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

July 13, 2018

Chattanooga Environmental Field Office
Division of Water Pollution Control
1301 Riverfront Parkway, #206
Chattanooga, Tennessee 37402-2013

TENNESSEE VALLEY AUTHORITY (TVA) - SEQUOYAH NUCLEAR PLANT (SQN) - NPDES PERMIT NO. TN0026450 - DISCHARGE MONITORING REPORT (DMR) FOR June 2018

Enclosed is the June 2018 Discharge Monitoring Report for Sequoyah Nuclear Plant. There were no exceedances during the reporting period. The final report for toxicity sampling conducted at outfall 101, from May 13 – May 18, 2018, is enclosed. No adverse effects were observed. The corrected May discharge monitoring report for outfall 101T is also enclosed. If you have any questions or need additional information, please contact Millicent Garland by email at mrmoore@tva.gov or by phone at (423) 843-6714.

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.

Sincerely,

Anthony L. Williams
Site Vice President
Sequoyah Nuclear Plant

Enclosures

cc (Enclosures):
U.S. Nuclear Regulatory Commission
Attn: Document Control Desk
Washington, DC 20555

IE25
NRR

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

Name TVA - SEQUOYAH NUCLEAR PLANT
Address P.O. BOX 2000
(INTEROFFICE OPS-5N-SQN)
SODDY - DAISY, TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN:Millicent Garland

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	18	06	01	To	18	06	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	********	********	**	********	********	39.9	04	0	30 / 30	RCORDR
00010 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	********	********	**Req. Mon. DAILY MAX**	DEG. C.		CONTI NUOUS	CALCTD
TEMPERATURE, WATER DEG. CENTIGRADE	SAMPLE MEASUREMENT	********	********	**	********	********	28.0	04	0	30 / 30	MODELD
00010 Z 0 INSTREAM MONITORING	PERMIT REQUIREMENT	********	********	****	********	********	**30.5 DAILY MX**	DEG. C.		CONTI NUOUS	CALCTD
TEMP. DIFF. BETWEEN SAMP. & UPSTRM DEG.C	SAMPLE MEASUREMENT	********	********	**	********	********	2.6	04	0	30 / 30	CALCTD
00016 1 S EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****.	********	********	**3.0 DAILY MX**	DEG. C.		CONTI NUOUS	CALCTD
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT	********	1863	03	********	********	********	**	0	30 / 30	RCORDR
50050 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	**Req. Mon. DAILY MAX**	MGD	********	********	********	****		CONTI NUOUS	RCORDR
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT		********	03	********	********	********	03	0	30 / 30	CALCTD
50050 1 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	**Req. Mon. MO AVG**	********	MGD	********	********	********	MGD		CONTI NUOUS	CALCTD
CHLORINE, TOTAL RESIDUAL	SAMPLE MEASUREMENT	********	********	**	********	0.016	0.035	19	0	23 / 30	GRAB
50060 1 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	********	**0.1 MO AVG**	**0.1 DAILY MAX**	MG/L		FIVE PER WEEK	CALCTD
TEMPERATURE - C, RATE OF CHANGE	SAMPLE MEASUREMENT	********	0.2	62	********	********		**	0	30 / 30	CALCTD
82234 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	**2.0 DAILY MX**	DEG C/HR	********	********	*******	****		CONTI NUOUS	CALCTD

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER			TELEPHONE		DATE		
Anthony L. Williams 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.	Site Vice President SIGNATURE OF PRINCIPAL EXECUTIVE OFFICER OR AUTHORIZED AGENT	423	843-7001	18	07	11
TYPED OR PRINTED			AREA CODE	NUMBER	YEAR	MO	DAY

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

No closed mode operation. The following injections occurred: Flogard MS 6236 (max calc. was 0.033 mg/L, limit -- 0.20 mg/L), Spectrus BD 1500 (max calc. was 0.037 mg/L, limit -- 2.0 mg/L).

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

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SODDY - DAISY, TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN:Millicent Garland

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	18	06	01	To	18	06	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	********	********	**	Monitoring Not Required	********	********	23			
TRP3B 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	**42.8 MINIMUM**	********	********	PERCENT		SEMI ANNUAL	COMPOS
IC25 STATRE 7DAY CHR PIMEPHALES	SAMPLE MEASUREMENT	********	********	**	Monitoring Not Required	********	********	23			
TRP6C 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	**42.8 MIMINUM**	********	********	PERCENT		SEMI ANNUAL	COMPOS
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

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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 not sampled in June 2018.

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Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN:Millicent Garland

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	18	06	01	To	18	06	30

MAJOR
(SUBR 01)
F - FINAL
LOW VOL. WASTE TREATMENT POND
EFFLUENT

*** NO DISCHARGE ☐ ***

Form Approved.
OMB No. 2040-0004

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PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
PH	SAMPLE MEASUREMENT	********	********	**	6.5	********	6.7	12	0	5 / 30	GRAB
00400 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	**	**6.0 MINIMUM**	********	**9.0 MAXIMUM**	SU		ONCE/ WEEK	GRAB
SOLIDS, TOTAL SUSPENDED	SAMPLE MEASUREMENT	********	********	**	********	<2.5	<2.5	19	0	1 / 30	GRAB
00530 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	**	********	**30.0 MO AVG**	**100.0 DAILY MX**	MG/L		ONCE/ MONTH	GRAB
OIL AND GREASE	SAMPLE MEASUREMENT	********	********	**	********	<5.0	<5.0	19	0	1 / 30	GRAB
00556 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	**	********	**15.0 MO AVG**	**20.0 DAILY MX**	MG/L		ONCE/ MONTH	GRAB
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT	1.022	1.114	03	********	********	********	**	0	4 / 30	INSTAN
50050 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	**Req. Mon. MO AVG**	**Req. Mon DAILY MX**	MGD	********	********	********	**		ONCE/ WEEK	INSTAN
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

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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)*

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SODDY - DAISY, TN 37384
Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN:Millicent Garland

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	18	06	01	To	18	06	30

MAJOR
(SUBR 01)
F - FINAL
RECYCLED COOLING WATER
EFFLUENT

*** NO DISCHARGE XX ***

Form Approved.
OMB No. 2040-0004

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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			
00010 1 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	**	********	********	**REPORT DAILY MX**	DEG C		CONTIN UOUS	CALCTD
TEMPERATURE, WATER DEG. CENTIGRADE	SAMPLE MEASUREMENT	********	********	**	********	********		04			
00010 Z 0 INSTREAM MONITORING	PERMIT REQUIREMENT	********	********	**	********	********	**30.5 DAILY MX**	DEG C		CONTIN UOUS	CALCTD
TEMP. DIFF. BETWEEN SAMP. & UPSTRM DEG.C	SAMPLE MEASUREMENT	********	********	**	********	********		04			
00016 1 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	**	********	********	**5 DAILY MX**	DEG C		CONTIN UOUS	CALCTD
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT	********		03	********	********	********	**			
50050 1 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	**Req. Mon. DAILY MX**	MGD	********	********	********	**		CONTIN UOUS	RCORDR
CHLORINE, TOTAL RESIDUAL	SAMPLE MEASUREMENT	********	********	**	********			19			
50060 1 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	**	********	**0.1 MO AVG**	**0.1 DAILY MX**	MG/L		Five per Week	CALCTD
TEMPERATURE - C, RATE OF CHANGE	SAMPLE MEASUREMENT	********		04	********	********	********	**			
82234 1 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	**2 DAILY MX**	DEG C	********	********	********	**		CONTIN UOUS	CALCTD
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

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TYPED OR PRINTED			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)*

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Facility TVA - SEQUOYAH NUCLEAR PLANT
Location HAMILTON COUNTY

ATTN:Millicent Garland

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	18	06	01	To	18	06	30

MAJOR
(SUBR 01)
F - FINAL
RECYCLED COOLING WATER
EFFLUENT

*** NO DISCHARGE XX ***

Form Approved.
OMB No. 2040-0004

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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	********	********	****	**42.8 MINIMUM**	********	********	PERCENT		SEMI ANNUAL	COMPOS
IC25 STATRE 7DAY CHR PIMEPHALES	SAMPLE MEASUREMENT	********	********	**		********	********	23			
TRP6C 1 0 0 EFFLUENT GROSS VALUE	PERMIT REQUIREMENT	********	********	****	**42.8 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										

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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

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Location HAMILTON COUNTY

ATTN:Millicent Garland

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	18	06	01	To	18	06	30

MAJOR
(SUBR 01)
F - FINAL
WASTEWATER & STORM WATER
EFFLUENT

*** NO DISCHARGE XX ***

Form Approved.
OMB No. 2040-0004

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		AVERAGE	MAXIMUM	UNITS	MINIMUM	AVERAGE	MAXIMUM	UNITS			
OXYGEN, DISSOLVED (DO)	SAMPLE MEASUREMENT	********	********	**		********	********	19			
00300 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	**2 MINIMUM**	********	********	MG/L		TWICE/ WEEK	GRAB
SOLIDS, TOTAL SUSPENDED	SAMPLE MEASUREMENT	********	********	**	********	********		19			
00530 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	********	********	**100 DAILY MX**	MG/L		TWICE/ WEEK	GRAB
SOLIDS, SETTLEABLE	SAMPLE MEASUREMENT	********	********	**	********	********		25			
00545 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	********	********	**1 DAILY MX**	ML/L		ONCE/ MONTH	GRAB
FLOW, IN CONDUIT OR THRU TREATMENT PLANT	SAMPLE MEASUREMENT			03	********	********	********	**			
50050 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	**Req. Mon. MO AVG**	**Req. Mon. DAILY MX**	MGD	********	********	********	*		ONCE/ BATCH	ESTIMA
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										

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TYPED OR PRINTED			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. No Discharge this Period

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Location HAMILTON COUNTY

ATTN:Millicent Garland

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	18	05	01	To	18	05	31

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	3 / 180	COMPOS
TRP3B 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	**42.8 MINIMUM**	********	********	PERCENT		SEMI ANNUAL	COMPOS
IC25 STATRE 7DAY CHR PIMEPHALES	SAMPLE MEASUREMENT	********	********	**	>100.0	********	********	23	0	3 / 180	COMPOS
TRP6C 1 0 EFFLUENT GROSS	PERMIT REQUIREMENT	********	********	****	**42.8 MIMINUM**	********	********	PERCENT		SEMI ANNUAL	COMPOS
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
	SAMPLE MEASUREMENT										
	PERMIT REQUIREMENT										
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	PERMIT REQUIREMENT										

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TYPED OR PRINTED			AREA CODE	NUMBER	YEAR	MO	DAY

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

Toxicity was sampled May 14 - 18, 2018. The final report will be included with the June DMR.

TENNESSEE VALLEY AUTHORITY
TOXICITY TEST REPORT

INTRODUCTION / EXECUTIVE SUMMARY

Report Date: June 01, 2018

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): 1,579
6. Receiving Stream: Tennessee River (TRM 483.6)
7. 1Q10: 3.491
8. Outfall Tested: 101
9. Dates Sampled: May 13 – 18, 2018
10. Average Flow on Days Sampled (MGD): 1805, 1810, 1788
11. Pertinent Site Conditions: Production / operation data will be provided upon request.
12. Test Dates: May 15 – 22, 2018
13. Test Type: Short-term Chronic Definitive
14. Test Species: Fathead Minnows (*Pimephales promelas*)
    Daphnids (*Ceriodaphnia dubia*)
15. Concentrations Tested (%):
    Pimephales promelas: UV treated Outfall 101: 10.7, 21.4, 42.8, 85.6, 100
    UV treated Intake: 100

    Ceriodaphnia dubia: Non-treated Outfall 101: 10.7, 21.4, 42.8, 85.6, 100
    Non-treated Intake: 100
16. Permit Limit Endpoint (%): Outfall 101: IC_{25} = 42.8%
17. Test Results: Outfall 101: *Pimephales promelas:* $IC_{25} > 100\%$
    Ceriodaphnia dubia: $IC_{25} > 100\%$

18. Facility Contact: Millicent Garland Phone #: (423) 843-6714

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

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

21. TVA Contact: Donald W. Snodgrass Phone #: (256) 386-2787

22. Notes: Exposures to samples collected May 13 – 18, 2018 from Outfall 101 resulted in no toxic effects to fathead minnows or daphnids. The resulting IC25 values, for both species, were >100 percent. Exposure of minnows and daphnids to intake samples resulted in no significant difference from the controls during this study period.

Independent Review by Kelley E. Keenan:

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) Last Used By
101	05-13-18 0755 to 05-14-18 0655	05-14-18 1305	2.9	<0.10	05-15-18 1105 05-16-18 1008
Intake	05-13-18 0812 to 05-14-18 0712	05-14-18 1305	2.2	<0.10	05-15-18 1105 05-16-18 1008
101	05-15-18 0722 to 05-16-18 0622	05-16-18 1240	1.0	<0.10	05-17-18 1013 05-18-18 1037
Intake	05-15-18 0740 to 05-16-18 0640	05-16-18 1240	1.2	<0.10	05-17-18 1013 05-18-18 1037
101	05-17-18 0657 to 05-18-18 0557	05-18-18 1315	1.1, 1.3†	<0.10	05-19-18 1027 05-20-18 1053 05-21-18 1006
Intake	05-17-18 0711 to 05-18-18 0611	05-18-18 1315	0.5	<0.10	05-19-18 1027 05-20-18 1053 05-21-18 1006

*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:	In-house Cultures	In-house Cultures
2. Age:	< 24-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):	05-15-18 1024 ET	05-15-18 1105 ET
7. Test Termination: (Date/Time):	05-22-18 0926 ET	05-22-18 1025 ET
8. Test Temperature: Outfall 101:	Mean = 24.6°C (24.2 – 25.1°C)	Mean = 24.9°C (24.6 – 25.2°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).

Independent Review by Kelley E. Keenan

TOXICITY TEST RESULTS (see Appendix C for Bench Sheets)

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

Conducted May 15 – 22, 2018 using effluent from Outfall 101.

Test Solutions (% Effluent)	Percent Surviving (time interval used – days)						
	1	2	3	4	5	6	7
Control, UV-treated	100	100	100	100	100	100	100
10.7%	100	100	100	100	100	100	100
21.4%	100	100	100	100	100	100	100
42.8%	100	100	100	100	100	100	100
85.6%	100	100	100	100	100	100	100
100.0%	100	100	100	100	100	100	100
Intake	100	100	100	100	100	98	98
Control, Non-treated	100	100	100	100	100	100	98

Test Solutions (% Effluent)	Mean Dry Weight (mg) (replicate number)				
	1	2	3	4	Mean
Control, UV-treated	0.688	0.614	0.653	0.763	0.680
10.7%	0.741	0.759	0.728	0.743	0.743
21.4%	0.728	0.743	0.719	0.750	0.735
42.8%	0.724	0.753	0.675	0.715	0.717
85.6%	0.729	0.756	0.630	0.705	0.705
100.0%	0.705	0.690	0.731	0.694	0.705
Intake	0.628	0.568	0.608	0.677	0.620
Control, Non-treated	0.736	0.759	0.644	0.602	0.685

IC_{25} Value: > 100% Permit Limit: 42.8% 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: < 1.0 TUc* Permit Limit: 2.3 TUc

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

Independent Review by Valley E. Keenan:

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 May 15 – 22, 2018 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
10.7%	100	100	100	100	100	100	100
21.4%	100	100	100	100	100	100	100
42.8%	100	100	100	100	100	100	100
85.6%	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	28	28	27	29	33	28	27	28	29	27	28.4
10.7%	31	33	32	32	29	29	33	29	31	32	31.1
21.4%	34	32	30	31	31	32	30	30	34	36	32.0
42.8%	32	35	32	37	34	37	34	30	33	31	33.5
85.6%	36	36	35	35	39	33	31	34	35	35	34.9
100.0%	37	33	37	39	36	39	36	34	34	37	36.2

IC_{25} Value: > 100% Permit Limit: 42.8% 95% Confidence Limits: Upper Limit: NA Lower Limit: NA	Calculated TU Estimates: < 1.0 TUc* Permit Limit: 2.3 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 May 15 – 22, 2018 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	30	29	26	30	30	29	31	27	30	28	29.0
Intake	35	33	33	34	34	36	32	33	34	35	33.9

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 = $100/ IC_{25}$

REFERENCE TOXICANT TEST RESULTS (see Appendix A and D)

Species	Date	Time	Duration	Toxicant	Results (IC_{25})
Pimephales promelas	May 08 – 15, 2018	0735	7 days	KCl	0.72 g/L
Ceriodaphnia dubia	May 08 – 15, 2018	0911	7-days	NaCl	1.08 g/L

Independent Review by Kelley F. Keenan

PHYSICAL/CHEMICAL SUMMARY

Water Chemistry Mean Values and Ranges for UV-treated *Pimephales promelas* and Non-treated *Ceriodaphnia dubia*, Sequoyah Nuclear Plant (SQN), Effluent Outfall 101 and Intake performed May 15-22, 2018.

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, Non-treated	24.7 24.6 - 24.8	24.5 24.3 - 24.6	7.8 7.6 - 8.0	7.3 6.3 - 8.0	7.95 7.85 - 8.04	7.68 7.49 - 7.84	307 300 - 315	61 60 - 61	83 82 - 84	- - - -
	Control, UV-treated	24.7 24.6 - 24.8	24.4 24.3 - 24.6	7.9 7.6 - 8.1	7.5 6.9 - 8.0	7.98 7.91 - 8.04	7.74 7.54 - 7.85	309 298 - 318	61 59 - 63	83 82 - 84	- - - -
	10.7%	24.8 24.7 - 24.8	24.5 24.3 - 24.6	7.9 7.7 - 8.1	7.5 7.0 - 7.9	8.00 7.93 - 8.05	7.68 7.49 - 7.84	293 286 - 300	- - - -	- - - -	- - - -
	21.4%	24.8 24.7 - 24.9	24.5 24.2 - 24.7	8.0 7.7 - 8.1	7.4 6.9 - 8.0	8.01 7.94 - 8.06	7.68 7.50 - 7.85	276 267 - 283	- - - -	- - - -	- - - -
	42.8%	24.8 24.7 - 24.9	24.4 24.2 - 24.5	8.0 7.7 - 8.1	7.4 6.9 - 8.0	8.01 7.94 - 8.06	7.71 7.56 - 7.84	242 237 - 246	- - - -	- - - -	- - - -
	85.6%	24.9 24.8 - 24.9	24.3 24.2 - 24.6	8.0 7.8 - 8.3	7.4 6.8 - 8.0	7.99 7.92 - 8.05	7.62 7.45 - 7.76	178 171 - 187	- - - -	- - - -	- - - -
	100%	24.9 24.8 - 25.0	24.4 24.2 - 24.6	8.1 7.7 - 8.3	7.4 6.7 - 8.1	7.98 7.91 - 8.04	7.61 7.43 - 7.74	154 146 - 162	58 54 - 62	63 60 - 64	< 0.10 < 0.10 - < 0.10
	Intake	25.0 24.9 - 25.1	24.5 24.2 - 24.7	8.1 7.8 - 8.3	7.4 6.9 - 8.2	7.98 7.89 - 8.04	7.60 7.31 - 7.79	154 143 - 161	56 56 - 56	60 60 - 60	< 0.10 < 0.10 - < 0.10
Ceriodaphnia dubia	Control, Non-treated	24.7 24.6 - 24.8	25.1 24.9 - 25.2	7.8 7.6 - 8.0	7.9 7.7 - 8.0	7.95 7.85 - 8.04	8.00 7.92 - 8.04	307 300 - 315	61 60 - 61	83 82 - 84	- - - -
	10.7%	24.8 24.7 - 24.9	24.9 24.8 - 25.2	7.9 7.8 - 8.0	7.9 7.8 - 8.0	7.99 7.91 - 8.07	8.00 7.92 - 8.05	287 280 - 294	- - - -	- - - -	- - - -
	21.4%	24.8 24.7 - 24.9	25.0 24.8 - 25.0	7.9 7.7 - 8.1	7.9 7.8 - 8.0	8.00 7.92 - 8.07	8.00 7.92 - 8.03	273 265 - 282	- - - -	- - - -	- - - -
	42.8%	24.8 24.7 - 24.9	24.9 24.8 - 25.1	7.9 7.7 - 8.1	7.9 7.8 - 8.0	8.00 7.93 - 8.07	7.99 7.91 - 8.03	241 237 - 250	- - - -	- - - -	- - - -
	85.6%	24.9 24.7 - 25.0	24.9 24.8 - 25.2	7.9 7.7 - 8.1	7.9 7.8 - 8.0	7.98 7.89 - 8.06	7.97 7.89 - 8.02	176 171 - 179	- - - -	- - - -	- - - -
	100%	24.9 24.8 - 25.0	24.9 24.7 - 25.1	8.0 7.8 - 8.2	7.9 7.8 - 8.1	7.98 7.88 - 8.04	7.95 7.88 - 8.00	151 138 - 156	58 58 - 58	64 60 - 68	< 0.10 < 0.10 - < 0.10
	Intake	24.9 24.7 - 25.0	24.8 24.8 - 25.0	8.0 7.8 - 8.2	8.0 7.8 - 8.1	7.97 7.91 - 8.03	7.97 7.88 - 8.04	154 147 - 159	57 56 - 58	61 60 - 64	< 0.10 < 0.10 - < 0.10

*Note: Total residual chlorine was performed on non-treated Outfall 101 and Intake samples.

Overall temperature (°C)	Average	Minimum	Maximum
Pimephales promelas	24.6	24.2	25.1
Ceriodaphnia dubia	24.9	24.6	25.2

SUMMARY / CONCLUSIONS

Exposures to samples collected May 13 – 18, 2018 from Outfall 101 resulted in no toxic effects to fathead minnows or daphnids. The resulting IC25 values, for both species, were >100 percent. Exposure of minnows and daphnids to intake samples resulted in no significant difference from the controls during this study period.

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*

   Samples used in the fathead minnow test were exposed to UV light for two minutes prior to introduction of test organisms. UV treatment is used to control interference of fish pathogens. This treatment method was approved on November 23, 2015 by the State of Tennessee in a letter from Jessica Murphy to Terry Cheek, Senior Manager of TVA Water Permits, Compliance, and Monitoring.

2. *Ceriodaphnia dubia*

   None

DEVIATIONS / MODIFICATIONS TO PRETEST CULTURE OR HOLDING OF TEST ORGANISMS

1. *Pimephales promelas*

   None

2. *Ceriodaphnia dubia*

   None

PHYSICAL AND CHEMICAL METHODS

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

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

3. Temperature was measured by SM 2550 B-2010.

4. Dissolved oxygen was measured by SM 4500-O G-2011.

5. The pH was measured by SM 4500-H+ B-2011.

6. Conductance was measured by SM 2510 B-2011.

7. Alkalinity was measured by SM 2320 B-2011.

8. Total hardness was measured by SM 2340 C-2011.

9. Total residual chlorine was measured by ORION 97-70-1977.

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. Standard Methods for the Examination of Water and Wastewater, 22nd Edition, 2012.

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

Independent Review by Kelley E. Keenan

Sequoyah Nuclear Plant Biomonitoring
May 15 – 22, 2018

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 Outfall 101
Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling,
February 6, 2005 – May 18, 2018

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
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	-	-	-	-	-	-	-

Table B-1. Sequoyah Nuclear Plant Outfall 101
Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling,
February 6, 2005 – May 18, 2018

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	MSW 101 mg/L Phosphate
11/12/2006	-	0.0055	-	-	-	-	-	-	-	-
11/13/2006	-	0.0068	-	-	-	-	-	-	0.037	-
11/14/2006	-	0.0143	-	-	-	-	-	-	0.037	-
11/15/2006	-	0.0068	-	-	-	-	-	-	0.037	-
11/16/2006	-	0.0267	-	-	-	-	-	-	0.037	-
11/17/2006	-	0.0222	-	-	-	-	-	-	-	-
11/26/2006	-	0.0188	-	-	-	-	-	-	-	-
11/27/2006	-	0.0138	-	-	-	-	-	-	-	-
11/28/2006	-	0.0120	-	-	-	-	-	-	-	-
11/29/2006	-	0.0288	-	-	-	-	-	-	-	-
11/30/2006	-	0.0376	-	-	-	-	-	-	-	-
12/01/2006	-	0.0187	-	-	-	-	-	-	-	-
05/28/07	-	-	-	-	-	-	-	-	-	0.015
05/29/07	-	-	-	-	-	-	-	-	0.036	0.015
05/30/07	-	0.0084	-	-	-	-	-	0.017	0.036	0.015
05/31/07	-	0.0103	-	-	-	-	-	-	0.036	0.015
06/01/07	-	0.0164	-	-	-	-	-	0.017	0.036	0.015
06/02/07	-	0.0305	-	-	-	-	-	-	-	0.015
12/02/07	-	0.0241	-	-	-	-	-	-	-	-
12/03/07	-	0.0128	-	-	-	-	-	-	-	-
12/04/07	-	0.0238	-	-	-	-	-	-	-	-
12/05/07	-	0.0158	-	-	-	-	-	-	-	-
12/06/07	-	0.0162	-	-	-	-	-	-	-	-
12/07/07	-	0.0175	-	-	-	-	-	-	-	
04/13/08	-	0.0039	-	-	-	-	-	-	-	-
04/14/08	-	0.0124	-	-	-	-	-	-	-	-
04/15/08	-	0.0229	-	-	-	-	-	-	-	-
04/16/08	-	0.0143	-	-	-	-	-	-	-	-
04/17/08	-	0.0120	-	-	-	-	-	-	-	-
04/18/08	-	0.0149	-	-	-	-	-	-	-	-
10/26/08	-	0.0260	-	-	-	-	-	-	-	-
10/27/08	-	0.0151	-	-	-	-	-	0.017	-	-
10/28/08	-	0.0172	-	-	-	-	-	-	0.041	-
10/29/08	-	0.0154	-	-	-	-	-	0.018	0.041	0.030
10/30/08	-	-	-	-	-	-	-	-	0.041	0.030
10/31/08	-	0.0086	-	-	-	-	-	-	0.041	0.030

Table B-1. Sequoyah Nuclear Plant Outfall 101
Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling,
February 6, 2005 – May 18, 2018

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	Spectrus CT1300 mg/L Quat	H-150M mg/L Quat	MSW 101 mg/L Phosphate
02/08/09	-	0.0197	-	-	-	-	-	0.017	-	-	-
02/09/09	-	0.0237	-	-	-	-	-	0.017	-	-	-
02/10/09	-	0.0104	-	-	-	-	-	0.021	-	-	-
02/11/09	-	0.0155	-	-	-	-	-	0.017	-	-	-
02/12/09	-	0.0106	-	-	-	-	-	0.017	-	-	-
02/13/09	-	-	-	-	-	-	-	-	-	-	-
05/10/09	-	0.0129	-	-	-	-	-	-	-	-	-
05/11/09	-	0.0415	-	-	-	-	-	-	-	0.0446	-
05/12/09	-	0.0053	-	-	-	-	-	-	-	0.0396	-
05/13/09	-	0.0049	-	-	-	-	-	-	-	0.0396	-
05/14/09	-	<0.0141	-	-	-	-	-	-	-	0.0397	-
05/15/09	-	<0.0160	-	-	-	-	-	-	-	-	-
11/15/09	-	0.025	-	-	-	-	-	-	-	-	-
11/16/09	-	0.0152	-	-	-	-	-	-	-	-	-
11/17/09	-	0.0255	-	-	-	-	-	-	-	-	-
11/18/09	-	0.0306	-	-	-	-	-	-	-	-	-
11/19/09	-	0.0204	-	-	-	-	-	-	-	-	-
11/20/09	-	0.0093	-	-	-	-	-	-	-	-	-
05/09/10	-	0.0192	-	-	-	-	-	-	-	-	-
05/10/10	-	0.0055	-	-	-	-	-	-	-	-	-
05/11/10	-	0.0100	-	-	-	-	-	-	0.039	-	-
05/12/10	-	0.0171	-	-	-	-	-	-	0.039	-	-
05/13/10	-	0.0041	-	-	-	-	-	-	0.039	-	-
05/14/10	-	0.0099	-	-	-	-	-	-	0.039	-	-

Table B-1. Sequoyah Nuclear Plant Outfall 101
Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling,
February 6, 2005 – May 18, 2018

Date	Sodium Hypo-chlorite mg/L TRC	Towerbrom mg/L TRC	PCL-222 mg/L Phos-phate	PCL-401 mg/L Copoly-mer	CL-363 mg/L DMAD	Cuprostat -PF mg/L Azole	H-130M mg/L Quat	Nalco 73551 mg/L EO/PO	Spectrus CT1300 mg/L Quat	H-150M mg/L Quat	MSW 101 mg/L Phos-phate	Floguard MS6236 mg/L Phosphate
10/31/10	-	-	-	-	-	-	-	-	-	-	-	-
11/01/10	-	0.0122	-	-	-	-	-	-	-	-	-	-
11/02/10	-	0.0112	-	-	-	-	-	-	-	-	-	-
11/03/10	-	0.0163	-	-	-	-	-	-	-	-	-	-
11/04/10	-	0.0107	-	-	-	-	-	-	-	-	-	-
11/05/10	-	0.0132	-	-	-	-	-	-	-	-	-	-
05/01/2011	-	-	-	-	-	-	-	-	-	-	-	-
05/02/2011	-	-	-	-	-	-	-	-	0.04	-	-	-
05/03/2011	-	-	-	-	-	-	-	-	0.04	-	-	-
05/04/2011	-	0.0155	-	-	-	-	-	-	0.04	-	-	-
05/05/2011	-	0.0179	-	-	-	-	-	-	0.04	-	-	-
05/06/2011	-	0.0089	-	-	-	-	-	-	-	-	-	-
11/06/2011	-	0.0168	-	-	-	-	-	-	-	-	-	-
11/07/2011	-	0.0225	-	-	-	-	-	-	-	-	-	-
11/08/2011	-	0.0141	-	-	-	-	-	-	-	-	-	-
11/09/2011	-	0.0239	-	-	-	-	-	-	-	-	-	-
11/10/2011	-	0.0242	-	-	-	-	-	-	-	-	-	-
11/11/2011	-	0.0231	-	-	-	-	-	-	-	-	-	-
05/06/2012	-	-	-	-	-	-	-	-	-	-	-	-
05/07/2012	-	-	-	-	-	-	-	-	-	-	-	-
05/08/2012	-	-	-	-	-	-	-	-	0.041	-	-	-
05/09/2012	-	0.0145	-	-	-	-	-	-	0.041	-	-	-
05/10/2012	-	0.0298	-	-	-	-	-	-	0.041	-	-	-
05/11/2012	-	0.0174	-	-	-	-	-	-	-	-	-	-
08/12/2012	-	-	-	-	-	-	-	-	-	-	-	0.029
08/13/2012	-	0.0256	-	-	-	-	-	0.028	0.037	-	-	0.029
08/14/2012	-	0.0209	-	-	-	-	-	-	0.037	-	-	0.029
08/15/2012	-	0.0279	-	-	-	-	-	0.028	-	-	-	0.029
08/16/2012	-	0.0076	-	-	-	-	-	-	-	-	-	0.029
08/17/2012	-	0.0446	-	-	-	-	-	-	-	-	-	0.032
05/12/2013	-	0.0099	-	-	-	-	-	-	-	-	-	-
05/13/2013	-		-	-	-	-	-	-	-	-	-	0.064
05/14/2013	-	0.0091	-	-	-	-	-	0.039	-	-	-	0.064
05/15/2013	-	0.0096	-	-	-	-	-	0.039	-	-	-	0.064
05/16/2013	-	0.0229	-	-	-	-	-	-	-	-	-	0.032
05/17/2013	-	0.0063	-	-	-	-	-	-	-	-	-	0.032
09/15/2013	-	-	-	-	-	-	-	-	-	-	-	0.03
09/16/2013	-	0.0072	-	-	-	-	-	-	0.0379	-	-	0.03
09/17/2013	-	0.0107	-	-	-	-	-	0.036	0.0379	-	-	0.03
09/18/2013	-	0.0217	-	-	-	-	-	0.036	0.0379	-	-	0.03
09/19/2013	-	0.0172	-	-	-	-	-	-	-	-	-	0.03
09/20/2013	-	0.0173	-	-	-	-	-	-	-	-	-	0.03

Table B-1. Sequoyah Nuclear Plant Outfall 101
Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling,
February 6, 2005 – May 18, 2018

Date	Sodium Hypo-chlorite mg/L TRC	Towerbrom mg/L TRC	PCL-222 mg/L Phos-phate	PCL-401 mg/L Copoly-mer	CL-363 mg/L DMAD	Cuprostat -PF mg/L Azole	H-130M mg/L Quat	Nalco 73551 mg/L EO/PO	Spectrus CT1300 mg/L Quat	H-150M mg/L Quat	MSW 101 mg/L Phos-phate	Floguard MS6236 mg/L Phosphate
05/04/2014	-	0.0118	-	-	-	-	-	-	-	-	-	-
05/05/2014	-	0.0112	-	-	-	-	-	-	-	-	-	-
05/06/2014	-	0.0096	-	-	-	-	-	-	-	-	-	-
05/07/2014	-	0.0164	-	-	-	-	-	-	-	-	-	-
05/08/2014	-	0.0235	-	-	-	-	-	-	-	-	-	-
05/09/2014	-	0.0110	-	-	-	-	-	-	-	-	-	-
09/07/2014	-	-	-	-	-	-	-	-	-	-	-	-
09/08/2014	-	-	-	-	-	-	-	-	0.04	-	-	-
09/09/2014	-	-	-	-	-	-	-	-	0.04	-	-	-
09/10/2014	-	-	-	-	-	-	-	-	0.04	-	-	-
09/11/2014	-	0.0070	-	-	-	-	-	-	-	-	-	-
09/12/2014	-	0.0074	-	-	-	-	-	-	-	-	-	-
08/09/2015		-							-			-
08/10/2015		0.0195							0.03			-
08/11/2015		0.0275							0.03			-
08/12/2015		0.0213							-			0.03
08/13/2015		0.0192							-			0.03
08/14/2015		0.0182							-			0.03
10/18/2015		0.0162										
10/19/2015		0.0125										
10/20/2015		0.0120										
10/21/2015		0.0130										
10/22/2015		0.0174										
10/23/2015		0.0156										
05/15/2016		-										
05/16/2016		0.0209										
05/17/2016		0.0210										
05/18/2016		0.0361										
05/19/2016		0.0254										
05/20/2016		0.0261										
07/31/2016		-							-			
08/01/2016		0.0091							0.03			
08/02/2016		0.0093							0.03			
08/03/2016		0.0209							0.03			
08/04/2016		-							-			
08/05/2016		-							-			
04/30/2017		-										
05/01/2017		0.0298										
05/02/2017		0.0218										
05/03/2017		0.0260										
05/04/2017		-										
05/05/2017		-										

Table B-1. Sequoyah Nuclear Plant Outfall 101
Diffuser Discharge Concentrations of Chemicals Used to Control Microbiologically Induced Corrosion and Mollusks, During Toxicity Test Sampling, February 6, 2005 – May 18, 2018

Date	Towerbrom mg/L TRC	Floguard MS 6237 mg/L Phosphate/Zinc	Spectrus BD1500 mg/L Quat	Nalco 73551 mg/L EO/PO	Spectrus CT1300 mg/L Quat	Floguard MS6236 mg/L Phosphate
07/23/2017	-	-	-			
07/24/2017	0.0124	0.04/0.01	-			
07/25/2017	0.0081	0.04/0.01	-			
07/26/2017	0.0232	0.04/0.01	0.03			
07/27/2017	0.0179	-	-			
07/28/2017	0.0296	-	0.03			
5/13/2018						
5/14/2018						
5/15/2018	0.0229		0.030			0.0275
5/16/2018	0.0159		0.030			0.0275
5/17/2018	0.0133		0.030			0.0275
5/18/2018						

Sequoyah Nuclear Plant Biomonitoring
May 15 – 22, 2018

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: Andy Panter, Marcus O. Moore, Lyndell Hamilton

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): Sonic Delivery

General Comments:
Took samples from 101 sampler @ 0747 on 5-14-18
Took samples from Int. sampler @ 0825 on 5-14-18
* 101 Backup sample was left on site
* Diss. Metals were collected & kept on site

Field Identification / Sample Description	Grab/Comp	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Project # 13337 Laboratory Use				
						Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time ET	Appearance
	---	Start	End	---	---									
SQN-101-TOX	Comp	5-13-18 0755 ET	5-14-18 0655 ET	~~2~~ 1 (2.5gal)	1805.00			X		180514.02	2.9°C	JA	1305	*
SQN-INT-TOX	Comp	5-13-18 0812 ET	5-14-18 0712 ET	1 (2.5 gal)	NA			X		180514.03	2.2°C	JA	1305	*

Sample Custody – Fill In From Top Down

* CUSTODY SEALS INTACT. SAMPLES RECEIVED IN GOOD CONDITION.

Relinquished By (Signature):	Date/Time	Received By (Signature):	Date/Time
TVA	5-14-18/0858 ET	BR Sikes SONIC DELIVERED	05-14-18 08:58 ET
BR Sikes SONIC DELIVERED	05-14-18/13:05 ET	ETS	05-14-18 1305 ET

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.

Whole Effluent Sample Receipt Log

*Sample temperature performed using Sample Receiving Thermometer: SN 160928622

Date Received	Time Received	Received by	Received from	*Sample Temp. (°C)	Project number	Sample number	Sample name and description	State	Comments
05-14-18	0906	K. Keenan	Fed - Ex	NA	13336	180514 .01	CORTROL IS3020	TN	
05-14-18	1305	J. Sumner	TVA Courier	2.9	13337	180514 .02	TVA - Sequoyah Nuclear Plant - 101	TN	
05-14-18	1305	J. Sumner	TVA Courier	2.2	13337	180514 .03	TVA - Sequoyah Nuclear Plant - Intake	TN	

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: Lyndell P. Williams, Marcus O. Moore

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): Sonic Delivery

General Comments:
Took samples from 101 sampler @ 0727 on 5-16-18
Took samples from INT sampler @ 0741 on 5-16-18
* 101 Backup samples was kept on site
* Diss. Metals were collected & kept on site

Field Identification / Sample Description	Grab/Comp	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Project # 13337 Laboratory Use				
		Start	End			Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time ET	Appearance
SQN-101-TOX	Comp	5-15-18 0722 ET	5-16-18 0622 ET	2 (2.5gal) 1 (Moore 5-16-18)	184.768			X		180516.04	1.0 °C	JJ	1240	#
SQN-INT-TOX	Comp	5-15-18 0740 ET	5-16-18 0640 ET	1 (2.5 gal)	NA			X		180516.05	1.2 °C	JJ	1240	#

Sample Custody – Fill In From Top Down

CUSTODY SEAL INTACT. SAMPLES RECEIVED IN GOOD CONDITION.

Relinquished By (Signature):		Date/Time	Received By (Signature):		Date/Time
M. O. M	TVA	5-16-18 / 0817 ET	B R Shirley	Sonic Delivery	05-16-18 / 0817 ET
B R Shirley	Sonic Delivery	05-16-18 / 12:40 ET	JJ	ETS	05-16-18 1240 ET

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.

Whole Effluent Sample Receipt Log

*Sample temperature performed using Sample Receiving Thermometer: SN 160928622

Date Received	Time Received	Received by	Received from	*Sample Temp. (°C)	Project number	Sample number	Sample name and description	State	Comments
05-16-18	0827	K. Keenan	S. Gouge	3.1	13340	180516 .01	Baxter Healthcare	NC	
05-16-18	0957	K. Keenan	Fed - Ex	2.3	13341	180516 .02	Enfield WWTP	NC	
05-16-18	0957	K. Keenan	Fed - Ex	2.3	13342	180516 .03	Scotland Neck WWTP	NC	
05-16-18	1240	J. Sumner	TVA Courier	1.0	13337	180516 .04	TVA - Sequoyah Nuclear Plant - 101	TN	
05-16-18	1240	J. Sumner	TVA Courier	1.2	13337	180516 .05	TVA - Sequoyah Nuclear Plant - Intake	TN	

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: Marcus O. Moore

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): Sonic Delivery

General Comments:
Took samples from 101 sampler @ 0650 on 5-18-18
Took samples from Int. sampler @ 0734 on 5-18-18

* Diss. Metals were collected + kept on site

Field Identification / Sample Description	Grab/Comp	Collection Date/Time		Container Number & Volume Collected	Flow (MGD)	Rain Event? (Mark as Appropriate)				Project # 13337 Laboratory Use				
	---	Start	End	---	---	Yes	If Yes, Inches	No	Trace	ETS Log Number	Arrival Temp. (°C)	By	Time ET	Appearance
SQN-101-TOX	Comp	5-17-18 0657 ET	5-18-18 0557 ET	2 (2.5gal)	1788.41				X	18051803	1.1, 1.3°C	JH	1315	*
SQN-INT-TOX	Comp	5-17-18 0711 ET	5-18-18 0611 ET	1 (2.5 gal)	NA				X	18051804	0.5°C	JH	1315	*

Sample Custody – Fill In From Top Down

* Custody seals intact. Samples received in good condition.

Relinquished By (Signature):		Date/Time	Received By (Signature):		Date/Time
	TVA	5-18-18/ 0815 ET	BR Skiles	Sonic Delivery	05-18-18/ 08:15 ET
BR Skiles	Sonic Delivery	05-18-18/ 1315 ET		ETS	05-18-18 1315 ET

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.

Whole Effluent Sample Receipt Log

*Sample temperature performed using Sample Receiving Thermometer: SN 160928622

Date Received	Time Received	Received by	Received from	*Sample Temp. (°C)	Project number	Sample number	Sample name and description	State	Comments
05-18-18	0948	J. Sumner	Fed - Ex	2.1	13341	180518 .01	Enfield WWTP	NC	
05-18-18	0948	J. Sumner	Fed - Ex	2.1	13342	180518 .02	Scotland Neck WWTP	NC	
05-18-18	1315	J. Sumner	TVA Courier	1.1/1.3	13337	180518 .03	TVA - Sequoyah Nuclear Plant - 101	TN	
05-18-18	1315	J. Sumner	TVA Courier	0.5	13337	180518 .04	TVA - Sequoyah Nuclear Plant - Intake	TN	

Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013 Method 1000.0)

Species: *Pimephales promelas*

Client: Tennessee Valley Authority
Facility: Sequoyah Nuclear Plant
NPDES #: TN0026450
Project #: 13337

County: Hamilton
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.7	21.4	42.8	85.6	100	Each concentration was UV-treated
Effluent volume (mL)	267.5	535	1070	2140	2500	for 2 minutes to remove pathogenic
Diluent volume (mL)	2232.5	1965	1430	360	0	Interferences.
Total volume (mL)	2500	2500	2500	2500	2500	

Test organism information:		*Test information:*	
Organism source:	In-house culture	Randomizing template:	LIGHT ORANGE
Age:	< 24-hours old	Incubator number and shelf location:	7B
Spawn date:	05-09-18	*Artemia* CHM number:	CHM984
Hatch dates and times:	05-14-18 1525 TO 05-15-18 0505	*Drying information for weight determination:*	
Transfer vessel information:	pH = 7.77 S.U. Temperature = 24.3 °C	Date / Time in oven:	05-22-18 0958
		Initial oven temperature:	60°C
Average transfer volume:	< 0.25 mL	Date / Time out of oven:	05-23-18 1000
		Final oven temperature:	60°C
		Total drying time:	24-HOURS

Daily feeding and renewal information:

Day	Date	Morning feeding		Afternoon feeding		Test initiation, renewal, or termination		Sample numbers used		MHSW batch used
		Time	Analyst	Time	Analyst	Time	Analyst	Outfall 101	Intake	
0	05-15-18	0510	JH	1210	JH	1024	JH	180514.02	180514.03	0510-18 A
1	05-16-18	0615	JH	1215	JH	0925	JH	180514.02	180514.03	05-10-18 A
2	05-17-18	0730	JH	1330	JH	0942	JH	180516.04	180516.05	05-16-18 A
3	05-18-18	0745	JH	1345	JH	0956	JH	180516.04	180516.05	05-16-18 A
4	05-19-18	0745	JH	1345	JH	0957	JH	180518.03	180518.04	05-16-18 B
5	05-20-18	0800	JH	1400	JH	1011	JH	180518.03	180518.04	05-16-18 B
6	05-21-18	0610	JH	1210	JH	0925	JH	180518.03	180518.04	05-16-18 B
7	05-22-18					0926	JH			

Control information: UV-CONTROL		Acceptance criteria	*Summary of test endpoints:*	
% Mortality:	0%	≤ 20%	7-day LC_{50}	>100%
Average weight per initial larvae:	0.680		NOEC	100%
Average weight per surviving larvae:	0.680	≥ 0.25mg/larvae	LOEC	>100%
			ChV	>100%
			IC_{25}	>100%

Independent Review by Kelley E. Keenan: JK

Species: *Pimephales promelas*
Client: TVA / Sequoyah Nuclear Plant, Outfall 101, UV-treated Date: 05-15-18

Survival and Growth Data

Day	CONTROL				10.7%				21.4%			
	A	B	C	D	E	F	G	H	I	J	K	L
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	10	10	10	10	10	10	10	10
7	10	10	10	10	10	10	10	10	10	10	10	10
A = Pan weight (mg) Tray color code:: grey Analyst: TS Date: 05-01-18	15.41	12.12	15.52	16.13	15.20	16.74	15.51	13.04	14.19	16.02	14.45	15.14
B = Pan + Larvae weight (mg) Analyst: JT Date: 05-23-18	22.29	18.26	22.05	23.76	22.61	24.33	22.79	20.47	21.47	23.45	21.64	22.64
C = Larvae weight (mg) = B – A Hand calculated. Analyst: JT	~~7.36~~ 6.88	~~7.59~~ 6.14	~~05-24-18 JT~~ 6.53	7.63	7.41	7.59	7.28	7.43	7.28	7.43	7.19	7.50
Weight per initial number of larvae (mg) = C / Initial number of larvae Hand calculated. Analyst: JT	0.688	0.614	0.653	0.763	0.741	0.759	0.728	0.743	0.728	0.743	0.719	0.750
Average weight per initial number of larvae (mg) / Percent reduction from control (%)	0.680				0.743		-9.37.		0.735		-8.27.	

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.

Comments:

Independent Review by Kelley E. Keenan: K

Species: *Pimephales promelas*
Client: TVA / Sequoyah Nuclear Plant, Outfall 101, UV-treated Date: 05-15-18

Survival and Growth Data

Day	42.8%				85.6%				100%			
	M	N	O	P	Q	R	S	T	U	V	W	X
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	10	10	10	10	10	10	10	10
7	10	10	10	10	10	10	10	10	10	10	10	10
A = Pan weight (mg) Tray color code:: grey Analyst: TS Date: 05-01-18	14.64	14.41	14.49	15.15	14.30	15.87	15.39	14.92	15.75	15.26	14.95	14.79
B = Pan + Larvae weight (mg) Analyst: JH Date: 05-23-18	21.88	21.94	21.24	22.30	21.59	23.43	21.69	21.97	22.80	22.16	22.26	21.73
C = Larvae weight (mg) = B – A Hand calculated. Analyst: JH	7.24	7.53	6.75	7.15	7.29	7.56	6.30	7.05	7.05	6.90	7.31	6.94
Weight per initial number of larvae (mg) = C / Initial number of larvae Hand calculated. Analyst: JH	0.724	0.753	0.675	0.715	0.729	0.756	0.630	0.705	0.705	0.690	0.731	0.694
Average weight per initial number of larvae (mg) / Percent reduction from control (%)	0.717		-5.5%		0.705		-3.8%		0.705		-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.

Comments:

Independent Review by Kelley E. Keenan: K

Species: *Pimephales promelas*
Client: TVA / Sequoyah Nuclear Plant, Outfall 101, UV-treated **Date:** 05-15-18

Survival and Growth Data

Day	100% Intake				Control – Non-treated							
	Y	Z	AA	BB	CC	DD	EE	FF	GG	HH	II	JJ
0	10	10	10	10	10	10	10	10				
1	10	10	10	10	10	10	10	10				
2	10	10	10	10	10	10	10	10				
3	10	10	10	10	10	10	10	10				
4	10	10	10	10	10	10	10	10				
5	10	10	10	10	10	10	10	10				
6	gid	10	10	10	10	10	10	10				
7	9	10	10	10	10	10	10	gid				
A = Pan weight (mg) Tray color code:: grey Analyst: TS Date: 05-01-18	15.78	15.56	15.64	15.39	14.05	15.02	15.05	15.69				
B = Pan + Larvae weight (mg) Analyst: JT Date: 05-23-18	22.06	21.24	21.72	22.16	21.41	22.61	21.49	21.71				
C = Larvae weight (mg) = B – A Hand calculated. Analyst: JT	6.28	5.68	6.08	6.77	7.36	7.59	6.44	6.02				
Weight per initial number of larvae (mg) = C / Initial number of larvae Hand calculated. Analyst: JT	0.628	0.568	0.608	0.677	0.736	0.759	0.644	0.602				
Average weight per initial number of larvae (mg) / Percent reduction from control (%)	0.620		8.77.		0.685		NOT APPLICABLE					

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.

Comments:

Independent Review by Kelley E. Keenan: K

Independent Review by Kelley E. Keenan

TVA / Sequoyah Nuclear Plant, Outfall 101
May 15-22, 2018

ETS
Environmental Testing Solutions, Inc.

Pimephales promelas Chronic Whole Effluent Toxicity Test
EPA-821-R-02-013, Method 1000.0

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

Project number: 13337

Concentration (%)	Replicate	Initial number of larvae	Final number of larvae	A = Pan weight (mg)	B = Pan + Larvae weight (mg)	Larvae weight (mg) = B - A	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 (%)
							Not for Compliance Assessment, Internal Laboratory QC							
Control, Non-treated	CC	10	10	14.05	21.41	7.36	0.736	0.702	7.7	0.736	**97.5**	**0.685**	**10.9**	**Not applicable**
	DD	10	10	15.02	22.61	7.59	0.759			0.759				
	EE	10	10	15.05	21.49	6.44	0.644			0.644				
	FF	10	9	15.69	21.71	6.02	0.669			0.602				
Control, UV-treated	A	10	10	15.41	22.29	6.88	0.688	0.680	9.3	0.688	**100.0**	**0.680**	**9.3**	**Not applicable**
	B	10	10	12.12	18.26	6.14	0.614			0.614				
	C	10	10	15.52	22.05	6.53	0.653			0.653				
	D	10	10	16.13	23.76	7.63	0.763			0.763				
10.7%	E	10	10	15.20	22.61	7.41	0.741	0.743	1.7	0.741	**100.0**	**0.743**	**1.7**	**-9.3**
	F	10	10	16.74	24.33	7.59	0.759			0.759				
	G	10	10	15.51	22.79	7.28	0.728			0.728				
	H	10	10	13.04	20.47	7.43	0.743			0.743				
21.4%	I	10	10	14.19	21.47	7.28	0.728	0.735	1.9	0.728	**100.0**	**0.735**	**1.9**	**-8.2**
	J	10	10	16.02	23.45	7.43	0.743			0.743				
	K	10	10	14.45	21.64	7.19	0.719			0.719				
	L	10	10	15.14	22.64	7.50	0.750			0.750				
42.8%	M	10	10	14.64	21.88	7.24	0.724	0.717	4.5	0.724	**100.0**	**0.717**	**4.5**	**-5.5**
	N	10	10	14.41	21.94	7.53	0.753			0.753				
	O	10	10	14.49	21.24	6.75	0.675			0.675				
	P	10	10	15.15	22.30	7.15	0.715			0.715				
85.6%	Q	10	10	14.30	21.59	7.29	0.729	0.705	7.7	0.729	**100.0**	**0.705**	**7.7**	**-3.8**
	R	10	10	15.87	23.43	7.56	0.756			0.756				
	S	10	10	15.39	21.69	6.30	0.630			0.630				
	T	10	10	14.92	21.97	7.05	0.705			0.705				
100%	U	10	10	15.75	22.80	7.05	0.705	0.705	2.6	0.705	**100.0**	**0.705**	**2.6**	**-3.8**
	V	10	10	15.26	22.16	6.90	0.690			0.690				
	W	10	10	14.95	22.26	7.31	0.731			0.731				
	X	10	10	14.79	21.73	6.94	0.694			0.694				
100% Intake	Y	10	9	15.78	22.06	6.28	0.698	0.638	9.4	0.628	**97.5**	**0.620**	**7.3**	**8.7**
	Z	10	10	15.56	21.24	5.68	0.568			0.568				
	AA	10	10	15.64	21.72	6.08	0.608			0.608				
	BB	10	10	15.39	22.16	6.77	0.677			0.677				

Outfall 101:
Dunnett's MSD value: 0.0648
PMSD: 9.5

Intake:
Dunnett's MSD value: 0.0757
PMSD: 11.1

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.

Lower PMSD bound determined by USEPA (10th percentile) = 12%
Upper PMSD bound determined by USEPA (90th percentile) = 30%

Lower and upper PMSD bounds were determined from the 10th and 90th percentile, respectively, of PMSD data from EPA's WET Interlaboratory Variability Study (USEPA, 2001a; USEPA, 2001b).

USEPA. 2001a, 2001b. Final Report: Interlaboratory Variability Study of EPA Short-term Chronic and Acute Whole Effluent Toxicity Test Methods, Volumes 1 and 2-Appendix. EPA-821-B-01-004 and EPA-821-B-01-005. US Environmental Protection Agency, Cincinnati, OH

Entered and Reviewed by

Environmental Testing Solutions, Inc.

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	5/15/2018	Test ID:	PpFRCR	Sample ID:	TVA / SQN Outfall 101
End Date:	5/22/2018	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:	May 2018	Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:	UV-treated				

Conc-%	1	2	3	4
UV-Control	0.6880	0.6140	0.6530	0.7630
Non-Control	0.7360	0.7590	0.6440	0.6020
10.7	0.7410	0.7590	0.7280	0.7430
21.4	0.7280	0.7430	0.7190	0.7500
42.8	0.7240	0.7530	0.6750	0.7150
85.6	0.7290	0.7560	0.6300	0.7050
100	0.7050	0.6900	0.7310	0.6940
Intake	0.6280	0.5680	0.6080	0.6770

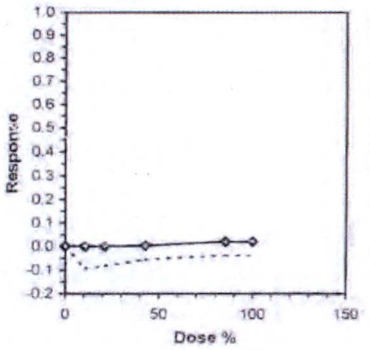
			Transform: Untransformed						1-Tailed		Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
UV-Control	0.6795	0.9916	0.6795	0.6140	0.7630	9.322	4				0.7191	1.0000
Non-Control	0.6853	1.0000	0.6853	0.6020	0.7590	10.871	4					
10.7	0.7428	1.0839	0.7428	0.7280	0.7590	1.711	4	-2.351	2.410	0.0648	0.7191	1.0000
21.4	0.7350	1.0726	0.7350	0.7190	0.7500	1.914	4	-2.063	2.410	0.0648	0.7191	1.0000
42.8	0.7168	1.0460	0.7168	0.6750	0.7530	4.494	4	-1.385	2.410	0.0648	0.7168	0.9968
85.6	0.7050	1.0288	0.7050	0.6300	0.7560	7.683	4	-0.948	2.410	0.0648	0.7050	0.9804
100	0.7050	1.0288	0.7050	0.6900	0.7310	2.618	4	-0.948	2.410	0.0648	0.7050	0.9804
Intake	0.6203	0.9051	0.6203	0.5680	0.6770	7.306	4					

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.95561	0.884	-0.0046	1.48441
Bartlett's Test indicates equal variances (p = 0.05)	11.2468	15.0863		
The control means are not significantly different (p = 0.91)	0.11761	2.44691		

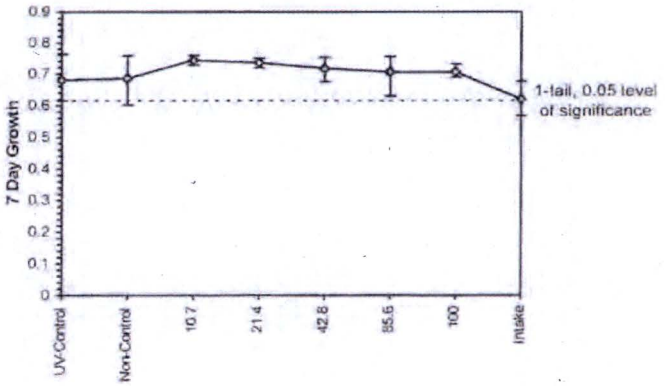
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test Treatments vs UV-Control	100	>100		1	0.06483	0.09541	0.0021	0.00145	0.25389	5, 18

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



Independent Review by Kelley E. Keenan

ETS
Environmental Testing Solutions, Inc.

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	5/15/2018	Test ID:	PpFRCR	Sample ID:	TVA / SQN Intake
End Date:	5/22/2018	Lab ID:	ETS-Envir. Testing Sol.	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:	May 2018	Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	PP-Pimephales promelas
Comments:	UV-treated				

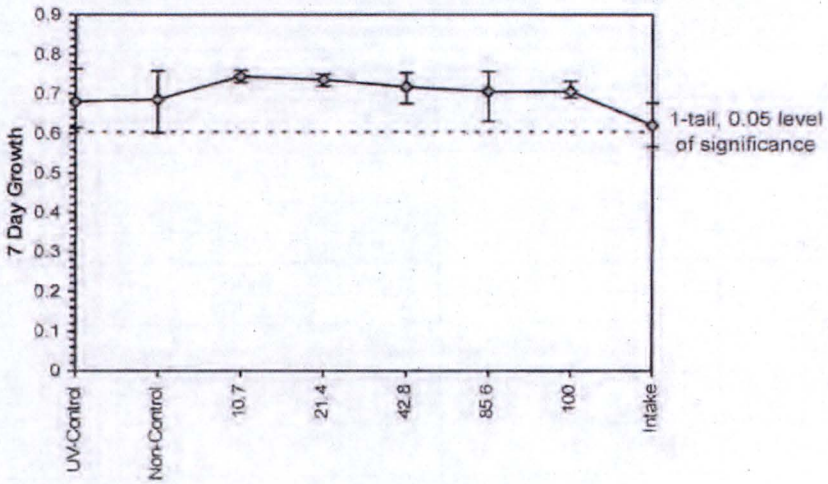
Conc-%	1	2	3	4
UV-Control	0.6880	0.6140	0.6530	0.7630
Non-Control	0.7360	0.7590	0.6440	0.6020
10.7	0.7410	0.7590	0.7280	0.7430
21.4	0.7280	0.7430	0.7190	0.7500
42.8	0.7240	0.7530	0.6750	0.7150
85.6	0.7290	0.7560	0.6300	0.7050
100	0.7050	0.6900	0.7310	0.6940
Intake	0.6280	0.5680	0.6080	0.6770

			Transform: Untransformed						1-Tailed	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
UV-Control	0.6795	0.9916	0.6795	0.6140	0.7630	9.322	4			
Non-Control	0.6853	1.0000	0.6853	0.6020	0.7590	10.871	4			
10.7	0.7428	1.0839	0.7428	0.7280	0.7590	1.711	4			
21.4	0.7350	1.0726	0.7350	0.7190	0.7500	1.914	4			
42.8	0.7168	1.0460	0.7168	0.6750	0.7530	4.494	4			
85.6	0.7050	1.0288	0.7050	0.6300	0.7560	7.683	4			
100	0.7050	1.0288	0.7050	0.6900	0.7310	2.618	4			
Intake	0.6203	0.9051	0.6203	0.5680	0.6770	7.306	4	1.521	1.943	0.0757

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.95581	0.749	0.46261	-0.5692
F-Test indicates equal variances (p = 0.60)	1.95382	47.4683		
The control means are not significantly different (p = 0.91)	0.11761	2.44691		

Hypothesis Test (1-tail, 0.05)	MSDu	MSDp	MSB	MSE	F-Prob	df
Homoscedastic t Test indicates no significant differences	0.07567	0.11136	0.00702	0.00303	0.17896	1, 6
Treatments vs UV-Control						

Dose-Response Plot



Entered and reviewed by

Independent Review by Kelley E. Keenan

Species: *Pimephales promelas* **Date:** 05-15-18

Client: TVA / Sequoyah Nuclear Plant, Outfall 101, UV-treated

Daily Chemistry:

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)					
		0		1		2	
	Analyst	MS	MS	MS	MS	MS	[illegible]
Concentration	**Parameter**			MS 05K			
CONTROL UV-treated	pH (S.U.)	7.97	7.85	~~7.89~~	7.82	7.91	7.65
	DO (mg/L)	8.0	7.9	(7.95) 8.0	7.6	7.8	7.4
	Conductivity (µmhos/cm)	309		308		311	
	*Alkalinity (mg $CaCO_3$/L)	60		[struck through]		59	
	*Hardness (mg $CaCO_3$/L)	82		[struck through]		84	
	*Temperature (°C)	24.7	24.4	24.7	24.3	24.6	24.6
10.7%	pH (S.U.)	7.97	7.78	7.98	7.75	7.93	7.58
	DO (mg/L)	8.0	7.9	8.0	7.6	7.8	7.4
	Conductivity (µmhos/cm)	293		295		291	
	*Temperature (°C)	24.7	24.4	24.8	24.4	24.7	24.5
21.4%	pH (S.U.)	7.98	7.76	7.98	7.71	7.94	7.59
	DO (mg/L)	8.1	8.0	8.0	7.6	7.8	7.3
	Conductivity (µmhos/cm)	274		276		277	
	*Temperature (°C)	24.8	24.4	24.8	24.2	24.8	24.7
42.8%	pH (S.U.)	7.98	7.78	7.98	7.72	7.94	7.65
	DO (mg/L)	8.1	8.0	8.1	7.7	7.9	7.2
	Conductivity (µmhos/cm)	241		244		238	
	*Temperature (°C)	24.8	24.5	24.8	24.4	24.8	24.4
85.6%	pH (S.U.)	7.95	7.73	7.96	7.64	7.92	7.53
	DO (mg/L)	8.3	8.0	8.1	7.7	7.9	6.8
	Conductivity (µmhos/cm)	174		171		177	
	*Temperature (°C)	24.9	24.2	24.9	24.4	24.9	24.4
100%	pH (S.U.)	7.94	7.72	7.96	7.65	7.91	7.50
	DO (mg/L)	8.3	8.1	8.2	7.7	8.0	6.7
	Conductivity (µmhos/cm)	152		146		146	
	*Alkalinity (mg $CaCO_3$/L)	58				54	
	*Hardness (mg $CaCO_3$/L)	60				64	
	*TR chlorine (mg/L)	<0.10				<0.10	
	*Temperature (°C)	25.0	24.6	24.9	24.5	24.9	24.5
100% Intake	pH (S.U.)	7.93	7.71	7.94	7.64	7.89	7.31
	DO (mg/L)	8.3	8.2	8.2	7.7	7.9	6.9
	Conductivity (µmhos/cm)	150		143		154	
	*Alkalinity (mg $CaCO_3$/L)	56				56	
	*Hardness (mg $CaCO_3$/L)	60				60	
	*TR chlorine (mg/L)	<0.10				<0.10	
	*Temperature (°C)	25.0	24.4	25.1	24.5	25.0	24.7
		Initial	Final	Initial	Final	Initial	Final

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1.
Alkalinity, hardness and total residual chlorine performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: ______. Total residual chlorine was performed on non-treated Outfall 101 and Intake samples.

Independent Review Kelley E. Keenan

Species: *Pimephales promelas*

Client: TVA / Sequoyah Nuclear Plant, Outfall 101, UV-treated Date: 05-15-18

Concentration	Parameter	Day 3 Analyst		Day 4 Analyst		Day 5 Analyst		Day 6 Analyst	
		W	BSC	BSC	W	W	MS	MS	MS
CONTROL UV-treated	pH (S.U.)	7.90	7.67	8.04	7.51	7.99	7.85	8.04	7.80
	DO (mg/L)	8.0	8.0	8.1	6.9	7.4	7.6	7.8	7.0
	Conductivity (µmhos/cm)	312		318		304		298	
	*Alkalinity (mg $CaCO_3$/L)			63					
	*Hardness (mg $CaCO_3$/L)			84					
	*Temperature (°C)	24.8	24.5	24.7	24.3	24.7	24.4	24.7	24.3
10.7%	pH (S.U.)	8.01	7.54	8.05	7.49	8.02	7.84	8.05	7.78
	DO (mg/L)	8.0	7.9	8.1	7.0	7.7	7.4	7.8	7.1
	Conductivity (µmhos/cm)	300		292		291		286	
	*Temperature (°C)	24.8	24.6	24.8	24.6	24.7	24.3	24.8	24.5
21.4%	pH (S.U.)	8.01	7.59	8.06	7.50	8.03	7.85	8.05	7.79
	DO (mg/L)	8.0	7.9	8.1	6.9	7.7	7.4	8.0	7.1
	Conductivity (µmhos/cm)	283		278		274		267	
	*Temperature (°C)	24.8	24.6	24.8	24.3	24.7	24.6	24.9	24.6
42.8%	pH (S.U.)	8.01	7.61	8.06	7.56	8.03	7.84	8.06	7.82
	DO (mg/L)	8.1	7.8	8.1	6.9	7.7	7.3	8.0	7.2
	Conductivity (µmhos/cm)	237		246		246		240	
	*Temperature (°C)	24.9	24.3	24.8	24.4	24.7	24.2	24.9	24.5
85.6%	pH (S.U.)	8.00	7.46	8.05	7.45	8.03	7.76	8.04	7.74
	DO (mg/L)	8.1	7.6	8.1	7.0	7.6	7.3	8.0	7.2
	Conductivity (µmhos/cm)	184		184		187		171	
	*Temperature (°C)	24.9	24.6	24.9	24.4	24.8	24.2	24.9	24.2
100%	pH (S.U.)	7.92	7.46	8.04	7.43	8.03	7.74	8.04	7.74
	DO (mg/L)	8.1	7.6	8.1	7.0	7.7	7.3	8.0	7.2
	Conductivity (µmhos/cm)	159		160		162		150	
	*Alkalinity (mg $CaCO_3$/L)			62					
	*Hardness (mg $CaCO_3$/L)			64					
	*TR chlorine (mg/L)			<0.10					
	*Temperature (°C)	25.0	24.3	25.0	24.5	24.8	24.4	25.0	24.2
100% Intake	pH (S.U.)	7.90	7.54	8.04	7.47	8.04	7.79	8.03	7.74
	DO (mg/L)	8.2	7.4	8.1	7.0	7.8	7.3	8.1	7.1
	Conductivity (µmhos/cm)	159		158		161		153	
	*Alkalinity (mg $CaCO_3$/L)			56					
	*Hardness (mg $CaCO_3$/L)			60					
	*TR chlorine (mg/L)			<0.10					
	*Temperature (°C)	25.0	24.6	24.9	24.3	25.0	24.5	25.0	24.2
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1. Alkalinity, hardness and total residual chlorine performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: W. Total residual chlorine was performed on non-treated Outfall 101 and Intake samples.

Independent Review by Kelley E. [illegible]

Species: Pimephales promelas
Client: TVA / Sequoyah Nuclear Plant, Outfall 101, UV-treated

Date: 05-15-18

Daily Chemistry:

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)					
		0		1		2	
	Analyst	JK	MS	MS	MS	MS	N
Concentration	Parameter						
Control Non-treated	pH (S.U.)	7.86	7.81	7.89	7.73	7.85	7.50
	DO (mg/L)	7.9	8.0	7.7	7.6	7.8	6.3
	Conductivity (µmhos/cm)	312		305		302	
	*Alkalinity (mg $CaCO_3$/L)	61		05-22-18 JK		60	
	*Hardness (mg $CaCO_3$/L)	82		76		84	
	*Temperature (°C)	24.8	24.3	24.6	24.4	24.6	24.6
		Initial	Final	Initial	Final	Initial	Final

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)							
		3		4		5		6	
	Analyst	N	BSL	BSL	N	N	MS	MS	MS
Concen-tration	Parameter					05052018			
Control Non-treated	pH (S.U.)	7.99	7.51	8.04	7.49	~~8.03~~	7.81	7.99	7.84
	DO (mg/L)	7.9	7.6	8.0	7.0	(8.03) 7.6	7.2	7.7	7.1
	Conductivity (µmhos/cm)	315		308		300		305	
	*Alkalinity (mg $CaCO_3$/L)	05-22-18 JK		61		05-22-18 JK		05-22-18 JK	
	*Hardness (mg $CaCO_3$/L)			84					
	*Temperature (°C)	24.7	24.5	24.7	24.4	24.6	24.5	24.8	24.6
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1. Alkalinity, hardness and total residual chlorine performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: N

Independent Review by Kelley E. Keenan: N

Independent Review by Kelley E. Keenan

ETS
Environmental Testing Solutions, Inc.

TVA / Sequoyah Nuclear Plant, Outfall 101, UV-treated
May 15-22, 2018

Pimephales promelas Chronic Whole Effluent Toxicity Test
EPA-821-R-02-013, Method 1000.0

Daily Chemical Analyses

Project number: 13337

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, Non-treated	pH (SU)	7.86	7.81	7.89	7.73	7.85	7.58	7.99	7.51	8.04	7.49	8.03	7.81	7.99	7.84
	DO (mg/L)	7.9	8.0	7.7	7.6	7.8	6.3	7.9	7.6	8.0	7.0	7.6	7.2	7.7	7.1
	Conductivity (µmhos/cm)	312		305		302		315		308		300		305	
	Alkalinity (mg/L $CaCO_3$)	61				60				61					
	Hardness (mg/L $CaCO_3$)	82				84				84					
	Temperature (°C)	24.8	24.3	24.6	24.4	24.6	24.6	24.7	24.5	24.7	24.4	24.6	24.5	24.8	24.6
Control, UV-treated	pH (SU)	7.97	7.85	7.95	7.82	7.91	7.65	7.98	7.67	8.04	7.54	7.99	7.85	8.04	7.80
	DO (mg/L)	8.0	7.9	8.0	7.6	7.8	7.4	8.0	8.0	8.1	6.9	7.6	7.6	7.8	7.0
	Conductivity (µmhos/cm)	309		308		311		312		318		304		298	
	Alkalinity (mg/L $CaCO_3$)	60				59				63					
	Hardness (mg/L $CaCO_3$)	82				84				84					
	Temperature (°C)	24.7	24.4	24.7	24.3	24.6	24.6	24.8	24.5	24.7	24.3	24.7	24.4	24.7	24.3
10.7%	pH (SU)	7.97	7.78	7.98	7.75	7.93	7.58	8.01	7.54	8.05	7.49	8.02	7.84	8.05	7.78
	DO (mg/L)	8.0	7.9	8.0	7.6	7.8	7.4	8.0	7.9	8.1	7.0	7.7	7.4	7.8	7.1
	Conductivity (µmhos/cm)	293		295		291		300		292		291		286	
	Temperature (°C)	24.7	24.4	24.8	24.4	24.7	24.5	24.8	24.6	24.8	24.6	24.7	24.3	24.8	24.5
21.4%	pH (SU)	7.98	7.76	7.98	7.71	7.94	7.59	8.01	7.59	8.06	7.50	8.03	7.85	8.05	7.79
	DO (mg/L)	8.1	8.0	8.0	7.6	7.8	7.3	8.0	7.8	8.1	6.9	7.7	7.4	8.0	7.1
	Conductivity (µmhos/cm)	274		276		277		283		278		274		267	
	Temperature (°C)	24.8	24.4	24.8	24.2	24.8	24.7	24.8	24.6	24.8	24.3	24.7	24.6	24.9	24.6
42.8%	pH (SU)	7.98	7.78	7.98	7.72	7.94	7.65	8.01	7.61	8.06	7.56	8.03	7.84	8.06	7.82
	DO (mg/L)	8.1	8.0	8.1	7.7	7.9	7.2	8.1	7.8	8.1	6.9	7.7	7.3	8.0	7.2
	Conductivity (µmhos/cm)	241		244		238		237		246		246		240	
	Temperature (°C)	24.8	24.5	24.8	24.4	24.8	24.4	24.9	24.3	24.8	24.4	24.7	24.2	24.9	24.5
85.6%	pH (SU)	7.95	7.73	7.96	7.64	7.92	7.53	8.00	7.46	8.05	7.45	8.03	7.76	8.04	7.74
	DO (mg/L)	8.3	8.0	8.1	7.7	7.9	6.8	8.1	7.6	8.1	7.0	7.8	7.3	8.0	7.2
	Conductivity (µmhos/cm)	174		171		177		184		184		187		171	
	Temperature (°C)	24.9	24.2	24.9	24.4	24.9	24.4	24.9	24.6	24.9	24.4	24.8	24.2	24.9	24.2
100%	pH (SU)	7.94	7.72	7.96	7.65	7.91	7.50	7.92	7.46	8.04	7.43	8.03	7.74	8.04	7.74
	DO (mg/L)	8.3	8.1	8.2	7.7	8.0	6.7	8.1	7.6	8.1	7.0	7.7	7.3	8.0	7.2
	Conductivity (µmhos/cm)	152		146		146		159		160		162		150	
	Alkalinity (mg/L $CaCO_3$)	58				54				62					
	Hardness (mg/L $CaCO_3$)	60				64				64					
	*Total Residual Chlorine (mg/L	<0.10				<0.10				<0.10					
	Temperature (°C)	25.0	24.6	24.9	24.5	24.9	24.5	25.0	24.3	25.0	24.5	24.8	24.4	25.0	24.2
100% Intake	pH (SU)	7.93	7.71	7.94	7.64	7.89	7.31	7.98	7.54	8.04	7.47	8.04	7.79	8.03	7.74
	DO (mg/L)	8.3	8.2	8.2	7.7	7.9	6.9	8.2	7.6	8.1	7.0	7.8	7.3	8.1	7.1
	Conductivity (µmhos/cm)	150		143		154		159		158		161		153	
	Alkalinity (mg/L $CaCO_3$)	56				56				56					
	Hardness (mg/L $CaCO_3$)	60				60				60					
	*Total Residual Chlorine (mg/L	<0.10				<0.10				<0.10					
	Temperature (°C)	25.0	24.4	25.1	24.5	25.0	24.7	25.0	24.6	24.9	24.3	25.0	24.5	25.0	24.2

*Note: Total residual chlorine was performed on non-treated Outfall 101 and Intake samples.

File: sqn101_051518chem
Entered by: T. Sleeper
Reviewed by: KK

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

Client: Tennessee Valley Authority
Facility: Sequoyah Nuclear Plant
NPDES #: TN0026450
Project #: 13327

County: Hamilton
Outfall: 101

Dilution preparation information:						*Comments:*
Dilution prep (%)	10.7	21.4	42.8	85.6	100	
Effluent volume (mL)	267.5	535	1070	2140	2500	
Diluent volume (mL)	2232.5	1965	1430	360	0	
Total volume (mL)	2500	2500	2500	2500	2500	

Test organism source information:											*Test information:*	
Organism age:	< 24-hours old										Randomizing template color:	BLUE
Date and times organisms were born between:	05-15-18 0600 TO 1046										Incubator number and shelf location:	2B1
Culture board:	05-08-18 A / 05-08-18 B											
Replicate number:	1	2	3	4	5	6	7	8	9	10	YWT batch:	04-16-18
Culture board cup number:	35	36	31	8	12	13	14	23	30	31		
Transfer vessel information:	pH = 7.93 S.U. Temperature = 24.8 °C										*Selenastrum* batch:	04-30-18
Average transfer volume (mL):	< 0.25 mL											

Daily renewal information:

Day	Date	Test initiation and feeding, renewal and feeding, or termination time	MHSW batch used	Sample numbers used Outfall 101	Sample numbers used Intake	Analyst
0	05-15-18	1105	05-10-18 A	180514.02	180514.03	JH
1	05-16-18	1008	05-10-18 A	180514.02	180514.03	JH
2	05-17-18	1013	05-16-18 A	180516.04	180516.05	JH
3	05-18-18	1037	05-16-18 A	180516.04	180516.05	JH
4	05-19-18	1027	05-16-18 B	180518.03	180518.04	JH
5	05-20-18	1053	05-16-18 B	180518.03	180518.04	JH
6	05-21-18	1006	05-16-18 B	180518.03	180518.04	JH
7	05-22-18	1025				JH

Control information:	Control-1	Control-2	Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	0%	≤ 20%	7-day LC_{50}	>100%
% Adults having 3rd Broods:	100%	100%	≥ 60% surviving adults	NOEC	100%
% Mortality:	0%	0%	≤ 20%	LOEC	>100%
Mean Offspring/Female:	28.4	29.0	≥ 15.0 offspring/female	ChV	>100%
% CV:	6.37%	5.47%	< 42.0 %	IC_{25}	>100%

Independent Review by Kelley E. Keenan

Species: *Ceriodaphnia dubia*
Client: TVA / Sequoyah Nuclear Plant, Outfall 101 Date: 05-15-18

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	3	4	4	3	4	4	5	3	3	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	9	11	10	11	13	10	10	10	12	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	16	13	13	15	16	14	12	15	14	14
Total young produced		28	28	27	29	33	28	27	28	29	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), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

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

CONC: **10.7%** *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	5	3	4	3	3	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	10	13	12	10	10	12	11	10	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	15	18	15	16	14	16	17	15	18	17
Total young produced		31	33	32	32	29	29	33	29	31	32
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	31.1
% Reduction from Control-1:	-9.5%

Independent Review by Kelley E. Keenan

Species: *Ceriodaphnia dubia*

Client: TVA / Sequoyah Nuclear Plant, Outfall 101 Date: 05-15-18

CONC: 21.4% *Survival and Reproduction Data*

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

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	32.0
% Reduction from Control-1:	-12.77%

CONC: 42.8% *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	6	4	5	5	4	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	13	11	13	12	12	14	12	10	11	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	16	19	15	19	18	18	17	16	18	17
Total young produced		32	35	32	37	34	37	34	30	33	31
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	33.5
% Reduction from Control-1:	-18.07%

Independent Review by Kelley E. Keenan:

Species: *Ceriodaphnia dubia*
Client: TVA / Sequoyah Nuclear Plant, Outfall 101 Date: 05-15-18

CONC: 85.6% *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	4	6	6	4	5	4	4	5	6	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	12	12	13	14	13	12	10	10	13
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	20	18	17	18	20	16	15	19	19	17
Total young produced		36	36	35	35	39	05-23-18 3 5 (33)	31	34	35	35
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	34.9
% Reduction from Control-1:	-22.9%

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	4	5	6	5	5	6	4	5	5
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	14	11	13	12	13	14	13	12	12	13
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	18	18	19	21	18	20	17	18	17	19
Total young produced		37	33	37	39	36	39	36	34	34	37
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	36.2
% Reduction from Control-1:	~~-27.2%~~ -27.5%

Independent Review by Kelley E. Keenan:

Species: *Ceriodaphnia dubia*
Client: TVA / Sequoyah Nuclear Plant, Outfall 101 Date: 05-15-18

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	4	4	3	4	3	3	5	3	4	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	10	12	9	12	10	10	10	11	12	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	16	13	14	14	17	16	16	13	14	13
Total young produced		30	29	26	30	30	29	31	27	30	28
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), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer)

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	29.0

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	5	3	5	6	4	4	6
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	13	12	11	13	12	10	10	13	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	18	15	17	18	16	19	16	19	17	19
Total young produced		35	33	33	34	34	36	32	33	34	35
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	33.9
% Reduction from Control-2:	-16.9%

Independent Review by Kelley E. Keenan:

Independent Review by Kelley E. Hannan

TVA / Sequoyah Nuclear Plant, Outfall 101
May 15-22, 2018

ETS
Environmental Testing Solutions, Inc.

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	3	4	4	3	4	4	5	3	3	3	36
5	9	11	10	11	13	10	10	10	12	10	106
6	0	0	0	0	0	0	0	0	0	0	0
7	16	13	13	15	16	14	12	15	14	14	142
Total	28	28	27	29	33	28	27	28	29	27	284

10.7%

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	5	3	4	3	3	3	38
5	12	10	13	12	10	10	12	11	10	12	112
6	0	0	0	0	0	0	0	0	0	0	0
7	15	18	15	16	14	16	17	15	18	17	161
Total	31	33	32	32	29	29	33	29	31	32	311

21.4%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	4	4	3	5	4	4	4	5	4	42
5	12	12	10	10	10	13	10	11	13	13	114
6	0	0	0	0	0	0	0	0	0	0	0
7	17	16	16	18	16	15	16	15	16	19	164
Total	34	32	30	31	31	32	30	30	34	36	320

42.8%

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	6	4	5	5	4	4	4	44
5	13	11	13	12	12	14	12	10	11	10	118
6	0	0	0	0	0	0	0	0	0	0	0
7	16	19	15	19	18	18	17	16	18	17	173
Total	32	35	32	37	34	37	34	30	33	31	335

85.6%

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	4	6	6	4	5	4	4	5	6	5	49
5	12	12	12	13	14	13	12	10	10	13	121
6	0	0	0	0	0	0	0	0	0	0	0
7	20	18	17	18	20	16	15	19	19	17	179
Total	36	36	35	35	39	33	31	34	35	35	349

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	4	5	6	5	5	6	4	5	5	50
5	14	11	13	12	13	14	13	12	12	13	127
6	0	0	0	0	0	0	0	0	0	0	0
7	18	18	19	21	18	20	17	18	17	19	185
Total	37	33	37	39	36	39	36	34	34	37	362

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	4	4	3	4	3	3	5	3	4	4	37
5	10	12	9	12	10	10	10	11	12	11	107
6	0	0	0	0	0	0	0	0	0	0	0
7	16	13	14	14	17	16	16	13	14	13	146
Total	30	29	26	30	30	29	31	27	30	28	290

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	5	3	5	6	4	4	6	47
5	12	13	12	11	13	12	10	10	13	10	116
6	0	0	0	0	0	0	0	0	0	0	0
7	18	15	17	18	18	19	16	19	17	19	176
Total	35	33	33	34	34	36	32	33	34	35	339

Reviewed and Approved by Test Director

Environmental Testing Solutions, Inc.

TVA / Sequoyah Nuclear Plant, Outfall 101
May 15-22, 2018
Chronic Whole Effluent Toxicity Test (EPA-821-R-02-013, Method 1002.0)
Ceriodaphnia dubia Chronic Whole Effluent Toxicity Test
EPA-821-R-02-013, Method 1002.0

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

Project number: 13337

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	28	28	27	29	33	28	27	28	29	27	100	28.4	6.3	Not applicable
10.7%	31	33	32	32	29	29	33	29	31	32	100	31.1	5.1	-9.5
21.4%	34	32	30	31	31	32	30	30	34	36	100	32.0	6.4	-12.7
42.8%	32	35	32	37	34	37	34	30	33	31	100	33.5	7.1	-18.0
85.6%	36	36	35	35	39	33	31	34	35	35	100	34.9	6.0	-22.9
100%	37	33	37	39	36	39	36	34	34	37	100	36.2	5.6	-27.5
Control - 2	30	29	26	30	30	29	31	27	30	28	100	29.0	5.4	Not applicable
100% Intake	35	33	33	34	34	36	32	33	34	35	100	33.9	3.5	-16.9

Outfall 101:
Dunnett's MSD value: 2.047
PMSD: 7.2

Intake:
Dunnett's MSD value: 1.080
PMSD: 3.7

MSD = Minimum Significant Difference
PMSD = Percent Minimum Significant Difference
PMSD is a measure of test precision. The PMSD is the minimum percent difference between the control and treatment that can be declared statistically significant in a whole effluent toxicity test.

Lower PMSD bound determined by USEPA (10th percentile) = 13%.
Upper PMSD bound determined by USEPA (90th percentile) = 47%.
Lower and upper PMSD bounds were determined from the 10th and 90th percentile, respectively, of PMSD data from EPA's WET Interlaboratory Variability Study (USEPA, 2001a; USEPA, 2001b).

USEPA. 2001a, 2001b. Final Report: Interlaboratory Variability Study of EPA Short-term Chronic and Acute Whole Effluent Toxicity Test Methods, Volumes 1 and 2-Appendix. EPA-821-B-01-004 and EPA-821-B-01-005. US Environmental Protection Agency, Cincinnati, OH.

ETS
Environmental Testing Solutions, Inc.

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	5/15/2018	Test ID:	CdFRCR	Sample ID:	TVA / SQN Outfall 101
End Date:	5/22/2018	Lab ID:	ETS-Envir. Testing Sol	Sample Type:	DMR-Discharge Monitoring Report
Sample Date:	May 2018	Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia
Comments:	Non-treated				

Conc-%	1	2	3	4	5	6	7	8	9	10
Control-1	28.000	28.000	27.000	29.000	33.000	28.000	27.000	28.000	29.000	27.000
Control-2	30.000	29.000	26.000	30.000	30.000	29.000	31.000	27.000	30.000	28.000
10.7	31.000	33.000	32.000	32.000	29.000	29.000	33.000	29.000	31.000	32.000
21.4	34.000	32.000	30.000	31.000	31.000	32.000	30.000	30.000	34.000	36.000
42.8	32.000	35.000	32.000	37.000	34.000	37.000	34.000	30.000	33.000	31.000
85.6	36.000	36.000	35.000	35.000	39.000	33.000	31.000	34.000	35.000	35.000
100	37.000	33.000	37.000	39.000	36.000	39.000	36.000	34.000	34.000	37.000
Intake	35.000	33.000	33.000	34.000	34.000	36.000	32.000	33.000	34.000	35.000

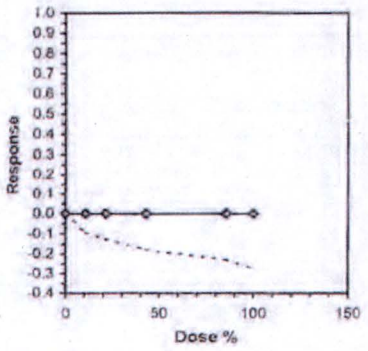
			Transform: Untransformed					1-Tailed			Isotonic	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean	N-Mean
Control-1	28.400	0.9793	28.400	27.000	33.000	6.255	10	*			32.683	1.0000
Control-2	29.000	1.0000	29.000	26.000	31.000	5.391	10					
10.7	31.100	1.0724	31.100	29.000	33.000	5.129	10	-3.017	2.287	2.047	32.683	1.0000
21.4	32.000	1.1034	32.000	30.000	36.000	6.421	10	-4.022	2.287	2.047	32.683	1.0000
42.8	33.500	1.1552	33.500	30.000	37.000	7.071	10	-5.698	2.287	2.047	32.683	1.0000
85.6	34.900	1.2034	34.900	31.000	39.000	5.957	10	-7.262	2.287	2.047	32.683	1.0000
100	36.200	1.2483	36.200	33.000	39.000	5.646	10	-8.715	2.287	2.047	32.683	1.0000
Intake	33.900	1.1690	33.900	32.000	36.000	3.532	10					

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.67338	1.035	0.37827	-0.0943
Bartlett's Test indicates equal variances (p = 0.90)	1.59049	15.0863		
The control means are not significantly different (p = 0.43)	0.80178	2.10092		

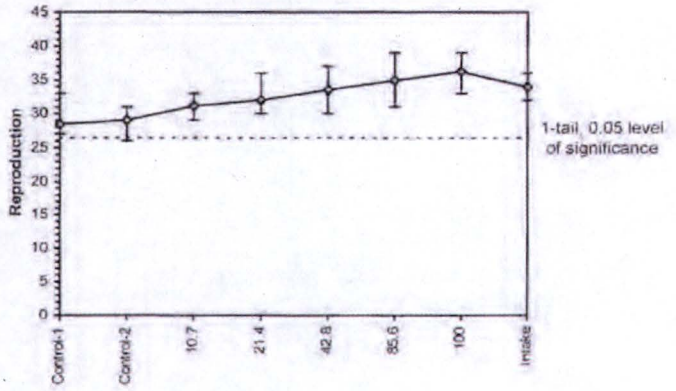
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test Treatments vs Control-1	100	>100		1	2.04668	0.07207	78.5367	4.00556	4.4E-11	5, 54

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



Independent Review by Kelley E. Keenan

TVA / Sequoyah Nuclear Plant, Outfall 101 - Intake
May 15-22, 2018

Environmental Testing Solutions, Inc.

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	5/15/2018	Test ID:	CdFRCR	Sample ID:	TVA / SQN Intake	
End Date:	5/22/2018	Lab ID:	ETS-Envir. Testing Sol	Sample Type:	DMR-Discharge Monitoring Report	
Sample Date:	May 2018	Protocol:	FWCHR-EPA-821-R-02-013	Test Species:	CD-Ceriodaphnia dubia	
Comments:	Non-treated					

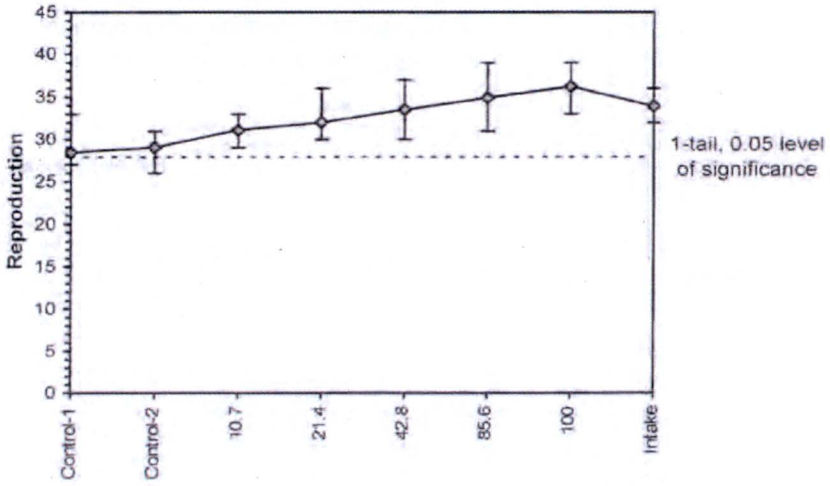
Conc-%	1	2	3	4	5	6	7	8	9	10
Control-1	28.000	28.000	27.000	29.000	33.000	28.000	27.000	28.000	29.000	27.000
Control-2	30.000	29.000	26.000	30.000	30.000	29.000	31.000	27.000	30.000	28.000
10.7	31.000	33.000	32.000	32.000	29.000	29.000	33.000	29.000	31.000	32.000
21.4	34.000	32.000	30.000	31.000	31.000	32.000	30.000	30.000	34.000	36.000
42.8	32.000	35.000	32.000	37.000	34.000	37.000	34.000	30.000	33.000	31.000
85.6	36.000	36.000	35.000	35.000	39.000	33.000	31.000	34.000	35.000	35.000
100	37.000	33.000	37.000	39.000	36.000	39.000	36.000	34.000	34.000	37.000
Intake	35.000	33.000	33.000	34.000	34.000	36.000	32.000	33.000	34.000	35.000

			Transform: Untransformed						1-Tailed	
Conc-%	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
Control-1	28.400	0.9793	28.400	27.000	33.000	6.255	10			
Control-2	29.000	1.0000	29.000	26.000	31.000	5.391	10			
10.7	31.100	1.0724	31.100	29.000	33.000	5.129	10			
21.4	32.000	1.1034	32.000	30.000	36.000	6.421	10			
42.8	33.500	1.1552	33.500	30.000	37.000	7.071	10			
85.6	34.900	1.2034	34.900	31.000	39.000	5.957	10			
100	36.200	1.2483	36.200	33.000	39.000	5.646	10			
Intake	33.900	1.1690	33.900	32.000	36.000	3.532	10	-7.869	1.734	1.080

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.94659	0.868	-0.4961	-0.2125
F-Test indicates equal variances (p = 0.44)	1.70543	6.54109		
The control means are not significantly different (p = 0.43)	0.80178	2.10092		

Hypothesis Test (1-tail, 0.05)	MSDu	MSDp	MSB	MSE	F-Prob	df
Homoscedastic t Test indicates no significant differences Treatments vs Control-2	1.07983	0.03724	120.05	1.93889	3.1E-07	1, 18

Dose-Response Plot



Entered and Reviewed by Jim Sumner

Independent Review by Kelley E. Keenen

Species: *Ceriodaphnia dubia* Date: 05-15-18

Client: TVA / Sequoyah Nuclear Plant, Outfall 101

Daily Chemistry:

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)					
		0		1		2	
	Analyst	MS	MS	MS	MS	MS	W
Concentration	Parameter						
CONTROL	pH (S.U.)	7.86	7.96	7.89	7.92	7.85	7.99
	DO (mg/L)	7.9	8.0	7.7	7.8	7.8	7.9
	Conductivity (µmhos/cm)	312		305		302	
	*Alkalinity (mg $CaCO_3$/L)	61		outlier		60	
	*Hardness (mg $CaCO_3$/L)	82		outlier		84	
	*Temperature (°C)	24.7	25.1	24.6	25.0	24.7	25.1
10.7%	pH (S.U.)	7.95	7.97	7.96	7.92	7.91	8.00
	DO (mg/L)	8.0	7.9	7.9	7.8	7.8	7.9
	Conductivity (µmhos/cm)	290		289		294	
	*Temperature (°C)	24.8	24.8	24.7	24.8	24.7	25.2
21.4%	pH (S.U.)	7.96	7.96	7.98	7.92	7.92	7.99
	DO (mg/L)	8.1	7.9	8.0	7.8	7.8	7.9
	Conductivity (µmhos/cm)	275		278		272	
	*Temperature (°C)	24.9	25.0	24.7	25.0	24.7	25.0
42.8%	pH (S.U.)	7.96	7.96	7.98	7.91	7.93	7.99
	DO (mg/L)	8.1	8.0	8.0	7.8	7.9	7.9
	Conductivity (µmhos/cm)	241		244		237	
	*Temperature (°C)	24.9	24.8	24.7	24.9	24.7	24.8
85.6%	pH (S.U.)	7.94	7.92	7.96	7.89	7.89	7.98
	DO (mg/L)	8.1	8.0	8.1	7.9	8.0	8.0
	Conductivity (µmhos/cm)	172		176		171	
	*Temperature (°C)	25.0	24.8	24.8	24.9	24.7	24.8
100%	pH (S.U.)	7.94	7.92	7.95	7.88	7.88	7.93
	DO (mg/L)	8.2	8.1	8.1	8.1	8.0	7.9
	Conductivity (µmhos/cm)	156		147		138	
	*Alkalinity (mg $CaCO_3$/L)	58				58	
	*Hardness (mg $CaCO_3$/L)	68				60	
	*TR chlorine (mg/L)	<0.10				<0.10	
	*Temperature (°C)	25.0	24.7	24.9	25.1	24.8	24.8
100% Intake	pH (S.U.)	7.92	7.92	7.95	7.88	7.91	7.94
	DO (mg/L)	8.2	8.1	8.1	8.1	8.0	7.9
	Conductivity (µmhos/cm)	147		149		152	
	*Alkalinity (mg $CaCO_3$/L)	56				58	
	*Hardness (mg $CaCO_3$/L)	60				64	
	*TR chlorine (mg/L)	<0.10				<0.10	
	*Temperature (°C)	24.9	24.8	24.8	24.8	24.7	24.9
		Initial	Final	Initial	Final	Initial	Final

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1. Alkalinity, hardness and total residual chlorine performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: X

Independent Review by Kelley E. Keenan: W

Species: *Ceriodaphnia dubia*

Client: TVA / Sequoyah Nuclear Plant, Outfall 101 Date: 05-15-18

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)							
		3		4		5		6	
	Analyst	[illegible]	BSL	BSL	[illegible]	[illegible]	MS	RS	MS
Concentration	Parameter								
CONTROL	pH (S.U.)	7.99	8.01	8.04	8.03	8.03	8.04	7.99	8.04
	DO (mg/L)	7.9	8.0	8.0	7.8	7.6	7.7	7.7	7.8
	Conductivity (µmhos/cm)	315		308		300		305	
	*Alkalinity (mg $CaCO_3$/L)	[illegible]		61		[illegible]		[illegible]	
	*Hardness (mg $CaCO_3$/L)			84					
	*Temperature (°C)	24.7	25.1	24.8	25.1	24.8	25.2	24.7	24.9
10.7%	pH (S.U.)	8.00	8.02	8.03	8.03	8.07	8.04	8.01	8.05
	DO (mg/L)	7.9	8.0	8.0	7.8	7.6	7.8	7.8	7.8
	Conductivity (µmhos/cm)	292		280		280		285	
	*Temperature (°C)	24.8	24.8	24.9	25.0	24.9	25.0	24.8	24.8
21.4%	pH (S.U.)	8.00	8.02	8.04	8.03	8.07	8.02	8.04	8.03
	DO (mg/L)	7.9	8.0	7.9	7.9	7.7	7.8	7.8	7.8
	Conductivity (µmhos/cm)	282		265		270		272	
	*Temperature (°C)	24.8	25.0	24.9	25.0	24.9	25.0	24.8	24.8
42.8%	pH (S.U.)	8.00	8.02	8.04	8.01	8.07	8.02	8.05	8.03
	DO (mg/L)	8.0	8.0	7.9 BSL 05-19-18 7.9		7.7	7.8	7.9	7.8
	Conductivity (µmhos/cm)	250		237		237		240	
	*Temperature (°C)	24.8	24.8	24.9	25.0	24.9	24.9	24.9	25.1
85.6%	pH (S.U.)	8.00	8.01	7.99	8.00	8.06	8.00	8.03	8.02
	DO (mg/L)	7.9	7.9	7.9	8.0	7.7	7.8	7.9	7.8
	Conductivity (µmhos/cm)	179		178		179		177	
	*Temperature (°C)	24.8	24.8	24.9	25.2	24.9	25.1	24.9	24.8
100%	pH (S.U.)	8.01	7.96	8.00	7.96	8.04	8.00	8.03	7.98
	DO (mg/L)	8.0	7.9	7.9	7.9	7.8	7.8	7.9	7.9
	Conductivity (µmhos/cm)	156		155		151		154	
	*Alkalinity (mg $CaCO_3$/L)			58					
	*Hardness (mg $CaCO_3$/L)			64					
	*TR chlorine (mg/L)			<0.10					
	*Temperature (°C)	24.9	25.1	25.0	24.9	25.0	24.9	24.9	25.0
100% Intake	pH (S.U.)	8.00	7.98	7.96	8.02	8.03	8.01	8.03	8.04
	DO (mg/L)	8.2	7.9	7.9	7.9	7.8	7.8	8.0	8.0
	Conductivity (µmhos/cm)	156		157		155		159	
	*Alkalinity (mg $CaCO_3$/L)			58					
	*Hardness (mg $CaCO_3$/L)			60					
	*TR chlorine (mg/L)			<0.10					
	*Temperature (°C)	24.8	24.8	25.0	24.8	24.9	25.0	25.0	24.8
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

Independent Review by Kelley E. Keenan: [illegible]

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1. Alkalinity, hardness and total residual chlorine performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: [illegible].

Independent Review by Kelley E. Hamann

TVA / Sequoyah Nuclear Plant, Outfall 101, Non-treated
May 15-22, 2018

ETS
Environmental Testing Solutions, Inc.

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

Daily Chemical Analyses

Project number: 13337

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)	7.86	7.96	7.89	7.92	7.85	7.99	7.99	8.01	8.04	8.03	8.03	8.04	7.99	8.04
	DO (mg/L)	7.9	8.0	7.7	7.8	7.8	7.9	7.9	8.0	8.0	7.8	7.6	7.7	7.7	7.8
	Conductivity (µmhos/cm)	312		305		302		315		308		300		305	
	Alkalinity (mg/L $CaCO_3$)	61				60				61					
	Hardness (mg/L $CaCO_3$)	82				84				84					
	Temperature (°C)	24.7	25.1	24.6	25.0	24.7	25.1	24.7	25.1	24.8	25.1	24.8	25.2	24.7	24.9
10.7%	pH (SU)	7.95	7.97	7.96	7.92	7.91	8.00	8.00	8.02	8.03	8.03	8.07	8.04	8.01	8.05
	DO (mg/L)	8.0	7.9	7.9	7.8	7.8	7.9	7.9	8.0	8.0	7.8	7.8	7.8	7.8	7.8
	Conductivity (µmhos/cm)	290		289		294		292		280		280		285	
	Temperature (°C)	24.8	24.8	24.7	24.8	24.7	25.2	24.8	24.8	24.9	25.0	24.9	25.0	24.8	24.8
21.4%	pH (SU)	7.96	7.96	7.98	7.92	7.92	7.99	8.00	8.02	8.04	8.03	8.07	8.02	8.04	8.03
	DO (mg/L)	8.1	7.9	8.0	7.8	7.8	7.9	7.9	8.0	7.9	7.9	7.7	7.8	7.8	7.8
	Conductivity (µmhos/cm)	275		278		272		282		265		270		272	
	Temperature (°C)	24.9	25.0	24.7	25.0	24.7	25.0	24.8	25.0	24.9	25.0	24.9	25.0	24.8	24.8
42.8%	pH (SU)	7.96	7.96	7.98	7.91	7.93	7.99	8.00	8.02	8.04	8.01	8.07	8.02	8.05	8.03
	DO (mg/L)	8.1	8.0	8.0	7.8	7.9	7.9	8.0	8.0	7.9	7.9	7.7	7.8	7.9	7.8
	Conductivity (µmhos/cm)	241		244		237		250		237		237		240	
	Temperature (°C)	24.9	24.8	24.7	24.9	24.7	24.8	24.8	24.8	24.9	25.0	24.9	24.9	24.9	25.1
85.6%	pH (SU)	7.94	7.92	7.96	7.89	7.89	7.98	8.00	8.01	7.99	8.00	8.06	8.00	8.03	8.02
	DO (mg/L)	8.1	8.0	8.1	7.9	8.0	8.0	7.9	7.9	7.9	8.0	7.7	7.8	7.9	7.8
	Conductivity (µmhos/cm)	172		176		171		179		178		179		177	
	Temperature (°C)	25.0	24.8	24.8	24.9	24.7	24.8	24.8	24.8	24.9	25.2	24.9	25.1	24.9	24.8
100%	pH (SU)	7.94	7.92	7.95	7.88	7.88	7.93	8.01	7.96	8.00	7.96	8.04	8.00	8.03	7.98
	DO (mg/L)	8.2	8.1	8.1	8.1	8.0	7.9	8.0	7.9	7.9	7.9	7.8	7.8	7.9	7.9
	Conductivity (µmhos/cm)	156		147		138		156		155		151		154	
	Alkalinity (mg/L $CaCO_3$)	58				58				58					
	Hardness (mg/L $CaCO_3$)	68				60				64					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	25.0	24.7	24.9	25.1	24.8	24.8	24.9	25.1	25.0	24.9	25.0	24.9	24.9	25.0
100% Intake	pH (SU)	7.92	7.92	7.95	7.88	7.91	7.94	8.00	7.98	7.96	8.02	8.03	8.01	8.03	8.04
	DO (mg/L)	8.2	8.1	8.1	8.1	8.0	7.9	8.2	7.9	7.9	7.9	7.8	7.8	8.0	8.0
	Conductivity (µmhos/cm)	147		149		152		156		157		155		159	
	Alkalinity (mg/L $CaCO_3$)	56				58				58					
	Hardness (mg/L $CaCO_3$)	60				64				60					
	Total Residual Chlorine (mg/L)	<0.10				<0.10				<0.10					
	Temperature (°C)	24.9	24.8	24.8	24.8	24.7	24.9	24.8	24.8	25.0	24.8	24.9	25.0	25.0	24.8

File: sqn101_051518chem
Entered by: T.Sleeper
Reviewed by:

Total Residual Chlorine (4500-Cl G-2011), Screening Whole Effluent Toxicity Samples

Matrix: Water, RL = 0.10 mg/L

Analyst	N
Date analyzed	05.15.18

DPD: CHM 1004

Positive and Negative Control:

Control Type	Sample ID	Result (✓)	
		Positive	Negative
Negative control	Deionized water		✓
Positive control	Tap water	✓	

Sample screening:

Sample number	Sample ID	Sample characteristics	Result (✓)	
			Positive	Negative
180514.02	TVA/SQN 101	no color, clear		✓
180514.03	↓ INTAKE	pale tan, clear		✓
180515.02	ENFIELD	no col'n, clear		✓
180515.01	CHATTANOOGA	pale tan, clear		✓

N 05.15.18

Note: All samples were analyzed in excess of EPA recommended holding time (15 minutes) unless otherwise noted. A positive result indicates the presence of total residual chlorine above detection (> 0.10 mg/L), which results in a pink to red color change with the addition of the DPD indicator. A negative result indicates the absence of total residual chlorine, which results in no color change with the addition of the DPD indicator.

Reviewed by	N
Date reviewed	05-15-18

Total Residual Chlorine (ORION-97-70-1977), Confirmation of Whole Effluent Toxicity Samples

Matrix: Water, RL = 0.10 mg/L, Meter: Accumet Model AB250 pH/mV/Ion Meter

Analyst	TS	Iodide reagent:	INR 829
Date analyzed	05-15-18	Acid reagent:	INR 835

Calibration:

	0.10 mg/L	1.00 mg/L	Slope (mV) (suggested range = 26 to 30 mV)
Reference standard #	INSS 1617	INSS 1617	30

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

Laboratory control sample:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1617	0.50	0.538	107.6 %

Duplicate sample precision:

Sample number	Sample ID	Total residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
180514.02	TVA | SQN 101	S <0.017	
L	Duplicate	D <0.015	05-15-18 TS

Sample measurements:

Sample number	Sample ID	Total residual chlorine (mg/L)
TV < 0.05 mg/L	Method Blank (MB)	<0.012
TV = 0.30 mg/L	Method Detection Limit spike sample (MDL_s)	0.320
180514.03	TVA | SQN INTAKE	<0.005

TS 05-15-18

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

Laboratory control sample:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1617	0.50	0.476	95.2%

Reviewed by [ ] Date reviewed 05.15.18

Total Residual Chlorine (4500-Cl G-2011), Screening Whole Effluent Toxicity Samples

Matrix: Water, RL = 0.10 mg/L

Analyst	TS
Date analyzed	05-17-18

DPD: CHM 1004

Positive and Negative Control:

Control Type	Sample ID	Result (✓)	
		Positive	Negative
Negative control	Deionized water		✓
Positive control	Tap water	✓	

Sample screening:

Sample number	Sample ID	Sample characteristics	Result (✓)	
			Positive	Negative
180516.04	TVA/SQN 101	no color, clear		✓
180516.05	↓ INTAKE	no color, clear		✓
180517.02	ENFIELD	no color, clear		✓
180517.01	CHATTANOOGA	light yellow, clear		✓

TS 05-17-18

Note: All samples were analyzed in excess of EPA recommended holding time (15 minutes) unless otherwise noted. A positive result indicates the presence of total residual chlorine above detection (> 0.10 mg/L), which results in a pink to red color change with the addition of the DPD indicator. A negative result indicates the absence of total residual chlorine, which results in no color change with the addition of the DPD indicator.

Reviewed by	
Date reviewed	05-17-18

Total Residual Chlorine (ORION-97-70-1977), Confirmation of Whole Effluent Toxicity Samples

Matrix: Water, RL = 0.10 mg/L, Meter: Accumet Model AB250 pH/mV/Ion Meter

Analyst	TS	Iodide reagent:	INR 829
Date analyzed	05-17-18	Acid reagent:	INR 835

Calibration:

	0.10 mg/L	1.00 mg/L	Slope (mV) (suggested range = 26 to 30 mV)
Reference standard #	INSS 1617	INSS 1617	30

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

Laboratory control sample:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1617	0.50	0.534	106.8%

Duplicate sample precision:

Sample number	Sample ID	Total residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
18O516.04	TVA|SQN 101	S < 0.018	
↓	Duplicate	D < 0.015	05-17-18 TS

Sample measurements:

Sample number	Sample ID	Total residual chlorine (mg/L)
TV < 0.05 mg/L	Method Blank (MB)	< 0.012
TV = 0.30 mg/L	Method Detection Limit spike sample (MDL_s)	0.313
18O516.05	TVA|SQN INTAKE	< 0.007

TS 05-17-18

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

Laboratory control sample:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1617	0.50	0.513	102.6%

Reviewed by JJ Date reviewed 05-17-18

Total Residual Chlorine (4500-Cl G-2011), Screening Whole Effluent Toxicity Samples

Matrix: Water, RL = 0.10 mg/L

Analyst: TS
Date analyzed: 05-19-18

DPD: CHM 1004

Positive and Negative Control:

Control Type	Sample ID	Result (✓)	
		Positive	Negative
Negative control	Deionized water		✓
Positive control	Tap water	✓	

Sample screening:

Sample number	Sample ID	Sample characteristics	Result (✓)	
			Positive	Negative
18OS18-03	TVA/SQN 101	no color, clear, particles		✓
18OS18-04	↓ INTAKE	no color, clear	* ✓	
18OS19-02	ENFIELD	light tan, cloudy		✓
18OS19-01	CHATTANOOGA	light yellow, cloudy		✓

05-19-18 TS

Note: All samples were analyzed in excess of EPA recommended holding time (15 minutes) unless otherwise noted. A positive result indicates the presence of total residual chlorine above detection (> 0.10 mg/L), which results in a pink to red color change with the addition of the DPD indicator. A negative result indicates the absence of total residual chlorine, which results in no color change with the addition of the DPD indicator.

* FALSE POSITIVE FOR TVA/SQN SAMPLE
POSSIBLE TEST INTERFERENCE.

Reviewed by: JJ
Date reviewed: 05-19-18

Total Residual Chlorine (ORION-97-70-1977), Confirmation of Whole Effluent Toxicity Samples

Matrix: Water, RL = 0.10 mg/L, Meter: Accumet Model AB250 pH/mV/Ion Meter

Analyst	TS	Iodide reagent:	INR 829
Date analyzed	05-19-18	Acid reagent:	INR 835

Calibration:

	0.10 mg/L	1.00 mg/L	Slope (mV) (suggested range = 26 to 30 mV)
Reference standard #	INSS 1617	INSS 1617	28

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

Laboratory control sample:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1617	0.50	0.542	108.4%

Duplicate sample precision:

Sample number	Sample ID	Total residual chlorine (mg/L)	%RPD = {(S - D) /[(S+D)/2]} x 100 (acceptable range = ± 10%)
180518.03	TVA|SQN 101	S < 0.012	
↓	Duplicate	D < 0.010	TS 05-19-18

Sample measurements:

Sample number	Sample ID	Total residual chlorine (mg/L)
TV < 0.05 mg/L	Method Blank (MB)	<0.014
TV = 0.30 mg/L	Method Detection Limit spike sample (MDL_s)	0.321
180518.04	TVA|SQN INTAKE	<0.004

TS 05-19-18

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

Laboratory control sample:

Reference standard number	True value (TV) (mg/L)	Measured value (MV) (mg/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1617	0.50	0.525	105.0%

Reviewed by JT Date reviewed 05-19-18

Analyst: BSL
Date analyzed: 05.12.18

Alkalinity (SM 2320 B-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L, Samples are titrated to pH = 4.5 S.U.

Titrant normality and multiplier determination:

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (N x 50000)/ 100 mL sample = N x 500
INR 836	INSS 1654	0.1	12.6	12.5	0.0200	10.0

Laboratory control standard (LCS):

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%)
INSS 1638	100	100	12.6	22.5	9.9	10.0	~~10.0~~ 99.0 BSL 05.12.18	99.0%

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%)
	EES# 051804MV1	50	22.5	25.9	3.4	(2) 10.0	S 68 ~~34~~ BSL 05.12.18	3.0%
	Duplicate (D)	1	25.9	29.2	3.3	(2) 1	D 64 33 BSL 05.12.18	3.0% 1.5% BSL 05.12.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Alkalinity (mg $CaCO_3$/L)
MB (TV < 2.5 mg/L)	Deionized water, pH = 5.46 s.u.	100	0.0	0.1	0.1	10.0	1.0
	EES# 051805 MVE	100	29.2	30.6	1.4		14
	EES# 051806 MV ML	50	30.6	38.9	8.3	(2)	170
05.07.18	SSW	100	38.9	42.2	3.3		33
05.11.18	"	"	42.2	45.5	3.3		33
05.07.18	MHSW		0.0	6.0	6.0		60
05.10.18 A	"		6.0	12.1	6.1		61
05.10.18 B	"		12.1	18.3	6.2		62
05.11.18	SaltSW		18.3	28.5	10.2		100
05.07.18A	"		28.5	39.8	11.3		110
05.07.18B	"		39.8	49.8	10.0		100
05.08.18	"		0.0	9.8	9.8		98
05.09.18	"		9.8	20.4	10.6		110
05.10.18	"		20.4	31.6	11.2		110
180327.01	cdAC DMR		31.6	37.9	6.3		63
	ppAC DMR		37.9	44.8	6.9		69
	ABAC DMR		0.0	10.8	10.8		110
	ABLK DMR		10.8	22.0	11.2		110
	MBAC DMR		22.0	32.8	10.8		110
	MBLK DMR		32.8	43.9	11.1		110

Laboratory control standard (LCS):

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%)
INSS	100	100						BSL 05.12.18

Reviewed by: [signature]
Date reviewed: 05.13.18

Analyst: BSL
Date analyzed: 05.12.18

Alkalinity (SM 2320 B-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L, Samples are titrated to pH = 4.5 S.U.

Titrant normality and multiplier determination:

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 = 0.0180 - 0.0220)~~	pH Factor or Multiplier = (N x 50000)/ 100 mL sample = N x 500
INR	INSS					BSL 05.12.18

Laboratory control standard (LCS):

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%)
INSS 1638	100	100	0.0	9.8	9.8	10.0	98	98.0%

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%)
180509.18	Washington	25	9.8	14.4	4.6	10.0 (4)	S 180	
↓	Duplicate (D)	↓	14.4	18.9	4.5	10.0 (4)	D 180	BSL 05.12.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Alkalinity (mg $CaCO_3$/L)
~~MB (TV < 2.5 mg/L)~~	~~Deionized water, pH = ___ S.U.~~	~~100~~				10.0	BSL 05.12.18
180510.08	Chemtrade	25	18.9	20.9	2.0 (4)		(80) ~~Hold~~ BSL 05.12.18
180508.20	TVA SHF Am	50	20.9	25.5	4.6 (2)		92
180508.21	↓ Am INT	↓	25.5	29.6	4.1 (2)		82
180508.18	↓ Pm	↓	29.6	34.1	4.5 (2)		90
180508.19	↓ Pm INT	↓	34.1	38.4	4.3 (2)		86
180508.01	TVA BFN 1	50	38.4	41.5	3.1 (2)		62
180509.24	↓ 2	↓	41.5	44.6	3.1 (2)		62
180511.18	↓ 3	↓	~~44.6~~	47.9	3.3 (2)		66
180508.02	TVA BFN INT 1	50	0.0	3.1	3.1 (2)		62
180509.25	↓ 2	↓	3.1	6.3	3.2 (2)		64
180511.19	↓ 3	↓	6.3	9.5	3.2 (2)		64
180507.03	Eastman 1	25	9.5	11.6	2.1 (4)		84
180509.22	↓ 2	↓	11.6	13.9	2.3 (4)		92
180511.18	↓ 3	↓	13.9	16.5	2.6 (4)		100
180507.04	Eastman RW 1	25	16.5	18.4	1.9 (4)		76
180509.23	↓ 2	↓	18.4	20.4	2.0 (4)		80
180511.19	↓ 3	↓	20.4	22.7	2.3 (4)		92
180507.03	Eastman Anti 1	25	22.7	24.6	1.9 (4)		76
180509.22	↓ 2	↓	24.6	26.9	2.3 (4)		92

Laboratory control standard (LCS):

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%)
INSS	100	100						BSL 05.12.18

Reviewed by: K Date reviewed: 05.13.18

Analyst	BSL
Date analyzed	05.12.18

Alkalinity (SM 2320 B-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L, Samples are titrated to pH = 4.5 S.U.

Titrant normality and multiplier determination:

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (N x 50000)/ 100 mL sample = N x 500
INR	INSS					BSL 05.12.18

Laboratory control standard (LCS):

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%)
INSS 1638	100	100	26.7	36.5	9.6	10.0	96	96.0%

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%)
180511.18	Eastman Anti 3	25	36.5	39.0	2.5	(4) 10.0	S 100	
〃	Duplicate (D)	〃	39.0	41.5	2.5	(4) 〃	D 100	BSL 05.12.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Alkalinity (mg $CaCO_3$/L)
MB (TV < 2.5 mg/L)	Deionized water, pH = ____ S.U.	100					BSL 05.12.18
180507.04	Eastman Anti RW 1	25	41.5	43.5	2.0	(4) 10.0	80
180509.23	〃 2	〃	43.5	45.6	2.1	(4)	84
180511.19	〃 3	〃	45.6	47.8	2.2	(4)	88
05.03.18A	MHSW Anti 1	100	0.0	6.1	6.1		61
05.03.18B	〃 2	〃	6.1	12.1	6.0		60
05.07.18	〃 3	〃	12.1	18.2	6.1		61
180507.01	S. Pine 1	100	18.2	22.7	4.5		45
180509.21	〃 2	〃	22.7	26.8	4.1		41
180511.15	〃 3	〃	26.8	31.2	4.4		44
180508.06	Rockingham 1	50	31.2	33.2	2.0	(2)	40
180510.02	〃 2	〃	33.2	38.0	4.8	(2)	96
180512.01	〃 3	〃	38.0	42.8	4.8	(2)	96
180508.07	Bogue 1	5	42.8	48.0	5.2	(20)	1000
180510.03	〃 2	〃	0.0	4.8	4.8	(20)	960
180512.02	〃 3	〃	4.8	9.3	4.5	(20)	900
180508.08	Engelhard 1	2	9.3	15.5	6.2	(50)	3100
180510.04	〃 2	〃	15.5	21.9	6.4	(50)	3200
180512.03	〃 3	〃	21.9	28.3	6.4	(50)	3200
180508.09	Pasqw tank 1	〃	28.3	32.3	4.0	(50)	2000

Laboratory control standard (LCS):

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%)
INSS	100	100						BSL 05.12.18

Reviewed by: [signature] Date reviewed: 05.13.18

Analyst: BSL
Date analyzed: 05.12.18

Alkalinity (SM 2320 B-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L, Samples are titrated to pH = 4.5 S.U.

Titrant normality and multiplier determination:

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (N x 50000)/ 100 mL sample = N x 500
INR	INSS					BSL 05.12.18

Laboratory control standard (LCS):

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%)
INSS 1038	100	100	32.3	41.9	9.6	10.0	96.0	96.0%

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%)
180510.05	Pasqua tank 2	2	0.0	3.9	3.9 (50)	10.0	S 2000 ~~1900~~	051318
↓	Duplicate (D)	1	3.9	7.8	3.9 (50)	10.0	D 2000 ~~1900~~	BSL 05.12.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Alkalinity (mg $CaCO_3$/L)
~~MB (TV <2.5 mg/L)~~	~~Deionized water, pH = ____ S.U.~~	100					BSL 05.12.18
180512.04	Pasqua tank 3	2	7.8	12.0	4.2 (50)	10.0	2100
180508.10	C. Beach 1	25	12.0	15.3	3.3 (4)	|	130
180510.06	| 2	|	15.3	18.3	3.0 (4)	|	120
180512.05	| 3	|	18.3	20.9	2.6 (4)	|	100
180508.11	Pinova 1	25	20.9	24.8	3.9 (4)	|	160
180510.07	| 2	|	24.8	28.4	3.6 (4)	|	140
180512.06	| 3	|	28.4	32.1	3.7 (4)	|	~~HBSL~~ 150 05.12.18
05.12.18A	Saltsw	100	32.1	42.6	10.5	051318	~~HBSL~~ 100
05.12.18 B	↓	↓	23.3	32.9	9.6	↓	~~960~~ 96 051318
							BSL 05.12.18

Laboratory control standard (LCS):

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%)
INSS 1038	100	100	32.9	42.6	9.7	10.0	97	97.0%

Reviewed by: K Date reviewed: 05.13.18

Analyst: BSL
Date analyzed: 05.19.18

Alkalinity (SM 2320 B-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L, Samples are titrated to pH = 4.5 S.U.

Titrant normality and multiplier determination:

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (N x 50000)/ 100 mL sample = N x 500
INR 836	INSS 1654	0.1	12.6	12.5	0.0200	10.0

Laboratory control standard (LCS):

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%)
INSS 1638	100	100	12.6	22.5	9.9	10.0	99	99.0%

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%)
05.16.18B	MHSW	100	22.5	28.6	6.1	10.0	S 61	
↓	Duplicate (D)	↓	28.6	34.7	6.1	↓	D 61	BSL 05.19.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Alkalinity (mg $CaCO_3$/L)
MB (TV < 2.5 mg/L)	Deionized water, pH = 5.62 S.U.	100	0.0	0.1	0.1	10.0	1.0
05.18.18A	MHSW		34.7	40.6	5.9		59
05.18.18B	↓		40.6	46.5	5.9		59
05.16.18A			0.0 46.5 BSL 05.19.18		6.0		60
180327.01	CCCK DMR		6.0	12.1	6.1		61
↓	PPCK DMR		12.1	18.4	6.3		63
05-16-18A	MHSW UV 1		18.4	24.4	6.0		60
05-16-18A	↓ 2		24.4	30.3	5.9		59
05-16-18B	3		30.3	36.6	6.3		63
180514.02	TVA SQN 101 1	50	36.6	39.5	2.9 (2)		58
180516.04	↓ 2		39.5	42.4	2.9 (2)		58
180518.03	3		42.4	45.3	2.9 (2)		58
180514.02	TVA SQN 101 UV 1		45.3	48.2	2.9 (2)		58
180516.04	↓ 2		0.0	2.7	2.7 (2)		54
180518.03	↓ 3		2.7	5.8	3.1 (2)		62
180514.03	TVA SQN INT 1		5.8	8.6	2.8 (2)		56
180516.05	↓ 2		8.6	11.5	2.9 (2)		58
180518.04	↓ 3		11.5	14.4	2.9 (2)		58
180514.03	TVA SQN INT UV 1		14.4	17.2	2.8 (2)		56
180516.05	↓ 2		17.2	20.0	2.8 (2)		56

Laboratory control standard (LCS):

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%)
INSS	100	100						BSL 05.19.18

Reviewed by: ↓ Date reviewed: 05.19.18

Analyst	BSL
Date analyzed	05.19.18

Alkalinity (SM 2320 B-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L, Samples are titrated to pH = 4.5 S.U.

Titrant normality and multiplier determination:

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 = 0.0180 – 0.0220)	pH Factor or Multiplier = (N x 50000)/ 100 mL sample = N x 500
INR	INSS					BSL 05.19.18

Laboratory control standard (LCS):

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%)
INSS 1638	100	100	20.0	29.7	9.7	10.0	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%)
	EES# 051B09 MVI	50	29.7	33.8	4.1 (2)	10.0	S 82	
	Duplicate (D)	↓	33.8	37.9	4.1 (2)	↓	D 82	BSL 05.19.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Alkalinity (mg $CaCO_3$/L)
MB (TV < 2.5 mg/L)	Deionized water, pH = ____ S.U.	100					BSL 05.19.18
	EES# 051B10 MVE	100	37.9	38.9	1.0	10.0	10
	↓ 051B11 MVML	25	38.9	42.7	3.8 (4)		150
180516.04	TVA SQW INT UV 3	~~100~~ 50	~~38.9~~ 42.7	45.5	2.8 (2)		56
180515.02	Enhvd 1	100	0.0	0.5	0.5		5.0
180517.02	↓ 2	↓	0.5	~~0.5~~ 2.4	1.9		19
180519.02	3	↓	2.4	3.8	1.4		14
180515.01	Chattanooga 1	25	3.8	9.0	5.2 (4)		210
180517.01	↓ 2	↓	9.0	14.3	5.3 (4)		210
180519.01	↓ 3	↓	14.3	20.4	6.1 (4)		240
							BSL 05.19.18

Laboratory control standard (LCS):

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%)
INSS 1638	100	100	20.4	30.0	9.6	10.0	96	96.0%

Reviewed by: [signature] Date reviewed: 05.19.18

Analyst	BSL
Date analyzed	05.12.18

Hardness (SM 2340 C-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (N x 50000)/ 50 mL sample = N x 1000
INR 803	INSS 1588	0.0	10.0	10.0	0.0200	20.0

Laboratory control standard (LCS):

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%)
INSS 1618	40	50	10.0	12.1	2.1	20.0	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%)
	S. Pine EH	50	12.1	~~13.8~~ 13.8	1.7	20.0	S 34	
	Duplicate (D)	1	13.8	15.5	1.7	1	D 34	BSL 05.12.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)	
MB (TV < 2.5 mg/L)	Deionized water	50	0.0	0.0	0.0	20.0	ND	
	S. Pine UP		15.5	16.6	1.1		22	
05.07.18	SSW		16.6	18.5	1.9		38	
05.11.18	↓		18.5	20.5	2.0		40	
05.07.18	MHSW		20.5	24.7	4.2		84	
05.10.18A	↓		24.7	28.5	4.1		82	
05.10.18B	↓		~~33.0~~ 28.5	33.0 BSL 05.12.18	4.2		84	
180327.01	CdAC DMR		33.0	37.2	4.2		84	
↓	PPAC DMR		37.2	41.4	4.2		84	
180510.08	Chemtrade		41.4	48.0	6.6		130	
~~———~~	~~Washington~~						BSL 05.12.18	
180508.20	TVA SHF AM	25	0.0	2.9	2.9		58 BSL 05.12.18 120	=116
180508.21	↓ AM INT	↓	2.9	5.5	2.6		52 100	=104
180508.18	↓ PM	↓	5.5	8.4	2.9		58 120	=116
180508.19	↓ PM INT	↓	8.4	11.0	2.6		52 100	=104
180508.01	TVA BFN 1	25	11.0	12.8	1.8		86 72	
180509.24	↓ 2	↓	12.8	14.4	1.6		64	
180511.18	↓ 3	↓	14.4	16.1	1.7		68	
180508.02	TVA BFN INT 1	25	16.1	17.7	1.6		64	
180509.25	↓ 2	↓	17.7	19.4	1.7		68	

Laboratory control standard (LCS):

Reference standard number	True value (TV) (mg $CaCO_3$/L)	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS	40	50						BSL 05.12.18

Reviewed by: [ ] Date reviewed: 05.12.18

Analyst	BSC
Date analyzed	05.12.18

Hardness (SM 2340 C-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (N x 50000)/ 50 mL sample = N x 1000
INR	INSS					BSL 05.12.18

Laboratory control standard (LCS):

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%)
INSS 1618	40	50	19.4	21.3	1.9	20.0	38	95.0%

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%)
180511.19	TVA BFN INT 3	25	21.3	23.0	1.7	(2) 20.0	S 68	
↓	Duplicate (D)	↓	23.0	24.7	1.7	(2) ↓	D 68	BSL 05.12.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)
MB ~~(TV < 2.5 mg/L)~~	~~Deionized water~~	50				20	BSL 05.12.18
180507.03	Eastman 1	25	24.7	27.0	2.3	(2) 20.0	92
180509.22	2		27.0	29.5	2.5		100
180511.18	3		29.5	32.5	3.0		120
180507.04	Eastman RW 1		32.5	34.3	1.8		72
180509.23	2		34.3	36.3	2.0		80
180511.19	3		36.3	38.6	2.3		92
05.03.18A	MHSW Anti 1	50	38.6	42.8	4.2		84
05.03.18B	2		42.8	46.9	4.1		82
05.07.18	3		46.9 0.0 BSL 05/12/18	4.1	4.1		82
180507.05	Eastman Anti 1	25	4.1	6.4	2.3	(2)	92
180509.22	2		6.4	9.0	2.6		100
180511.18	3		9.0	11.8	2.8		110
180507.04	Eastman RW Anti 1		11.8	13.7	1.9		76
180509.23	2		13.7	15.8	2.1		84
180511.19	3		15.8	18.1	2.3		92
180507.01	Spine 1	50	18.1	19.9	1.8		36
180509.21	2		19.9	21.6	1.7		34
180511.15	3		21.6	23.5	1.9		38
180508.06	Rockingham 1		23.5	27.3	3.8		76

Laboratory control standard (LCS):

Reference standard number	True value (TV) (mg $CaCO_3$/L)	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS	40	50						BSL 05.12.18

Reviewed by: [signature] Date reviewed: 05.12.18

Analyst	BSL
Date analyzed	05.12.18

Hardness (SM 2340 C-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (*N* x 50000)/ 50 mL sample = *N* x 1000
INR	INSS					BSL 05.12.18

Laboratory control standard (LCS):

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%)
INSS 1618	40	50	27.3	29.3	2.0	20.0	40	100.0%

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%)
							S	
	Duplicate (D)						D 05.12.18	

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)
MB (TV < 2.5 mg/L)	Deionized water	50					05.12.18 BSL
180510.02	Rockingham 2	50	29.3	37.9	8.6	20.0	170
180512.01	↓ 3	↓	37.9	46.3	8.4	↓	170
180509.18	Washington	25	35.8	38.9	3.1	(2) ↓	120
							BSL 05.12.18

Laboratory control standard (LCS):

Reference standard number	True value (TV) (mg $CaCO_3$/L)	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1618	40	50	30.9	40.9	2.0	20.0	40	100.0%

Reviewed by: JL Date reviewed: 05.12.18

Analyst	BSL
Date analyzed	05.19.18

Hardness (SM 2340 C-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (N x 50000)/ 50 mL sample = N x 1000
INR 803	INSS 1588	0.0	10.0	10.0	0.0200	20.0

Laboratory control standard (LCS):

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%)
INSS 1618	40	50	10.0	12.0	2.0	20.0	40.8 BSL 05.19.18	100.0%

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%)
05.16.18B	MHSW	50	12.0	16.2	4.2	20.0	S 84	
↓	Duplicate (D)	↓	16.2	20.4	4.2	↓	D 84	BSL — 05.19.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)
MB (TV < 2.5 mg/L)	Deionized water	50	0.0	0.0	0.0	~~0.0~~ 20.0 BSL 05.19.18	ND
05.18.18A	MHSW	↓	20.4	24.6	4.2	↓	84
05.18.18B	↓	↓	24.6	28.8	4.2	↓	84
05.16.18A	MHSW	↓	28.8	33.0	4.2	↓	84
18 0327.01	CACR DMR	↓	33.0	37.1	4.1	↓	82
↓	PPCR DMR	↓	37.1	41.3	4.2	↓	84
05.10.18A	MHSWUV 1	↓	41.3	45.4	4.1	↓	82
05-16-18A	↓ 2	↓	45.4	49.6	4.2	↓	84
05-16-18B	↓ 3	↓	0.0	4.2	4.2	↓	84
18 0514.02	TVA-SQN 101 1	25	4.2	5.9	1.7 (2)	↓	68
18 0516.04	↓ 2	↓	5.9	7.4	1.5 (2)	↓	60
18 0518.03	↓ 3	↓	7.4	9.0	1.6 (2)	↓	64
18 0514.02	TVA SQN 101W 1	↓	9.0	10.5	1.5 (2)	↓	60
18 0516.04	↓ 2	↓	10.5	12.1	1.6 (2)	↓	64
18 0518.03	↓ 3	↓	12.1	13.7	1.6 (2)	↓	64
18 0514.03	TVA SQN INT 1	↓	13.7	15.2	1.5 (2)	↓	60
18 0516.05	↓ 2	↓	15.2	16.8	1.6 (2)	↓	64
18 0518.04	↓ 3	↓	16.8	18.3	1.5 (2)	↓	60
18 0514.03	TVA-SQN INTUV 1	↓	18.3	19.8	1.5 (2)	↓	60
18 0516.05	↓ 2	↓	19.8	21.3	1.5 (2)	↓	60

Laboratory control standard (LCS):

Reference standard number	True value (TV) (mg $CaCO_3$/L)	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS	40	50						BSL 05.19.18

Reviewed by: [signature] Date reviewed: 05.19.18

Analyst	BSL
Date analyzed	05.19.18

Hardness (SM 2340 C-2011)

Matrix: Water, RL = 5.0 mg $CaCO_3$/L

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 = 0.0180 - 0.0220)	pH Factor or Multiplier = (*N* x 50000)/ 50 mL sample = *N* x 1000
INR	INSS					BSL 05.19.18

Laboratory control standard (LCS):

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%)
INSS 1618	40	50	21.3	23.3	2.0	20.0	40	100.0%

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%)
18 0518.04	TVASOPINTON 3	25	23.3	24.8	1.5 (2)	20.0	S 60	
↓	Duplicate (D)	↓	24.8	26.3	1.5 (2)	↓	D 60	BSL 05.19.18

Sample measurements:

Sample number	Sample ID	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)
~~MB (TV < 2.5 mg/L)~~	Deionized water	50					BSL 05.19.18
18 0515.02	Enfield 1	50	26.3	27.2	0.9	20.0	18
18 0517.02	↓ 2	↓	27.2	28.4	1.2	↓	24
18 0519.02	3		37.5 ~~28.5~~	39.0	1.5		30
18 0515.01	Chattanooga 1	25	28.4	33.0	4.6 (2)		180
18 0517.01	2	↓	33.0	37.4	4.4 (2)		180
18 0519.01	↓ 3		39.0	43.0	4.0 (2)	↓	160
							BSL 05.19.18

Laboratory control standard (LCS):

Reference standard number	True value (TV) (mg $CaCO_3$/L)	Sample volume (mL)	Begin mL	End mL	Total mL	Multiplier	Hardness (mg $CaCO_3$/L)	% RS = MV / TV x 100 (acceptable range = 90 to 110%)
INSS 1618	40	50	43.0	45.0	2.0	20.0	40	100.0%

Reviewed by: [signature] Date reviewed: 05.19.18

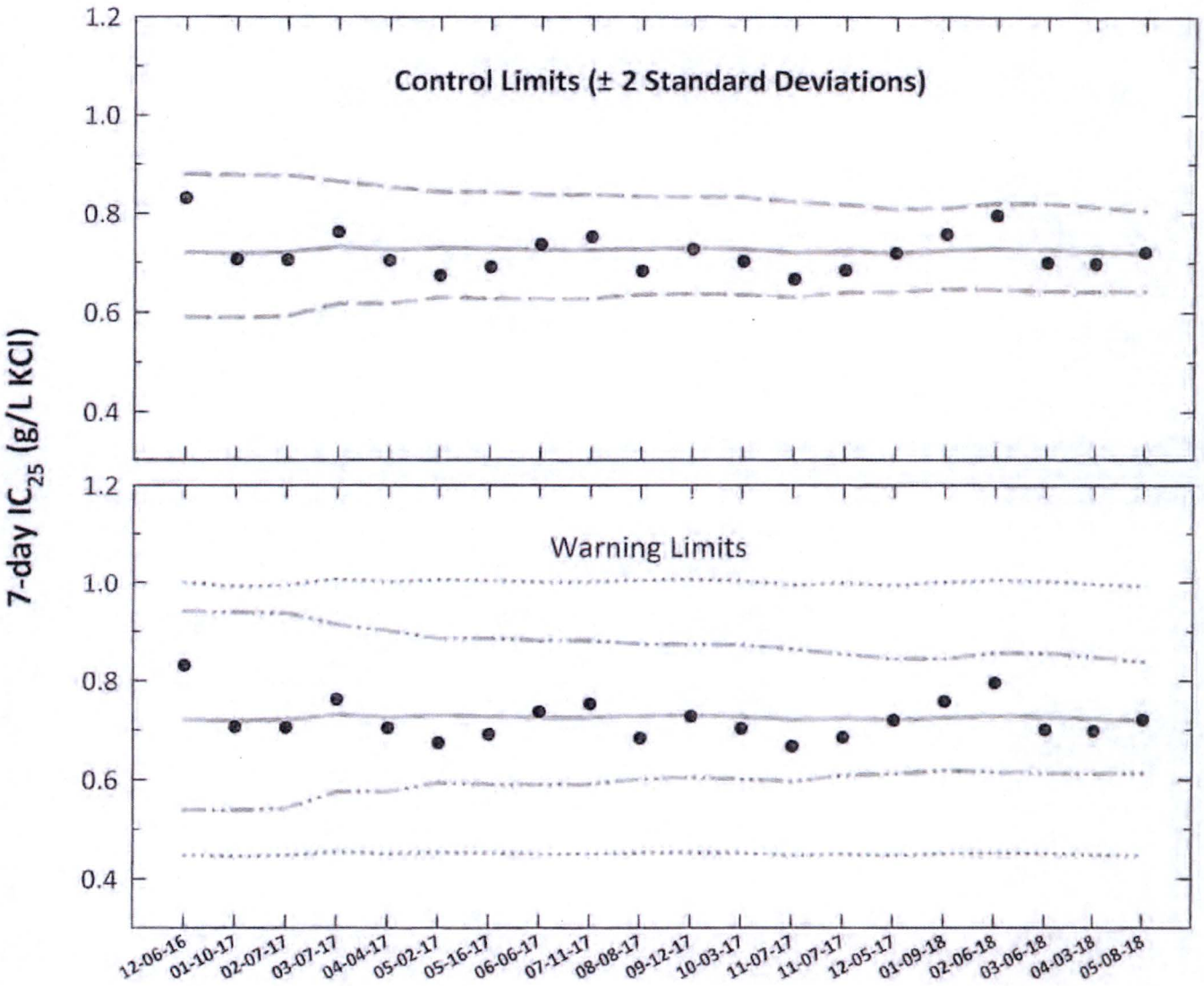
Sequoyah Nuclear Plant Biomonitoring
May 15 – 22, 2018

Appendix D

Reference Toxicant Test and
Control Chart

Environmental Testing Solutions, Inc.

Pimephales promelas
Chronic Reference Toxicant Control Chart
Source: In-house Culture



- ● **7-day IC_{25}** = 25% inhibition concentration. An estimation of the potassium chloride concentration which would cause a 25% reduction in *Pimephales* growth (calculated using ToxCalc).
- ——— **Central Tendency** (mean logarithmic IC_{25} converted to anti-logarithmic values)
- – – – **Control Limits** (mean logarithmic IC_{25} ± 2 standard deviations converted to anti-logarithmic values)
- — · — · **Laboratory Warning Limits** (mean logarithmic IC_{25} ± 2 coefficent of variations converted to anti-logarithmic values)
- ····· **USEPA Warning Limits** (mean logarithmic IC_{25} ± $S_{A.75}$ converted to anti-logarithmic values, $S_{A.75}$ = 75th percentile of CVs reported nationally by USEPA)

Independent Review by Kelley E. Keenan

Entered and Reviewed By Jim Sumner

Environmental Testing Solutions, Inc.

Pimephales promelas

Chronic Reference Toxicant Control Chart

Source: In-house Culture

Test number	Test date	7-day IC_{25} ToxCal Determination (g/L KCl)	Log_{10} Conversion			Anti-logarithmic Values (g/L KCl)						
			7-day IC_{25}	CT	S	CT	Control Limits		Laboratory Calculated CV Warning Limits		75th Percentile CV Warning Limits	
							CT - 2S	CT + 2S	CT - 2CV	CT + 2CV	CT - $S_{A,75}$	CT + $S_{A,75}$
1	**12-06-16**	**0.8303**	-0.0808	-0.1423	0.0433	0.7206	0.5904	0.8795	0.5399	0.9411	0.4468	1.0009
2	**01-10-17**	**0.7066**	-0.1508	-0.1433	0.0432	0.7189	0.5892	0.8773	0.5384	0.9392	0.4457	0.9921
3	**02-07-17**	**0.7052**	-0.1517	-0.1421	0.0426	0.7209	0.5924	0.8772	0.5426	0.9378	0.4469	0.9948
4	**03-07-17**	**0.7620**	-0.1181	-0.1362	0.0365	0.7308	0.6178	0.8646	0.5761	0.9139	0.4531	1.0086
5	**04-04-17**	**0.7043**	-0.1523	-0.1392	0.0351	0.7258	0.6174	0.8531	0.5765	0.9012	0.4500	1.0015
6	**05-02-17**	**0.6740**	-0.1714	-0.1371	0.0316	0.7292	0.6305	0.8434	0.5939	0.8858	0.4521	1.0063
7	**05-16-17**	**0.6906**	-0.1608	-0.1382	0.0320	0.7275	0.6278	0.8431	0.5904	0.8864	0.4511	1.0040
8	**06-06-17**	**0.7367**	-0.1327	-0.1393	0.0314	0.7256	0.6280	0.8384	0.5911	0.8810	0.4499	1.0013
9	**07-11-17**	**0.7519**	-0.1238	-0.1393	0.0314	0.7257	0.6280	0.8385	0.5911	0.8811	0.4499	1.0014
10	**08-08-17**	**0.6829**	-0.1656	-0.1379	0.0294	0.7280	0.6357	0.8336	0.6012	0.8731	0.4513	1.0046
11	**09-12-17**	**0.7271**	-0.1384	-0.1371	0.0292	0.7294	0.6377	0.8342	0.6037	0.8730	0.4522	1.0065
12	**10-03-17**	**0.7024**	-0.1534	-0.1382	0.0293	0.7274	0.6355	0.8326	0.6011	0.8720	0.4510	1.0038
13	**11-07-17**	**0.6666**	-0.1761	-0.1421	0.0289	0.7210	0.6310	0.8238	0.5961	0.8636	0.4470	0.9949
14	**11-07-17**	**0.6847**	-0.1645	-0.1404	0.0265	0.7237	0.6406	0.8175	0.6089	0.8533	0.4487	0.9987
15	**12-05-17**	**0.7191**	-0.1432	-0.1424	0.0251	0.7205	0.6418	0.8089	0.6112	0.8432	0.4467	0.9943
16	**01-09-18**	**0.7574**	-0.1207	-0.1397	0.0244	0.7249	0.6478	0.8113	0.6185	0.8441	0.4495	1.0004
17	**02-06-18**	**0.7951**	-0.0996	-0.1377	0.0260	0.7283	0.6461	0.8211	0.6154	0.8557	0.4516	1.0051
18	**03-06-18**	**0.7002**	-0.1548	-0.1387	0.0263	0.7266	0.6437	0.8201	0.6126	0.8552	0.4505	1.0027
19	**04-03-18**	**0.6973**	-0.1566	-0.1411	0.0256	0.7225	0.6423	0.8128	0.6115	0.8474	0.4480	0.9971
20	**05-08-18**	**0.7204**	-0.1424	-0.1429	0.0243	0.7196	0.6435	0.8048	0.6138	0.8380	0.4462	0.9931

Note: **7-day IC_{25}** = 25% inhibition concentration. An estimation of the potassium chloride concentration that would cause a 25% reduction in *Pimephales* growth (calculated using ToxCalc).

CT = Central tendency of the IC_{25} values.

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

Control Limits = Mean logarithmic IC_{25} ± 2 standard deviations converted to anti-logarithmic values.

Warning Limits = Mean logarithmic IC_{25} ± 2CV or $S_{A,75}$ converted to anti-logarithmic values.

$S_{A,75}$ = Standard deviation corresponding to the 75th percentile of CVs reported nationally by USEPA ($S_{A,75}$ = 0.38).

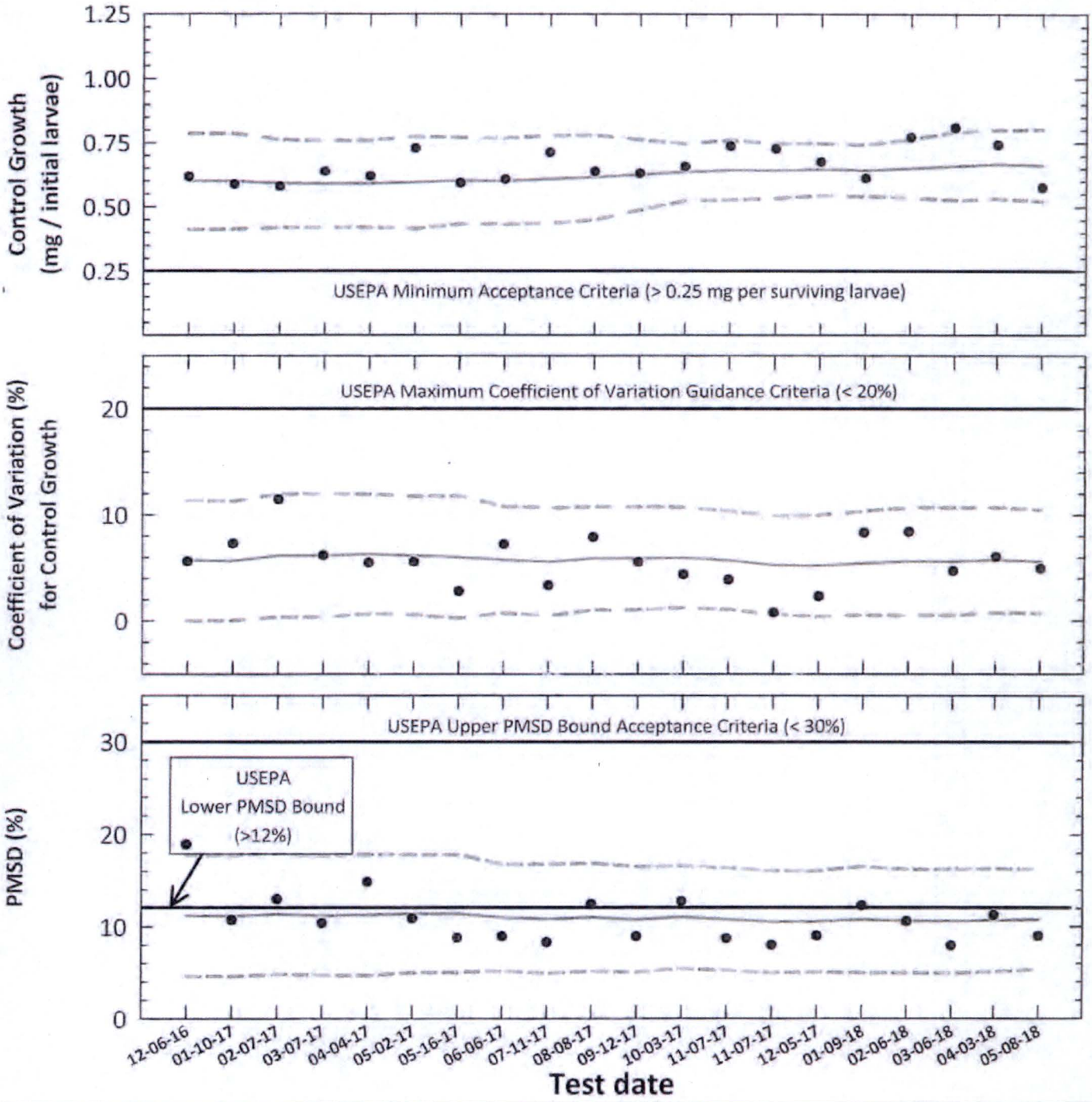
CV = Coefficient of variation.

ETS
Environmental Testing Solutions, Inc.

Pimephales promelas

Chronic Reference Toxicant Testing, Test Acceptability Criteria

Organism Source: In-house Culture



- **Control Growth, Coefficient of Variation (CV) or Percent Minimum Significant Difference (PMSD)**
  PMSD is the percent minimum significant difference between the control and treatment that can be declared statistically significant. The lower PMSD bound represents a practical limit to the sensitivity of the test method and is not a minimum acceptance criteria.

Central Tendency (mean Control Growth, CV or PMSD)

95% Confidence Interval (mean Control Growth, CV or PMSD ± 2 Standard Deviations)

Independent Review by Kelley E. Keenan

Entered and Reviewed by Jim Sumner

Pimephales promelas

Chronic Reference Toxicant Testing, Test Acceptability Criteria

Source: In-house Culture

Test number	Test date	ToxCal Determination					Control Growth (mg/initial larvae)			Control Growth CV (%)			Test PMSD (%)		
		Control Survival (%)	Control Growth Mean (mg/initial larvae)	Control Growth CV (%)	Test MSD	Test PMSD (%)	CT	95% Confidence Interval CT - 2S	CT + 2S	CT	95% Confidence Interval CT - 2S	CT + 2S	CT	95% Confidence Interval CT - 2S	CT + 2S
1	12-06-16	100	0.617	5.6	0.1167	18.9	0.599	0.413	0.786	5.7	0.0	11.4	11.2	4.6	17.8
2	01-10-17	100	0.587	7.3	0.0629	10.7	0.600	0.414	0.786	5.7	0.0	11.3	11.2	4.6	17.7
3	02-07-17	100	0.579	11.5	0.0751	13.0	0.591	0.420	0.763	6.2	0.3	12.0	11.4	4.8	17.9
4	03-07-17	100	0.638	6.2	0.0662	10.4	0.590	0.421	0.759	6.2	0.4	12.0	11.2	4.7	17.7
5	04-04-17	100	0.620	5.5	0.0919	14.8	0.591	0.422	0.761	6.3	0.6	11.9	11.3	4.7	17.9
6	05-02-17	100	0.729	5.6	0.0792	10.9	0.596	0.416	0.776	6.2	0.6	11.8	11.4	5.0	17.8
7	05-16-17	100	0.593	2.8	0.0521	8.8	0.603	0.434	0.771	6.0	0.2	11.8	11.4	5.0	17.8
8	06-06-17	100	0.608	7.2	0.0542	8.9	0.601	0.434	0.768	5.8	0.7	10.8	11.0	5.2	16.8
9	07-11-17	100	0.713	3.3	0.0589	8.3	0.609	0.438	0.781	5.6	0.5	10.7	10.8	4.9	16.8
10	08-08-17	100	0.638	7.9	0.0791	12.4	0.616	0.451	0.781	5.9	1.0	10.8	11.0	5.1	16.9
11	09-12-17	100	0.631	5.6	0.0562	8.9	0.626	0.489	0.764	5.9	1.0	10.8	10.8	5.0	16.5
12	10-03-17	100	0.658	4.4	0.0838	12.7	0.637	0.525	0.748	6.0	1.2	10.7	11.0	5.4	16.6
13	11-07-17	100	0.738	3.9	0.0641	8.7	0.645	0.528	0.761	5.7	1.1	10.4	10.8	5.2	16.4
14	11-07-17	100	0.727	0.8	0.0580	8.0	0.642	0.535	0.749	5.3	0.6	10.0	10.5	5.0	16.1
15	12-05-17	100	0.676	2.3	0.0607	9.0	0.647	0.545	0.750	5.2	0.4	10.0	10.6	5.0	16.1
16	01-09-18	100	0.612	8.3	0.0752	12.3	0.643	0.543	0.744	5.4	0.5	10.4	10.8	5.0	16.6
17	02-06-18	100	0.772	8.4	0.0815	10.6	0.651	0.538	0.765	5.6	0.5	10.7	10.6	5.0	16.2
18	03-06-18	100	0.810	4.7	0.0640	7.9	0.660	0.526	0.794	5.6	0.5	10.7	10.6	4.9	16.3
19	04-03-18	100	0.743	6.1	0.0836	11.3	0.668	0.534	0.802	5.8	0.8	10.7	10.7	5.1	16.3
20	05-08-18	100	0.576	4.9	0.0516	9.0	0.663	0.523	0.803	5.6	0.7	10.5	10.8	5.3	16.3

Note: Control Survival = USEPA minimum test acceptability criteria ≥ 80% survival.

Control Mean Growth = USEPA minimum test acceptability criteria ≥ 0.25 mg/surviving larvae.

CV = Coefficient of variation for control growth.

USEPA maximum CV guidance criteria (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.

Lower PMSD bound determined by USEPA (10th percentile) > 12%.

The lower PMSD bound represents a practical limit to the sensitivity of the test method and is not a minimum acceptance criteria.

Upper PMSD bound acceptance criteria determined by USEPA (90th percentile) < 30%.

CT = Central tendency of the growth, CV or PMSD values.

S = Standard deviation of the growth, CV or PMSD values.

Environmental Testing Solutions, Inc.

Potassium Chloride Chronic Reference Toxicant Test
(EPA-822-R-02-013 Method 1000.0)
Species: *Pimephales promelas*

PpKClCR Test Number: 39

Dilution preparation information:							*Comments:*
KCl Stock INSS number:			INSS 1685				
Stock preparation:			50 g KCl/L: Dissolve 50 g KCl in 1-L Milli-Q water.				
Dilution prep (mg/L)	300	450	600	750	900	1050	
Stock volume (mL)	9	13.5	18	22.5	27	31.5	
Diluent volume (mL)	1491	1486.5	1482	1477.5	1473	1468.5	
Total volume (mL)	1500	1500	1500	1500	1500	1500	

Test organism information:		*Test information:*	
Organism source:	In-house culture	Randomizing template:	LIGHT ORANGE
Age:	< 24-hours old	Incubator number and shelf location:	7B
Spawn date:	05-02-18	*Artemia* CHM number:	CHM984
Hatch dates and times:	05-07-18 1630 TO 05-08-18 0500	*Drying information for weight determination:*	
Transfer vessel information:	pH = 7.80 S.U. Temperature = 24.5 °C	Date / Time in oven:	05-15-18 0755
		Initial oven temperature:	60°C
Average transfer volume:	< 0.25 mL	Date / Time out of oven:	05-16-18 0755
		Final oven temperature:	60°C
		Total drying time:	24-HOURS

Daily feeding and renewal information:

Day	Date	Morning feeding		Afternoon feeding		Test initiation, renewal, or termination		MHSW batch used
		Time	Analyst	Time	Analyst	Time	Analyst	
0	05-08-18	0515	JL	1115	JL	0735	JL	05-03-18 A
1	05-09-18	0600	JL	1200	JL	0800	JL	05-03-18 B
2	05-10-18	0600	JL	1200	JL	0800	JL	05-03-18 B
3	05-11-18	0600	JL	1200	JL	0800	JL	05-03-18 B
4	05-12-18	0630	JL	1230	JL	0832	JL	05-07-18
5	05-13-18	0630	JL	1230	JL	0830	JL	05-07-18
6	05-14-18	0600	JL	1200	JL	0800	JL	05-07-18
7	05-15-18					0635	JL	

Control information:		Acceptance criteria
% Mortality:	0%	≤ 20%
Average weight per initial larvae:	0.576	
Average weight per surviving larvae:	0.576	≥ 0.25 mg/larvae

Summary of test endpoints:	
7-day LC_{50}	853.9
NOEC	600
LOEC	750
ChV	670.8
IC_{25}	720.4

Independent Review by Kelley E. Keenan: JL

Environmental Testing Solutions, Inc.

Species: ***Pimephales promelas***

PpKClCR Test Number: 39

Survival and Growth Data

Day	Control				300 mg KCl/L				450 mg KCl/L			
	A	B	C	D	E	F	G	H	I	J	K	L
0	10	10	10	10	10	10	10	10	10	10	10	10
1	10	10	10	10	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10	10	10	10	10
4	10	10	10	10	10	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10	10	10	10	10
6	10	10	10	10	10	10	10	10	10	10	10	10
7	10	10	10	10	10	10	10	10	10	10	10	10
A = Pan weight (mg) Tray color code:: forest green Analyst: TS Date: 04-24-18	14.83	15.22	14.92	15.34	15.45	15.89	16.59	14.02	15.66	15.24	15.22	14.91
B = Pan + Larvae weight (mg) Analyst: TS Date: 05-17-18	20.45	20.75	21.09	21.05	22.24	22.19	23.13	20.95	21.39	21.43	20.87	21.24
C = Larvae weight (mg) = B – A Hand calculated. Analyst: JI	5.62	5.53	6.17	5.71	6.79	6.30	6.54	6.93	5.73	6.19	5.65	6.33
Weight per initial number of larvae (mg) = C / Initial number of larvae Hand calculated. Analyst: JI	0.562	0.553	0.617	0.571	0.679	0.630	0.654	0.693	0.573	0.619	0.565	0.633
Average weight per initial number of larvae (mg)	0.576		0.664		-15.37.		0.598		-3.87.			
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.

Comments:

Independent Review by Kelley E. Keenan

Environmental Testing Solutions, Inc.

Species: ***Pimephales promelas*** PpKClCR Test Number: 39

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	10	10	10	10	10	10	8 2d	9 1d	9 1d	8 2d
2	10	10	10	10	10	9 1d	10	10	8	9	9	8
3	10	10	10	10	10	8 1d	10	10	7 1d	9	9	8
4	10	10	10	10	10	8	10	10	7	8 1d	8 1d	8
5	10	10	10	10	9 1d	8	9 1d	9 1d	6 1d	6 2d	8	7 1d
6	10	10	10	9 1d	8 1d	7 1d	8 1d	9	5 1d	4 2d	5 3d	5 2d
7	10	10	10	9	7 1d	7	8	8 1d	4 1d	4	4 1d	4 1d
A = Pan weight (mg) Tray color code:: forest green Analyst: TS Date: 04-24-18	14.60	15.83	14.26	15.21	15.86	15.84	15.40	15.62	15.25	14.73	15.05	15.70
B = Pan + Larvae weight (mg) Analyst: TS Date: 05-17-18	20.70	21.50	20.62	20.79	20.01	19.79	20.34	19.92	17.77	17.20	17.59	18.24
C = Larvae weight (mg) = B – A Hand calculated. Analyst: JT	6.10	5.67	6.36	5.58	4.15	3.95	4.94	4.30	2.52	2.47	2.54	2.54
Weight per initial number of larvae (mg) = C / Initial number of larvae Hand calculated. Analyst: JT	0.610	0.567	0.636	0.558	0.415	0.395	0.494	0.430	0.252	0.247	0.254	0.254
Average weight per initial number of larvae (mg)	0.593				0.434				0.252			
Percent reduction from control (%)		-3.07.				24.77.				56.37.		

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.

Comments:

Independent Review by Kelley E. Keenan: K

ETS Environmental Testing Solutions, Inc.

Species: ***Pimephales promelas***

PpKClCR Test Number: 39

Survival and Growth Data

Day	1050 mg KCl/L			
	Y	Z	AA	BB
0	10	10	10	10
1	6 4d	6 4d	5 5d	6 4d
2	6	6	5	5 1d
3	6	6	5	5
4	6	6	4 1d	4 1d
5	3 3d	4 2d	3 1d	4
6	3	4	2 4d	3 1d
7	2 1d	1 3d	1 1d	1 2d
A = Pan weight (mg) Tray color code:: forest green Analyst: TS Date: 04-24-18	14.90	15.22	15.07	15.33
B = Pan + Larvae weight (mg) Analyst: TS Date: 05-17-18	16.17	15.78	15.73	15.90
C = Larvae weight (mg) = B – A Hand calculated. Analyst: JH	1.27	0.56	0.66	0.57
Weight per initial number of larvae (mg) = C / Initial number of larvae Hand calculated. Analyst: JH	0.127	0.056	0.066	0.057
Average weight per initial number of larvae (mg)	0.077		86.72	
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.

Comments:

Independent Review by Kelley E. Keenan: K

Environmental Testing Solutions, Inc.

Pimephales promelas Chronic Reference Toxicant Test
EPA-821-R-02-013, Method 1000.0

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

Test number: 39
Test dates: May 08-15, 2018

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) = B - A	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	14.83	20.45	5.62	0.562	0.576	4.9	0.562	100.0	0.576	4.9	Not applicable
	B	10	10	15.22	20.75	5.53	0.553			0.553				
	C	10	10	14.92	21.09	6.17	0.617			0.617				
	D	10	10	15.34	21.05	5.71	0.571			0.571				
300	E	10	10	15.45	22.24	6.79	0.679	0.664	4.2	0.679	100.0	0.664	4.2	-15.3
	F	10	10	15.89	22.19	6.30	0.630			0.630				
	G	10	10	16.59	23.13	6.54	0.654			0.654				
	H	10	10	14.02	20.95	6.93	0.693			0.693				
450	I	10	10	15.66	21.39	5.73	0.573	0.598	5.6	0.573	100.0	0.598	5.6	-3.8
	J	10	10	15.24	21.43	6.19	0.619			0.619				
	K	10	10	15.22	20.87	5.65	0.565			0.565				
	L	10	10	14.91	21.24	6.33	0.633			0.633				
600	M	10	10	14.60	20.70	6.10	0.610	0.608	4.9	0.610	97.5	0.593	6.2	-3.0
	N	10	10	15.83	21.50	5.67	0.567			0.567				
	O	10	10	14.26	20.62	6.36	0.636			0.636				
	P	10	9	15.21	20.79	5.58	0.620			0.558				
750	Q	10	7	15.86	20.01	4.15	0.593	0.578	6.0	0.415	75.0	0.434	9.9	24.7
	R	10	7	15.84	19.79	3.95	0.564			0.395				
	S	10	8	15.40	20.34	4.94	0.618			0.494				
	T	10	8	15.62	19.92	4.30	0.538			0.430				
900	U	10	4	15.25	17.77	2.52	0.630	0.629	1.3	0.252	40.0	0.252	1.3	56.3
	V	10	4	14.73	17.20	2.47	0.618			0.247				
	W	10	4	15.05	17.59	2.54	0.635			0.254				
	X	10	4	15.70	18.24	2.54	0.635			0.254				
1050	Y	10	2	14.90	16.17	1.27	0.635	0.606	8.1	0.127	12.5	0.077	44.4	86.7
	Z	10	1	15.22	15.78	0.56	0.560			0.056				
	AA	10	1	15.07	15.73	0.66	0.660			0.066				
	BB	10	1	15.33	15.90	0.57	0.570			0.057				

Dunnett's MSD value: 0.0516 MSD = Minimum Significant Difference
PMSD: 9.0 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.
Lower PMSD bound determined by USEPA (10th percentile) = 12%
Upper PMSD bound determined by USEPA (90th percentile) = 30%.
Lower and upper PMSD bounds were determined from the 10th and 90th percentile, respectively, of PMSD data from EPA's WET interlaboratory Variability Study (USEPA, 2001a; USEPA, 2001b).
The lower PMSD bound represents a practical limit to the sensitivity of the test method and is not a minimum acceptance criteria.

USEPA. 2001a, 2001b. Final Report: Interlaboratory Variability Study of EPA Short-term Chronic and Acute Whole Effluent Toxicity Test Methods, Volumes 1 and 2-Appendix. EPA-821-B-01-004 and EPA-821-B-01-005. US Environmental Protection Agency, Cincinnati, OH.

ETS
Environmental Testing Solutions, Inc.

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Survival

Start Date:	5/8/2018	Test ID:	PpKCICR	Sample ID:	REF-Ref Toxicant
End Date:	5/15/2018	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	1.0000	1.0000	1.0000	1.0000
300	1.0000	1.0000	1.0000	1.0000
450	1.0000	1.0000	1.0000	1.0000
600	1.0000	1.0000	1.0000	0.9000
750	0.7000	0.7000	0.8000	0.8000
900	0.4000	0.4000	0.4000	0.4000
1050	0.2000	0.1000	0.1000	0.1000

			Transform: Arcsin Square Root					Rank	1-Tailed	Number	Total
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	Sum	Critical	Resp	Number
D-Control	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4			0	40
300	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4	18.00	10.00	0	40
450	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4	18.00	10.00	0	40
600	0.9750	0.9750	1.3713	1.2490	1.4120	5.942	4	16.00	10.00	1	40
*750	0.7500	0.7500	1.0492	0.9912	1.1071	6.383	4	10.00	10.00	10	40
*900	0.4000	0.4000	0.6847	0.6847	0.6847	0.000	4	10.00	10.00	24	40
*1050	0.1250	0.1250	0.3572	0.3218	0.4636	19.861	4	10.00	10.00	35	40

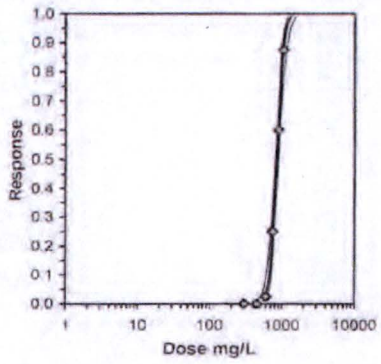
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.87142	0.896	-0.2893	2.51678
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	600	750	670.82	
Treatments vs D-Control				

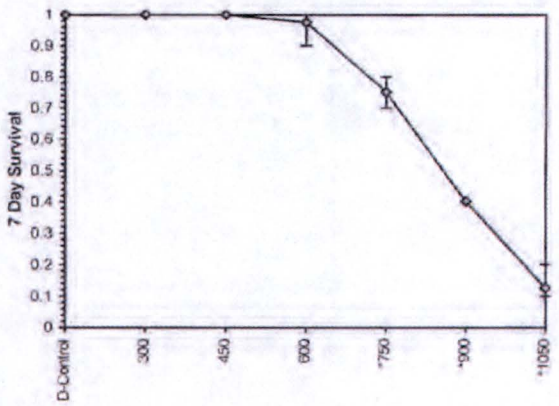
Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	12.5981	1.67874	9.30775	15.8884	0	0.07613	9.48773	0.99929	2.9314	0.07938	3
Intercept	-31.93	4.91886	-41.571	-22.289							
TSCR											

Point	Probits	mg/L	95% Fiducial Limits	
EC01	2.674	558.141	477.111	613.98
EC05	3.355	632.177	562.953	679.848
EC10	3.718	675.58	614.186	718.592
EC15	3.964	706.535	650.845	746.563
EC20	4.158	732.145	681.06	770.106
EC25	4.326	754.855	707.608	791.441
EC40	4.747	815.253	775.803	851.554
EC50	5.000	853.891	816.5	893.659
EC60	5.253	894.36	856.198	941.279
EC75	5.674	965.921	920.361	1032.97
EC80	5.842	995.882	945.635	1073.49
EC85	6.036	1031.98	975.257	1123.55
EC90	6.282	1079.27	1013.04	1190.82
EC95	6.645	1153.36	1070.6	1299.41
EC99	7.326	1306.36	1185.27	1533.44



Dose-Response Plot



Independent Review by Kelley E. Keenan

Environmental Testing Solutions, Inc.

Statistical Analyses

Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	5/8/2018	Test ID:	PpKCICR	Sample ID:	REF-Ref Toxicant
End Date:	5/15/2018	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:					

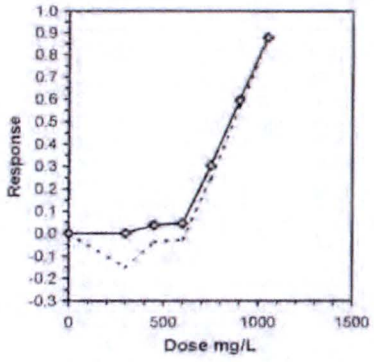
Conc-mg/L	1	2	3	4
D-Control	0.5620	0.5530	0.6170	0.5710
300	0.6790	0.6300	0.6540	0.6930
450	0.5730	0.6190	0.5650	0.6330
600	0.6100	0.5670	0.6360	0.5580
750	0.4150	0.3950	0.4940	0.4300
900	0.2520	0.2470	0.2540	0.2540
1050	0.1270	0.0560	0.0660	0.0570

			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	0.5758	1.0000	0.5758	0.5530	0.6170	4.944	4				0.6199	1.0000
300	0.6640	1.1533	0.6640	0.6300	0.6930	4.190	4	-3.919	2.290	0.0516	0.6199	1.0000
450	0.5975	1.0378	0.5975	0.5650	0.6330	5.617	4	-0.966	2.290	0.0516	0.5975	0.9639
600	0.5928	1.0295	0.5928	0.5580	0.6360	6.190	4	-0.755	2.290	0.0516	0.5928	0.9562
750	0.4335	0.7529	0.4335	0.3950	0.4940	9.874	4				0.4335	0.6993
900	0.2518	0.4373	0.2518	0.2470	0.2540	1.312	4				0.2518	0.4061
1050	0.0765	0.1329	0.0765	0.0560	0.1270	44.400	4				0.0765	0.1234

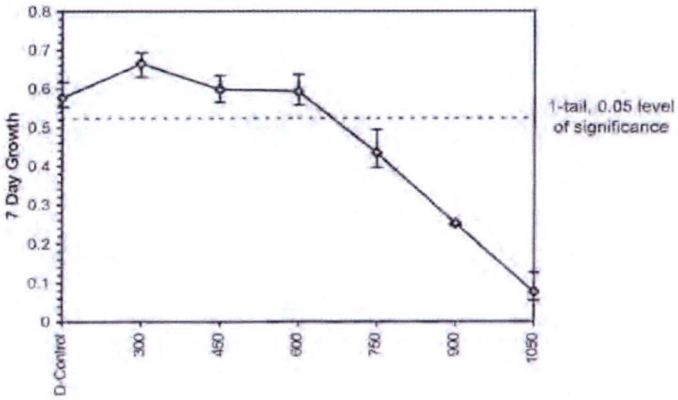
Auxiliary Tests					Statistic		Critical		Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.89619		0.844		0.24992	-1.5798
Bartlett's Test indicates equal variances (p = 0.96)					0.28117		11.3449			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	600	>600			0.05157	0.08957	0.00602	0.00101	0.01008	3, 12
Treatments vs D-Control										

Linear Interpolation (200 Resamples)

Point	mg/L	SD	95% CL(Exp)		Skew
IC05	603.64	78.83	281.16	639.87	-0.8198
IC10	632.84	15.23	592.01	666.71	-2.6811
IC15	662.03	11.96	623.04	701.48	0.1775
IC20	691.22	12.82	658.00	736.94	0.4492
IC25	720.42	14.65	686.25	770.90	0.4973
IC40	800.82	9.95	769.45	827.68	-0.1471
IC50	851.98	5.42	835.65	867.63	-0.1673



Dose-Response Plot



Independent Review by Kelley E. Keenan

Environmental Testing Solutions, Inc.

Species: ***Pimephales promelas*** PpKClCR Test Number: 39

Daily Chemistry:

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)					
		0		1		2	
	Analyst	MS	MS	MS	[illegible]	[illegible]	MS
Concentration	Parameter						
CONTROL	pH (S.U.)	7.94	7.82	7.88	7.69	8.01	7.75
	DO (mg/L)	7.7	7.7	7.8	6.9	7.7	7.4
	Conductivity (µmhos/cm)	317		310		324	
	*Alkalinity (mg $CaCO_3$/L)	59		59		[illegible]	
	*Hardness (mg $CaCO_3$/L)	86		86		[illegible]	
	*Temperature (°C)	24.6	24.6	24.7	24.7	24.8	24.4
300 mg KCl/L	pH (S.U.)	8.03	7.81	7.97	7.63	8.07	7.73
	DO (mg/L)	7.7	7.6	7.8	7.0	7.7	7.4
	Conductivity (µmhos/cm)	862		856		845	
	*Temperature (°C)	24.7	24.6	24.7	24.3	24.9	24.6
450 mg KCl/L	pH (S.U.)	8.02	7.82	7.97	7.61	8.04	7.79
	DO (mg/L)	7.8	7.5	7.9	7.0	7.7	7.4
	Conductivity (µmhos/cm)	1100		1110		1080	
	*Temperature (°C)	24.6	24.2	24.8	24.5	24.9	24.6
600 mg KCl/L	pH (S.U.)	8.02	7.78	7.96	7.57	8.04	7.75
	DO (mg/L)	7.8	7.5	7.9	6.9	7.7	7.5
	Conductivity (µmhos/cm)	1380		1350		1350	
	*Temperature (°C)	24.6	24.4	24.7	24.5	24.9	24.2
750 mg KCl/L	pH (S.U.)	8.02	7.78	7.95	7.56	8.05	7.69
	DO (mg/L)	7.9	7.5	8.0	6.8	7.8	7.5
	Conductivity (µmhos/cm)	1620		1630		1620	
	*Temperature (°C)	24.6	24.6	24.7	24.5	24.8	24.6
900 mg KCl/L	pH (S.U.)	8.00	7.80	7.95	7.56	8.06	7.70
	DO (mg/L)	7.9	7.5	8.1	7.0	7.9	7.5
	Conductivity (µmhos/cm)	1870		1900		1890	
	*Temperature (°C)	24.6	24.3	24.7	24.6	24.6	24.5
1050 mg KCl/L	pH (S.U.)	8.01	7.78	7.95	7.57	8.07	7.76
	DO (mg/L)	7.9	7.6	8.1	6.8	8.0	7.7
	Conductivity (µmhos/cm)	2160		2180		2190	
	*Temperature (°C)	24.6	24.5	24.7	24.5	24.8	24.5
		Initial	Final	Initial	Final	Initial	Final

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1.
Alkalinity and hardness performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: [illegible]

Independent Review by Kelley E. Keenan: [illegible]

Environmental Testing Solutions, Inc.

Species: ***Pimephales promelas***

PpKClCR Test Number: 39

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)							
		3		4		5		6	
	Analyst	MS	BSL	BSL	N	N	N	N	MS
Concentration	Parameter								
CONTROL	pH (S.U.)	7.94	7.80	7.95	7.69	8.03	7.65	8.00	7.83
	DO (mg/L)	7.7	7.9	7.6	7.0	7.7	7.7	7.8	7.4
	Conductivity (µmhos/cm)	305		299		306		300	
	*Alkalinity (mg $CaCO_3$/L)			60					
	*Hardness (mg $CaCO_3$/L)			84					
	*Temperature (°C)	24.7	24.5	24.7	24.6	24.6	24.4	24.8	24.4
300 mg KCl/L	pH (S.U.)	8.04	7.74	8.04	7.57	8.04	7.60	8.07	7.85
	DO (mg/L)	7.8	7.9	7.9	7.0	7.6	7.6	7.8	7.4
	Conductivity (µmhos/cm)	848		857		843		825	
	*Temperature (°C)	24.8	24.3	24.8	24.3	24.6	24.5	24.9	24.6
450 mg KCl/L	pH (S.U.)	8.03	7.76	8.05	7.59	8.04	7.66	8.05	7.84
	DO (mg/L)	7.8	7.8	8.0	6.9	7.7	7.6	7.9	7.6
	Conductivity (µmhos/cm)	1110		1110		1110		1070	
	*Temperature (°C)	24.8	24.4	24.7	24.6	24.6	24.5	24.9	24.3
600 mg KCl/L	pH (S.U.)	8.03	7.69	8.05	7.54	8.04	7.64	8.05	7.83
	DO (mg/L)	7.8	7.7	8.0	7.0	7.7	7.7	7.9	7.5
	Conductivity (µmhos/cm)	1380		1390		1380		1360	
	*Temperature (°C)	24.7	24.4	24.7	24.5	24.6	24.6	24.9	24.3
750 mg KCl/L	pH (S.U.)	8.03	7.70	8.05	7.50	8.05	7.59	8.04	7.80
	DO (mg/L)	7.9	7.6	8.0	6.7	7.8	7.7	7.9	7.5
	Conductivity (µmhos/cm)	1610		1630		1640		1620	
	*Temperature (°C)	24.9	24.6	24.7	24.5	24.7	24.3	24.9	24.5
900 mg KCl/L	pH (S.U.)	8.02	7.71	8.04	7.63	8.06	7.62	8.03	7.81
	DO (mg/L)	7.9	7.6	8.0	7.0	7.8	7.6	7.9	7.5
	Conductivity (µmhos/cm)	1860		1930		1950		1900	
	*Temperature (°C)	24.8	24.3	24.8	24.7	24.7	24.7	24.9	24.6
1050 mg KCl/L	pH (S.U.)	8.02	7.71	8.04	7.73	8.07	7.66	8.04	7.85
	DO (mg/L)	8.0	7.6	8.0	7.3	7.8	7.7	8.0	7.5
	Conductivity (µmhos/cm)	2150		2100		2110		2110	
	*Temperature (°C)	24.8	24.3	24.8	24.4	24.7	24.3	25.0	24.6
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

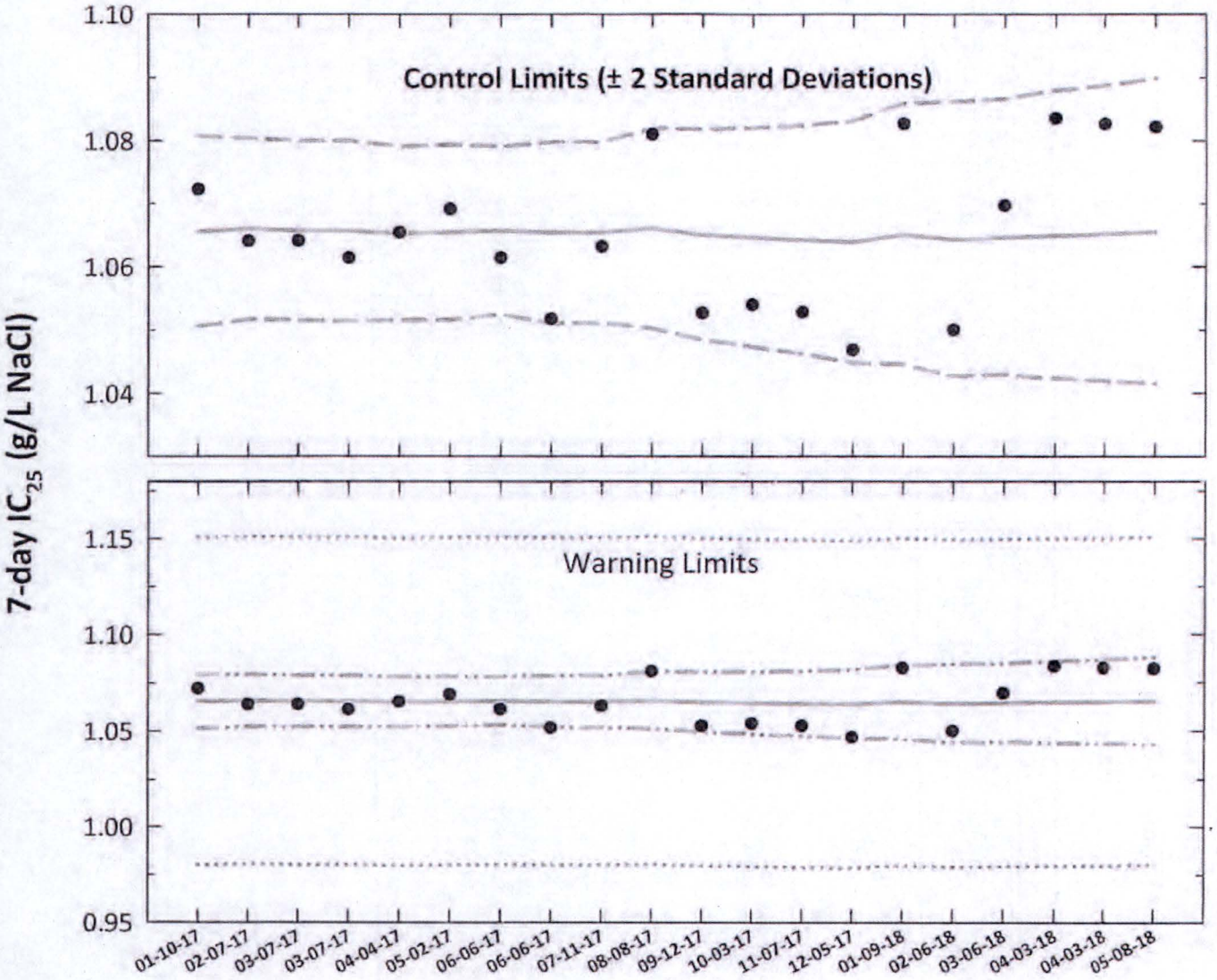
Independent Review by Kelley E. K. N

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1.

*Alkalinity and hardness performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: _____

ETS
Environmental Testing Solutions, Inc.

Ceriodaphnia dubia
Chronic Reference Toxicant Control Chart
Source: In-house Culture



Test date

- ● **7-day IC_{25}** = 25% inhibition concentration. An estimation of the sodium chloride concentration which would cause a 25% reduction in *Ceriodaphnia* reproduction (calculated using ToxCalc).
- ——— **Central Tendency** (mean logarithmic IC_{25} converted to anti-logarithmic values)
- – – – **Control Limits** (mean logarithmic IC_{25} ± 2 standard deviations converted to anti-logarithmic values)
- —·—·— **Laboratory Warning Limits** (mean logarithmic IC_{25} ± 2 coefficent of variations converted to anti-logarithmic values)
- ····· **USEPA Warning Limits** (mean logarithmic IC_{25} ± $S_{A.10}$ converted to anti-logarithmic values, $S_{A.10}$ = 10th percentile of CVs reported nationally by USEPA)

Independent Review by Kelley E. Keenan

Entered and Reviewed by Jim Sumner

Environmental Testing Solutions, Inc.

Ceriodaphnia dubia

Chronic Reference Toxicant Control Chart

Source: In-house Culture

Test number	Test date	7-day IC_{25} ToxCal Determination (g/L NaCl)	Log_{10} Conversion			Anti-logarithmic Values (g/L NaCl)						
			7-day IC_{25}	CT	S	CT	Control Limits		Laboratory Calculated CV Warning Limits		10th Percentile CV Warning Limits	
							CT - 2S	CT + 2S	CT - 2CV	CT + 2CV	CT - $S_{A.10}$	CT + $S_{A.10}$
1	01-10-17	1.0723	0.0303	0.0276	0.0031	1.0656	1.0506	1.0808	1.0515	1.0796	0.9803	1.1508
2	02-07-17	1.0641	0.0270	0.0278	0.0029	1.0660	1.0517	1.0804	1.0526	1.0794	0.9807	1.1513
3	03-07-17	1.0641	0.0270	0.0276	0.0029	1.0657	1.0516	1.0800	1.0524	1.0789	0.9804	1.1509
4	03-07-17	1.0614	0.0259	0.0276	0.0029	1.0656	1.0515	1.0800	1.0524	1.0789	0.9804	1.1509
5	04-04-17	1.0654	0.0275	0.0274	0.0028	1.0652	1.0516	1.0791	1.0524	1.0781	0.9800	1.1505
6	05-02-17	1.0691	0.0290	0.0275	0.0028	1.0654	1.0516	1.0794	1.0525	1.0783	0.9802	1.1506
7	06-06-17	1.0614	0.0259	0.0276	0.0027	1.0657	1.0524	1.0791	1.0532	1.0781	0.9804	1.1509
8	06-06-17	1.0518	0.0219	0.0275	0.0029	1.0653	1.0511	1.0798	1.0520	1.0787	0.9801	1.1506
9	07-11-17	1.0631	0.0266	0.0275	0.0029	1.0654	1.0512	1.0798	1.0520	1.0787	0.9802	1.1506
10	08-08-17	1.0810	0.0338	0.0277	0.0032	1.0660	1.0502	1.0820	1.0512	1.0807	0.9807	1.1513
11	09-12-17	1.0527	0.0223	0.0274	0.0034	1.0650	1.0485	1.0818	1.0495	1.0805	0.9798	1.1502
12	10-03-17	1.0540	0.0228	0.0272	0.0035	1.0646	1.0474	1.0821	1.0484	1.0807	0.9794	1.1497
13	11-07-17	1.0528	0.0223	0.0270	0.0037	1.0642	1.0463	1.0824	1.0473	1.0810	0.9790	1.1493
14	12-05-17	1.0469	0.0199	0.0269	0.0039	1.0638	1.0449	1.0832	1.0460	1.0817	0.9787	1.1489
15	01-09-18	1.0826	0.0345	0.0274	0.0042	1.0651	1.0446	1.0859	1.0458	1.0843	0.9799	1.1503
16	02-06-18	1.0500	0.0212	0.0270	0.0044	1.0642	1.0427	1.0862	1.0440	1.0844	0.9791	1.1494
17	03-06-18	1.0697	0.0293	0.0272	0.0045	1.0646	1.0430	1.0867	1.0443	1.0849	0.9794	1.1498
18	04-03-18	1.0835	0.0348	0.0273	0.0047	1.0649	1.0424	1.0880	1.0437	1.0861	0.9797	1.1501
19	04-03-18	1.0827	0.0345	0.0274	0.0048	1.0651	1.0420	1.0888	1.0434	1.0869	0.9799	1.1503
20	05-08-18	1.0822	0.0343	0.0275	0.0049	1.0655	1.0415	1.0899	1.0430	1.0879	0.9802	1.1507

Note: **7-day IC_{25}** = 25% inhibition concentration. An estimation of the sodium chloride concentration that would cause a 25% reduction in *Ceriodaphnia* reproduction (calculated using ToxCalc).

CT = Central tendency of the IC_{25} values.

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

Control Limits = Mean logarithmic IC_{25} ± 2 standard deviations converted to anti-logarithmic values.

Warning Limits = Mean logarithmic IC_{25} ± 2CV or $S_{A.10}$ converted to anti-logarithmic values.

$S_{A.10}$ = Standard deviation corresponding to the 10th percentile of CVs reported nationally by USEPA ($S_{A.10}$ = 0.08).

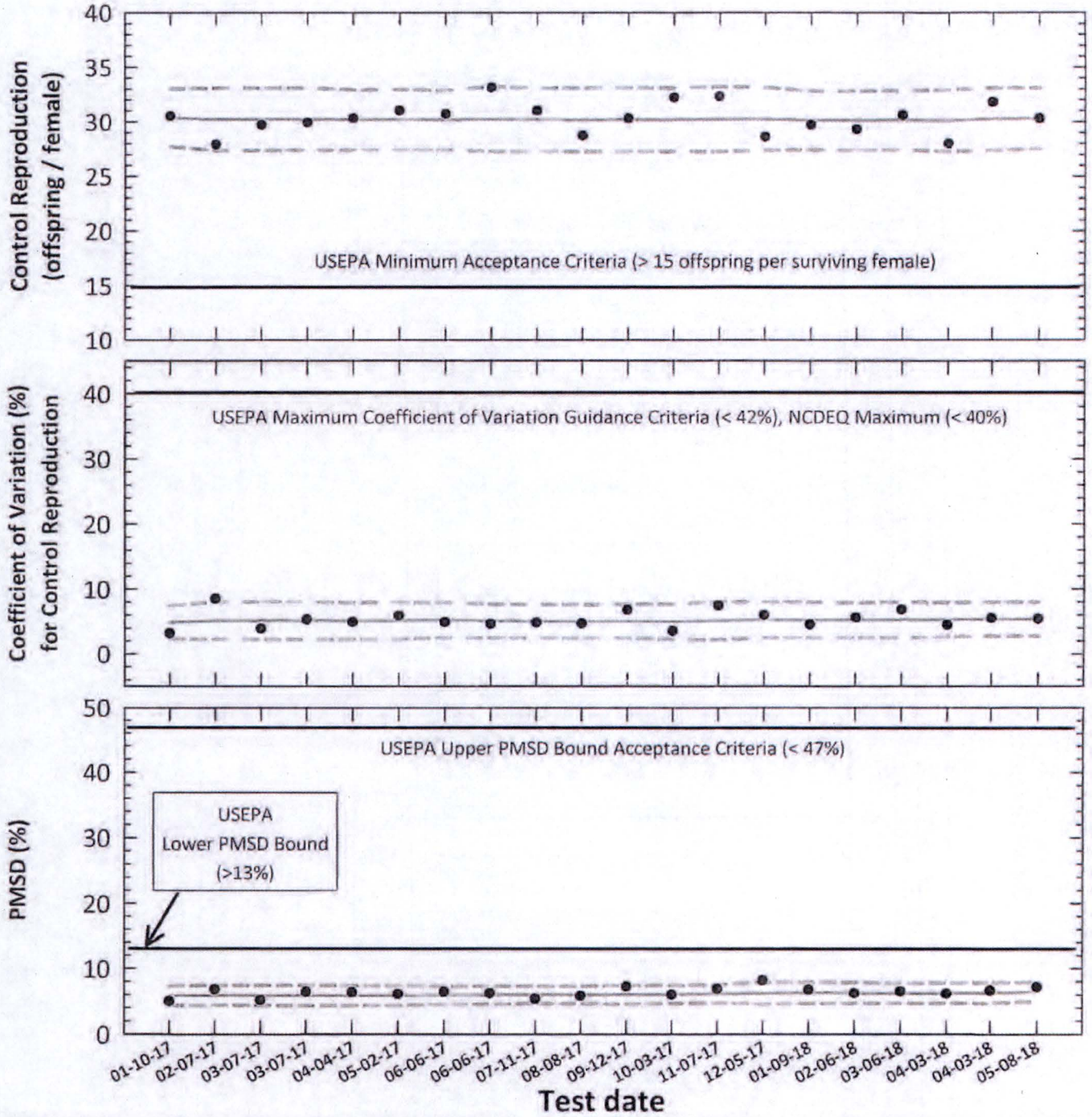
CV = Coefficient of variation.

Entered and Reviewed by JH Sumner

Ceriodaphnia dubia

Chronic Reference Toxicant Testing, Test Acceptability Criteria

Organism Source: In-house Culture



- **Control Reproduction, Coefficient of Variation (CV) or Percent Minimum Significant Difference (PMSD)**
  PMSD is the percent minimum significant difference between the control and treatment that can be declared statistically significant. The lower PMSD bound represents a practical limit to the sensitivity of the test method and is not a minimum acceptance criteria.

Central Tendency (mean Control Reproduction, CV or PMSD)

95% Confidence Interval (mean Control Reproduction, CV or PMSD ± 2 Standard Deviations)

Independent Review by Kelley E. Keenan

Entered and Reviewed by Jim Sumner

Ceriodaphnia dubia

Chronic Reference Toxicant Testing, Test Acceptability Criteria

Source: In-house Culture

Test number	Test date	ToxCal Determination					Control Reproduction (offspring/female)			Control Reproduction CV (%)			Test PMSD (%)		
		Control Survival (%)	Control Reproduction Mean (offspring/female)	Control Reproduction CV (%)	Test MSD	Test PMSD (%)	CT	95% Confidence Interval CT - 2S	95% Confidence Interval CT + 2S	CT	95% Confidence Interval CT - 2S	95% Confidence Interval CT + 2S	CT	95% Confidence Interval CT - 2S	95% Confidence Interval CT + 2S
1	01-10-17	100	30.5	3.2	1.524	5.0	30.4	27.7	33.0	4.8	2.3	7.4	5.8	4.3	7.3
2	02-07-17	100	27.9	8.5	1.895	6.8	30.2	27.4	33.0	5.1	2.2	8.0	5.9	4.4	7.4
3	03-07-17	100	29.7	3.9	1.530	5.2	30.2	27.4	33.0	5.1	2.2	8.0	5.8	4.3	7.4
4	03-07-17	100	29.9	5.3	1.918	6.4	30.2	27.4	33.1	5.1	2.2	8.0	5.9	4.3	7.4
5	04-04-17	100	30.3	4.9	1.924	6.3	30.2	27.4	32.9	5.1	2.2	7.9	5.8	4.3	7.3
6	05-02-17	100	31.0	5.9	1.894	6.1	30.2	27.4	32.9	5.1	2.3	8.0	5.8	4.3	7.3
7	06-06-17	100	30.7	4.9	1.968	6.4	30.2	27.4	32.9	5.2	2.5	7.9	5.9	4.6	7.3
8	06-06-17	100	33.1	4.6	2.050	6.2	30.2	27.3	33.2	5.1	2.5	7.7	5.9	4.6	7.3
9	07-11-17	100	31.0	4.8	1.661	5.4	30.2	27.3	33.1	5.0	2.5	7.6	5.9	4.6	7.3
10	08-08-17	100	28.7	4.7	1.668	5.8	30.1	27.2	33.1	5.0	2.4	7.5	5.9	4.6	7.3
11	09-12-17	100	30.3	6.8	2.191	7.2	30.2	27.3	33.1	5.0	2.4	7.7	6.0	4.5	7.4
12	10-03-17	100	32.2	3.5	1.926	6.0	30.2	27.3	33.1	4.9	2.2	7.7	6.0	4.6	7.4
13	11-07-17	100	32.3	7.4	2.215	6.9	30.2	27.3	33.2	5.1	2.1	8.0	6.1	4.7	7.5
14	12-05-17	100	28.6	6.0	2.344	8.2	30.3	27.4	33.1	5.2	2.5	8.0	6.2	4.5	7.9
15	01-09-18	100	29.7	4.5	2.015	6.8	30.1	27.4	32.8	5.2	2.5	8.0	6.3	4.7	7.9
16	02-06-18	100	29.3	5.6	1.824	6.2	30.1	27.4	32.8	5.2	2.5	7.9	6.2	4.7	7.8
17	03-06-18	100	30.6	6.8	1.995	6.5	30.1	27.4	32.8	5.2	2.4	8.0	6.2	4.7	7.8
18	04-03-18	100	28.0	4.5	1.729	6.2	30.1	27.3	32.9	5.3	2.6	7.9	6.2	4.7	7.8
19	04-03-18	100	31.8	5.5	2.108	6.6	30.2	27.4	33.1	5.3	2.7	7.9	6.3	4.7	7.8
20	05-08-18	100	30.3	5.4	2.172	7.2	30.3	27.5	33.1	5.3	2.7	8.0	6.4	4.9	7.8

Note: Control Survival = USEPA minimum test acceptability criteria ≥ 80% survival.

Control Mean Reproduction = USEPA minimum test acceptability criteria ≥ 15 offspring/surviving female.

CV = Coefficient of variation for control reproduction.

USEPA maximum CV guidance criteria (90th percentile) < 42%. NCDEQ maximum CV acceptance criteria < 40%.

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.

Lower PMSD bound determined by USEPA (10th percentile) > 13%.

The lower PMSD bound represents a practical limit to the sensitivity of the test method and is not a minimum acceptance criteria.

Upper PMSD bound acceptance criteria determined by USEPA (90th percentile) <47%.

CT = Central tendency of the reproduction, CV or PMSD values.

S = Standard deviation of the reproduction, CV or PMSD values.

ETS Environmental Testing Solutions, Inc.

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

CdNaClCR #: 215

Dilution preparation information:						*Comments:*
NaCl Stock INSS number:		INSS 1677				
Stock preparation:		100 g NaCl/L: Dissolve 50 g NaCl in 500 mL Milli-Q 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 source information:											*Test information:*	
Organism age:	< 24-hours old										Randomizing template color:	RED
Date and times organisms were born between:	05-01-18 0530-TO 0900										Incubator number and shelf location:	2 B7
Culture board:	05-01-18 A											
Replicate number:	1	2	3	4	5	6	7	8	9	10	YWT batch:	04-16-18
Culture board cup number:	4	7	8	9	13	15	17	18	22	23		
Transfer vessel information:	pH = 7.90 S.U. Temperature = 24.9°C										*Selenastrum* batch:	04-30-18
Average transfer volume (mL):	< 0.25 mL											

Daily renewal information:

Day	Date	Test initiation and feeding, renewal and feeding, or termination time	MHSW batch used	Analyst
0	05-08-18	0911	05-03-18 A	JK
1	05-09-18	0857	05-03-18 B	JH
2	05-10-18	0855	05-03-18 B	JK
3	05-11-18	0852	05-03-18 B	JH
4	05-12-18	0928	05-07-18	JK
5	05-13-18	0923	05-07-18	JH
6	05-14-18	0852	05-07-18	JH
7	05-15-18	0812		JH

Control information:		Acceptance criteria	*Summary of test endpoints:*	
% of Male Adults:	0%	≤ 20%	7-day LC_{50}	> 1400
% Adults having 3rd Broods:	100%	≥ 80%	NOEC	1000
% Mortality:	0%	≤ 20%	LOEC	1200
Mean Offspring/Female:	30.3	≥ 15.0 offspring/female	ChV	1095.5
% CV:	5.4%	< 40.0 %	IC_{25}	1082.2

Independent Review by Kelley E. Keenan

Species: *Ceriodaphnia dubia* CdNaClCR #: 215

CONTROL *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	3	3	3	4	4	3	4	4	5	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	13	12	10	12	10	10	11	11	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	15	17	13	16	16	18	15	17	14	15
Total young produced		29	33	28	30	32	31	29	32	30	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), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	30.3

600 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	3	5	4	4	3	4	3	5	3	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	12	11	12	12	13	10	13	10	10	11
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	16	18	15	15	17	19	16	17	17	15
Total young produced		31	34	31	31	33	33	32	32	30	29
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	31.6
% Reduction from Control:	-4.37

Independent Review by Kelley E. Keenan: K

Species: *Ceriodaphnia dubia* CdNaClCR #: 215

800 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	5	3	4	5	5	3	3	3	5	4
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	10	10	10	12	11	12	13	10	12
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	18	19	16	18	15	18	14	16	16	16
Total young produced		34	32	30	33	32	32	29	32	31	32
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	31.7
% Reduction from Control:	-4.67

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	5	3	5	4	4	4	3	3	3
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	11	10	10	11	10	12	12	12	10	10
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	18	14	14	17	16	17	15	17	19	17
Total young produced		33	29	27	33	30	33	31	32	32	30
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	31.0
% Reduction from Control:	-2.37

Species: *Ceriodaphnia dubia* CdNaClCR #: 215

1200 mg NaCl/L *Survival and Reproduction Data*

Day		Replicate number									
		1	2	3	4	5	6	7	8	9	10
1	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
2	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
3	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
4	Young produced	3	3	2	4	2	2	1	3	3	1
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	8	7	5	8	6	10	7	9	5	6
	Adult mortality	L	L	L	L	L	L	L	L	L	L
6	Young produced	0	0	0	0	0	0	0	0	0	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	8	0	6	0	0	0	7	0	9	0
Total young produced		19	10	13	12	8	12	15	12	17	7
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	12.5
% Reduction from Control:	58.7%

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	1	2	1	1	3	1	2	1	1	1
	Adult mortality	L	L	L	L	L	L	L	L	L	L
5	Young produced	0	0	2	0	0	0	0	4	2	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	0
	Adult mortality	L	L	L	L	L	L	L	L	L	L
7	Young produced	0	2	0	0	0	0	0	0	0	0
Total young produced		1	4	3	1	3	1	2	5	3	1
Final Adult Mortality		L	L	L	L	L	L	L	L	L	L

Note: Adult mortality (L = live, D = dead), SB = split brood (single brood split between two days), CO = carry over (offspring carried over with adult during transfer).

Concentration:	
% Mortality:	0%
Mean Offspring/Female:	2.4
% Reduction from Control:	92.1%

Independent Review by Kelley E. Keenan

Independent Review by Kelley E. Keenan

ETS

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	3	3	3	4	4	3	4	4	5	3	36
5	11	13	12	10	12	10	10	11	11	11	111
6	0	0	0	0	0	0	0	0	0	0	0
7	15	17	13	16	16	18	15	17	14	15	156
Total	29	33	28	30	32	31	29	32	30	29	303

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	5	3	5	4	4	4	3	3	3	38
5	11	10	10	11	10	12	12	12	10	10	108
6	0	0	0	0	0	0	0	0	0	0	0
7	18	14	14	17	16	17	15	17	19	17	164
Total	33	29	27	33	30	33	31	32	32	30	310

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	3	5	4	4	3	4	3	5	3	3	37
5	12	11	12	12	13	10	13	10	10	11	114
6	0	0	0	0	0	0	0	0	0	0	0
7	16	18	15	15	17	19	16	17	17	15	165
Total	31	34	31	31	33	33	32	32	30	29	316

1200 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	3	3	2	4	2	2	1	3	3	1	24
5	8	7	5	8	6	10	7	9	5	6	71
6	0	0	0	0	0	0	0	0	0	0	0
7	8	0	6	0	0	0	7	0	9	0	30
Total	19	10	13	12	8	12	15	12	17	7	125

800 mg NaCl/L

Day	Replicate number										Total
	1	2	3	4	5	6	7	8	9	10	
1	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0
4	5	3	4	5	5	3	3	3	5	4	40
5	11	10	10	10	12	11	12	13	10	12	111
6	0	0	0	0	0	0	0	0	0	0	0
7	18	19	16	18	15	18	14	16	16	16	166
Total	34	32	30	33	32	32	29	32	31	32	317

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	1	2	1	1	3	1	2	1	1	1	14
5	0	0	2	0	0	0	0	4	2	0	8
6	0	0	0	0	0	0	0	0	0	0	0
7	0	2	0	0	0	0	0	0	0	0	2
Total	1	4	3	1	3	1	2	5	3	1	24

Entered and Reviewed by Jim Sumner

Environmental Testing Solutions, Inc.

Ceriodaphnia dubia Chronic Reference Toxicant Test
EPA-821-R-02-013, Method 1002.0

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

Test number: CdNaClCR #215

Test dates: May 08-15, 2018

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	29	33	28	30	32	31	29	32	30	29	100	30.3	5.4	Not applicable
600	31	34	31	31	33	33	32	32	30	29	100	31.6	4.8	-4.3
800	34	32	30	33	32	32	29	32	31	32	100	31.7	4.5	-4.6
1000	33	29	27	33	30	33	31	32	32	30	100	31.0	6.5	-2.3
1200	19	10	13	12	8	12	15	12	17	7	100	12.5	30.0	58.7
1400	1	4	3	1	3	1	2	5	3	1	100	2.4	59.6	92.1

Dunnett's MSD value: 2.172

PMSD: 7.2

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.

Lower PMSD bound determined by USEPA (10th percentile) = 13%.

Upper PMSD bound determined by USEPA (90th percentile) = 47%.

Lower and upper PMSD bounds were determined from the 10th and 90th percentile, respectively, of PMSD data from EPA's WET Interlaboratory Variability Study (USEPA, 2001a; USEPA, 2001b). The lower PMSD bound represents a practical limit to the sensitivity of the test method and is not a minimum acceptance criteria.

USEPA. 2001a, 2001b. Final Report: Interlaboratory Variability Study of EPA Short-term Chronic and Acute Whole Effluent Toxicity Test Methods, Volumes 1 and 2-Appendix. EPA-821-B-01-004 and EPA-821-B-01-005. US Environmental Protection Agency, Cincinnati, OH.

Statistical Analyses

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	5/8/2018	Test ID:	CdNaClCR	Sample ID:	REF-Ref Toxicant
End Date:	5/15/2018	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	29.000	33.000	28.000	30.000	32.000	31.000	29.000	32.000	30.000	29.000
600	31.000	34.000	31.000	31.000	33.000	33.000	32.000	32.000	30.000	29.000
800	34.000	32.000	30.000	33.000	32.000	32.000	29.000	32.000	31.000	32.000
1000	33.000	29.000	27.000	33.000	30.000	33.000	31.000	32.000	32.000	30.000
1200	19.000	10.000	13.000	12.000	8.000	12.000	15.000	12.000	17.000	7.000
1400	1.000	4.000	3.000	1.000	3.000	1.000	2.000	5.000	3.000	1.000

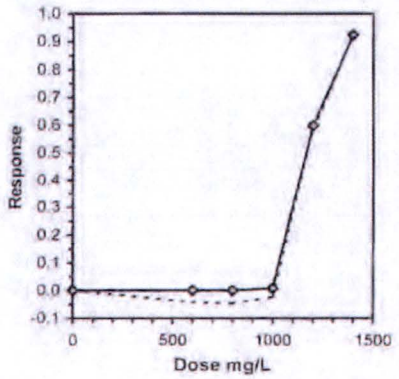
			Transform: Untransformed					Rank	1-Tailed	Isotonic	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	Sum	Critical	Mean	N-Mean
D-Control	30.300	1.0000	30.300	28.000	33.000	5.401	10			31.200	1.0000
600	31.600	1.0429	31.600	29.000	34.000	4.764	10	127.00	75.00	31.200	1.0000
800	31.700	1.0462	31.700	29.000	34.000	4.474	10	128.50	75.00	31.200	1.0000
1000	31.000	1.0231	31.000	27.000	33.000	6.452	10	118.50	75.00	31.000	0.9936
*1200	12.500	0.4125	12.500	7.000	19.000	29.993	10	55.00	75.00	12.500	0.4006
*1400	2.400	0.0792	2.400	1.000	5.000	59.577	10	55.00	75.00	2.400	0.0769

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.61747	1.035	0.14429	1.55997
Bartlett's Test indicates unequal variances (p = 8.62E-03)	15.446	15.0863		

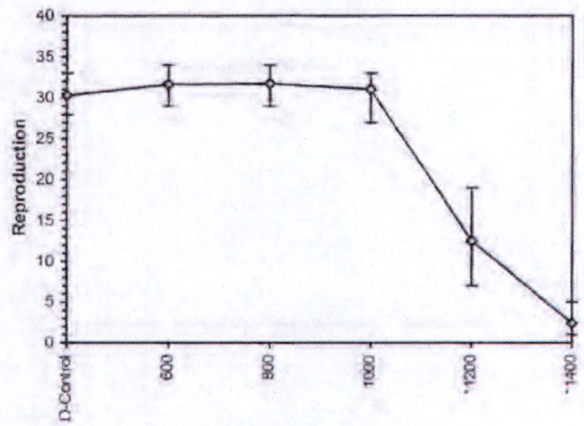
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	1000	1200	1095.45	
Treatments vs D-Control				

Linear Interpolation (200 Resamples)

Point	mg/L	SD	95% CL		Skew
IC05	1014.7	7.02626	1001.38	1018.62	-3.9420
IC10	1031.57	4.98651	1018.85	1037.46	-0.7722
IC15	1048.43	5.27534	1036.35	1056.35	-0.3960
IC20	1065.3	5.75815	1053.84	1075.32	-0.0539
IC25	1082.16	6.39113	1070.95	1094.63	0.1933
IC40	1132.76	8.83718	1118.91	1152.13	0.4884
IC50	1166.49	10.7131	1150.17	1191.85	0.5230



Dose-Response Plot



Independent Review by Kelley E. Keenan

ETS
Environmental Testing Solutions, Inc.

Statistical Analyses

Used for PMSD calculation only.

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date:	5/8/2018	Test ID:	CdNaCICR	Sample ID:	REF-Ref Toxicant
End Date:	5/15/2018	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	29.000	33.000	28.000	30.000	32.000	31.000	29.000	32.000	30.000	29.000
600	31.000	34.000	31.000	31.000	33.000	33.000	32.000	32.000	30.000	29.000
800	34.000	32.000	30.000	33.000	32.000	32.000	29.000	32.000	31.000	32.000
1000	33.000	29.000	27.000	33.000	30.000	33.000	31.000	32.000	32.000	30.000
1200	19.000	10.000	13.000	12.000	8.000	12.000	15.000	12.000	17.000	7.000
1400	1.000	4.000	3.000	1.000	3.000	1.000	2.000	5.000	3.000	1.000

			Transform: Untransformed						1-Tailed	
Conc-mg/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD
D-Control	30.300	1.0000	30.300	28.000	33.000	5.401	10			
600	31.600	1.0429	31.600	29.000	34.000	4.764	10	-1.369	2.287	2.172
800	31.700	1.0462	31.700	29.000	34.000	4.474	10	-1.474	2.287	2.172
1000	31.000	1.0231	31.000	27.000	33.000	6.452	10	-0.737	2.287	2.172
*1200	12.500	0.4125	12.500	7.000	19.000	29.993	10	18.744	2.287	2.172
*1400	2.400	0.0792	2.400	1.000	5.000	59.577	10	29.379	2.287	2.172

Auxiliary Tests					Statistic		Critical		Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)					0.61747		1.035		0.14429	1.55997
Bartlett's Test indicates unequal variances (p = 8.62E-03)					15.446		15.0863			
Hypothesis Test (1-tail, 0.05)	**NOEC**	**LOEC**	**ChV**	**TU**	**MSDu**	**MSDp**	**MSB**	**MSE**	**F-Prob**	**df**
Dunnett's Test	1000	1200	1095.45		2.17155	0.07167	1602.35	4.50926	5.2E-40	5, 54
Treatments vs D-Control										

Entered and Reviewed by Jim Summer

Independent Review by Kelley E. Keenan:

Species: *Ceriodaphnia dubia* CdNaClCR #: 215

Daily Chemistry:

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)					
		0		1		2	
	Analyst	MS	MS	MS	N	N	MS
Concentration	Parameter						
CONTROL	pH (S.U.)	7.94	7.95	7.88	8.01	8.01	8.00
	DO (mg/L)	7.7	7.8	7.8	7.8	7.7	7.8
	Conductivity (µmhos/cm)	317		310		324	
	*Alkalinity (mg $CaCO_3$/L)	59		59		08/16/18	
	*Hardness (mg $CaCO_3$/L)	86		86			
	*Temperature (°C)	24.8	25.0	24.7	25.1	24.8	25.1
600 mg NaCl/L	pH (S.U.)	8.00	7.94	7.91	7.95	8.06	7.96
	DO (mg/L)	7.7	7.8	7.8	7.7	7.6	7.8
	Conductivity (µmhos/cm)	1430		1390		1400	
	*Temperature (°C)	24.9	24.8	24.8	24.8	24.9	25.0
800 mg NaCl/L	pH (S.U.)	8.00	7.92	7.93	7.96	8.04	7.96
	DO (mg/L)	7.8	7.8	7.9	7.8	7.6	7.8
	Conductivity (µmhos/cm)	1800		1810		1790	
	*Temperature (°C)	24.8	24.8	24.8	24.8	24.9	25.2
1000 mg NaCl/L	pH (S.U.)	7.99	7.93	7.93	7.95	8.04	7.94
	DO (mg/L)	7.9	7.8	7.9	7.8	7.6	7.8
	Conductivity (µmhos/cm)	2180		2160		2160	
	*Temperature (°C)	25.0	25.0	24.8	25.1	24.9	25.2
1200 mg NaCl/L	pH (S.U.)	7.99	7.92	7.93	7.94	8.04	7.94
	DO (mg/L)	8.0	8.0	7.9	7.8	7.7	7.7
	Conductivity (µmhos/cm)	2490		2500		2530	
	*Temperature (°C)	24.9	24.9	24.8	24.9	24.9	25.0
1400 mg NaCl/L	pH (S.U.)	7.98	7.92	7.93	7.95	8.03	7.94
	DO (mg/L)	8.1	8.0	8.0	7.9	7.7	7.7
	Conductivity (µmhos/cm)	2900		2890		2890	
	*Temperature (°C)	24.9	24.9	24.8	25.0	24.9	25.0
		Initial	Final	Initial	Final	Initial	Final

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1. Alkalinity and hardness performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: N

Independent review by Kelley E. Keenan: K

Species: *Ceriodaphnia dubia* CdNaClCR #: 215

		Day (Analyst identified for each day, performed pH, D.O. and conductivity measurements only.)							
		3		4		5		6	
	Analyst	MS	BSL	BSL	JL	JL	JL	JL	MS
Concentration	Parameter								
CONTROL	pH (S.U.)	7.94	7.85	7.95	8.04	8.08	8.01	8.00	7.84
	DO (mg/L)	7.7	8.0	7.6	7.8	7.7	7.9	7.8	7.9
	Conductivity (µmhos/cm)	305		299		306		300	
	*Alkalinity (mg $CaCO_3$/L)	05-16-18 JL		60		05-16-18 JL		05-16-18 JL	
	*Hardness (mg $CaCO_3$/L)			84					
	*Temperature (°C)	24.8	25.1	24.8	25.0	24.7	25.0	24.9	25.2
600 mg NaCl/L	pH (S.U.)	8.00	7.85	7.91	8.01	8.01	7.97	8.02	7.87
	DO (mg/L)	7.7	8.0	7.7	7.8	7.8	7.9	7.9	7.9
	Conductivity (µmhos/cm)	1440		1390		1300		1390	
	*Temperature (°C)	24.9	24.8	24.8	24.9	24.8	25.2	25.0	24.9
800 mg NaCl/L	pH (S.U.)	8.01	7.88	7.91	8.00	8.03	7.97	8.03	7.87
	DO (mg/L)	7.8	8.0	7.8	7.8	7.8	7.9	7.9	7.9
	Conductivity (µmhos/cm)	1850		1800		1760		1830	
	*Temperature (°C)	24.9	24.8	24.9	24.9	24.7	25.0	25.0	25.0
1000 mg NaCl/L	pH (S.U.)	8.01	7.89	7.89	7.98	8.03	7.97	8.04	7.87
	DO (mg/L)	7.8	7.9	7.8	7.8	7.9	7.9	8.0	7.9
	Conductivity (µmhos/cm)	2210		2160		2130		2200	
	*Temperature (°C)	24.8	24.9	24.8	25.1	24.7	25.0	25.0	24.9
1200 mg NaCl/L	pH (S.U.)	8.00	7.91	7.88	7.97	8.03	7.97	8.04	7.87
	DO (mg/L)	8.0	7.9	7.9	7.8	7.9	8.0	8.0	8.1
	Conductivity (µmhos/cm)	2580		2510		2500		2570	
	*Temperature (°C)	24.8	25.1	24.8	24.8	24.7	25.1	25.0	25.1
1400 mg NaCl/L	pH (S.U.)	8.01	8.00	7.87	7.96	8.02	7.96	8.04	7.89
	DO (mg/L)	8.0	7.8	8.0	7.8	7.9	8.0	8.0	8.1
	Conductivity (µmhos/cm)	2970		2850		2800		2960	
	*Temperature (°C)	24.9	24.9	24.8	24.8	24.9	25.1	25.0	24.8
		Initial	Final	Initial	Final	Initial	Final	Initial	Final

*Temperatures performed at the time of test initiation, renewal or termination by the analyst identified in the Daily Renewal Information table located on Page 1. Alkalinity and hardness performed by the analyst identified on the bench sheet specific for each analysis and transcribed to this bench sheet by: JL

Independent Review by Kelley E. Keenum: K