0.689	0.647	7.3	0.689	97.5	0.630	7.3	0.4
	Z	10	9	14.35	20.42	6.07	0.674			0.607				
	AA	10	10	15.23	21.65	6.42	0.642			0.642				
	BB	10	10	14.79	20.62	5.83	0.583			0.583				

Outfall 101:
Dunnett's MSD value: ~~0.0693~~ 0.0633
PMSD: ~~11.0~~ 10.0

Intake:
Dunnett's MSD value: 0.0624
PMSD: 9.9

MSD = Minimum Significant Difference
PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 14.0% 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.

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 16-23, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/16/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN 101, Non-treated
End Date:	11/23/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

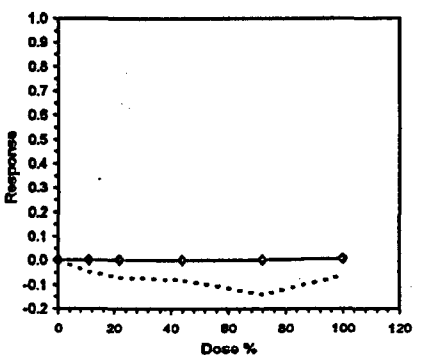
Conc-%	1	2	3	4
D-Control	0.6630	0.6530	0.5660	0.6480
10.98	0.6170	0.6630	0.6770	0.6890
22	0.6290	0.6990	0.7040	0.6710
43.9	0.6890	0.6360	0.7010	0.7040
72	0.6870	0.7130	0.7040	0.7750
100	0.6180	0.7050	0.6610	0.6990

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6325	1.0000	0.6325	0.5660	0.6630	7.078	4				0.6744	1.0000
10.98	0.6615	1.0458	0.6615	0.6170	0.6890	4.764	4	-1.105	2.410	0.0633	0.6744	1.0000
22	0.6758	1.0684	0.6758	0.6290	0.7040	5.088	4	-1.647	2.410	0.0633	0.6744	1.0000
43.9	0.6825	1.0791	0.6825	0.6360	0.7040	4.640	4	-1.904	2.410	0.0633	0.6744	1.0000
72	0.7198	1.1379	0.7198	0.6870	0.7750	5.332	4	-3.323	2.410	0.0633	0.6744	1.0000
100	0.6708	1.0605	0.6708	0.6180	0.7050	5.994	4	-1.457	2.410	0.0633	0.6708	0.9946

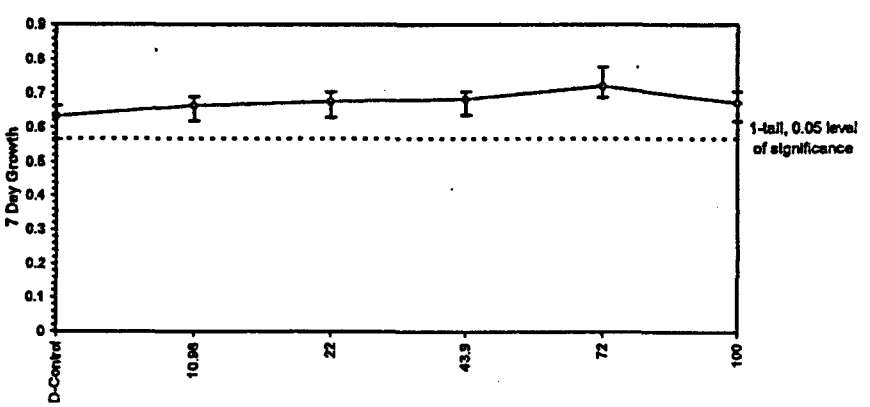
Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.92524302		0.884		-0.56720585	-0.7081248
Bartlett's Test indicates equal variances (p = 0.99)					0.532381773		15.08627224			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	100	>100		1	0.063273977	0.100037909	0.003245742	0.001378625	0.082390115	5, 18
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



TVA / Sequoyah Nuclear Plant, Intake
Non-treated
November 16-23, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/16/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN Intake, Non-treated
End Date:	11/23/2005	Lab ID:	BTS-Envir. Testing Sol	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.6630	0.6530	0.5660	0.6480
100	0.6890	0.6070	0.6420	0.5830

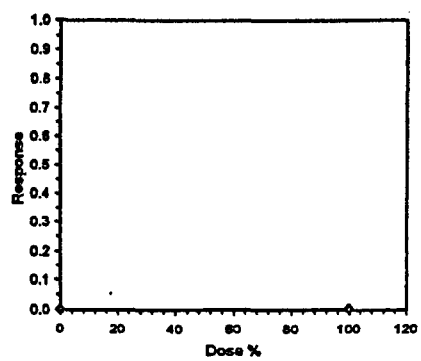
			Transform: Untransformed						1-Tailed		Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6325	1.0000	0.6325	0.5660	0.6630	7.078	4				0.6325	1.0000
100	0.6303	0.9964	0.6303	0.5830	0.6890	7.307	4	0.070	1.943	0.0624	0.6303	0.9964

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.946636319	0.749	-0.42796383	-0.74660924
F-Test indicates equal variances (p = 0.96)	1.058165669	47.46722794		

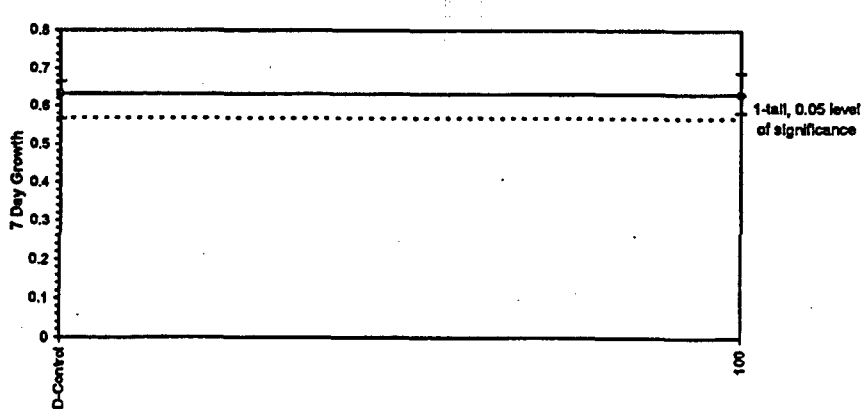
Hypothesis Test (1-tail, 0.05)	MSDu	MSDp	MSB	MSE	F-Prob	df
Homoscedastic t Test indicates no significant differences	0.062403399	0.0986615	1.0125E-05	0.002062625	0.946420491	1, 6
Treatments vs D-Control						

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



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

Client: TVA
Facility: Sequoyah Nuclear Plant
NPDES #: TN 0026450
Project #: 7220

County: Hamilton
Treatment: Non-treated
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.98	22	43.9	72	100	
Effluent volume (mL)	274.5	550	1097.5	1800	2500	
Diluent volume (mL)	2225.5	1950	1402.5	700	0	
Total volume (mL)	2500	2500	2500	2500	2500	

Test organism information:		*Test information:*	
Organism age:	< 24-HOURS OLD	Randomizing template:	GREEN & WHITE
Date and times organisms were born between:	11-16-05 0940 TO 1112	Incubator number and shelf location:	ZC1 #2
Organism source:	11-08-05 A-F	YCT batch:	09-30-05
Transfer bowl information:	pH = 7.72 Temperature = 25.1°C	*Selenastrum* batch:	11-11-05

Daily renewal information:

Day	Date	Test initiation, renewal, or termination time	Control water batch used	Sample numbers used	Analyst
0	11-16-05	1237	11-11-05 A	051115.04 & 05	dl
1	11-17-05	1143	11-11-05 B	051115.04 & 05	dl
2	11-18-05	1200	11-11-05 B	051117.034 04	dl
3	11-19-05	1152	11-17-05 A	051117.034 04	dl
4	11-20-05	1216	11-17-05 A	051119.12 & 13	dl
5	11-21-05	1221	11-17-05 B	051119.12 & 13	dl
6	11-22-05	1218	11-17-05 B	051119.12 & 13	dl
7	11-23-05	1151			dl

Control information:	1	2	Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	0%	≤ 20%	7-day LC50	> 100%
% Adults having 3rd Broods:	100%	100%	≥ 80%	NOEC	100%
% Mortality:	0%	0%	≤ 20%	LOEC	> 100%
Mean Offspring/Female:	30.2	31.3	≥ 15.0 offspring/female	ChV	> 100%
% CV:	9.2%	5.7%	< 40.0 %	IC25	> 100%

6.9% dm 12-13-05

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated Date: 11-16-05

CONTROL - 1 *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	6	6	5	4	5	6	4	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	10	13	10	11	10	10	12	11	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	9	0	0	0	0	0	*2	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	14	15	18	13	16	16	13	12	16	13
Total young produced		29	31	36	27	32	32	29	28	31	27
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L
X for 3rd Broods		X	X	X	X	X	X	X	X	X	X

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	30.2

CONC: 10.98% *Survival and Reproduction Data*

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

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	31.9
% Reduction from Control:	-5.6%

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated **Date:** 11-16-05

CONC: 22% *Survival and Reproduction Data*

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

Note: Adult mortality (L = live, D = dead) *SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	32.3
% Reduction from Control:	-7.0%

CONC: 43.9% *Survival and Reproduction Data*

Day		Replicate number 1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	10	5	4	4	5	4	4	5	5	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	0	0	0	11	0	10	0	11	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	13	10	12	10	0	12	0	12	0	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	18	15	17	15	14	18	16	16	17	16
Total young produced		37	30	33	29	30	34	30	33	33	33
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	32.2
% Reduction from Control:	-6.6%

Species: ***Ceriodaphnia dubia***
Client: Sequoyah Nuclear Plant - Non-treated **Date:** 11-16-05

CONC: 72% ***Survival and Reproduction Data***

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	5	4	5	5	6	5	5	4	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	0	10	10	14	11	12	14	10	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	12	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	19	14	15	17	19	17	18	14	15	19
Total young produced		35	31	29	32	38	35	35	33	29	36
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	33.2
% Reduction from Control:	-9.9%

CONC: 100% ***Survival and Reproduction Data***

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	6	6	4	4	4	6	5	5	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	0	11	11	13	10	11	11	*1	*2
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	13	0	0	0	0	0	0	*11	*10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	18	20	19	19	15	18	16	16	19	18
Total young produced		35	39	36	34	32	32	33	32	36	35
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

*SPLIT BROODS

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	34.4
% Reduction from Control:	-13.9%

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated **Date:** 11-16-05

CONTROL - 2 ***Survival and Reproduction Data***

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	6	4	4	5	5	5	6	5	4	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	0	0	11	0	11	12	11	13	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	11	10	12	0	10	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	16	14	14	13	15	14	16	16	17	16
Total young produced		33 (38)	28	30	29	30	30	34	32	34	33
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	31.3

CONC: 100% Intake ***Survival and Reproduction Data***

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	6	5	6	5	5	6	5	4	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	0	11	12	11	0	14	11	15	13
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	14	14	0	0	0	11	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	16	20	19	17	17	20	18	19	18	16
Total young produced		35	40	35	35	33	36	38	35	37	34
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	35.8
% Reduction from Control:	-14.4%

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 16-23, 2005

Verification of *Ceriodaphnia* Reproduction Totals

Control-1

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	6	6	5	4	5	6	4	4	4	4	48
5	0	10	13	10	11	10	10	12	11	10	97
6	9	0	0	0	0	0	2	0	0	0	11
7	14	15	18	13	16	16	13	12	16	13	146
Total	29	31	36	27	32	32	29	28	31	27	302

10.98%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	5	5	4	5	5	5	6	5	5	50
5	10	0	11	0	10	11	12	0	12	14	80
6	0	12	0	10	0	0	0	11	0	0	33
7	16	18	16	16	15	18	14	14	15	14	156
Total	31	35	32	30	30	34	31	31	32	33	319

22%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	6	5	5	4	5	4	6	4	5	5	49
5	9	0	12	11	1	0	0	11	0	0	44
6	0	12	0	0	11	12	12	0	10	10	67
7	19	15	15	17	16	15	15	17	19	15	163
Total	34	32	32	32	33	31	33	32	34	30	323

43.9%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	6	5	4	4	5	4	4	5	5	5	47
5	0	0	0	0	11	0	10	0	11	0	32
6	13	10	12	10	0	12	0	12	0	12	81
7	18	15	17	15	14	18	16	16	17	16	162
Total	37	30	33	29	30	34	30	33	33	33	322

72%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	5	4	5	5	6	5	5	4	5	49
5	11	0	10	10	14	11	12	14	10	12	104
6	0	12	0	0	0	0	0	0	0	0	12
7	19	14	15	17	19	17	18	14	15	19	167
Total	35	31	29	32	38	34	35	33	29	36	332

100%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	6	6	4	4	4	6	5	5	5	50
5	12	0	11	11	13	10	11	11	1	2	82
6	0	13	0	0	0	0	0	0	11	10	34
7	18	20	19	19	15	18	16	16	19	18	178
Total	35	39	36	34	32	32	33	32	36	35	344

Control-2

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	6	4	4	5	5	5	6	5	4	5	49
5	0	0	0	11	0	11	12	11	13	12	70
6	11	10	12	0	10	0	0	0	0	0	43
7	16	14	14	13	15	14	16	16	17	16	151
Total	33	28	30	29	30	30	34	32	34	33	313

100% Intake

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	6	5	6	5	5	6	5	4	5	52
5	0	0	11	12	11	0	14	11	15	13	87
6	14	14	0	0	0	11	0	0	0	0	39
7	16	20	19	17	17	20	18	19	18	16	180
Total	35	40	35	35	33	36	38	35	37	34	358

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 16-23, 2005

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

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

Project number: 2220

Reveiwed by:

Concentration (%)	Replicate number										Survival (%)	Average reproduction (offspring/female)	Coefficient of variation (%)	Percent reduction from pooled controls (%)
	1	2	3	4	5	6	7	8	9	10				
Control - 1	29	31	36	27	32	32	29	28	31	27	100	30.2	9.2	Not applicable
10.98%	31	35	32	30	30	34	31	31	32	33	100	31.9	5.2	-5.6
22%	34	32	32	32	33	31	33	32	34	30	100	32.3	3.9	-7.0
43.9%	37	30	33	29	30	34	30	33	33	33	100	32.2	7.6	-6.6
72%	35	31	29	32	38	34	35	33	29	36	100	33.2	9.0	-9.9
100%	35	39	36	34	32	32	33	32	36	35	100	34.4	6.6	-13.9
Control - 2	33	28	30	29	30	30	34	32	34	33	100	31.3	6.9	Not applicable
100% Intake	35	40	35	35	33	36	38	35	37	34	100	35.8	5.7	-14.4

Outfall 101:
Dunnett's MSD value: 2.362
PMSD: 7.8

Intake:
Dunnett's MSD value: 1.632
PMSD: 5.2

MSD = Minimum Significant Difference
PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, Inc. chronic toxicity tests when a toxicant reduces *Ceriodaphnia* reproduction by 9.7% from the control.

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

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

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

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

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 16-23, 2005

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	11/16/2005	Test ID:	CdFRCR	Sample ID:	TVA / SQN 101, Non-treated
End Date:	11/23/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia
Comments:					

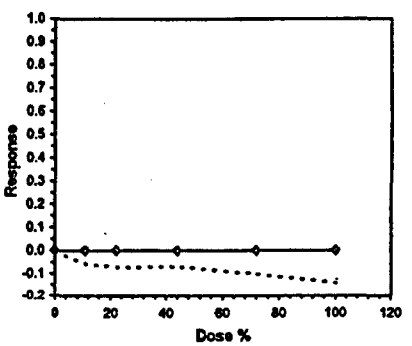
Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	29.000	31.000	36.000	27.000	32.000	32.000	29.000	28.000	31.000	27.000
10.98	31.000	35.000	32.000	30.000	30.000	34.000	31.000	31.000	32.000	33.000
22	34.000	32.000	32.000	32.000	33.000	31.000	33.000	32.000	34.000	30.000
43.9	37.000	30.000	33.000	29.000	30.000	34.000	30.000	33.000	33.000	33.000
72	35.000	31.000	29.000	32.000	38.000	34.000	35.000	33.000	29.000	36.000
100	35.000	39.000	36.000	34.000	32.000	32.000	33.000	32.000	36.000	35.000

			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	30.200	1.0000	30.200	27.000	36.000	9.208	10				32.367	1.0000
10.98	31.900	1.0563	31.900	30.000	35.000	5.214	10	-1.645	2.287	2.362	32.367	1.0000
22	32.300	1.0695	32.300	30.000	34.000	3.875	10	-2.033	2.287	2.362	32.367	1.0000
43.9	32.200	1.0662	32.200	29.000	37.000	7.579	10	-1.936	2.287	2.362	32.367	1.0000
72	33.200	1.0993	33.200	29.000	38.000	8.958	10	-2.904	2.287	2.362	32.367	1.0000
100	34.400	1.1391	34.400	32.000	39.000	6.601	10	-4.065	2.287	2.362	32.367	1.0000

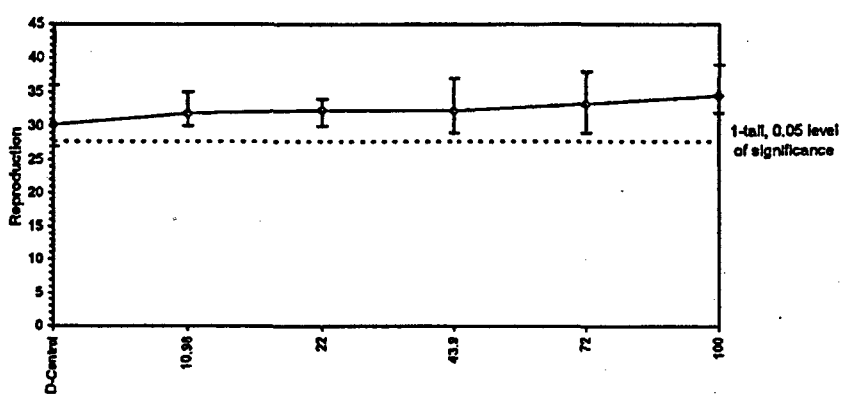
Auxiliary Tests					Statistic		Critical		Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)					0.714224041		1.035		0.403227399	0.084524151
Bartlett's Test indicates equal variances (p = 0.15)					8.037992477		15.08627224			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	100	>100		1	2.36247902	0.078227782	19.54666667	5.337037037	0.006318601	5, 54
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



sqn_11-16-05data

TVA / Sequoyah Nuclear Plant, Intake
Non-treated
November 16-23, 2005

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	11/16/2005	Test ID:	CdFRCR	Sample ID:	TVA / SQN Intake, Non-treated
End Date:	11/23/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia
Comments:					

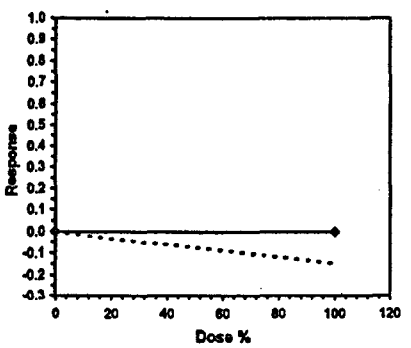
Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	33.000	28.000	30.000	29.000	30.000	30.000	34.000	32.000	34.000	33.000
100	35.000	40.000	35.000	35.000	33.000	36.000	38.000	35.000	37.000	34.000

			Transform: Untransformed						1-Tailed		Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	31.300	1.0000	31.300	28.000	34.000	6.910	10				33.550	1.0000
100	35.800	1.1438	35.800	33.000	40.000	5.709	10	-4.782	1.734	1.632	33.550	1.0000

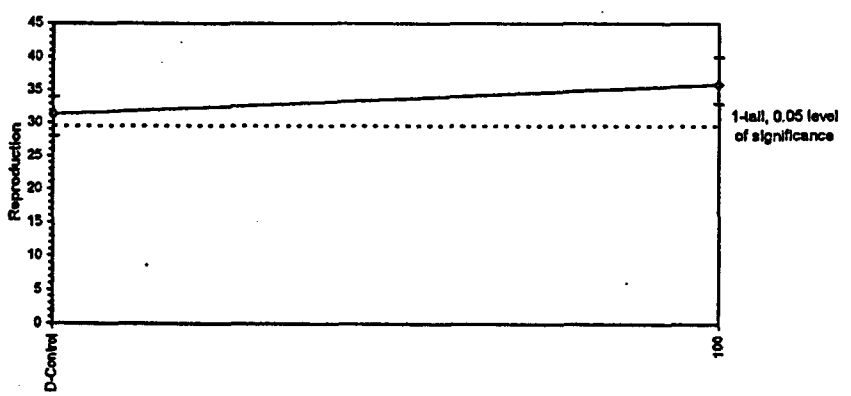
Auxiliary Tests	Statistic	Critical			Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.958691359	0.868			0.350417145	-0.71197525
F-Test indicates equal variances (p = 0.87)	1.119680882	6.541089535				
Hypothesis Test (1-tail, 0.05)	MSDu	MSDp	MSB	MSE	F-Prob	df
Homoscedastic t Test indicates no significant differences	1.631822565	0.052134906	101.25	4.427777778	1.5E-04	1, 18
Treatments vs D-Control						

Linear Interpolation (200 Resamples)

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



Dose-Response Plot



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

Client: TVA
Facility: Sequoyah Nuclear Plant
NPDES #: TN 0026450
Project #: 2220

County: Hamilton
Treatment: UV-treated
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.98	22	43.9	72	100	Each concentration was treated
Effluent volume (mL)	274.5	550	1097.5	1800	2500	for 2 minutes with a UV sterilizer
Diluent volume (mL)	2225.5	1950	1402.5	700	0	to remove pathogenic
Total volume (mL)	2500	2500	2500	2500	2500	interferences.

Test organism information:		*Test information:*	
Organism age:	22.75 TO 24.25 HOURS OLD	Randomizing template:	BLUE
Date and times organisms were born between:	11-15-05 1330 TO 1500	Incubator number:	3B
Organism source:	ABS BATCH #b 11-15-05	*Artemia* lot number:	BG18041T
Transfer bowl information:	pH = 8.10 Temperature = 24.2 °C	Total drying time:	24 HOURS
		Date / Time in:	11-23-05 1350
Average transfer volume:	10.4 mL	Date / Time out:	11-24-05 1356
		Oven temperature:	60°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	MHS batch used	Sample numbers used	Analyst
0	11-16-05	—x	1500	1344	11-11-05A	051115.04 & 05	dl
1	11-17-05	0900	1506	1317	11-11-05B	051115.04 & 05	dl
2	11-18-05	0912	1520	1300	11-11-05B	051117.03 & 04	dl
3	11-19-05	0847	1452	1248	11-17-05A	051117.03 & 04	dl
4	11-20-05	0905	1506	1254	11-17-05A	051119. 12 & 13	dl
5	11-21-05	0908	1511	1247	11-17-05B	051119. 12 & 13	dl
6	11-22-05	0960	1500	1250	11-17-05B	051119. 12 & 13	dl
7	11-23-05			1322			KEK

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	0%	≤ 20%	7-day LC_{50}	>100%
Average weight per initial larvae:	0.660		NOEC	100%
Average weight per surviving larvae:	0.660	≥ 0.25 mg/larvae	LOEC	>100%
			ChV	>100%
			IC_{25}	>100%

Species: *Pimephales promelas* **Date:** 11-16-05

Client: TVA / Sequoyah Nuclear Plant - UV-treated

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	ISM 10	10	10	10	LG 10	10	ISM 10	9d	10	10	10	ISM 10
A = Pan weight (mg) Color identification: Lt. Blue Analyst: JN	14.92	14.62	14.73	16.02	14.70	15.09	16.28	16.00	14.63	16.02	16.21	14.69
B = Pan + Larvae weight (mg) Analyst: KKleman	20.54	21.80	21.61	22.72	22.02	21.65	21.79	21.47	21.01	22.44	22.59	20.40
Larvae weight (mg) = A – B	5.62	7.18	6.88	6.70	7.32	6.56	5.51	5.47	6.38	6.42	6.38	5.71
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.562	0.718	0.688	0.670	0.732	0.656	0.551	0.547	0.638	0.642	0.638	0.571
Average weight per initial number of larvae (mg)	0.660				0.622		~~5.6%~~ 5.8%		0.622		5.6%	
Percent reduction from control (%)												

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Species: *Pimephales promelas*

Client: TVA / Sequoyah Nuclear Plant - UV-treated **Date:** 11-16-05

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	ISM 10	10	2LG 10	ISM 10	10	10	ISM 10	10	10	10	10	ISM 10
A = Pan weight (mg) Color identification: Lt. Blue Analyst: JN	14.75	16.12	16.08	15.92	15.06	16.37	15.17	16.29	14.82	14.78	14.73	14.97
B = Pan + Larvae weight (mg) Analyst: KKennard	20.00	22.14	23.84	21.55	21.23	25.15	20.82	22.67	20.99	21.68	20.84	20.61
Larvae weight (mg) = A – B	5.25	6.02	7.76	5.63	6.17	6.78	5.65	6.38	6.17	6.90	6.11	5.64
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.525	0.602	0.776	0.563	0.617	0.678	0.565	0.638	0.617	0.690	0.611	0.564
Average weight per initial number of larvae (mg)	0.617				0.625				0.621			
Percent reduction from control (%)	6.5%				5.3%				5.9%			

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Species: ***Pimephales promelas***

Client: TVA / Sequoyah Nuclear Plant - UV-treated **Date:** 11-16-05

Survival and Growth Data

Day		100% Intake			
		Y	Z	AA	BB
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) Color identification: Lt Blue Analyst: JN		15.02	14.87	19.66	14.83
B = Pan + Larvae weight (mg) Analyst: KKiernan		21.04	21.05	20.87	20.63
Larvae weight (mg) = A – B		6.02	6.18	6.21	5.80
Weight per initial number of larvae (mg) = C / Initial number of larvae		0.602	0.618	0.621	0.580
Average weight per initial number of larvae (mg)	Percent reduction from control (%)	0.605		8.2%	

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: JA

Comments:

TVA / Sequoyah Nuclear Plant, Outfall 101
UV-treated
November 16-23, 2005

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

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

Project number: 2220 **Reveiwed by:** cw

							Not for Compliance Assessment, Internal Laboratory QC							
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 / Surviving number of larvae (mg)	Mean weight / Surviving number of larvae (mg)	Coefficient of variation (Mean weight per surviving number of larvae) (%)	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight / Initial number of larvae (mg)	Coefficient of variation (Mean weight per initial number of larvae) (%)	Percent reduction from control (%)
Control	A	10	10	14.92	20.54	5.62	0.562	0.660	10.3	0.562	100.0	0.660	10.3	Not applicable
	B	10	10	14.62	21.80	7.18	0.718			0.718				
	C	10	10	14.73	21.61	6.88	0.688			0.688				
	D	10	10	16.02	22.72	6.70	0.670			0.670				
10.98%	E	10	10	14.70	22.02	7.32	0.732	0.637	12.0	0.732	97.5	0.622	14.4	5.8
	F	10	10	15.09	21.65	6.56	0.656			0.656				
	G	10	10	16.28	21.79	5.51	0.551			0.551				
	H	10	9	16.00	21.47	5.47	0.608			0.547				
22%	I	10	10	14.63	21.01	6.38	0.638	0.622	5.5	0.638	100.0	0.622	5.5	5.6
	J	10	10	16.02	22.44	6.42	0.642			0.642				
	K	10	10	16.21	22.59	6.38	0.638			0.638				
	L	10	10	14.69	20.40	5.71	0.571			0.571				
43.9%	M	10	10	14.75	20.00	5.25	0.525	0.617	18.0	0.525	100.0	0.617	18.0	6.5
	N	10	10	16.12	22.14	6.02	0.602			0.602				
	O	10	10	16.08	23.84	7.76	0.776			0.776				
	P	10	10	15.92	21.55	5.63	0.563			0.563				
72%	Q	10	10	15.06	21.23	6.17	0.617	0.625	7.5	0.617	100.0	0.625	7.5	5.3
	R	10	10	16.37	23.15	6.78	0.678			0.678				
	S	10	10	15.17	20.82	5.65	0.565			0.565				
	T	10	10	16.29	22.67	6.38	0.638			0.638				
100%	U	10	10	14.82	20.99	6.17	0.617	0.621	8.4	0.617	100.0	0.621	8.4	5.9
	V	10	10	14.78	21.68	6.90	0.690			0.690				
	W	10	10	14.73	20.84	6.11	0.611			0.611				
	X	10	10	14.97	20.61	5.64	0.564			0.564				
100% Intake	Y	10	10	15.02	21.04	6.02	0.602	0.605	3.1	0.602	100.0	0.605	3.1	8.2
	Z	10	10	14.87	21.05	6.18	0.618			0.618				
	AA	10	10	14.66	20.87	6.21	0.621			0.621				
	BB	10	10	14.83	20.63	5.80	0.580			0.580				

Outfall 101:
Dunnett's MSD value: 0.1224
PMSD: 18.6

Intake:
Dunnett's MSD value: 0.0685
PMSD: 10.4

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, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 14.0% 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.

TVA / Sequoyah Nuclear Plant, Outfall 101
UV-treated
November 16-23, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/16/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN 101, UV-treated
End Date:	11/23/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.5620	0.7180	0.6880	0.6700
10.98	0.7320	0.6560	0.5510	0.5470
22	0.6380	0.6420	0.6380	0.5710
43.9	0.5250	0.6020	0.7760	0.5630
72	0.6170	0.6780	0.5650	0.6380
100	0.6170	0.6900	0.6110	0.5640

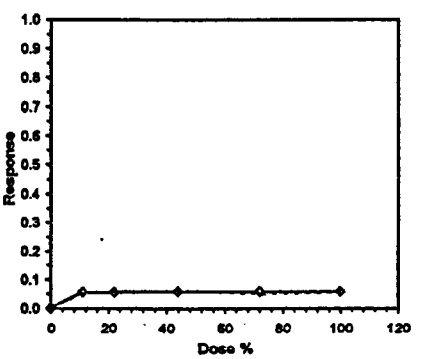
			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6595	1.0000	0.6595	0.5620	0.7180	10.303	4				0.6595	1.0000
10.98	0.6215	0.9424	0.6215	0.5470	0.7320	14.368	4	0.748	2.410	0.1224	0.6219	0.9429
22	0.6223	0.9435	0.6223	0.5710	0.6420	5.499	4	0.733	2.410	0.1224	0.6219	0.9429
43.9	0.6165	0.9348	0.6165	0.5250	0.7760	17.986	4	0.846	2.410	0.1224	0.6205	0.9409
72	0.6245	0.9469	0.6245	0.5650	0.6780	7.534	4	0.689	2.410	0.1224	0.6205	0.9409
100	0.6205	0.9409	0.6205	0.5640	0.6900	8.387	4	0.768	2.410	0.1224	0.6205	0.9409

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.958492696		0.884		0.561262513	0.354520295
Bartlett's Test indicates equal variances (p = 0.45)					4.741466045		15.08627224			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSE**	**MSE**	**F-Prob**	**df**
Dunnett's Test	100	>100		1	0.122449555	0.185670288	0.001013242	0.005163097	0.959918439	5, 18
Treatments vs D-Control										

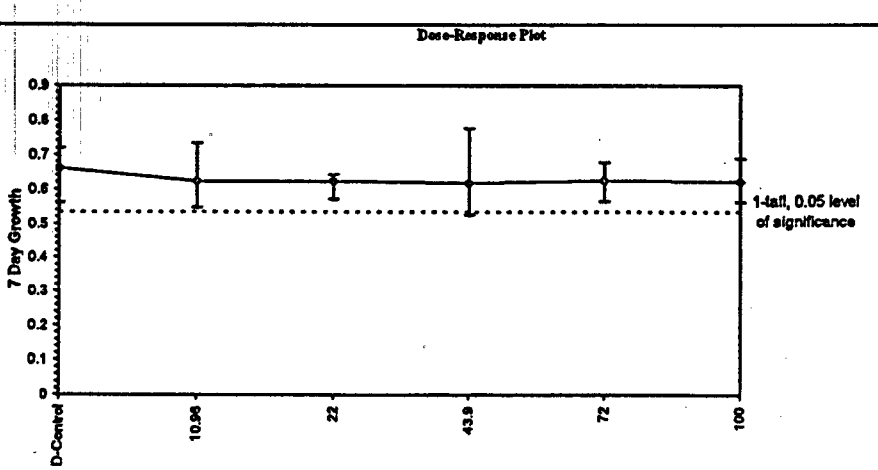
Linear Interpolation (200 Resamples)

Point	%	SD	95% CL(Exp)	Skew
IC05*	9.6230			
IC10	>100			
IC15	>100			
IC20	>100			
IC25	>100			
IC40	>100			
IC50	>100			

* indicates IC estimate less than the lowest concentration



Dose-Response Plot



TVA / Sequoyah Nuclear Plant, Intake
UV-treated
November 16-23, 2005

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/16/2005	Test ID:	PpFRCR	Sample ID:	TVA / SQN Intake, UV-treated
End Date:	11/23/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

Conc-%	1	2	3	4
D-Control	0.5620	0.7180	0.6880	0.6700
100	0.6020	0.6180	0.6210	0.5800

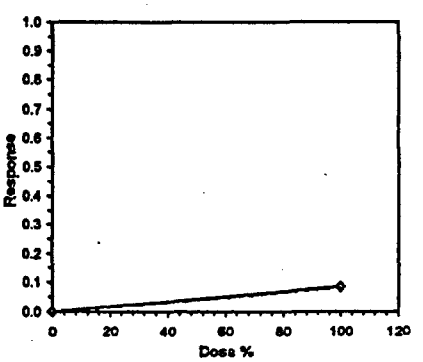
			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
D-Control	0.6595	1.0000	0.6595	0.5620	0.7180	10.303	4				0.6595	1.0000
100	0.6053	0.9177	0.6053	0.5800	0.6210	3.104	4	1.539	1.943	0.0685	0.6053	0.9177

Auxiliary Tests	Statistic	Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.884777725	0.749		-1.38060549	2.902723251
F-Test indicates equal variances (p = 0.05)	13.08240891	47.46722794			
Hypothesis Test (1-tail, 0.05)	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Homoscedastic t Test indicates no significant differences	0.068494809	0.103858695	0.005886125	0.002484958	0.174713358	1, 6
Treatments vs D-Control						

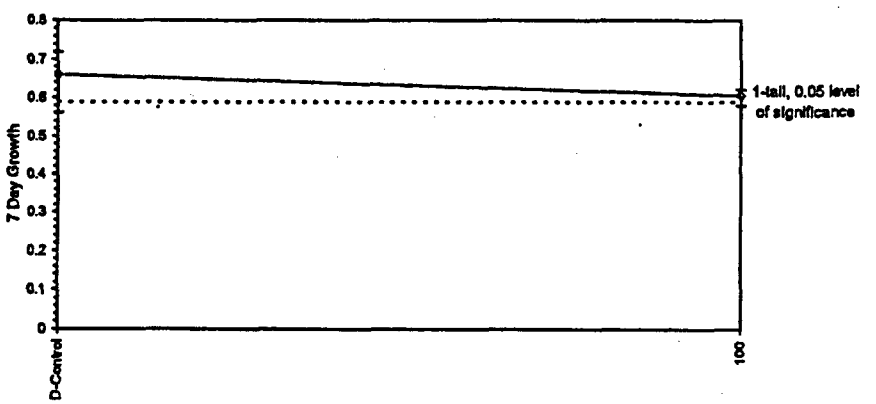
Linear Interpolation (200 Resamples)

Point	%	SD	95% CL(Exp)	Skew
IC05*	60.783			
IC10	>100			
IC15	>100			
IC20	>100			
IC25	>100			
IC40	>100			
IC50	>100			

* indicates IC estimate less than the lowest concentration



Dose-Response Plot



sqn_11-16-05data

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 16-23, 2005

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

Daily Chemical Analyses

Project number: 2220 **Reviewed by:** CM

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.68	7.92	7.99	8.02	8.05	7.86	8.03	7.86	7.99	7.90	8.00	7.92	7.98	7.71
	DO (mg/L)	7.7	7.7	7.7	7.5	7.7	7.4	7.8	7.7	7.8	7.3	7.9	7.5	7.8	7.0
	Conductivity (µmhos/cm)	310		317		318		314		321		321		307	
	Alkalinity (mg/L $CaCO_3$)	61		62				61				58			
	Hardness (mg/L $CaCO_3$)	95		95				92				96			
	Temperature (°C)	24.9	24.7	24.8	24.6	25.0	24.8	25.1	24.7	24.9	24.8	24.9	24.7	24.7	24.9
10.98%	pH (SU)	7.92	7.86	8.00	8.00	8.10	7.77	8.03	7.80	8.03	7.87	8.02	7.87	8.05	7.62
	DO (mg/L)	7.4	7.5	7.9	7.5	7.6	7.2	7.8	7.5	8.0	7.4	8.0	7.5	7.7	6.9
	Conductivity (µmhos/cm)	298		300		303		292		300		292		288	
	Temperature (°C)	25.1	24.9	24.8	24.4	24.9	24.7	25.1	24.6	25.0	24.8	24.8	24.9	24.8	24.7
22%	pH (SU)	7.93	7.87	8.00	7.96	8.10	7.73	8.02	7.84	8.04	7.85	8.03	7.87	8.05	7.66
	DO (mg/L)	7.4	7.5	7.8	7.6	7.7	6.9	7.7	7.6	7.9	7.3	7.9	7.3	7.8	6.7
	Conductivity (µmhos/cm)	284		281		287		276		288		280		276	
	Temperature (°C)	25.1	24.9	24.9	24.7	24.9	24.7	25.0	24.6	25.0	24.6	24.8	24.9	24.8	24.9
43.9%	pH (SU)	7.93	7.89	8.00	7.93	8.11	7.79	8.00	7.86	8.04	7.93	8.03	7.93	8.04	7.67
	DO (mg/L)	7.5	7.4	7.7	7.3	7.6	7.2	7.6	7.5	7.9	7.3	7.9	7.3	7.8	6.8
	Conductivity (µmhos/cm)	256		259		270		250		260		253		248	
	Temperature (°C)	25.1	24.7	24.9	24.6	25.1	24.9	25.2	24.6	25.0	24.6	24.8	24.8	24.8	24.8
72%	pH (SU)	7.94	7.90	8.01	7.97	8.10	7.79	7.99	7.84	8.05	7.95	8.03	7.93	8.03	7.68
	DO (mg/L)	7.5	7.3	7.8	7.5	7.6	7.2	7.6	7.5	7.8	7.3	7.9	7.5	7.8	6.9
	Conductivity (µmhos/cm)	219		223		217		212		222		221		214	
	Temperature (°C)	25.1	24.7	25.0	24.6	25.1	24.8	25.2	24.9	24.9	24.7	24.8	24.8	24.9	25.0
100%	pH (SU)	7.95	7.88	8.02	7.95	8.10	7.76	7.98	7.84	8.06	7.96	8.03	7.93	8.03	7.68
	DO (mg/L)	7.5	7.3	7.8	7.7	7.7	6.9	7.6	7.5	7.7	7.3	8.0	7.5	7.8	6.7
	Conductivity (µmhos/cm)	183		186		178		175		183		183		180	
	Alkalinity (mg/L $CaCO_3$)	63				62				65					
	Hardness (mg/L $CaCO_3$)	72				70				74					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	25.1	24.7	25.0	24.6	25.2	24.8	25.2	24.7	24.9	24.7	24.8	24.8	24.8	24.9
100% Intake	pH (SU)	7.94	7.88	8.01	7.97	8.09	7.77	7.95	7.87	8.04	7.93	8.02	7.94	8.03	7.68
	DO (mg/L)	7.8	7.4	7.8	7.7	7.6	6.9	7.7	7.5	7.8	7.2	7.9	7.3	7.8	6.9
	Conductivity (µmhos/cm)	184		187		177		173		183		182		180	
	Alkalinity (mg/L $CaCO_3$)	64				61				63					
	Hardness (mg/L $CaCO_3$)	76				72				76					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	24.9	24.8	25.0	24.6	25.0	24.8	25.2	24.7	25.1	24.7	24.9	24.8	24.9	24.9

Species: *Pimephales promelas* Date: 11-16-05
Client: TVA / Sequoyah Nuclear Plant – Non-treated

Daily Chemistry:

		Day 0		Day 1 KEK		Day 2	
	Analyst	AAB	AAB	AAB	AAB	AAB	JN
Concentration	Parameter						
CONTROL HHSW	pH (S.U.)	7.68	7.92	7.99	8.02	8.05	7.86
	DO (mg/L)	7.7	7.7	7.7	7.5	7.7	7.4
	Conductivity (µmhos/cm)	310		317		318	
	Alkalinity (mg $CaCO_3$/L)	61		(62)			
	Hardness (mg $CaCO_3$/L)	95		(95)		K	
	Temperature (°C)	24.9	24.7	24.8	24.6	25.0	24.8
10.98%	pH (S.U.)	7.92	7.86	8.00	8.00	8.10	7.77
	DO (mg/L)	7.4	7.5	7.9	7.5	7.6	7.2
	Conductivity (µmhos/cm)	298		300		303	
	Temperature (°C)	25.1	24.9	24.8	24.4	24.9	24.7
22%	pH (S.U.)	7.93	7.87	8.00	7.96	8.10	7.73
	DO (mg/L)	7.4	7.5	7.8	7.6	7.7	6.9
	Conductivity (µmhos/cm)	284		281		287	
	Temperature (°C)	25.1	24.9	24.9	24.7	24.9	24.7
43.9%	pH (S.U.)	7.93	7.89	8.00	7.93	8.11	7.79
	DO (mg/L)	7.5	7.4	7.7	7.3	7.6	7.2
	Conductivity (µmhos/cm)	256		259		270	
	Temperature (°C)	25.1	24.7	24.9	24.6	25.1	24.9
72%	pH (S.U.)	7.94	7.90	8.01	7.97	8.10	7.79
	DO (mg/L)	7.5	7.3	7.8	7.5	7.6	7.2
	Conductivity (µmhos/cm)	219		223		217	
	Temperature (°C)	25.1	24.7	25.0	24.6	25.1	24.8
100%	pH (S.U.)	7.95	7.88	8.02	7.95	8.10	7.76
	DO (mg/L)	7.5	7.3	7.8	7.7	7.7	6.9
	Conductivity (µmhos/cm)	183		186		178	
	Alkalinity (mg $CaCO_3$/L)	63				62	
	Hardness (mg $CaCO_3$/L)	72				70	
	TR chlorine (mg/L)	<0.10				<0.10	
	Temperature (°C)	25.1	24.7	25.0	24.6	25.2	24.8
100% Intake	pH (S.U.)	7.94	7.88	8.01	7.97	8.09	7.77
	DO (mg/L)	7.8	7.4	7.8	7.7	7.6	6.9
	Conductivity (µmhos/cm)	184		187		177	
	Alkalinity (mg $CaCO_3$/L)	64				61	
	Hardness (mg $CaCO_3$/L)	76				72	
	TR chlorine (mg/L)	<0.10				<0.10	
	Temperature (°C)	24.9	24.8	25.0	24.6	25.0	24.8
		Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas* Date: 11-16-05
Client: TVA / Sequoyah Nuclear Plant – Non-treated

		Day							
		3		4		5		6	
	Analyst	JN	JN	JN	KEK	KEK	KEK	KEK	KEK
Concentration	Parameter								
CONTROL mHSW	pH (S.U.)	8.03	7.86	7.99	7.90	8.00	7.92	7.98	7.71
	DO (mg/L)	7.8	7.7	7.8	7.3	7.9	7.5	7.8	7.0
	Conductivity (µmhos/cm)	314		321		321		307	
	Alkalinity (mg $CaCO_3$/L)	61				(58)			
	Hardness (mg $CaCO_3$/L)	92				(96)			
	Temperature (°C)	25.1	24.7	24.9	24.8	24.9	24.7	24.7	24.9
10.98%	pH (S.U.)	8.03	7.80	8.03	7.87	8.02	7.87	8.05	7.67
	DO (mg/L)	7.8	7.5	8.0	7.4	8.0	7.5	7.7	6.9
	Conductivity (µmhos/cm)	292		300		292		288	
	Temperature (°C)	25.1	24.6	25.0	24.8	24.8	24.9	24.8	24.7
22%	pH (S.U.)	8.02	7.84	8.04	7.85	8.03	7.87	8.05	7.66
	DO (mg/L)	7.7	7.6	7.9	7.3	7.9	7.3	7.8	6.7
	Conductivity (µmhos/cm)	276		288		280		276	
	Temperature (°C)	25.0	24.6	25.0	24.6	24.8	24.9	24.8	24.9
43.9%	pH (S.U.)	8.00	7.86	8.04	7.93	8.03	7.93	8.04	7.67
	DO (mg/L)	7.6	7.5	7.9	7.3	7.9	7.3	7.8	6.9
	Conductivity (µmhos/cm)	250		260		253		248	
	Temperature (°C)	25.2	24.6	25.0	24.6	24.8	24.8	24.8	24.8
72%	pH (S.U.)	7.99	7.84	8.05	7.95	8.03	7.93	8.03	7.68
	DO (mg/L)	7.6	7.5	7.8	7.3	7.9	7.5	7.8	6.9
	Conductivity (µmhos/cm)	212		222		221		214	
	Temperature (°C)	25.2	24.9	24.9	24.7	24.8	24.8	24.9	25.0
100%	pH (S.U.)	7.98	7.84	8.06	7.96	8.03	7.93	8.03	7.68
	DO (mg/L)	7.6	7.5	7.7	7.3	8.0	7.5	7.8	6.7
	Conductivity (µmhos/cm)	175		183		183		180	
	Alkalinity (mg $CaCO_3$/L)			65					
	Hardness (mg $CaCO_3$/L)			74					
	TR Chlorine (mg/L)			<0.10					
	Temperature (°C)	25.2	24.7	24.9	24.7	24.8	24.8	24.8	24.9
100% Intake	pH (S.U.)	7.95	7.87	8.04	7.93	8.02	7.94	8.03	7.68
	DO (mg/L)	7.7	7.5	7.8	7.2	7.9	7.3	7.8	6.9
	Conductivity (µmhos/cm)	173		183		182		180	
	Alkalinity (mg $CaCO_3$/L)			63					
	Hardness (mg $CaCO_3$/L)			76					
	TR chlorine (mg/L)			<0.10					
	Temperature (°C)	25.2	24.7	25.1	24.7	24.9	24.8	24.9	24.9
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

TVA / Sequoyah Nuclear Plant, Outfall 101
Non-treated
November 16-23, 2005

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

Daily Chemical Analyses

Project number: 2220

Reviewed by: Cer

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.68	7.93	7.99	8.04	8.05	8.07	8.03	7.98	7.99	7.98	8.00	8.03	7.98	8.00
	DO (mg/L)	7.7	7.5	7.7	7.8	7.7	8.0	7.8	7.8	7.8	7.9	7.9	7.8	7.8	7.8
	Conductivity (µmhos/cm)	310		317		318		314		321		321		307	
	Alkalinity (mg/L $CaCO_3$)	61		62				61				58			
	Hardness (mg/L $CaCO_3$)	95		95				92				96			
	Temperature (°C)	24.9	25.0	24.7	25.1	24.9	25.2	24.9	25.2	24.8	24.9	24.9	24.9	24.8	25.3
10.98%	pH (SU)	7.92	7.96	8.00	8.03	8.10	8.09	8.03	8.01	8.03	7.98	8.02	8.03	8.05	8.01
	DO (mg/L)	7.4	7.5	7.9	7.9	7.6	8.2	7.8	7.8	8.0	8.0	8.0	8.0	7.7	7.8
	Conductivity (µmhos/cm)	298		300		303		292		300		292		288	
	Temperature (°C)	24.9	25.2	24.9	25.3	24.8	25.2	25.0	25.1	24.8	25.2	24.8	24.9	24.7	25.3
22%	pH (SU)	7.93	7.99	8.00	8.01	8.10	8.11	8.02	8.00	8.04	7.99	8.03	8.02	8.05	8.05
	DO (mg/L)	7.4	7.6	7.8	7.9	7.7	8.3	7.7	7.8	7.9	8.0	7.9	7.9	7.8	7.9
	Conductivity (µmhos/cm)	284		281		287		276		288		280		276	
	Temperature (°C)	24.8	25.2	24.9	25.2	24.9	25.2	24.9	25.1	25.0	25.2	24.8	24.9	24.7	25.4
43.9%	pH (SU)	7.93	7.97	8.00	8.05	8.11	8.11	8.00	8.00	8.04	8.00	8.03	8.03	8.04	8.06
	DO (mg/L)	7.5	7.6	7.7	8.0	7.6	8.3	7.6	7.7	7.9	7.9	7.9	7.9	7.8	7.9
	Conductivity (µmhos/cm)	256		259		270		250		260		253		248	
	Temperature (°C)	24.8	25.1	24.9	25.2	25.0	25.2	24.9	25.0	25.0	24.9	24.8	24.8	24.9	25.2
72%	pH (SU)	7.94	7.99	8.01	8.09	8.10	8.14	7.99	8.03	8.05	8.00	8.03	8.06	8.03	8.09
	DO (mg/L)	7.5	7.5	7.8	8.0	7.6	8.1	7.6	7.8	7.8	7.8	7.9	7.8	7.8	8.0
	Conductivity (µmhos/cm)	219		223		217		212		222		221		214	
	Temperature (°C)	24.7	25.1	25.0	25.2	25.1	25.0	24.9	25.3	24.9	25.0	24.7	24.9	24.9	25.3
100%	pH (SU)	7.95	8.04	8.02	8.10	8.10	8.16	7.98	8.05	8.06	8.01	8.03	8.08	8.03	8.10
	DO (mg/L)	7.5	7.5	7.8	8.1	7.7	8.3	7.6	7.8	7.7	7.7	8.0	7.8	7.8	8.1
	Conductivity (µmhos/cm)	183		186		178		175		183		183		180	
	Alkalinity (mg/L $CaCO_3$)	63				62				65					
	Hardness (mg/L $CaCO_3$)	72				70				74					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	24.7	25.1	25.1	25.2	25.1	25.1	24.8	25.1	25.0	25.1	24.9	25.0	24.9	25.0
100% Intake	pH (SU)	7.94	7.98	8.01	8.09	8.09	8.13	7.95	8.04	8.04	8.02	8.02	8.07	8.03	8.11
	DO (mg/L)	7.8	7.4	7.8	8.1	7.6	8.1	7.7	7.7	7.8	7.9	7.9	8.0	7.8	8.1
	Conductivity (µmhos/cm)	184		187		177		173		183		182		180	
	Alkalinity (mg/L $CaCO_3$)	64				61				63					
	Hardness (mg/L $CaCO_3$)	76				72				76					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	24.9	25.2	24.9	25.2	24.9	25.2	24.9	25.0	24.9	25.1	24.9	24.8	24.8	25.3

Species: *Ceriodaphnia dubia* **Date:** 11-16-05
Client: Sequoyah Nuclear Plant - Non-treated

Daily Chemistry:

		Day 0		Day 1 KEK		Day 2 KEK		
	Analyst	AAB	AAB	AAB	AAB	AAB	JN	
Concentration	Parameter							
CONTROL	pH (S.U.)	7.68	7.93	7.99	8.04	8.05	8.07	
	DO (mg/L)	7.7	7.5	7.7	7.8	7.7	~~7.5~~	(8.0)
	Conductivity (µmhos/cm)	310		317		318		
	Alkalinity (mg $CaCO_3$/L)	61		62				
	Hardness (mg $CaCO_3$/L)	95		95				
	Temperature (°C)	24.9	25.0	24.7	25.1	24.9	25.2	
10.98%	pH (S.U.)	7.92	7.96	8.00	8.03	8.10	8.09	
	DO (mg/L)	7.4	7.5	7.9	7.9	7.6	~~7.5~~	(8.2)
	Conductivity (µmhos/cm)	298		300		303		
	Temperature (°C)	24.9	25.2	24.9	25.3	24.8	25.2	
22%	pH (S.U.)	7.93	7.99	8.00	8.01	8.10	8.11	
	DO (mg/L)	7.4	7.6	7.8	7.9	7.7	~~7.9~~	(8.3)
	Conductivity (µmhos/cm)	284		281		287		
	Temperature (°C)	24.8	25.2	24.9	25.2	24.9	25.2	
43.9%	pH (S.U.)	7.93	7.97	8.00	8.05	8.11	8.11	
	DO (mg/L)	7.5	7.6	7.7	8.0	7.6	~~7.7~~	(8.3)
	Conductivity (µmhos/cm)	256		259		270		
	Temperature (°C)	24.8	25.1	24.9	25.2	25.0	25.2	
72%	pH (S.U.)	7.94	7.99	8.01	8.09	8.10	8.14	
	DO (mg/L)	7.5	7.5	7.8	8.0	7.6	~~7.6~~	(8.1)
	Conductivity (µmhos/cm)	219		223		217		
	Temperature (°C)	24.7	25.1	25.0	25.2	25.1	25.0	
100%	pH (S.U.)	7.95	8.04	8.02	8.10	8.10	8.16	
	DO (mg/L)	7.5	7.5	7.8	8.1	7.7	~~7.5~~	(8.3)
	Conductivity (µmhos/cm)	183		186		178		
	Alkalinity (mg $CaCO_3$/L)	63				62		
	Hardness (mg $CaCO_3$/L)	72				70		
	TR chlorine (mg/L)	<0.10				<0.10		
	Temperature (°C)	24.7	25.1	25.1	25.2	25.1	25.1	
100% Intake	pH (S.U.)	7.94	7.98	8.01	8.09	8.09	8.13	
	DO (mg/L)	7.8	7.4 ~~8.4~~	7.8	8.1	7.6	~~7.5~~	(8.1)
	Conductivity (µmhos/cm)	184		187		177		
	Alkalinity (mg $CaCO_3$/L)	64				61		
	Hardness (mg $CaCO_3$/L)	76				72		
	TR chlorine (mg/L)	<0.10				<0.10		
	Temperature (°C)	24.9	25.2	24.9	25.2	24.9	25.2	
		Initial	Final	Initial	Final	Initial	Final	

Species: *Ceriodaphnia dubia*
Client: Sequoyah Nuclear Plant - Non-treated Date: 11-16-05

		Day							
		3		4		5		6	
	Analyst	JN	JN	JN	KEL	KEL	KEL	KEL	KEL
Concentration	Parameter								
CONTROL MHS	pH (S.U.)	8.03	7.98	~~8.03~~ (K)	7.98	8.00	8.03	7.98	8.00
	DO (mg/L)	7.8	7.8	7.8	7.9	7.9	7.8	7.8	7.8
	Conductivity (µmhos/cm)	314		321		321		307	
	Alkalinity (mg $CaCO_3$/L)	61		\		(58)		\	
	Hardness (mg $CaCO_3$/L)	92		K		(96) K		↓	
	Temperature (°C)	24.9	25.2	24.8	24.9	24.9	24.9	24.8	25.3
10.98%	pH (S.U.)	8.03	8.01	8.03	7.98	8.02	8.03	8.05	8.01
	DO (mg/L)	7.8	7.8	8.0	8.0	8.0	8.0	7.7	7.8
	Conductivity (µmhos/cm)	292		300		292		288	
	Temperature (°C)	25.0	25.1	24.8	25.2	24.8	24.9	24.7	25.3
22%	pH (S.U.)	8.02	8.00	8.04	7.99	8.03	8.02	8.05	8.05
	DO (mg/L)	7.7	7.8	7.9	8.0	7.9	7.9	7.8	7.9
	Conductivity (µmhos/cm)	276		288		280		276	
	Temperature (°C)	24.9	25.1	25.0	25.2	24.8	24.9	24.7	25.4
43.9%	pH (S.U.)	8.00	8.00	8.04	8.00	8.03	8.03	8.04	8.06
	DO (mg/L)	7.6	7.7	7.9	7.9	7.9	7.9	7.8	7.9
	Conductivity (µmhos/cm)	250		260		253		248	
	Temperature (°C)	24.9	25.0	25.0	24.9	24.8	24.8	24.9	25.2
72%	pH (S.U.)	7.99	8.03	8.05	8.00	8.03	8.06	8.03	8.09
	DO (mg/L)	7.6	7.8	7.8	7.8	7.9	7.8	7.8	8.0
	Conductivity (µmhos/cm)	212		222		221		214	
	Temperature (°C)	24.9	25.3	24.9	25.0	24.7	24.9	24.9	25.3
100%	pH (S.U.)	7.98	8.05	8.06	8.01	8.03	8.08	8.03	8.10
	DO (mg/L)	7.6	7.8	7.7	7.7	8.0	7.8	7.8	8.1
	Conductivity (µmhos/cm)	175		183		183		180	
	Alkalinity (mg $CaCO_3$/L)			65					
	Hardness (mg $CaCO_3$/L)			74					
	TR Chlorine (mg/L)			<0.10					
	Temperature (°C)	24.8	25.1	25.0	25.1	24.9	25.0	24.9	25.0
100% Intake	pH (S.U.)	7.95	8.04	8.04	8.02	8.02	8.07	8.03	8.11
	DO (mg/L)	7.7	7.7	7.8	7.9	7.9	8.0	7.8	8.1
	Conductivity (µmhos/cm)	173		183		182		180	
	Alkalinity (mg $CaCO_3$/L)			63					
	Hardness (mg $CaCO_3$/L)			76					
	TR chlorine (mg/L)			<0.10					
	Temperature (°C)	24.9	25.0	24.9	25.1	24.9	24.8	24.8	25.3
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

TVA / Sequoyah Nuclear Plant, Outfall 101
UV-treated
November 16-23, 2005

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

Daily Chemical Analyses

Project number: 2220

Reviewed by: JHunnicutt

Concentration	Parameter	Day 0		Day 1		Day 2		Day 3		Day 4		Day 5		Day 6	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Control	pH (SU)	7.92	7.91	7.96	7.98	8.11	7.74	8.02	7.85	8.04	7.85	7.99	7.81	8.02	7.66
	DO (mg/L)	7.8	7.5	7.6	7.5	7.6	7.3	7.9	7.6	7.7	7.4	7.9	7.3	7.8	7.0
	Conductivity (µmhos/cm)	302		314		311		305		305		310		290	
	Temperature (°C)	25.0	24.8	24.9	24.6	24.9	24.8	25.1	24.9	24.9	24.8	24.9	24.9	24.9	24.9
10.98%	pH (SU)	7.92	7.91	7.96	7.96	8.12	7.77	8.02	7.86	8.05	7.87	7.99	7.81	8.05	7.70
	DO (mg/L)	7.7	7.5	7.6	7.4	7.6	7.5	7.9	7.4	7.8	7.4	8.0	7.0	7.8	7.0
	Conductivity (µmhos/cm)	294		293		304		300		305		296		289	
	Temperature (°C)	25.2	24.9	25.1	24.6	25.1	24.9	25.2	24.9	25.1	24.8	25.0	25.2	25.0	24.8
22%	pH (SU)	7.93	7.92	7.97	7.98	8.12	7.86	8.02	7.89	8.06	7.86	7.98	7.91	8.05	7.66
	DO (mg/L)	7.7	7.6	7.6	7.5	7.5	7.6	7.9	7.6	7.9	7.4	7.9	7.0	7.8	6.8
	Conductivity (µmhos/cm)	279		279		289		288		291		283		275	
	Temperature (°C)	25.2	24.9	25.1	24.6	25.1	24.9	25.3	25.1	25.1	24.9	24.9	25.1	25.0	24.8
43.9%	pH (SU)	7.93	7.92	7.98	7.98	8.12	7.79	8.02	7.86	8.06	7.85	7.99	7.90	8.05	7.65
	DO (mg/L)	7.7	7.6	7.7	7.4	7.6	7.4	7.9	7.5	7.9	7.3	7.8	7.4	7.8	6.8
	Conductivity (µmhos/cm)	254		256		274		259		261		256		247	
	Temperature (°C)	25.2	24.9	25.1	24.5	25.2	24.7	25.2	24.8	25.1	24.9	24.9	24.7	25.1	24.8
72%	pH (SU)	7.94	7.94	7.99	8.02	8.12	7.79	8.00	7.87	8.07	7.87	7.99	7.89	8.06	7.71
	DO (mg/L)	7.7	7.5	7.7	7.4	7.6	7.2	7.9	7.4	7.9	7.3	7.8	7.4	7.8	6.9
	Conductivity (µmhos/cm)	217		221		224		220		224		220		217	
	Temperature (°C)	25.3	24.8	25.2	24.6	25.2	24.9	25.1	24.8	25.0	24.9	24.9	24.8	25.1	24.8
100%	pH (SU)	7.94	7.94	7.99	8.03	8.12	7.73	7.99	7.86	8.07	7.89	8.00	7.93	8.06	7.74
	DO (mg/L)	7.8	7.6	7.8	7.4	7.7	7.0	8.0	7.2	7.8	7.3	7.9	7.2	7.8	7.1
	Conductivity (µmhos/cm)	187		188		182		183		183		186		183	
	Temperature (°C)	25.3	24.7	25.2	24.3	25.2	24.6	25.2	24.8	25.2	25.1	24.9	24.8	25.0	25.0
100% Intake	pH (SU)	7.95	8.01	7.90	8.02	8.12	7.83	7.98	7.89	8.08	7.98	8.05	7.88	8.05	7.70
	DO (mg/L)	7.8	7.8	7.5	7.5	7.6	7.2	8.3	7.5	7.9	7.4	7.8	7.4	7.9	7.2
	Conductivity (µmhos/cm)	179		191		176		180		180		184		182	
	Temperature (°C)	25.4	24.8	25.0	24.5	25.0	24.8	25.2	24.9	25.1	25.1	24.9	25.0	24.9	24.8

Species: *Pimephales promelas* **Date:** 11-16-05
Client: TVA / Sequoyah Nuclear Plant – UV-treated

Daily Chemistry:

		Day					
		0		1 KDL		2	
	Analyst	AAB	AAB	AAB	AAB	AAB	JN
Concentration	Parameter						
CONTROL MHSW UV	pH (S.U.)	7.92	7.91	7.96	7.98	8.11	7.74
	DO (mg/L)	7.8	7.5	7.6	7.5	7.6	7.3
	Conductivity (µmhos/cm)	302		314		311	
	Temperature (°C)	25.0	24.8	24.9	24.6	24.9	24.8
10.98%	pH (S.U.)	7.92	7.91	7.96	7.96	8.12	7.77
	DO (mg/L)	7.7	7.5	7.6	7.4	7.6	7.5
	Conductivity (µmhos/cm)	294		293		304	
	Temperature (°C)	25.2	24.9	25.1	24.6	25.1	24.9
22%	pH (S.U.)	7.93	7.92	7.97	7.98	8.12	7.86
	DO (mg/L)	7.7	7.6	7.6	7.5	7.5	7.6
	Conductivity (µmhos/cm)	279		279		289	
	Temperature (°C)	25.2	24.9	25.1	24.6	25.1	24.9
43.9%	pH (S.U.)	7.93	7.92	7.98	7.98	8.12	7.79
	DO (mg/L)	7.7	7.6	7.7	7.4	7.6	7.4
	Conductivity (µmhos/cm)	254		256		274	
	Temperature (°C)	25.2	24.9	25.1	24.5	25.2	24.7
72%	pH (S.U.)	7.94	7.94	7.99	8.02	8.12	7.79
	DO (mg/L)	7.7	7.5	7.7	7.4	7.6	7.2
	Conductivity (µmhos/cm)	217		221		224	
	Temperature (°C)	25.3	24.8	25.2	24.6	25.2	24.9
100%	pH (S.U.)	7.94	7.94	7.99	8.03	8.12	7.75
	DO (mg/L)	7.8	7.6	7.8	7.4	7.7	7.0
	Conductivity (µmhos/cm)	187		188		182	
	Temperature (°C)	25.3	24.7	25.2	24.3	25.2	24.6
100% Intake	pH (S.U.)	7.95	8.01	7.90	8.02	8.12	7.83
	DO (mg/L)	7.8	7.8	7.5	7.5	7.6	7.2
	Conductivity (µmhos/cm)	179		191		176	
	Temperature (°C)	25.4	24.8	25.0	24.5	25.0	24.8
		Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas* **Date:** 11-16-05
Client: TVA / Sequoyah Nuclear Plant – UV-treated

		Day							
		3		4		5		6	
	Analyst	JN	JN	JN	MEK	MEK	MEK	MEK	MEK
Concentration	Parameter								
CONTROL MHSW UV	pH (S.U.)	8.02	7.85	8.04	7.85	7.99	7.81	8.02	7.66
	DO (mg/L)	7.9	7.6	7.7	7.4	7.9	7.3	7.8	7.0
	Conductivity (µmhos/cm)	305		305		310		290	
	Temperature (°C)	25.1	24.9	24.9	24.8	24.9	24.9	24.9	24.9
10.98%	pH (S.U.)	8.02	7.86	8.05	7.89	7.99	7.81	8.05	7.70
	DO (mg/L)	7.9	7.4	7.8	7.4	8.0	7.0	7.8	7.0
	Conductivity (µmhos/cm)	300		305		296		289	
	Temperature (°C)	25.2	24.9	25.1	24.8	25.0	25.2	25.0	24.8
22%	pH (S.U.)	8.02	7.89	8.06	7.86	7.98	7.91	8.05	7.66
	DO (mg/L)	7.9	7.6	7.9	7.4	7.9	7.0	7.8	6.8
	Conductivity (µmhos/cm)	288		291		283		275	
	Temperature (°C)	25.3	25.1	25.1	24.9	24.9	25.1	25.0	24.8
43.9%	pH (S.U.)	8.02	7.86	8.06	7.85	7.99	7.90	8.05	7.65
	DO (mg/L)	7.9	7.5	7.9	7.3	7.8	7.4	7.8	6.8
	Conductivity (µmhos/cm)	259		261		256		247	
	Temperature (°C)	25.2	24.8	25.1	24.9	24.9	24.7	25.1	24.8
72%	pH (S.U.)	8.00	7.87	8.07	7.87	7.99	7.89	8.06	7.71
	DO (mg/L)	7.9	7.4	7.9	7.3	7.8	7.4	7.8	6.9
	Conductivity (µmhos/cm)	220		224		220		217	
	Temperature (°C)	25.1	24.8	25.0	24.9	24.9	24.8	25.1	24.8
100%	pH (S.U.)	7.99	7.86	8.07	7.89	8.00	7.93	8.06	7.74
	DO (mg/L)	8.0	7.7	7.8	7.3	7.9	7.2	7.8	7.1
	Conductivity (µmhos/cm)	183		183		186		183	
	Temperature (°C)	25.2	24.8	25.2	25.1	24.9	24.8	25.0	25.0
100% Intake	pH (S.U.)	7.98	7.89	8.08	7.98	8.05	7.88	~~4.88~~ 8.05	7.70
	DO (mg/L)	8.3	7.5	7.9	7.4	7.8	7.4	7.9	7.2
	Conductivity (µmhos/cm)	180		180		184		182	
	Temperature (°C)	25.2	24.9	25.1	25.1	24.9	25.0	24.9	24.8
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

Total Residual Chlorine
(EPA Method 330.5)

Matrix: Water, MDL = 0.10 mg/L

Meter: Accumet Model AR25 pH/Ion Meter

Analyst: AAB

Date analyzed: 11-16-05

Iodide reagent: INR178 19 & 20

Acid reagent: INR90 1 & 2

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	IN55325	IN55325

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%)
IN55325	0.50	0.531	106.2 %

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
05111506	T.A. Stevens	pale yellow, slightly cloudy	S<0.00274	
	Duplicate		D<0.00322	—

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00317
051115.03	Coats American	light brown, slightly cloudy, particles	0.229
051116.02	Johnson Co. public util	pale yellow, clear	<0.00332
051116.01	Apex	pale yellow, clear	<0.00836
051116.04	Smith Creek	pale beige clear	<0.00265
051116.03	South Cary	pale beige, clear	<0.00234
051116.09	Roseboro	light beige, clear	<0.00954
051116.08	Rockingham	light beige, clear	<0.00303
051116.09	J P Stevens	light brownish yellow slightly cloudy	<0.0000621
05111607	Monroe	light beige slightly cloudy	<0.000661

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%)
IN55325	0.50	0.524	104.8%

Reviewed by: KW

Date reviewed: 11-16-05

Total Residual Chlorine
(EPA Method 330.5)

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

Analyst	AAB
Date analyzed	11-16-05

Iodide reagent:	INR178 19820
Acid reagent:	INA170 1012

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	IN55325	IN55325

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%)
IN55325	0.50	0.497	99.4%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
051116.05	Raleigh Reuse	pale yellow, clear	S <0.000865	
051116.06	Duplicate		D <0.000942	—

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00264
051116.11	Morehead City	pale yellow, cloudy particles	<0.0198
05111504	TVA-SQN-101	clear, no color particles	<0.00208
051115.05	TVA-SQN-INT	clear, no color particles	<0.00119
			AAB

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%)
IN55325	0.50	0.523	104.6%

Reviewed by	KGL
Date reviewed	11.16.05

Total Residual Chlorine
(EPA Method 330.5)

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

Analyst	AAB
Date analyzed	11-18-05

Iodide reagent:	IN2178 (20 of 20)
Acid reagent:	IN2196 (1 of 2)

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INSS325	INSS325

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%)
INSS325	0.50	0.537	107.4%

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
051117.03	TVA-SQN-101	clear, no color	S <0.00361	
1	Duplicate		D <0.00342	-

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		<0.00532
051117.04	TVA-SQN-INT	clear, no, color	<0.00223
			AAB

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%)
INSS325	0.50	0.467	93.4%

Reviewed by	KW
Date reviewed	11.18.05

Total Residual Chlorine
(EPA Method 330.5)

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

Analyst	JN
Date analyzed	11-20-05

Iodide reagent:	INR 192
Acid reagent:	INR 190

Calibration:

	0.10 mg/L	1.00 mg/L
Reference standard number	INSS 325	INSS 325

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%)
INSS 325	0.50	0.491	98.2 %

Duplicate sample precision:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
051119.11	Raleigh rinse pp	no color, clear particles	S 0.0000764	
↓	Duplicate		D 0.0000381	JN

Sample measurements:

Sample number	Sample ID	Sample characteristics	Residual chlorine (mg/L)
	Blank (should be = < 0.10 mg/L)		< 0.000503
051119.12	TVA-SON- 101	no color, clear	< 0.0000586
051119.13	TVA-SON INT	no color, clear	< 0.0000971

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%)
JNSS 325	0.50	0.477	95.4 %

Reviewed by	KCL
Date reviewed	11-26-05

Environmental Testing Solutions, Inc.

Alkalinity
(EPA Method 310.1)

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

Analyst	KEN
Date analyzed	11.13.05

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%)
INR183	100	100	9.6	19.3	9.7	10.4	101	101.1

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%)
							S	
	Duplicate						D	

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)

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%)

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
11.07-05A	salt H_2O	100	19.3	30.6	11.3	10.4	120
11.07.05B	↓	↓	30.6	42.0	11.4	↓	120
11.12.05	↓	↓	28.4	39.0	10.6	↓	110
				KEN			

Reviewed by: JL

Date reviewed: 11-15-05

Environmental Testing Solutions, Inc.

Alkalinity
(EPA Method 310.1)

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

Analyst: STK
Date analyzed: 11-20.05

Titrate samples to pH = 4.50 S.U.

Time started: 1301
Time ended: 1349

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
5.2	INR184	INR157	0.1	12.9	12.8	0.0195	9.8

Bill 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%)
INR183	100	100	12.9	23.2	10.3	9.8	101	101%.

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%)
11-15-05	SSW H2O	100	23.2	26.5	3.3	9.8	S 32	
↓	Duplicate	100	26.5	29.9	3.4	9.8	D 33	3.1%.

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)
INR183	50	100	26.5	34.6	8.1	9.8	79

Sample alkalinity (B) (mg $CaCO_3$/L)	Measured spike value (MV) MV = A - B (mg $CaCO_3$/L)	% R = MV / SV x 100 (acceptable range = 75 to 125%)
33	46	92%.

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Alkalinity (mg $CaCO_3$/L)
11-17-05 A	MHS H2O	100	34.6	40.8	6.2	9.8	61
11-17-05 B	MHS H2O		0.5	6.4	5.9		58
11-15-05	Salt H2O		6.4	20.3	13.9		140
051116.06	Raleigh 1		20.3	28.9	8.6		84
051118.01	↓ 2		28.9	37.4	8.5		83
051119.11	↓ 3		37.4	45.3	7.9		77
051115.03	Canea 1		0.0	9.1	9.1		89
051118.05	↓ 2		9.1	18.1	9.0		88
051115.01	Foxwood 1	↓	18.1	21.5	3.4	↓	33

Reviewed by: JL
Date reviewed: 11-24-05

Alkalinity
(EPA Method 310.1)

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

Analyst: KEK
Date analyzed: 11-20.05

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%)
INR183	100	100	21.5	31.1	9.6	9.8	94	94%

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%)
051117.01	Forwood 2	100	31.3	43.0	11.7	9.8	S 120	
↓	Duplicate	100	0.0	11.6	11.6	9.8	D 110	8.7%

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)
INR183	50	~~50~~ 100	0.0	16.6	16.6	9.8	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)
051119.01	Forwood 3	100	16.6	21.8	5.2	9.8	51
051115.04	TVA SSN 101 1		21.8	28.2	6.4		63
051117.03	↓ 2		28.3	34.6	6.3		62
051119.12	↓ 3		34.7	41.3	6.6		65
051115.05	TVA SSN INT 1		1.2	7.7	6.5		64
051117.04	↓ 2		7.8	14.0	6.2		61
051119.13	↓ 3	↓	14.0	20.4	6.4		63
051112.04	Hercules 1	50	20.4	26.2	5.8	(2)	110
~~051117.~~	↓ 2	↓	26.2	31.6	5.4	↓	110
051115.02							
051117.02	Hercules 3	50	31.6	39.2	7.6	(2)	9.8 150

Reviewed by: [signature]

Date reviewed: 11.24.05

Environmental Testing Solutions, Inc.

Total Hardness
(EPA Method 130.2)

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

Time started: 1004
Time ended: 1031

Analyst	KEN
Date analyzed	11-13-05

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
INR175	INR123	0.0	9.9	9.9	0.0202	20.2

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%)
INSS309	40	50	9.9	12.0	2.1	20.2	42	105.0%

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%)
11.08.05B	MHS H2O	50	16.8	21.3	4.5	20.2	S 91	
↓	Duplicate	50	21.3	25.6	4.3	↓	D 87	4.5%

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)
INSS309	40	50	21.3	27.6	6.3	20.2	130

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%)
87	43	107.5%

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	20.2	ND
11.08.05A	MHS H2O	↓	12.0	16.8	4.8	↓	97
11.11.05A	↓	↓	27.6	32.3	4.7	↓	95
11.11.05B	↓	↓	32.3	37.0	4.7	↓	95
11.08.05	SSW 1-H2O	↓	37.0	39.0	2.0	↓	40
051108.01	OWASA 1	↓	0.2	2.7	2.5	↓	50
051110.01	↓ 2	↓	2.7	5.1	2.4	↓	48
051112.01	↓ 3	↓	5.1	7.9	2.8	↓	57
051109.13	Marshall WWTP	↓	7.9	12.5	4.6	↓	93
051108.03	Int. Paper	25	12.5	17.3	4.8	12 ↓	190

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

Reviewed by: JL

Date reviewed: 11-15-05

Environmental Testing Solutions, Inc.

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

Analyst	KM
Date analyzed	11.20.05

Time started: 1102
Time ended: 1126

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
INR175	INR123	0.0	10.0	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%)
INSS309	40	50	10.0	12.2	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%)
11.15.05	SSW H_2O	50	12.6	14.8	2.2	20	S 44	
↓	Duplicate	50	14.8	17.0	2.2	20	D 44	—

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)
INSS309	40	50	14.8	19.1	4.3	20	86

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

Sample measurements:

Sample number	Sample ID	Sample volume (ml)	Begin ml	End ml	Total ml	Multiplier	Hardness (mg $CaCO_3$/L)
TV=ND	Blank (should be = 0 mg $CaCO_3$/L)	50	0.0	0.0	0.0	20	ND
11.17.05A	MHS H_2O		19.1	23.7	4.6		92
11.17.05B	MHS H_2O		23.7	28.5	4.8		96
051116.06	Raleigh 1		28.5	31.6	3.1		62
051118.01	↓ 2		31.6	34.8	3.2		64
051119.11	↓ 3		34.8	38.3	3.5		70
051115.03	CArver 1		38.3	41.5	3.2		64
051118.05	↓ 2		41.5	45.0	3.5		70
051115.01	Foxwood 1		0.0	3.5	3.5		70
051117.61	↓ 2	↓	3.5	10.5	7.0	↓	140

Note: If >15ml of titrant is used, sample must be diluted. Reviewed by: [signature] Date reviewed: 11-24-05

Environmental Testing Solutions, Inc.

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

Analyst	JCN
Date analyzed	11.20.05

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%)
INSS309	40	50	10.5	12.5	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%)
051119.01	Foxwood 3	50	12.5	18.2	5.7	20	S 110	
↓	Duplicate	50	18.2	23.5	5.3	20	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 hardness (A) (mg $CaCO_3$/L)
INSS309	40	50	18.2	25.5	7.3	20	150

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%)
110	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)						K
051115.04	TVA SQN 101 1	50	25.5	29.1	3.6	20	72
051117.03	↓ 2		29.1	32.6	3.5		70
051119.12	↓ 3		32.6	36.3	3.7		74
051115.05	TVA SQN INT 1		36.3	40.1	3.8		76
051117.04	↓ 2		40.1	43.7	3.6		72
051119.13	↓ 3		43.7	47.5	3.8		76

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

Reviewed by: [signature]

Date reviewed: 11-24-05

Sequoyah Nuclear Plant Biomonitoring
November 16-23, 2005

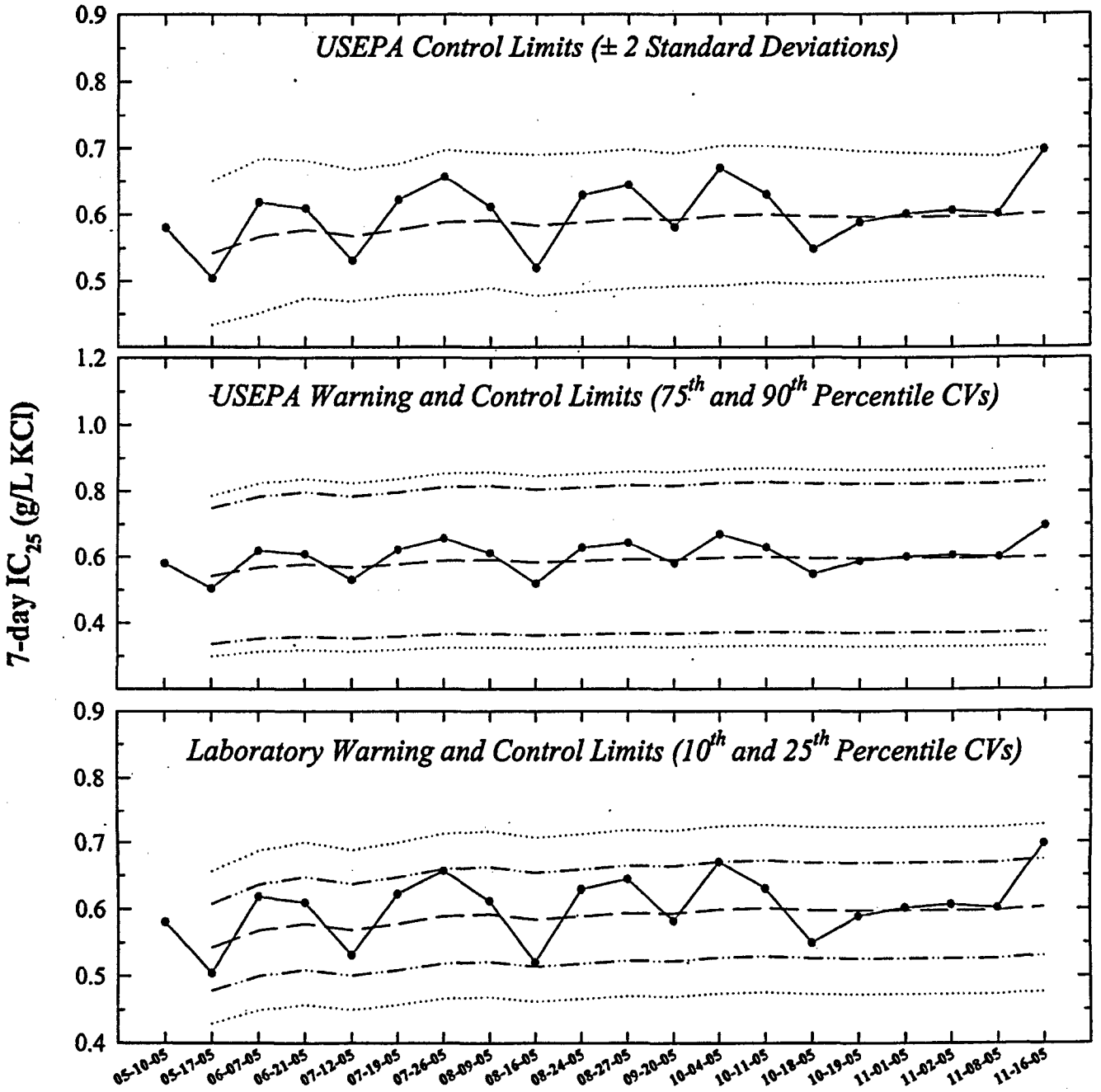
Appendix D

Reference Toxicant Test and Control Chart

Environmental Testing Solutions, Inc.

Pimephales promelas

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



Test date

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

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

—·—·— **Warning Limits** (mean $IC_{25} \pm S_{A.10}$ or $S_{A.75}$)

····· **Control Limits** (mean $IC_{25} \pm S_{A.25}$, $S_{A.90}$, or 2 Standard Deviations)

Environmental Testing Solutions, Inc.

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

Test number	Test date	7-day IC_{25} (g/L KCl)	CT (g/L KCl)	S	State and USEPA Control Limits CT - 2S	CT + 2S	$S_{A.10}$	Laboratory Warning Limits CT - $S_{A.10}$	CT + $S_{A.10}$	$S_{A.25}$	Laboratory Control Limits CT - $S_{A.25}$	CT + $S_{A.25}$	$S_{A.75}$	USEPA Warning Limits CT - $S_{A.75}$	CT + $S_{A.75}$	$S_{A.90}$	USEPA Control Limits CT - $S_{A.90}$	CT + $S_{A.90}$	CV
1	05-10-05	0.58																	
2	05-17-05	0.50	0.54	0.05	0.43	0.65	0.07	0.48	0.61	0.11	0.43	0.66	0.21	0.34	0.75	0.24	0.30	0.79	0.10
3	06-07-05	0.62	0.57	0.06	0.45	0.68	0.07	0.50	0.64	0.12	0.45	0.69	0.22	0.35	0.78	0.26	0.31	0.82	0.10
4	06-21-05	0.61	0.58	0.05	0.47	0.68	0.07	0.51	0.65	0.12	0.46	0.70	0.22	0.36	0.80	0.26	0.32	0.84	0.09
5	07-12-05	0.53	0.57	0.05	0.47	0.67	0.07	0.50	0.64	0.12	0.45	0.69	0.22	0.35	0.78	0.26	0.31	0.82	0.09
6	07-19-05	0.62	0.58	0.05	0.48	0.68	0.07	0.51	0.65	0.12	0.46	0.70	0.22	0.36	0.80	0.26	0.32	0.84	0.09
7	07-26-05	0.66	0.59	0.05	0.48	0.70	0.07	0.52	0.66	0.12	0.47	0.71	0.22	0.37	0.81	0.27	0.32	0.85	0.09
8	08-09-05	0.61	0.59	0.05	0.49	0.69	0.07	0.52	0.66	0.12	0.47	0.72	0.22	0.37	0.82	0.27	0.33	0.86	0.09
9	08-16-05	0.52	0.58	0.05	0.48	0.69	0.07	0.51	0.65	0.12	0.46	0.71	0.22	0.36	0.81	0.26	0.32	0.85	0.09
10	08-24-05	0.63	0.59	0.05	0.48	0.69	0.07	0.52	0.66	0.12	0.46	0.71	0.22	0.36	0.81	0.26	0.32	0.85	0.09
11	08-27-05	0.64	0.59	0.05	0.49	0.70	0.07	0.52	0.66	0.12	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.86	0.09
12	09-20-05	0.58	0.59	0.05	0.49	0.69	0.07	0.52	0.66	0.12	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.86	0.08
13	10-04-05	0.67	0.60	0.05	0.49	0.70	0.07	0.53	0.67	0.13	0.47	0.72	0.23	0.37	0.83	0.27	0.33	0.87	0.09
14	10-11-05	0.63	0.60	0.05	0.50	0.70	0.07	0.53	0.67	0.13	0.47	0.73	0.23	0.37	0.83	0.27	0.33	0.87	0.09
15	10-18-05	0.55	0.60	0.05	0.50	0.70	0.07	0.53	0.67	0.13	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.87	0.09
16	10-19-05	0.59	0.60	0.05	0.50	0.70	0.07	0.53	0.67	0.13	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.87	0.08
17	11-01-05	0.60	0.60	0.05	0.50	0.69	0.07	0.53	0.67	0.13	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.87	0.08
18	11-02-05	0.61	0.60	0.05	0.50	0.69	0.07	0.53	0.67	0.13	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.87	0.08
19	11-08-05	0.60	0.60	0.05	0.51	0.69	0.07	0.53	0.67	0.13	0.47	0.72	0.23	0.37	0.82	0.27	0.33	0.87	0.08
20	11-16-05	0.70	0.60	0.05	0.50	0.70	0.07	0.53	0.68	0.13	0.48	0.73	0.23	0.37	0.83	0.27	0.33	0.87	0.08

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

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

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

Laboratory Control and Warning Limits

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

$S_{A.10}$ = Standard deviation corresponding to the 10th percentile CV. ($S_{A.10}$ = 0.12)

$S_{A.25}$ = Standard deviation corresponding to the 25th percentile CV. ($S_{A.25}$ = 0.21)

USEPA Control and Warning Limits

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

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

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

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

Organisms obtained from Aquatic BioSystems, Inc.

11-16-05

Environmental Testing Solutions, Inc.

Precision of Endpoint Measurements

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

Test number	Test date	Control Survival (%)	Control Mean Growth (mg/larvae)	CT for Control Growth (mg/larvae)	CV (%)	CT for Control Growth CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	05-10-05	100	0.772		10.7		0.10	13.5	
2	05-17-05	100	0.858	0.815	6.0	8.4	0.06	7.4	10.5
3	06-07-05	100	0.848	0.826	3.3	6.7	0.09	10.1	10.4
4	06-21-05	97.5	0.622	0.775	7.2	6.8	0.09	14.5	11.4
5	07-12-05	100	0.765	0.773	5.6	6.6	0.10	13.4	11.8
6	07-19-05	100	0.596	0.743	2.9	5.9	0.06	9.7	11.4
7	07-26-05	100	0.807	0.753	11.5	6.7	0.11	13.2	11.7
8	08-09-05	100	0.622	0.736	5.5	6.6	0.11	17.0	12.3
9	08-16-05	100	0.599	0.721	12.7	7.3	0.11	18.9	13.1
10	08-24-05	100	0.802	0.729	14.9	8.0	0.14	17.4	13.5
11	08-27-05	100	0.678	0.724	4.0	7.7	0.11	16.5	13.8
12	09-20-05	100	0.814	0.732	8.4	7.7	0.11	13.3	13.7
13	10-04-05	100	0.785	0.736	4.5	7.5	0.06	7.3	13.2
14	10-11-05	100	0.555	0.723	4.1	7.2	0.11	19.4	13.7
15	10-18-05	100	0.741	0.724	4.9	7.1	0.04	5.9	13.2
16	10-19-05	100	0.556	0.714	5.9	7.0	0.08	14.6	13.3
17	11-01-05	100	0.694	0.713	1.2	6.7	0.09	12.4	13.2
18	11-02-05	100	0.852	0.720	5.5	6.6	0.12	14.5	13.3
19	11-08-05	100	0.754	0.722	12.3	6.9	0.15	20.0	13.6
20	11-16-05	100	0.648	0.718	13.5	7.2	0.14	21.4	14.0

Note:

CV = Coefficient of variation for control growth.
On average, the CV for control growth is 7.2% in Environmental Testing Solutions, Inc. *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, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 14.0% from the control.
Lower PMSD bound determined by USEPA (10th percentile) = 9.4%.
Upper PMSD bound determined by USEPA (90th percentile) = 35%.

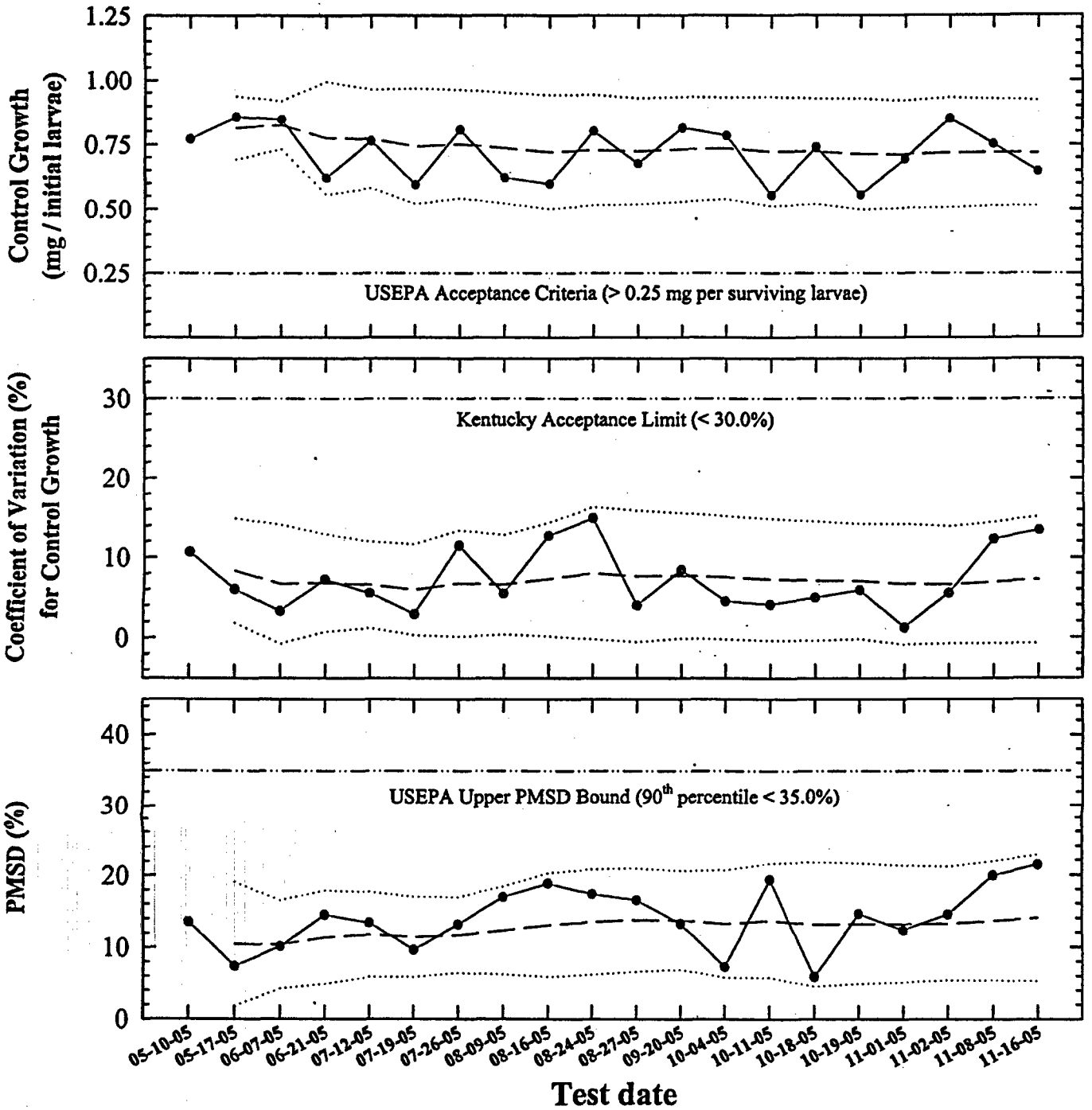
CT = Central Tendancy (mean Control Growth, CV, or PMSD)

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

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

Environmental Testing Solutions, Inc.

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



—●— **Control Reproduction, Coefficient of Variation (CV), or Percent Minimum Significant Difference (PMSD)** PMSD is the minimum significant difference between the control and treatment that can be declared statistically significant.

— — · **Central Tendency** (mean Control Growth, CV, or PMSD)

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

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

PpKClCR Test Number: 84

Dilution preparation information:						*Comments:*
KCl CHM number:		CHM 188				* FIRST TEST USING
Stock preparation:		50 g KCl/L: Dissolve 50 g KCl in 1-L Deionized water				SOLO-CUP LIDS.
Dilution prep (mg/L)	300	450	600	750	900	
Stock volume (mL)	6	9	12	15	18	
Diluent volume (mL)	994	991	988	985	982	
Total volume (mL)	1000	1000	1000	1000	1000	

Test organism information:		*Test information:*	
Organism age:	23.25 TO 24.75 HOURS OLD	Randomizing template:	PURPLE
Date and times organisms were born between:	11-15-05 1330 TO 1500	Incubator number and shelf location:	3E
Organism source:	ABS BATCH Pp 11-15-05	*Artemia* lot number:	BG1804T
Transfer bowl information:	pH = 8.10 SU Temperature = 24.2 °C	Total drying time:	24-HOURS
		Date / Time in: 11-23-05	1350
Average transfer volume:	10.4 mL	Date / Time out: 11-24-05	1356
		Oven temperature:	60°C

Daily feeding and renewal information:

Day	Date	Morning feeding time	Afternoon feeding time	Test initiation, renewal, or termination time	MHS batch used	Analyst
0	11-16-05	~~0900~~	1500	1416	11-11-05 A	JH
1	11-17-05	0900	1506	1330	11-11-05 B	JH
2	11-18-05	0912	1520	1336	11-11-05 B	JH
3	11-19-05	0847	1452	1320	11-17-05 BA	JH
4	11-20-05	0905	1506	1332	11-17-05 BA	JH
5	11-21-05	0908	1511	1319	11-17-05 B	JH
6	11-22-05	0900	1500	1323	11-17-05 B	JH
7	11-23-05			1341		JH/KEK

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	0%	≤ 20%	7-day LC_{50}	796.2
Average weight per initial larvae:	0.648		NOEC	600
Average weight per surviving larvae:	0.648	≥ 0.25 mg/larvae	LOEC	750
			ChV	670.8
			IC_{25}	697.9

Species: ***Pimephales promelas*** PpKClCR Test Number: 84

Survival and Growth Data

Day	Control				300 mg KCl/L				450 mg KCl/L			
	A	B	C	D	E	F	G	H	I	J	K	L
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	8 2nd	10	10	10	10	10	10	10
7	1SM 10	10	10	10	8	1LG 10	10	10	10	10	10	1LG 10
A = Pan weight (mg) Tray color code:: Gold Analyst: JN	14.68	16.13	16.12	14.99	15.07	16.16	15.87	16.26	JN ~~14.90~~ 14.89	14.29	16.31	15.98
B = Pan + Larvae weight (mg) Analyst: KKeenan	20.08	23.62	22.85	21.29	20.37	23.88	22.62	23.69	22.41	21.04	22.30	23.95
Larvae weight (mg) = A – B	5.40	7.49	6.73	6.30	5.30	7.72	6.75	7.43	7.52	6.75	5.99	7.97
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.540	0.749	0.673	0.630	0.530	0.772	0.675	0.743	0.752	0.675	0.599	0.797
Average weight per initial number of larvae (mg)	0.648				0.680				0.706			
Percent reduction from control (%)					-4.9%				-8.9%			

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed: JN

Comments:

Species: ***Pimephales promelas*** PpKClCR Test Number: 84

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	10	9^{1d}	10	10	9^{1d}	8^{2d}	10	8^{2d}	8^{2d}	7^{3d}	9^{1d}	9^{1d}
2	10	9	10	10	6^{3d}	7^{1d}	10	8	8	3^{4d}	5^{4d}	7^{2d}
3	10	9	9^{1d}	10	6	7	10	7^{1d}	5^{3d}	1^{2d}	5	6^{1d}
4	10	9	9	9^{1d}	6	7	10	7	4^{1d}	1	5	5^{1d}
5	10	9	9	9	6	6^{1d}	10	7	4	1	5	5
6	10	9	9	9	5^{1d}	6	8^{2d}	7	4	1	3^{2d}	5
7	10	9	9 lg	9	5	6	6^{2d}	7	3^{3d}	1	3^{3d}	5
A = Pan weight (mg) Tray color code:: Gold Analyst: JN	14.59	15.99	14.69	14.78	16.20	14.56	14.65	16.18	16.14	14.82	15.06	14.63
B = Pan + Larvae weight (mg) Analyst: McKeenan	21.28	21.92	21.81	21.52	20.12	18.89	18.97	20.68	17.60	15.48	16.22	18.05
Larvae weight (mg) = A – B	6.69	5.93	7.12	6.74	3.92	4.33	4.32	4.50	1.46	0.66	1.16	3.42
Weight per initial number of larvae (mg) = C / Initial number of larvae	0.669	0.593	0.712	0.674	0.392	0.433	0.432	0.450	0.146	0.066	0.116	0.342
Average weight per initial number of larvae (mg)	0.662				0.427				0.168			
Percent reduction from control (%)			-2.7%				34.1%				74.7%	

Comment codes: c = clear, d = dead, fg = fungus, k = killed, m = missing, sk = sick, sm = unusually small, lg = unusually large, d&r = decanted and returned, w = wounded.

Calculations and data reviewed:

Comments:

Environmental Testing Solutions, Inc.

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

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

Test number: PpKClCR # 124 (#84 at 351 Depot St.)

Test dates: November 16-23, 2005

Reviewed by: [signature]

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 / Surviving number of larvae (mg)	Mean weight/ Surviving number of larvae (mg)	Coefficient of variation (Mean weight per surviving number of larvae) (%)	Weight / Initial number of larvae (mg)	Mean survival (%)	Mean weight/ Initial number of larvae (mg)	Coefficient of variation (%)	Percent reduction from control (%)
Control	A	10	10	14.68	20.08	5.40	0.540	0.648	13.5	0.540	100.0	0.648	13.5	Not applicable
	B	10	10	16.13	23.62	7.49	0.749			0.749				
	C	10	10	16.12	22.85	6.73	0.673			0.673				
	D	10	10	14.99	21.29	6.30	0.630			0.630				
300	E	10	8	15.07	20.37	5.30	0.663	0.713	7.4	0.530	95.0	0.680	15.9	-4.9
	F	10	10	16.16	23.88	7.72	0.772			0.772				
	G	10	10	15.87	22.62	6.75	0.675			0.675				
	H	10	10	16.26	23.69	7.43	0.743			0.743				
450	I	10	10	14.89	22.41	7.52	0.752	0.706	12.4	0.752	100.0	0.706	12.4	-8.9
	J	10	10	14.29	21.04	6.75	0.675			0.675				
	K	10	10	16.31	22.30	5.99	0.599			0.599				
	L	10	10	15.98	23.95	7.97	0.797			0.797				
600	M	10	10	14.59	21.28	6.69	0.669	0.717	8.9	0.669	92.5	0.662	7.5	-2.2
	N	10	9	15.99	21.92	5.93	0.659			0.593				
	O	10	9	14.69	21.81	7.12	0.791			0.712				
	P	10	9	14.78	21.52	6.74	0.749			0.674				
750	Q	10	5	16.20	20.12	3.92	0.784	0.717	8.1	0.392	60.0	0.427	5.8	34.1
	R	10	6	14.56	18.89	4.33	0.722			0.433				
	S	10	6	14.65	18.97	4.32	0.720			0.432				
	T	10	7	16.18	20.68	4.50	0.643			0.450				
900	U	10	3	16.14	17.60	1.46	0.487	0.554	25.7	0.146	30.0	0.168	72.2	74.2
	V	10	1	14.82	15.48	0.66	0.660			0.066				
	W	10	3	15.06	16.22	1.16	0.387			0.116				
	X	10	5	14.63	18.05	3.42	0.684			0.342				

Dunnett's MSD value: 0.1387 MSD = Minimum Significant Difference

PMSD: 21.4 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, Inc. chronic toxicity tests when a toxicant reduces *Pimephales* growth by 14.0% 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.

Organisms obtained from Aquatic BioSystems, Inc.

Environmental Testing Solutions, Inc.
Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	11/16/2005	Test ID:	PpKCICR	Sample ID:	REF-Ref Toxicant
End Date:	11/23/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	KCL-Potassium chloride
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:					

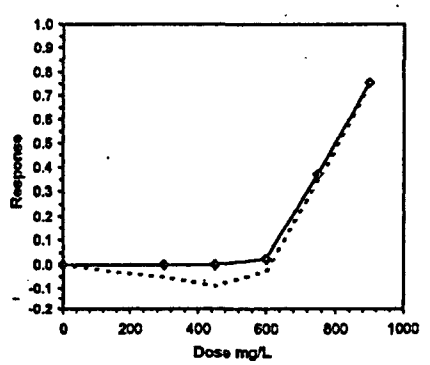
Conc-mg/L	1	2	3	4
D-Control	0.5400	0.7490	0.6730	0.6300
300	0.5300	0.7720	0.6750	0.7430
450	0.7520	0.6750	0.5990	0.7970
600	0.6690	0.5930	0.7120	0.6740
750	0.3920	0.4330	0.4320	0.4500
900	0.1460	0.0660	0.1160	0.3420

			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	0.6480	1.0000	0.6480	0.5400	0.7490	13.458	4				0.6779	1.0000
300	0.6800	1.0494	0.6800	0.5300	0.7720	15.875	4	-0.528	2.290	0.1387	0.6779	1.0000
450	0.7058	1.0891	0.7058	0.5990	0.7970	12.354	4	-0.954	2.290	0.1387	0.6779	1.0000
600	0.6620	1.0216	0.6620	0.5930	0.7120	7.530	4	-0.231	2.290	0.1387	0.6620	0.9765
750	0.4268	0.6586	0.4268	0.3920	0.4500	5.763	4				0.4268	0.6295
900	0.1675	0.2585	0.1675	0.0660	0.3420	72.193	4				0.1675	0.2471

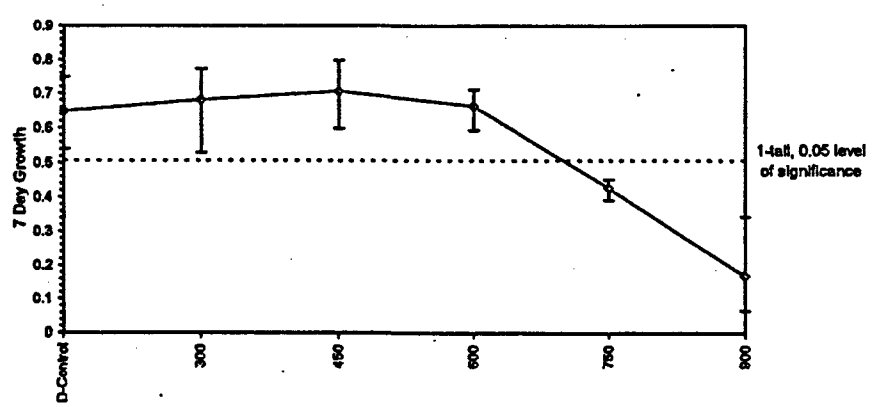
Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.943732083		0.844		-0.53452712	-0.61984917
Bartlett's Test indicates equal variances (p = 0.70)					1.444379091		11.34486675			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	600	>600			0.138692184	0.214031149	0.002485396	0.007336063	0.797709525	3, 12
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

Point	mg/L	SD	95% CL(Exp)		Skew
IC05	611.46	94.63	0.00	629.74	-2.9101
IC10	633.08	34.11	539.15	653.40	-7.5504
IC15	654.69	15.15	591.12	677.05	-1.2863
IC20	676.30	13.56	616.70	700.70	-0.8554
IC25	697.91	12.65	642.86	724.51	-0.6090
IC40	761.57	10.96	724.92	790.45	-0.0580
IC50	800.80	15.61	765.31	865.13	0.9336



Dose-Response Plot



Organisms obtained from Aquatic BioSystems, Inc.

11-16-05

Species: ***Pimephales promelas*** PpKClCR Test Number: 84

Daily Chemistry:

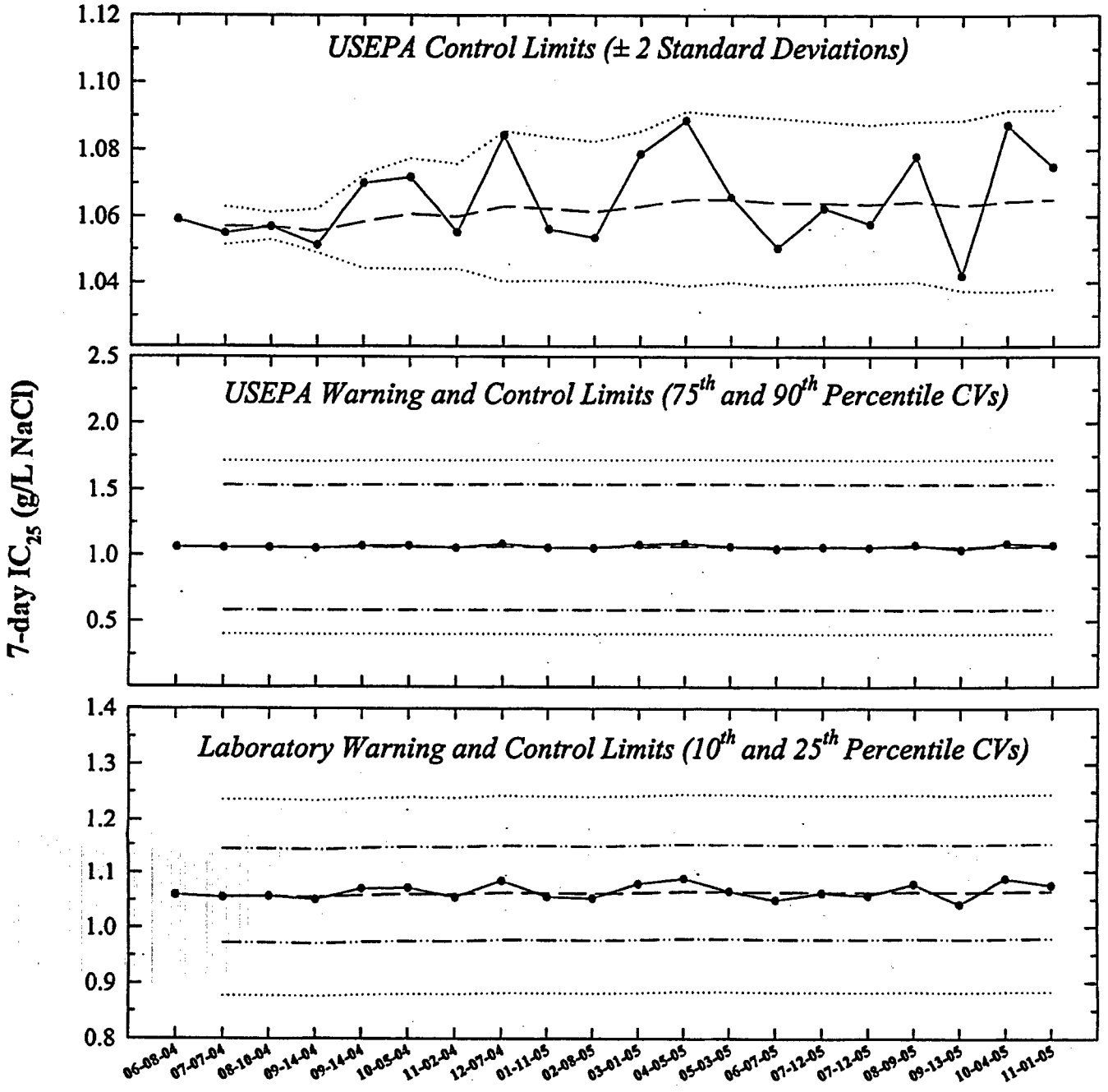
		Day					
		0		1	KER	KER	2
	Analyst	[redacted]	[redacted]	[redacted]	AAB	AAB	JN
Concentration	Parameter						
CONTROL	pH (S.U.)	7.86	7.88	7.99	7.96	8.05	7.77
	DO (mg/L)	7.4	7.5	7.7	7.6	7.7	7.4
	Conductivity (µmhos/cm)	310	[redacted]	317	[redacted]	318	[redacted]
	Alkalinity (mg $CaCO_3$/L)	61	[redacted]	62	[redacted]	[redacted]	[redacted]
	Hardness (mg $CaCO_3$/L)	95	[redacted]	95	[redacted]	[redacted]	[redacted]
	Temperature (°C)	24.9	24.7	24.8	24.5	24.9	24.9
300 mg KCl/L	pH (S.U.)	7.89	7.87	7.96	7.97	8.08	7.77
	DO (mg/L)	7.5	7.4	7.6	7.6	7.6	7.3
	Conductivity (µmhos/cm)	842	[redacted]	839	[redacted]	841	[redacted]
	Temperature (°C)	24.9	24.9	24.9	24.8	24.9	25.1
450 mg KCl/L	pH (S.U.)	7.90	7.85	7.99	7.95	8.09	7.78
	DO (mg/L)	7.5	7.2	7.7	7.7	7.6	7.3
	Conductivity (µmhos/cm)	1120	[redacted]	1110	[redacted]	1110	[redacted]
	Temperature (°C)	24.9	24.7	24.9	24.7	24.9	25.1
600 mg KCl/L	pH (S.U.)	7.90	7.86	7.98	7.94	8.08	7.82
	DO (mg/L)	7.5	7.4	7.7	7.6	7.7	7.4
	Conductivity (µmhos/cm)	1380	[redacted]	1380	[redacted]	1390	[redacted]
	Temperature (°C)	24.9	24.7	24.9	24.8	24.9	24.8
750 mg KCl/L	pH (S.U.)	7.91	7.85	7.98	7.97	8.08	7.75
	DO (mg/L)	7.6	7.5	7.8	7.6	7.6	7.2
	Conductivity (µmhos/cm)	1730	[redacted]	1670	[redacted]	1690	[redacted]
	Temperature (°C)	25.1	24.7	25.0	24.8	25.0	24.7
900 mg KCl/L	pH (S.U.)	7.92	7.86	7.99	7.97	8.08	7.78
	DO (mg/L)	7.6	7.6	7.9	7.7	7.6	7.3
	Conductivity (µmhos/cm)	1900	[redacted]	1930	[redacted]	1930	[redacted]
	Temperature (°C)	25.0	24.7	24.8	24.8	24.9	24.8
STOCK	Conductivity (µmhos/cm)	75700	[redacted]	[redacted]	[redacted]	76,100	[redacted]
		Initial	Final	Initial	Final	Initial	Final

Species: *Pimephales promelas* PpKClCR Test Number: 84

		Day							
		3		4		5		6	
	Analyst	JN	JN	JN	KEK	KEK	KEK	KEK	KEK
Concentration	Parameter								
CONTROL MHS	pH (S.U.)	7.95	~~7.77~~ 7.8	7.99	7.87	8.00	7.79	7.98	7.61
	DO (mg/L)	7.8	7.4	7.8	7.2	7.9	7.2	7.8	7.0
	Conductivity (µmhos/cm)	314		321		321		307	
	Alkalinity (mg $CaCO_3$/L)	61				58			
	Hardness (mg $CaCO_3$/L)	92				96			
	Temperature (°C)	24.9	25.1 7.82	24.7	24.8	24.8	25.0	24.7	24.8
300 mg KCl/L	pH (S.U.)	8.00	~~7.77~~	8.02	7.86	8.00	7.86	8.00	7.58
	DO (mg/L)	7.8	7.3	7.7	7.3	7.9	7.2	7.9	7.0
	Conductivity (µmhos/cm)	840		860		859		833	
	Temperature (°C)	25.0	24.8 7.80	24.9	25.1	24.9	24.7	24.7	24.9
450 mg KCl/L	pH (S.U.)	8.00	~~7.75~~	8.00	7.79	8.01	7.85	8.00	7.62
	DO (mg/L)	7.8	7.2	7.9	7.2	7.8	7.4	7.8	6.9
	Conductivity (µmhos/cm)	JN ~~1060~~ 1100		JN ~~1070~~ 1110		1130		1160	
	Temperature (°C)	25.0	24.8 7.83	24.9	25.1	24.9	24.6	24.6	24.9
600 mg KCl/L	pH (S.U.)	8.02	~~7.82~~	8.01	7.84	8.02	7.83	8.00	7.61
	DO (mg/L)	7.7	7.2	7.9	7.4	7.8	7.3	7.8	7.0
	Conductivity (µmhos/cm)	1370		1350		1370		1420	
	Temperature (°C)	24.9	24.9 7.81	24.9	24.9	24.7	24.8	24.6	24.9
750 mg KCl/L	pH (S.U.)	8.02	~~7.73~~	8.01	7.84	8.02	7.80	8.01	7.59
	DO (mg/L)	7.7	7.2	7.9	7.5	7.8	7.3	7.7	6.9
	Conductivity (µmhos/cm)	1650		~~1620~~ 1600		1710		1730	
	Temperature (°C)	24.9	24.9 7.82	24.9	24.9	24.9	24.8	24.6	25.0
900 mg KCl/L	pH (S.U.)	8.02	~~7.78~~	8.01	7.87	8.02	7.85	8.00	7.57
	DO (mg/L)	7.9	7.1	7.9	7.4	7.8	7.4	7.8	6.9
	Conductivity (µmhos/cm)	1890		~~1860~~ 1920		1950		1980	
	Temperature (°C)	24.8	24.9	24.9	24.9	24.9	24.7	24.6	25.0
STOCK	Conductivity (µmhos/cm)	—K		71300		—K		75600	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

Environmental Testing Solutions, Inc.

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



Test date

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

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

—·—·— **Warning Limits** (mean $IC_{25} \pm S_{A.10}$ or $S_{A.75}$)

····· **Control Limits** (mean $IC_{25} \pm S_{A.25}$, $S_{A.90}$, or 2 Standard Deviations)

Environmental Testing Solutions, Inc.

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

Test number	Test date	7-day IC_{25} (g/L NaCl)	CT (g/L NaCl)	S	State and USEPA Control Limits		$S_{A.10}$	Laboratory Warning Limits		$S_{A.25}$	Laboratory Control Limits		$S_{A.75}$	USEPA Warning Limits		$S_{A.90}$	USEPA Control Limits		CV
					CT - 2S	CT + 2S		CT - $S_{A.10}$	CT + $S_{A.10}$		CT - $S_{A.25}$	CT + $S_{A.25}$		CT - $S_{A.75}$	CT + $S_{A.75}$		CT - $S_{A.90}$	CT + $S_{A.90}$	
1	06-08-04	1.06																	
2	07-07-04	1.06	1.06	0.00	1.05	1.06	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.66	0.40	1.71	0.00
3	08-10-04	1.06	1.06	0.00	1.05	1.06	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.66	0.40	1.71	0.00
4	09-14-04	1.05	1.06	0.00	1.05	1.06	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.65	0.40	1.71	0.00
5	09-14-04	1.07	1.06	0.01	1.04	1.07	0.08	0.97	1.14	0.18	0.88	1.24	0.48	0.58	1.53	0.66	0.40	1.71	0.01
6	10-05-04	1.07	1.06	0.01	1.04	1.08	0.08	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
7	11-02-04	1.06	1.06	0.01	1.04	1.08	0.08	0.98	1.14	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
8	12-07-04	1.08	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
9	01-11-05	1.06	1.06	0.01	1.04	1.08	0.08	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
10	02-08-05	1.05	1.06	0.01	1.04	1.08	0.08	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
11	03-01-05	1.08	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
12	04-05-05	1.09	1.07	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.73	0.01
13	05-03-05	1.07	1.07	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.73	0.01
14	06-07-05	1.05	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.72	0.01
15	07-12-05	1.06	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.59	1.54	0.66	0.40	1.72	0.01
16	07-12-05	1.06	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.59	1.54	0.66	0.40	1.72	0.01
17	08-09-05	1.08	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.72	0.01
18	09-13-05	1.04	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.24	0.48	0.58	1.54	0.66	0.40	1.72	0.01
19	10-04-05	1.09	1.06	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.72	0.01
20	11-01-05	1.08	1.07	0.01	1.04	1.09	0.09	0.98	1.15	0.18	0.88	1.25	0.48	0.59	1.54	0.66	0.40	1.73	0.01

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

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

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

Laboratory Control and Warning Limits

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

$S_{A.10}$ = Standard deviation corresponding to the 10th percentile CV. ($S_{A.10}$ = 0.08)

$S_{A.25}$ = Standard deviation corresponding to the 25th percentile CV. ($S_{A.25}$ = 0.17)

USEPA Control and Warning Limits

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

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

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

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

Environmental Testing Solutions, Inc.

Precision of Endpoint Measurements

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

Test number	Test date	Control Survival (%)	Control Mean Reproduction (offspring/female)	CT for Control Mean Reproduction (offspring/female)	CV (%)	CT for Control Reproduction CV (%)	MSD	PMSD (%)	CT for PMSD (%)
1	06-08-04	100	32.9		6.8		3.0	9.0	
2	07-07-04	100	33.3	33.1	6.3	6.6	2.6	7.8	8.4
3	08-10-04	100	27.4	31.2	5.8	6.3	2.3	8.6	8.4
4	09-14-04	100	28.7	30.6	4.9	6.0	2.4	8.2	8.4
5	09-14-04	100	28.8	30.2	7.5	6.3	3.0	10.5	8.8
6	10-05-04	100	29.6	30.1	8.2	6.6	2.8	9.6	8.9
7	11-02-04	100	30.5	30.2	8.5	6.8	2.7	8.8	8.9
8	12-07-04	100	31.8	30.4	4.9	6.6	2.5	7.9	8.8
9	01-11-05	100	31.0	30.4	9.4	6.9	3.1	10.0	8.9
10	02-08-05	100	30.0	30.4	6.8	6.9	2.4	8.0	8.8
11	03-01-05	100	29.9	30.4	6.6	6.9	2.9	9.9	8.9
12	04-05-05	100	32.4	30.5	6.9	6.9	3.1	9.6	9.0
13	05-03-05	100	29.9	30.5	7.0	6.9	2.9	9.8	9.1
14	06-07-05	100	30.4	30.5	5.0	6.7	2.6	8.5	9.0
15	07-12-05	100	30.4	30.5	7.5	6.8	2.7	8.8	9.0
16	07-12-05	100	31.1	30.5	7.2	6.8	3.2	10.2	9.1
17	08-09-05	100	28.3	30.4	7.3	6.8	2.9	10.3	9.2
18	09-13-05	100	27.9	30.2	7.3	6.9	3.9	13.9	9.4
19	10-04-05	100	27.0	30.1	5.8	6.8	3.0	11.1	9.5
20	11-01-05	100	28.4	30.0	10.3	7.0	3.8	13.2	9.7

Note:

CV = Coefficient of variation for control reproduction.
On average, the CV for control reproduction is 7.0% in Environmental Testing Solutions, Inc. *Ceriodaphnia* chronic toxicity tests.
Lower CV bound determined by USEPA (10th percentile) = 8.9%.
Upper CV bound determined by USEPA (90th percentile) = 42%

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference
PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, Inc. chronic toxicity tests when a toxicant reduces *Ceriodaphnia* reproduction by 9.7% from the control.
Lower PMSD bound determined by USEPA (10th percentile) = 11%.
Upper PMSD bound determined by USEPA (90th percentile) = 37%.

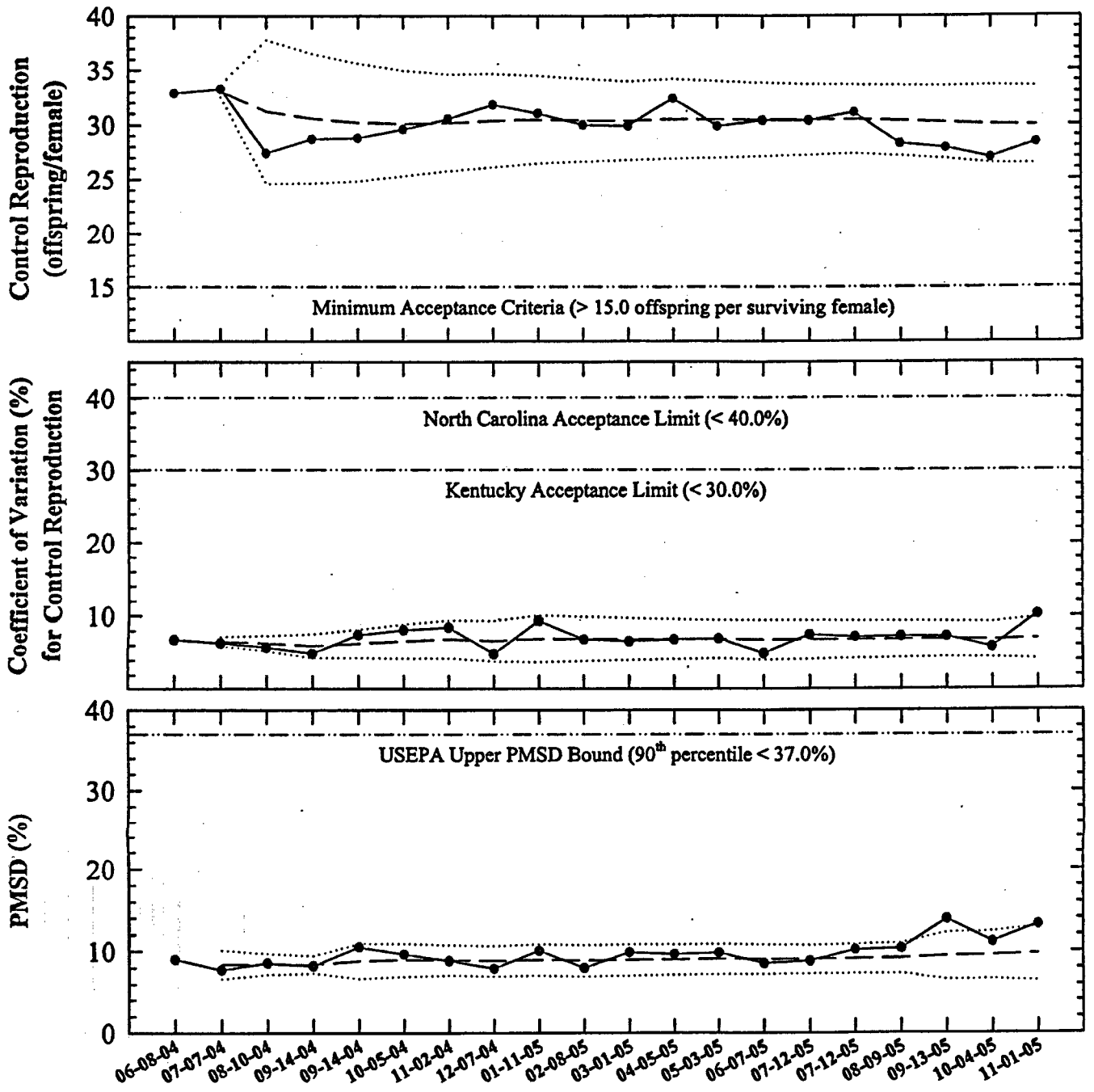
CT = Central Tendancy (Mean Control Reproduction, CV, or PMSD)

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

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

Environmental Testing Solutions, Inc.

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



—•— **Control Reproduction, Coefficient of Variation (CV), or Percent Minimum Significant Difference (PMSD)** PMSD is the minimum significant difference between the control and treatment that can be declared statistically significant.

— — · **Central Tendency** (mean Control Reproduction, CV, or PMSD)

····· **Control Limits** (mean Control Reproduction, CV, or PMSD ± 2 Standard Deviations)

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

CdNaCLCR #: 42

Dilution preparation information:						*Comments:*
NaCl CHM number:		CHM 120				
Stock preparation:		100 g NaCl/l (dissolve 50 g NaCl in 500 ml deionized water)				
Dilution prep (mg/L)	600	800	1000	1200	1400	
Stock volume (mL)	9	12	15	18	21	
Diluent volume (mL)	1491	1488	1485	1482	1479	
Total volume (mL)	1500	1500	1500	1500	1500	

Test organism information:		*Test information:*	
Organism age:	< 24-hours old	Randomizing template:	Yellow
Date and times organisms were born between:	11-01-05 0731 TO 1000	Incubator number and shelf location:	~~2B~~ 3E1
Organism source:	10-25-05 A-F	YCT batch:	09-30-05
Transfer bowl information:	pH = 7.91 SU Temperature = 24.8°C	*Selenastrum* batch:	10-16-05

Daily renewal information:

Day	Date	Test initiation, renewal, or termination time	MHS water batch used	Analyst
0	11-01-05	1101	10-24-05 B	dl
1	11-02-05	1006	10-24-05 B	dl
2	11-03-05	1004	10-31-05 A	dl
3	11-04-05	1008	10-31-05 B	dl
4	11-05-05	1010	11-03-05 A ~~10-31-05 B~~	dl
5	11-06-05	1016	11-03-05 B	dl
6	11-07-05	1003	11-05-05 A	dl
7	11-08-05	1057		dl

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	≤ 20%	7-day LC50	> 1400
% Adults having 3rd Broods:	100%	≥ 80%	NOEC	1000
% Mortality:	0%	≤ 20%	LOEC	1200
Mean Offspring/Female:	28.4	≥ 15.0 offspring/female	ChV	1095.4
% CV:	10.3%	< 40.0 %	IC25	1075.0

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

CONTROL

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	4	4	3	4	3	4	3	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	12	10	12	10	*2	0	11	0	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	13	0	0	0	0	12	11	0	11	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	14	18	12	14	12	12	11	14	13	13
Total young produced		31	34	26	30	25	30	25	29	27	27
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L
X for 3rd Broods		X	X	X	X	X	X	X	X	X	X

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	28.4

600 mg NaCl/L

Survival and Reproduction Data

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	5	4	4	4	3	4	4	3	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	11	*1	11	11	10	10	10	10	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	13	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	12	13	15	12	14	17	12	15	15	16
Total young produced		28	29	33	27	29	30	26	29	28	33
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead) *SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	29.2
% Reduction from Control:	-2.87%

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

800 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	5	4	4	3	3	4	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	14	10	11	10	12	9	10	0	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	1*	0	0	0	0	10	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	13	11	13	11	16	14	14	12	14	12
Total young produced		29	30	27	27	29	29	27	26	28	27
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	27.9
% Reduction from Control:	1.8%

1000 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	3	3	4	3	4	4	4	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	*1	13	0	12	0	9	9	0	10	9
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	10	0	10	0	11	0	0	9	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	17	14	13	14	14	11	10	10	13	12
Total young produced		32	31	26	29	29	23	23	23	27	24
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

*SPLIT BROOD

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	26.7
% Reduction from Control:	6.0%

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

1200 mg NaCl/L ***Survival and Reproduction Data***

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	4	3	1	2	5	2	2	1	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	5	5	8	6	5	8	4	0	8	6
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	4	0	0	0	0	0	3	0	0
	Adult mortality	L	L	L	X	L	L	L	L	L	L
7	Young produced	13	11	0	12	0	0	0	0	6	0
Total young produced		22	24	11	19	7	13	6	5	15	9
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	13.1
% Reduction from Control:	53.9%

1400 mg NaCl/L ***Survival and Reproduction Data***

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	0	2	0	0	3	3	0	0	1
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	2	3	7	3	4	4	5	9	4	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	7
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	1	5	5	9	3	0	6	0	5	0
Total young produced		7	8	14	12	7	7	14	9	9	8
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	9.5
% Reduction from Control:	66.5%

Environmental Testing Solutions, Inc.

Verification of *Ceriodaphnia* Reproduction Totals

Control

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	4	4	3	4	3	4	3	4	37
5	0	12	10	12	10	2	0	11	0	10	67
6	13	0	0	0	0	12	11	0	11	0	47
7	14	18	12	14	12	12	11	14	13	13	133
Total	31	34	26	30	25	30	25	29	27	27	284

1000 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	3	3	4	3	4	4	4	3	36
5	1	13	0	12	0	9	9	0	10	9	63
6	10	0	10	0	11	0	0	9	0	0	40
7	17	14	13	14	14	11	10	10	13	12	128
Total	32	31	26	29	29	23	23	23	27	24	267

600 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	5	4	4	4	3	4	4	3	5	40
5	12	11	1	11	11	10	10	10	10	12	98
6	0	0	13	0	0	0	0	0	0	0	13
7	12	13	15	12	14	17	12	15	15	16	141
Total	28	29	33	27	29	30	26	29	28	33	292

1200 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	4	3	1	2	5	2	2	1	3	27
5	5	5	8	6	5	8	4	0	8	6	55
6	0	4	0	0	0	0	0	3	0	0	7
7	13	11	0	12	0	0	0	0	6	0	42
Total	22	24	11	19	7	13	6	5	15	9	131

800 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	5	4	4	3	3	4	4	4	4	39
5	12	14	10	11	10	12	9	10	0	11	99
6	0	0	0	1	0	0	0	0	10	0	11
7	13	11	13	11	16	14	14	12	14	12	130
Total	29	30	27	27	29	29	27	26	28	27	279

1400 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	0	2	0	0	3	3	0	0	1	13
5	2	3	7	3	4	4	5	9	4	0	41
6	0	0	0	0	0	0	0	0	0	7	7
7	1	5	5	9	3	0	6	0	5	0	34
Total	7	8	14	12	7	7	14	9	9	8	95

Environmental Testing Solutions, Inc.

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

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

Test number: CdNaClCR #71 (#42 at 351 Depot St.)

Test dates: November 01-08, 2005

Reveiwed by:

Concentration (mg/L NaCl)	Replicate number										Survival (%)	Average reproduction (offspring/female)	Coefficient of variation (%)	Percent reduction from control (%)
	1	2	3	4	5	6	7	8	9	10				
Control	31	34	26	30	25	30	25	29	27	27	100	28.4	10.3	Not applicable
600	28	29	33	27	29	30	26	29	28	33	100	29.2	7.9	-2.8
800	29	30	27	27	29	29	27	26	28	27	100	27.9	4.6	1.8
1000	32	31	26	29	29	23	23	23	27	24	100	26.7	12.9	6.0
1200	22	24	11	19	7	13	6	5	15	9	100	13.1	51.6	53.9
1400	7	8	14	12	7	7	14	9	9	8	100	9.5	29.5	66.5

Dunnett's MSD value: 3.751

PMSD: 13.2

MSD = Minimum Significant Difference

PMSD = Percent Minimum Significant Difference

PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test. On average, a significant difference occurs for Environmental Testing Solutions, Inc. chronic toxicity tests when a toxicant reduces *Ceriodaphnia* reproduction by 9.7% from the control.

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

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

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

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

Environmental Testing Solutions, Inc.

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	11/1/2005	Test ID:	CdNaClCR	Sample ID:	REF-Ref Toxicant
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol	Sample Type:	NACL-Sodium chloride
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia

Comments:

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	31.000	34.000	26.000	30.000	25.000	30.000	25.000	29.000	27.000	27.000
600	28.000	29.000	33.000	27.000	29.000	30.000	26.000	29.000	28.000	33.000
800	29.000	30.000	27.000	27.000	29.000	29.000	27.000	26.000	28.000	27.000
1000	32.000	31.000	26.000	29.000	29.000	23.000	23.000	23.000	27.000	24.000
1200	22.000	24.000	11.000	19.000	7.000	13.000	6.000	5.000	15.000	9.000
1400	7.000	8.000	14.000	12.000	7.000	7.000	14.000	9.000	9.000	8.000

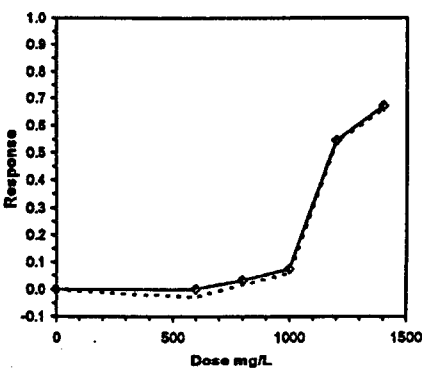
			Transform: Untransformed					Rank	1-Tailed	Isotonic	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	Sum	Critical	Mean	N-Mean
D-Control	28.400	1.0000	28.400	25.000	34.000	10.259	10			28.800	1.0000
600	29.200	1.0282	29.200	26.000	33.000	7.876	10	113.00	75.00	28.800	1.0000
800	27.900	0.9824	27.900	26.000	30.000	4.612	10	102.00	75.00	27.900	0.9688
1000	26.700	0.9401	26.700	23.000	32.000	12.860	10	90.00	75.00	26.700	0.9271
*1200	13.100	0.4613	13.100	5.000	24.000	51.579	10	55.00	75.00	13.100	0.4549
*1400	9.500	0.3345	9.500	7.000	14.000	29.461	10	55.00	75.00	9.500	0.3299

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.74299985	1.035	0.57349241	1.15846555
Bartlett's Test indicates unequal variances (p = 1.19E-04)	25.3599415	15.0862722		

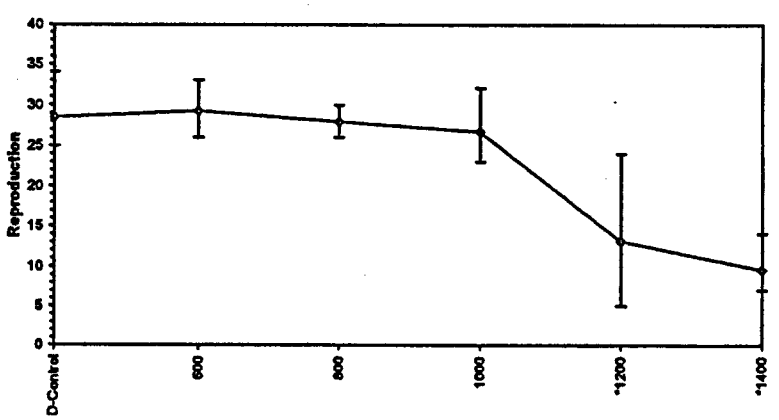
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	1000	1200	1095.44512	
Treatments vs D-Control				

Linear Interpolation (200 Resamples)

Point	mg/L	SD	95% CL		Skew
IC05	890	110.963805	675.69375	1017.77749	-0.6514
IC10	1011.47059	45.5475413	869.819444	1037.68214	-1.6247
IC15	1032.64706	20.514908	983.356136	1059.1445	-1.5647
IC20	1053.82353	17.4425533	1018.4389	1080.63192	-0.1269
IC25	1075	18.7206497	1041.48686	1116.95268	0.2515
IC40	1138.52941	26.3589432	1100.74816	1215.26767	1.1061
IC50	1180.88235	36.6868776	1138.88223	1289.27212	1.1729



Dose-Response Plot



Environmental Testing Solutions, Inc.
Statistical Analyses

Used for PMSD calculation only.			Ceriodaphnia Survival and Reproduction Test-Reproduction			
Start Date:	11/1/2005	Test ID:	CdNaClCR	Sample ID:	REF-Ref Toxicant	
End Date:	11/8/2005	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	NACL-Sodium chloride	
Sample Date:		Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia	
Comments:						

Conc-mg/L	1	2	3	4	5	6	7	8	9	10
D-Control	31.000	34.000	26.000	30.000	25.000	30.000	25.000	29.000	27.000	27.000
600	28.000	29.000	33.000	27.000	29.000	30.000	26.000	29.000	28.000	33.000
800	29.000	30.000	27.000	27.000	29.000	29.000	27.000	26.000	28.000	27.000
1000	32.000	31.000	26.000	29.000	29.000	23.000	23.000	23.000	27.000	24.000
1200	22.000	24.000	11.000	19.000	7.000	13.000	6.000	5.000	15.000	9.000
1400	7.000	8.000	14.000	12.000	7.000	7.000	14.000	9.000	9.000	8.000

			Transform: Untransformed						1-Tailed	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	28.400	1.0000	28.400	25.000	34.000	10.259	10			
600	29.200	1.0282	29.200	26.000	33.000	7.876	10	-0.488	2.287	3.751
800	27.900	0.9824	27.900	26.000	30.000	4.612	10	0.305	2.287	3.751
1000	26.700	0.9401	26.700	23.000	32.000	12.860	10	1.036	2.287	3.751
*1200	13.100	0.4613	13.100	5.000	24.000	51.579	10	9.328	2.287	3.751
*1400	9.500	0.3345	9.500	7.000	14.000	29.461	10	11.523	2.287	3.751

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)					0.74299985		1.035		0.57349241	1.15846555
Bartlett's Test indicates unequal variances (p = 1.19E-04)					25.3599415		15.0862722			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	1000	1200	1095.44512		3.7506703	0.13206586	767.706667	13.4518519	2.5E-20	5, 54
Treatments vs D-Control										

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

Daily Chemistry:

		Day					
		0		1		2	
	Analyst	AAB	AAB	AAB	AAB	AAB	AAB
Concentration	Parameter						
CONTROL MHSW	pH (S.U.)	7.73	7.86	7.79	7.74	7.67	7.69
	DO (mg/L)	7.9	8.0	7.7	7.4	7.5	7.4
	Conductivity (µmhos/cm)	314		315		306	
	Alkalinity (mg $CaCO_3$/L)	59				58	
	Hardness (mg $CaCO_3$/L)	92				87	
	Temperature (°C)	24.7	24.9	24.6	25.2	24.5	24.8
600 mg NaCl/L	pH (S.U.)	7.77	7.77	7.77	7.63	7.59	7.61
	DO (mg/L)	7.9	8.0	8.0	7.5	7.6	7.5
	Conductivity (µmhos/cm)	1590		1620		~~1700~~ AAB (1470)	
	Temperature (°C)	24.8	24.9	24.6	25.0	24.7	24.9
800 mg NaCl/L	pH (S.U.)	7.76	7.75	7.75	7.61	7.66	7.60
	DO (mg/L)	8.2	7.9	8.2	7.5	7.5	7.5
	Conductivity (µmhos/cm)	1910		1930		1830	
	Temperature (°C)	24.7	24.8	24.4	25.0	24.7	25.1
1000 mg NaCl/L	pH (S.U.)	7.74	7.73	7.75	7.58	7.66	7.58
	DO (mg/L)	8.2	7.9	8.2	7.6	7.7	7.5
	Conductivity (µmhos/cm)	2160		2170		2130 AAB (2230)	
	Temperature (°C)	24.7	24.9	24.4	24.9	24.8	25.1
1200 mg NaCl/L	pH (S.U.)	7.73	7.74	7.74	7.58	7.64	7.59
	DO (mg/L)	8.2	7.9	8.1	7.3	7.6	7.5
	Conductivity (µmhos/cm)	2590		2570		2580	
	Temperature (°C)	24.7	25.1	24.4	25.0	24.8	25.0
1400 mg NaCl/L	pH (S.U.)	7.72	7.73	7.74	7.56	7.65	7.62
	DO (mg/L)	8.3	8.0	8.1	7.4	7.6	7.6
	Conductivity (µmhos/cm)	2930		2950		2990	
	Temperature (°C)	24.7	25.0	24.5	24.9	24.7	25.1
STOCK	Conductivity (µmhos/cm)	126,000		—"		—"	
		Initial	Final	Initial	Final	Initial	Final

Species: *Ceriodaphnia dubia*

CdNaCLCR #: 42

		Day							
		3		4 JN		JN 5		6	
	Analyst	AAB	KEK	KEK	KEK	KEK	AAB	AAB	JN
Concentration	Parameter								
CONTROL MHSW	pH (S.U.)	7.53	7.64	7.87	7.57	7.70	8.12	8.10	7.95
	DO (mg/L)	7.4	7.5	7.7	7.5	7.5	7.7	7.7	7.7
	Conductivity (µmhos/cm)	310		314		313		310	
	Alkalinity (mg $CaCO_3$/L)	59		58		58		58	
	Hardness (mg $CaCO_3$/L)	97		95		93		85	
	Temperature (°C)	24.8	25.0	24.9	24.8	24.7	25.2	24.9	25.1
600 mg NaCl/L	pH (S.U.)	7.62	7.60	7.64	7.58	7.58	8.04	8.11	7.97
	DO (mg/L)	7.5	7.5	7.5	7.6	7.5	7.6	7.7	7.8
	Conductivity (µmhos/cm)	1450		1470		1450		1480	
	Temperature (°C)	24.7	25.2	25.0	24.7	24.6	25.3	24.8	25.1
800 mg NaCl/L	pH (S.U.)	7.61	7.59	7.65	7.58	7.61	8.04	8.11	7.96
	DO (mg/L)	7.6	7.6	7.6	7.6	7.5	7.6	7.6	7.9
	Conductivity (µmhos/cm)	1870		1910		1870		1830	
	Temperature (°C)	24.7	25.2	25.0	24.7	24.6	25.3	24.9	25.3
1000 mg NaCl/L	pH (S.U.)	7.60	7.58	7.64	7.59	7.61	8.02	8.10	7.94
	DO (mg/L)	7.6	7.5	7.7	7.5	7.6	7.6	7.7	7.9
	Conductivity (µmhos/cm)	2210		2320		2290		2270	
	Temperature (°C)	24.7	25.0	25.0	24.7	24.6	25.0	25.0	25.3
1200 mg NaCl/L	pH (S.U.)	7.59	7.59	7.64	7.56	7.61	8.01	8.09	7.96
	DO (mg/L)	7.6	7.5	7.9	~~7.8~~ 7.5	7.7	7.6	7.7	7.9
	Conductivity (µmhos/cm)	2550		2670		2680		2530	
	Temperature (°C)	24.7	25.2	25.0	24.9	24.6	25.0	24.8	25.0
1400 mg NaCl/L	pH (S.U.)	7.62	7.58	7.64	7.56	7.62	8.02	8.08	7.95
	DO (mg/L)	7.7	7.6	7.6	7.4	7.6	7.7	7.8	7.9
	Conductivity (µmhos/cm)	2980		3030		3010		3010	
	Temperature (°C)	24.6	25.2	25.0	24.7	24.6	25.1	24.9	25.0
STOCK	Conductivity (µmhos/cm)	—		127000		—		—	
		Initial	Final	Initial	Final	Initial	* Final	* Initial	* Final

* Higher overall pH due to new probe.

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*
Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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

TN0026450	103 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
LOW VOL. WASTE TREATMENT POND
EFFLUENT
*** NO DISCHARGE ☐ ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
PH	SAMPLE MEASUREMENT	********	********	**	7.0	********	8.1	12	0	14 / 30	GRAB
00400 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	**	6.0 MINIMUM	********	9.0 MAXIMUM	SU		THREE/ WEEK	GRAB
SOLIDS, TOTAL SUSPENDED	SAMPLE MEASUREMENT	81	131	26	********	9	14	19	0	5 / 30	GRAB
00530 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	380 MO AVG	1250 DAILY MX	LBS/DY	********	30 MO AVG	100 DAILY MX	MG/L		WEEKLY	GRAB
OIL AND GREASE	SAMPLE MEASUREMENT	<45	<48	26	********	<5	<5	19	0	5 / 30	GRAB
00556 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	190 MO AVG	250 DAILY MX	LBS/DY	********	15 MO AVG	20 DAILY MX	MG/L		WEEKLY	GRAB
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT	1.105	1.459	03	********	********	********	**	0	30 / 30	TOTALZ
50050 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	REPORT MO AVG	REPORT DAILY MX	MGD	********	********	********	**		SEE PERMIT	TOTALZ
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER			TELEPHONE		DATE		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*
Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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

TN0026450	107 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
METAL CLEANING WASTE POND
EFFLUENT

*** NO DISCHARGE XX ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
PH	SAMPLE MEASUREMENT	********	********	**		********		12			
00400 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	6.0 MINIMUM	********	9.0 MAXIMUM	SU		DAILY	GRAB
SOLIDS, TOTAL SUSPENDED	SAMPLE MEASUREMENT	********	********	**	********	********		19			
00530 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	30 DAILY MX	MG/L		DAILY	COMPOS
OIL AND GREASE	SAMPLE MEASUREMENT	********	********	**	********	********		19			
00556 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	15 DAILY MX	MG/L		DAILY	GRAB
PHOSPHORUS, TOTAL (AS P)	SAMPLE MEASUREMENT	********	********	**	********	********		19			
00665 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	1.0 DAILY MX	MG/L		DAILY	COMPOS
COPPER, TOTAL (AS CU)	SAMPLE MEASUREMENT	********	********	**	********	********		19			
01042 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	1.0 DAILY MX	MG/L		DAILY	COMPOS
IRON, TOTAL (AS FE)	SAMPLE MEASUREMENT	********	********	**	********	********		19			
01045 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	1.0 DAILY MX	MG/L		DAILY	COMPOS
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT			03	********	********	********	**			
50050 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	REPORT MO AVG	REPORT DAILY MX	MGD	********	********	********	****		DAILY	CALCTD

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER			TELEPHONE		DATE		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*
No Discharge this Period

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*
Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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

TN0026450	110 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
RECYCLED COOLING WATER
EFFLUENT
*** NO DISCHARGE XX ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
TEMPERATURE, WATER DEG. CENTIGRADE	SAMPLE MEASUREMENT	********	********	04	********	********		04			
00010 Z 0 0 INSTREAM MONITORING	PERMIT REQUIREMENT	********	********	DEG C	********	********	38.3 DAILY MX	DEG C		DAILY	GRAB-4
PH	SAMPLE MEASUREMENT	********	********	**		********		12			
00400 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	6.0 MINIMUM	********	9.0 MAXIMUM	SU		WEEKLY	GRAB
SOLIDS, TOTAL SUSPENDED	SAMPLE MEASUREMENT	********	********	**	********	********		19			
00530 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	30 DAILY MX	MG/L		DAILY	COMPOS
OIL AND GREASE	SAMPLE MEASUREMENT	********	********	**	********	********		19			
00556 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	15 DAILY MX	MG/L		DAILY	GRAB
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT			03	********	********	********	**			
50050 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	REPORT MO AVG	REPORT DAILY MX	MGD	********	********	********	****		DAILY	CALCTD
CHLORINE, TOTAL RESIDUAL	SAMPLE MEASUREMENT	********	********	**	********	********		19			
50060 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	0.10 DAILY MX	MG/L		WEEKLY	GRAB-4
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER			TELEPHONE		DATE		
J. Randy Douet 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.	Stephanie A Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*
No Discharge this Period

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*
Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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

TN0026450	110 T
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
RECYCLED COOLING WATER
EFFLUENT
*** NO DISCHARGE [XX] ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
IC25 STATRE 7DAY CHR CERIODAPHNIA	SAMPLE MEASUREMENT	********	********	**		********	********	23			
TRP3B 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	45.2 MINIMUM	********	********	PERCENT		SEMI ANNUAL	COMPOS
IC25 STATRE 7DAY CHR PIMEPHALES	SAMPLE MEASUREMENT	********	********	**		********	********	23			
TRP6C 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	45.2 MINIMUM	********	********	PERCENT		SEMI ANNUAL	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		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*
No Discharge this Period

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

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

ATTN: Stephanie A. Howard

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

TN0026450	116 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
BACKWASH
EFFLUENT

*** NO DISCHARGE ☐ ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
DEBRIS, FLOATING (SEVERITY)	SAMPLE MEASUREMENT	********	********	**	********	********	0	9A	0	1 / 30	VISUAL
01345 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	REPORT MO TOTAL	PASS=0 FAIL=1		SEE PERMIT	VISUAL
OIL AND GREASE VISUAL	SAMPLE MEASUREMENT	********	0	94	********	********	********	**	0	1 / 30	VISUAL
84066 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	REPORT MO TOTAL	YES=1 NO=0	********	********	********	****		SEE PERMIT	VISUAL
	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		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*

Operations performs visual inspections for floating debris and oil and grease during all backwashes.

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

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

ATTN: Stephanie A. Howard

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

TN0026450	117 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
BACKWASH
EFFLUENT

*** NO DISCHARGE ☐ ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
DEBRIS, FLOATING (SEVERITY)	SAMPLE MEASUREMENT	********	********	**	********	********	0	9A	0	1 / 30	VISUAL
01345 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	REPORT MO TOTAL	PASS=0 FAIL=1		SEE PERMIT	VISUAL
OIL AND GREASE VISUAL	SAMPLE MEASUREMENT	********	0	94	********	********	********	**	0	1 / 30	VISUAL
84066 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	REPORT MO TOTAL	YES=1 NO=0	********	********	********	****		SEE PERMIT	VISUAL
	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		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*

Operations performs visual inspections for floating debris and oil and grease during all backwashes.

PERMITTEE NAME/ADDRESS *(Include Facility Name/Location if Different)*
Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE SB-2A)
SODDY - DAISY TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN: Stephanie A. Howard

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

TN0026450	118 G
PERMIT NUMBER	DISCHARGE NUMBER

MONITORING PERIOD

	YEAR	MO	DAY		YEAR	MO	DAY
From	05	11	01	To	05	11	30

MAJOR
(SUBR 01)
F - FINAL
WASTEWATER & STORM WATER
EFFLUENT
*** NO DISCHARGE [XX] ***

Form Approved.
OMB No. 2040-0004

NOTE: Read instructions before completing this form.

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
OXYGEN, DISSOLVED (DO)	SAMPLE MEASUREMENT	********	********	**		********	********	19			
00300 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	2.0 DAILY MN	********	********	MG/L		TWICE/ WEEK	GRAB
SOLIDS, TOTAL SUSPENDED	SAMPLE MEASUREMENT	********	********	**	********	********		19			
00530 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	100 DAILY MX	MG/L		TWICE/ WEEK	GRAB
SOLIDS, SETTLEABLE	SAMPLE MEASUREMENT	********	********	**	********	********		25			
00545 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	********	1.0 DAILY MX	ML/L		ONCE/ MONTH	GRAB
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT			03	********	********	********	**			
50050 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	REPORT MO AVG	REPORT DAILY MX	MGD	********	********	********	*		ONCE/ BATCH	ESTIMA
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER			TELEPHONE		DATE		
J. Randy Douet 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.	Stephanie A. Howard Principal Environmental Engineer	423	843-6700	05	12	14
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)*
During this reporting period, there has been no flow from the Dredge Pond other than that resulting from rainfall.