

Jonathan Abelson  
Stantec  
300 N. Lake Avenue  
Pasadena, CA 91101

December 28, 2018

Dear Jon:

I have enclosed a copy of our report “A Toxicity Evaluation of an Ambient Water Sample Collected for the East San Gabriel Valley CIMP” for the sample collected December 6, 2018. The results of this evaluation are summarized below.

**Chronic Effects of East San Gabriel Valley CIMP Ambient Water on *Ceriodaphnia dubia***

There was 90% survival in the ambient water sample, indicating that the sample was not toxic to *C. dubia* survival. The TST analysis of the 100% sample reproduction data shows that the reproduction response in the ambient water sample *was* statistically less than the response at the Lab Control treatment; in this scenario, the 2010 EPA TST document states, “the test result is Fail and the sample is declared toxic”.

If you have any questions regarding the performance and interpretation of this test, please feel free to contact my colleague Stephen Clark or myself at (707) 207-7760.

Sincerely,

Stevi Vasquez  
Project Manager

Cc: Mitch Mysliwec, Larry Walker Associates  
Ed Othmer, Stantec



Pacific EcoRisk is accredited in accordance with NELAP (ORELAP ID 4043). Pacific EcoRisk certifies that the test results reported herein conform to the most current NELAP requirements for parameters for which accreditation is required and available. Any exceptions to NELAP requirements are noted, where applicable, in the body of the report. This report shall not be reproduced, except in full, without the written consent of Pacific EcoRisk. This testing was performed under Lab Order 29693.

# **A Toxicity Evaluation of an Ambient Water Sample Collected for the East San Gabriel Valley CIMP**

Sample collected December 6, 2018

Submitted To:

Stantec  
300 N. Lake Avenue  
Pasadena, CA 91101

Prepared By:

Pacific EcoRisk  
2250 Cordelia Rd.  
Fairfield, CA 94534

**December 2018**



# A Toxicity Evaluation of an Ambient Water Sample Collected for the East San Gabriel Valley CIMP

Sample collected December 6, 2018

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Appendix C	Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the <i>Ceriodaphnia dubia</i>

## 1. INTRODUCTION

Under contract to Stantec, Pacific EcoRisk (PER) performed a chronic toxicity evaluation of an ambient water sample collected from the East San Gabriel Valley (ESGV) watershed. The sample was collected to address compliance requirements in the ESGV CIMP. The chronic toxicity evaluation consists of performing the US EPA short-term chronic 3-brood (6-8 day) survival and reproduction test with the crustacean *Ceriodaphnia dubia*.

This chronic toxicity test was performed on an ambient water sample collected by a Stantec field team on December 6, 2018. This report describes the performance and results of this toxicity test.

## 2. SAMPLE COLLECTION AND HANDLING

On December 6, Stantec staff collected an ambient water sample from one ESGV monitoring station (Table 1). The ambient water sample was collected into an appropriately cleaned sample container. The ambient water sample was transported, on ice and under chain-of-custody, to the PER laboratory facility in Fairfield, CA within 24 hours of collection. Upon receipt at the testing laboratory, an aliquot of the sample was analyzed to determine initial water quality characteristics (Table 2). The remainder of the sample was stored at 0-6°C and used to initiate testing within 36 hours of collection.

The chain-of-custody record for the collection and delivery of the sample is presented in Appendix A.

Table 1. Collection of the ESGV ambient water sample.		
Sample ID Code	Sample Collection Date	Sample Receipt Date
ESGV-23-LOW-4	12/6/18 (0520)	12/7/18 (1125)

Table 2. Initial water quality characteristics of the ESGV ambient water sample.							
Sample ID	Temp. (°C)	pH	D.O. (mg/L)	Alkalinity (mg/L)	Hardness (mg/L)	Conductivity (µS/cm)	Total Ammonia (mg/L N)
ESGV-23-LOW-4	0.8	7.72	11.1	39	32	141	<1.0

### 3. TOXICITY TESTING PROCEDURES

The ESGV ambient water sample for this event was tested for toxicity using the following US EPA short-term chronic toxicity test:

- 3-brood (6-8 day) survival and reproduction test with the crustacean *Ceriodaphnia dubia*.

This testing followed the guidelines established in the US EPA manual “Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, Fourth Edition” (EPA-821-R-02-013).

#### 3.1 Survival and Reproduction Toxicity Testing with *Ceriodaphnia dubia*

The short-term chronic *Ceriodaphnia* test consists of exposing individual females to the ambient water sample for the length of time it takes for the Lab Control treatment females to produce 3 broods (typically 6-8 days), after which effects on survival and reproduction are evaluated. The specific procedures used in this test are described below.

The Lab Water Control medium for this test consisted of modified EPA synthetic moderately hard water, prepared by addition of reagent grade chemicals to Type 1 lab water. The ambient water sample was tested at the 100% concentration only. As an additional QA measure, and in response to the observation of low hardness in the sample (see Table 2), a “Hardness Control” treatment, consisting of Lab Water Control medium adjusted to the hardness of the water sample using Type 1 lab water, was also prepared and tested. Each treatment consisted of 200 mL of test solution to which the alga *Selenastrum capricornutum* and Yeast-Cerophyll®-Trout Food (YCT) had been added as food for the test organisms. “New” water quality characteristics (pH, dissolved oxygen [D.O.], and conductivity) were measured on these food-amended test solutions prior to use in the test.

There were 10 replicates for each test treatment, each replicate consisting of 15 mL of test solution in a 30-mL plastic cup. The test was initiated by allocating one neonate (<24 hours old, and within 8 hours of age) *C. dubia*, obtained from in-house laboratory cultures, into each replicate cup. The test replicate cups were placed into a temperature-controlled room at 25°C, under cool-white fluorescent lighting on a 16L:8D photoperiod.

Each day of the test, fresh test solutions were prepared and characterized as before, and a new set of replicate cups was prepared. The original test replicate cups were examined, with surviving original individual organisms being transferred to the corresponding new cup. The contents of each of the remaining old replicate cups was carefully examined and the number of neonate offspring produced by each original organism was determined, after which the “old” water quality characteristics (pH, D.O., and conductivity) were measured for the old media from one randomly-selected replicate at each treatment.

After it was determined that  $\geq 60\%$  of the *C. dubia* in the Control had produced their third brood of offspring, the test was terminated. The resulting survival and reproduction data were analyzed to evaluate any impairment caused by the ambient water. All statistical analyses were performed using CETIS™ (TidePool Scientific Software, McKinleyville, CA).

### **3.1.1 Reference Toxicant Testing of the *Ceriodaphnia dubia***

The reference toxicant test was performed similarly to the ambient water test, except that test solutions comprised of Lab Water Control medium spiked with NaCl at test concentrations of 500, 1000, 1500, 2000, and 2500 mg/L were used. The resulting test response data were statistically analyzed to determine key concentration-response point estimates. All statistical analyses were performed using CETIS. These response endpoints were then compared to the typical response ranges established by the mean  $\pm$  2 SD of the point estimates generated by the most recent previous reference toxicant tests performed by this lab.

## 4. RESULTS

### 4.1 Effects of ESGV Ambient Water on *Ceriodaphnia dubia*

The results of this test are summarized below in Table 3. There was 90% survival in the ambient water sample, indicating that the sample was not toxic to *C. dubia* survival. The reproduction data were analyzed using the TST analytical method. The reproduction response in the 100% ambient water sample *was* statistically less than the response at the Lab Control treatment; in this scenario, the 2010 EPA TST document states, “the test result is Fail and the sample is declared toxic”. The test data and summary of statistical analyses are presented in Appendix B.

Table 3. Effects of ESGV ambient water on <i>Ceriodaphnia dubia</i> .		
Treatment	Mean % Survival	Mean Reproduction (# neonates /female)
Hardness Control	100	26.4
Lab Water Control	100	26.6
100%	90	23.3
Summary of Statistics		
TST Analysis =		Fail: Sample is toxic

## 5. AQUATIC TOXICITY DATA QUALITY CONTROL

Three QC measures were assessed during the toxicity testing:

- Maintenance of acceptable test conditions;
- Negative control testing; and
- Positive control (reference toxicant) testing.

### 5.1 Maintenance of Acceptable Test Conditions

Due to equipment malfunction, the sample was stored for <24 hours at a temperature exceeding the method recommended sample storage temperature of 0-6°C. Otherwise, all test conditions (e.g., pH, D.O., etc.) were within acceptable limits for this test. All analyses were performed according to laboratory Standard Operating Procedures.

### 5.2 Negative Control Testing

The response at the Lab Control treatment was within acceptable limits.

### 5.3 Positive Control Testing

#### 5.3.1 Reference Toxicant Toxicity to *Ceriodaphnia dubia*

The results of this test are summarized below in Table 4. The survival EC<sub>50</sub> and reproduction IC<sub>50</sub> were both consistent with the typical response ranges established by the reference toxicant test database for this species, indicating that these organisms were responding to toxic stress in a typical and consistent fashion. The test data and summary of statistical analyses for this test are presented in Appendix C.

Table 4. Reference toxicant testing: Effects of NaCl on <i>Ceriodaphnia dubia</i> .		
NaCl Treatment (mg/L)	Mean % Survival	Mean Reproduction (# neonates/female)
Lab Water Control	100	30.3
500	100	29.8
1000	100	<b>27.9*</b>
1500	90	<b>17.1*</b>
2000	80	<b>5.1*</b>
2500	<b>0*</b>	-
Summary of Statistics		
Survival EC <sub>50</sub> or Reproduction IC <sub>50</sub> =	2050 mg/L NaCl	1580 mg/L NaCl
Typical Response Range (mean $\pm$ 2 SD)	1775 - 2230 mg/L NaCl	1355 - 1840 mg/L NaCl

\* The response at this test treatment was significantly less than the Lab Control treatment response at  $p < 0.05$ .



## **6. SUMMARY AND CONCLUSIONS**

The results for the toxicity evaluation of the ambient water sample collected for the East San Gabriel Valley CIMP are summarized below.

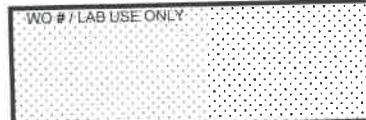
### **Chronic Effects of East San Gabriel Valley CIMP Ambient Water on *Ceriodaphnia dubia***

There was 90% survival in the ambient water sample, indicating that the sample was not toxic to *C. dubia* survival. The TST analysis of the 100% sample reproduction data shows that the reproduction response in the ambient water sample *was* statistically less than the response at the Lab Control treatment; in this scenario, the 2010 EPA TST document states, “the test result is Fail and the sample is declared toxic”.

## **Appendix A**

### **Chain-of-Custody Record for the Collection and Delivery of the East San Gabriel Valley CIMP Ambient Water Sample**

# CHAIN OF CUSTODY RECORD



DATE: 12/06/18  
PAGE: 1 OF 1

LABORATORY CLIENT: <b>Stantec</b>					CLIENT PROJECT NAME / NUMBER: <b>ESGV CIMP 224501167</b>					P.O. NO.: <b>224501167.200.0201</b>														
ADDRESS: <b>300 N Lake Ave, Suite 400</b>					PROJECT CONTACT: <b>Jonathan Abelson</b>					SAMPLER(S): (PRINT) <b>JEA</b>														
CITY: <b>Pasadena</b>			STATE: <b>CA</b>		ZIP: <b>91101</b>			<b>REQUESTED ANALYSES</b> Please check box or fill in blank as needed.																
TEL: <b>(626) 568-6090</b>			E-MAIL: <b><a href="mailto:jonathan.abelson@stantec.com">jonathan.abelson@stantec.com</a></b>																					
TURNAROUND TIME (Rush surcharges may apply to any TAT not "STANDARD"):										(EPA-821-R-02-013) Ceriodaphnia, chronic														
<input type="checkbox"/> SAME DAY <input type="checkbox"/> 24 HR <input type="checkbox"/> 48 HR <input type="checkbox"/> 72 HR <input type="checkbox"/> 5 DAYS <input checked="" type="checkbox"/> STANDARD																								
<input type="checkbox"/> COELT EDF				GLOBAL ID:			LOG CODE:																	
SPECIAL INSTRUCTIONS:																								
Please PDF signed COCs upon completion of sample login to Mitch Mysliwiec at MitchM@lwa.com. Send Lab Reports to jennyb@lwa.com, MitchM@lwa.com, and jonathan.abelson@stantec.com  Send invoices to SAPInvoices@stantec.com and jonathan.abelson@stantec.com																								
LAB USE ONLY	SAMPLE ID	SAMPLING		MATRIX	NO. OF CONT.	Unpreserved	Preserved	Field Filtered																
		DATE	TIME																					
	ESGV-23-LOW-4	12/6/2018	5:20	SW	2	x			x															
Relinquished by: (Signature)						Received by: (Signature/Affiliation)						Date: 12/6/18		Time: 8:00										
Relinquished by: (Signature)						Received by: (Signature/Affiliation)						Date: 12/7/18		Time: 125										
Relinquished by: (Signature)						Received by: (Signature/Affiliation)						Date:		Time:										

## **Appendix B**

### **Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of the East San Gabriel Valley CIMP Ambient Water Sample to *Ceriodaphnia dubia***

# CETIS Summary Report

Report Date: 17 Dec-18 10:56 (p 1 of 1)

Test Code: ESGV\_1218CD\_C1 | 09-9726-5971

Ceriodaphnia Survival and Reproduction Test						Pacific EcoRisk					
Batch ID:	07-5081-1716	Test Type:	Reproduction-Survival (7d)		Analyst:	Ashleigh Findley					
Start Date:	07 Dec-18 14:46	Protocol:	EPA-821-R-02-013 (2002)		Diluent:	Not Applicable					
Ending Date:	13 Dec-18 14:55	Species:	Ceriodaphnia dubia		Brine:	Not Applicable					
Duration:	6d 0h	Source:	In-House Culture		Age:	1					
Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project					
ESGV_23CD_C1	04-8599-6663	07 Dec-18 14:46	07 Dec-18 14:46	n/a (25 °C)	Stantec	29693					
ESGV_23CD_HAR	20-7333-8489	07 Dec-18 14:46	07 Dec-18 14:46	n/a (24.7 °C)							
ESGV-23-LOW-4	03-3375-4819	06 Dec-18 05:20	07 Dec-18 11:25	33h (0.8 °C)							
Sample Code	Material Type	Sample Source		Station Location		Lat/Long					
ESGV_23CD_C1	Lab Control	East San Gabriel Valley CIMP		LABQA							
ESGV_23CD_HAR	Lab Control	East San Gabriel Valley CIMP									
ESGV-23-LOW-4	Ambient Sample	East San Gabriel Valley CIMP		LOW-4							
Single Comparison Summary											
Analysis ID	Endpoint	Comparison Method			P-Value	Comparison Result					
13-6729-5428	Reproduction	TST-Welch's t Test			1.5E-04	ESGV_23CD_HARD passed reproduction					
16-2914-0443	Reproduction	TST-Welch's t Test			0.2033	ESGV-23-LOW-4 failed reproduction					
Reproduction Summary											
Sample	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
ESGV_23CD_C1	LW	10	26.6	24.3	28.9	21	32	1.02	3.24	12.18%	0.00%
ESGV_23CD_HAR	HC	10	26.4	23.8	29	22	32	1.15	3.63	13.74%	0.75%
ESGV-23-LOW-4		10	23.3	14.8	31.8	3	35	3.77	11.9	51.19%	12.41%
Reproduction Detail											
Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
ESGV_23CD_C1	LW	28	31	32	27	24	27	26	21	25	25
ESGV_23CD_HAR	HC	22	26	32	31	22	24	29	28	23	27
ESGV-23-LOW-4		16	34	35	31	18	31	6	34	3	25

# CETIS Analytical Report

Report Date: 17 Dec-18 10:56 (p 1 of 2)  
Test Code: ESGV\_1218CD\_C1 | 09-9726-5971

## Ceriodaphnia Survival and Reproduction Test Pacific EcoRisk

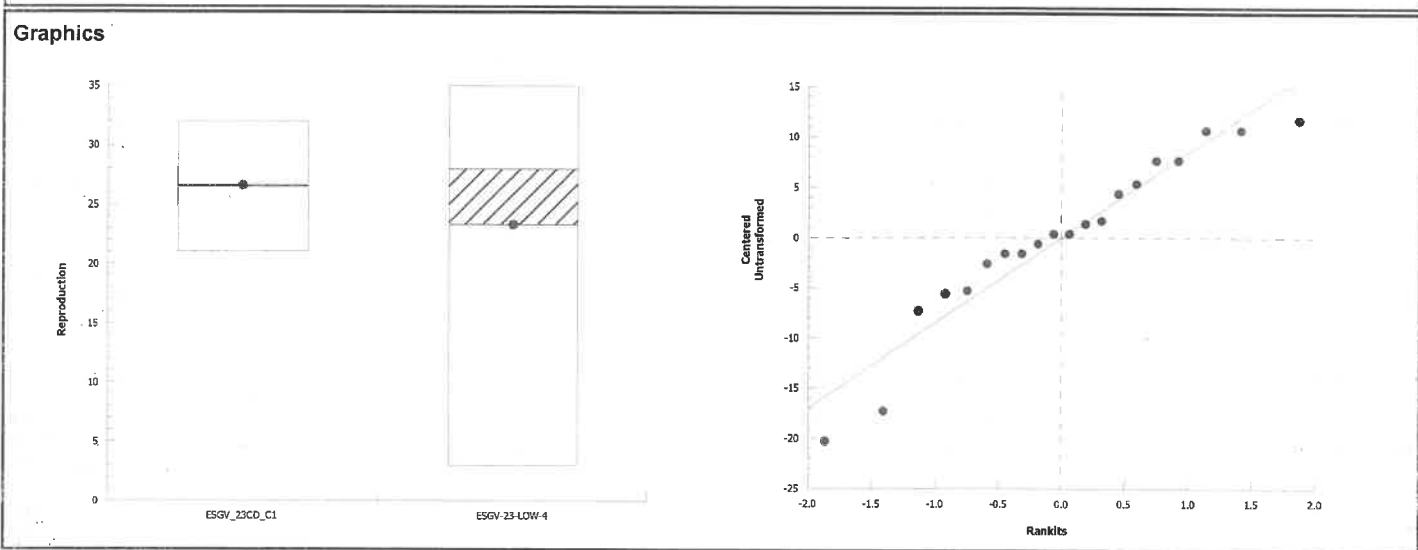
Analysis ID: 16-2914-0443	Endpoint: Reproduction	CETIS Version: CETISv1.9.2
Analyzed: 17 Dec-18 10:56	Analysis: Parametric Bioequivalence-Two Sample	Official Results: Yes

TST-Welch's t Test								
Sample I	vs	Sample II	Test Stat	Critical	DF	P-Type	P-Value	Decision( $\alpha$ :20%)
Lab Water Control		ESGV-23-LOW-4	0.87	0.883	9	CDF	0.2033	Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision( $\alpha$ :5%)
Between	54.45	54.45	1	0.713	0.4095	Non-Significant Effect
Error	1374.5	76.3611	18			
Total	1428.95		19			

Distributional Tests					
Attribute	Test	Test Stat	Critical	P-Value	Decision( $\alpha$ :1%)
Variances	Variance Ratio F Test	13.6	6.54	6.3E-04	Unequal Variances
Distribution	Shapiro-Wilk W Normality Test	0.928	0.866	0.1426	Normal Distribution

Reproduction Summary											
Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
ESGV_23CD_C1	LW	10	26.6	24.3	28.9	26.5	21	32	1.02	12.18%	0.00%
ESGV-23-LOW-4		10	23.3	14.8	31.8	28	3	35	3.77	51.19%	12.41%



Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test DataClient: Stantec: East San Gabriel Valley CIMPMaterial: ESGV-23-LOW-4Test Date: 12/7/18Project #: 29693Test ID: 81055Randomization: 10.4.1Control Water: Modified EPAMH

	Day	pH		D.O.		Cond. (µS/cm)	Temp (°C)	Survival / Reproduction										SIGN-OFF		
		New	Old	New	Old			A	B	C	D	E	F	G	H	I	J	Date:	New WQ:	Test Init.:
Lab Water Control	0	7.70		8.1		367	25.0	0	0	0	0	0	0	0	0	0	0	12/7/18	SR	TF
	1	7.71	7.56	8.10	6.3	371	25.3	0	0	0	0	0	0	0	0	0	0	12/8/18	SMC	Time: 1446
	2	7.64	7.95	8.14	6.6	370	25.4	0	0	0	0	0	0	0	0	0	0	12/8/18	SMC	Counts: SMC
	3	7.84	7.78	9.0	7.7	366	24.7	0	0	0	0	0	4	4	3	4	0	12/10/18	Old WQ: PM	Time: 1401
	4	7.82	7.83	9.2	6.8	365	24.4	5	5	5	5	3	0	9	0	0	4	12/11/18	Old WQ: PM	Counts: TK
	5	7.86	7.96	9.2	8.0	373	24.4	9	11	12	10	9	10	0	5	9	0	12/11/18	Old WQ: PM	Time: 1245
	6	-	7.43	-	7.4	399	24.3	14	15	15	12	12	13	13	12	13		12/13/18	Old WQ: PM	Counts: TK
	7																			Time: 1328
	8																			Time: 1302
Total=								28	31	32	27	24	27	26	21	25	25	Mean Neonates/Female = 26.6		
	Day	pH		D.O.		Cond. (µS/cm)	Temp (°C)	Survival / Reproduction										SAMPLE ID		
		New	Old	New	Old			A	B	C	D	E	F	G	H	I	J			
100%	0	7.75		9.8		143	24.6	0	0	0	0	0	0	0	0	0	0	51570		
	1	7.70	7.55	10.6	6.4	139	24.6	0	0	0	0	0	0	0	0	0	0	51570		
	2	7.62	7.83	10.3	5.5	144	24.8	0	0	0	0	0	0	0	0	0	0	51570		
	3	7.66	7.71	9.7	7.8	149	24.2	0	0	0	0	0	4	4	0	0	0	51570		
	4	7.78	7.73	10.2	6.9	149	24.4	5	5	6	6	1	0	0	4	3	4	51570		
	5	7.70	8.00	10.9	7.9	154	24.3	11	11	10	11	7	10	0	9	0	6	51570		
	6	-	7.38	-	7.3	164	25.2	14	13	16	15	10	17	26	21	X/0	15	-		
	7																			
	8																			
Total=								16	34	35	31	18	31	6	34	X/3	25	Mean Neonates/Female = 23.3		

# CETIS Analytical Report

Report Date: 17 Dec-18 10:56 (p 2 of 2)

Test Code: ESGV\_1218CD\_C1 | 09-9726-5971

## Ceriodaphnia Survival and Reproduction Test Pacific EcoRisk

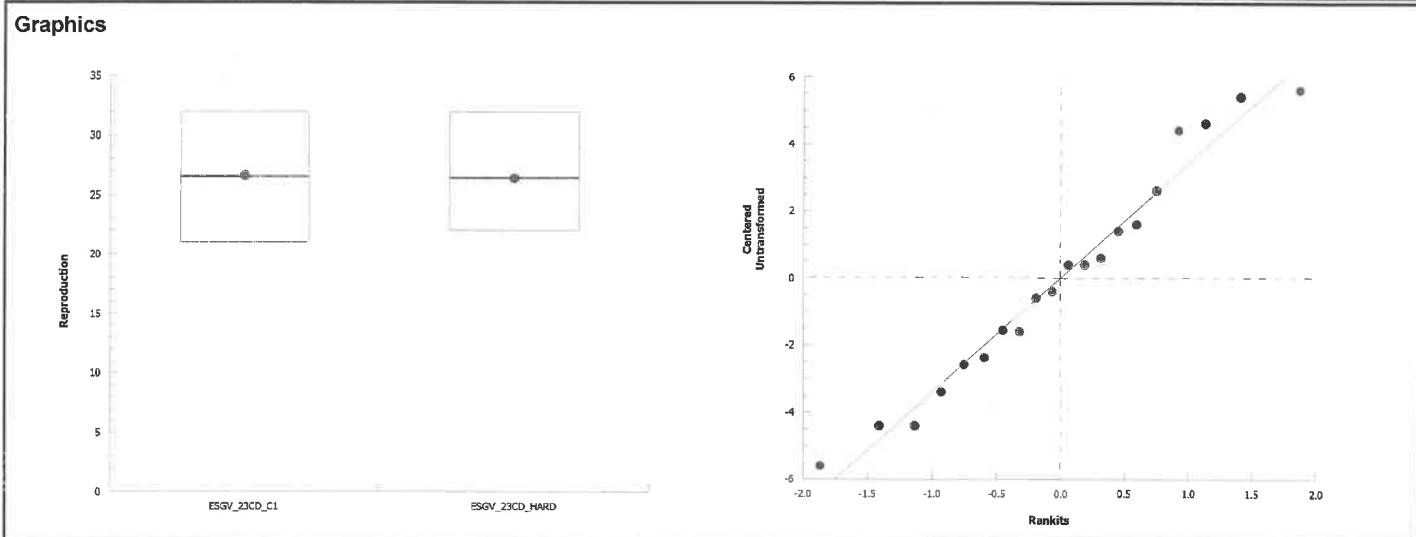
Analysis ID: 13-6729-5428	Endpoint: Reproduction	CETIS Version: CETISv1.9.2
Analyzed: 17 Dec-18 10:56	Analysis: Parametric Bioequivalence-Two Sample	Official Results: Yes

TST-Welch's t Test								
Sample I	vs	Sample II	Test Stat	Critical	DF	P-Type	P-Value	Decision( $\alpha$ :20%)
Lab Water Control		Hardness Control*	4.67	0.866	15	CDF	1.5E-04	Non-Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision( $\alpha$ :5%)
Between	0.2	0.2	1	0.0169	0.8980	Non-Significant Effect
Error	212.8	11.8222	18			
Total	213		19			

Distributional Tests					
Attribute	Test	Test Stat	Critical	P-Value	Decision( $\alpha$ :1%)
Variances	Variance Ratio F Test	1.25	6.54	0.7413	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.962	0.866	0.5811	Normal Distribution

Reproduction Summary											
Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
ESGV_23CD_C1	LW	10	26.6	24.3	28.9	26.5	21	32	1.02	12.18%	0.00%
ESGV_23CD_HAR	HC	10	26.4	23.8	29	26.5	22	32	1.15	13.74%	0.75%





Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: Stantec: East San Gabriel Valley CIMP Material: Hardness Control Test Date: 12/7/18  
 Project #: 29693 Test ID: 81055 Randomization: 10.4.1 Control Water: Mod EPAMH + DI @ hardness 32

	Day	pH		D.O.		Cond. (µS/cm)	Temp (°C)	Survival / Reproduction										SIGN-OFF		
		New	Old	New	Old			A	B	C	D	E	F	G	H	I	J			
Hardness Control	0	7.54		8.7		135	24.7	0	0	0	0	0	0	0	0	0	0	Date: 12/7/18 New WQ: RB Sol'n Prep: SR	Test Init.: TF	Time: 1446
	1	7.54	7.41	9.9	6.9	132	24.7	0	0	0	0	0	0	0	0	0	0	Date: 12/8/18 New WQ: BM Sol'n Prep: SMC	Old WQ: BM	Counts: SMC Time: 1401
	2	7.53	7.65	9.8	5.2	132	24.9	0	0	0	0	0	0	0	0	0	0	Date: 12/9/18 New WQ: DM Sol'n Prep: TK	Old WQ: BM	Counts: TK Time: 1245
	3	7.57	7.57	9.6	7.6	133	24.0	0	0	0	0	0	5	4	0	0	0	Date: 12/10/18 New WQ: GR Sol'n Prep: TK	Old WQ: SL	Counts: SL Time: 1328
	4	7.72	7.56	10.6	5.7	133	24.8	4	5	5	5	5	0	0	4	4	4	Date: 12/11/18 New WQ: SL Sol'n Prep: GR	Old WQ: TF	Counts: GR Time: 1302
	5	7.62	7.97	11.7	7.3	141	24.6	7	8	11	11	7	8	9	8	10	9	Date: 12/12/18 New WQ: TA Sol'n Prep: K6	Old WQ: WB	Counts: K6 Time: 1320
	6	—	7.25	—	7.3	142	25.0	11	13	16	15	10	11	16	16	9	14	Date: 12/13/18 New WQ: — Sol'n Prep: —	Old WQ: TA	Counts: GR Time: 1455
	7																	Date: New WQ: — Sol'n Prep: Old WQ: —	Counts: — Time: —	
	8																	Date: Old WQ: — Time: —		
Total=							22	26	32	31	22	24	29	28	23	27	Mean Neonates/Female = 26.4			

## **Appendix C**

### **Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Ceriodaphnia dubia***

# CETIS Summary Report

Report Date: 20 Dec-18 13:01 (p 1 of 2)

Test Code: 81022 | 08-4916-6117

Ceriodaphnia Survival and Reproduction Test							Pacific EcoRisk				
Batch ID:	16-3128-6289		Test Type:	Reproduction-Survival (7d)			Analyst:	Ashleigh Findley			
Start Date:	04 Dec-18 15:51		Protocol:	EPA-821-R-02-013 (2002)			Diluent:	Laboratory Water			
Ending Date:	11 Dec-18 13:40		Species:	Ceriodaphnia dubia			Brine:	Not Applicable			
Duration:	6d 22h		Source:	In-House Culture			Age:	1			
Sample ID:	03-0731-4373		Code:	NaCl			Client:	Reference Toxicant			
Sample Date:	04 Dec-18 15:51		Material:	Sodium chloride			Project:	29680			
Receipt Date:	04 Dec-18 15:51		Source:	Reference Toxicant							
Sample Age:	n/a (24.5 °C)		Station:	In House							
Multiple Comparison Summary											
Analysis ID	Endpoint		Comparison Method			NOEL	LOEL	TOEL	TU	PMSD ✓	
17-6091-0067	Reproduction		Steel Many-One Rank Sum Test			500	1000	707.1		13.1%	
14-3767-3457	Survival		Fisher Exact/Bonferroni-Holm Test			2000	> 2000	n/a		n/a	
Point Estimate Summary											
Analysis ID	Endpoint		Point Estimate Method			Level	mg/L	95% LCL	95% UCL	TU	✓
16-9668-1931	Reproduction		Linear Interpolation (ICPIN)	IC5	767	457	1020				
				IC10	1030	877	1080				
				IC15	1100	1050	1170				
				IC20	1170	1110	1280				
				IC25	1240	1170	1380				
				IC40	1450	1330	1590				
				IC50	1580	1440	1690				
20-2985-9813	Survival		Regression: Log-Normal (Probit)	EC5	1560	1020	1780				
				EC10	1660	1180	1860				
				EC15	1730	1290	1920				
				EC20	1790	1380	1970				
				EC25	1830	1470	2020				
				EC40	1970	1690	2170				
				EC50	2050	1810	2300				
Reproduction Summary											
Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	LW	10	30.3	28.8	31.8	27	33	0.651	2.06	6.79%	0.00%
500		10	29.8	28.9	30.7	28	32	0.389	1.23	4.13%	1.65%
1000		10	27.9	26.8	29	26	30	0.482	1.52	5.46%	7.92%
1500		10	17.1	12.1	22.1	3	26	2.19	6.94	40.56%	43.56%
2000		10	5.1	1.67	8.53	0	14	1.52	4.79	94.01%	83.17%
2500		10	0	0	0	0	0	0	0		100.00%
Survival Summary											
Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	LW	10	1.000	1.000	1.000	1.000	1.000	0.000	0.000	0.00%	0.00%
500		10	1.000	1.000	1.000	1.000	1.000	0.000	0.000	0.00%	0.00%
1000		10	1.000	1.000	1.000	1.000	1.000	0.000	0.000	0.00%	0.00%
1500		10	0.900	0.674	1.000	0.000	1.000	0.100	0.316	35.14%	10.00%
2000		10	0.800	0.498	1.000	0.000	1.000	0.133	0.422	52.70%	20.00%
2500		10	0.000	0.000	0.000	0.000	0.000	0.000	0.000		100.00%

# CETIS Summary Report

Report Date: 20 Dec-18 13:01 (p 2 of 2)  
 Test Code: 81022 | 08-4916-6117

Ceriodaphnia Survival and Reproduction Test											Pacific EcoRisk
Reproduction Detail											
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	LW	33	32	29	27	30	31	29	31	28	33
500		30	30	29	32	29	31	29	31	28	29
1000		28	29	27	27	26	29	30	26	27	30
1500		9	22	22	21	26	14	21	16	17	3
2000		9	0	5	1	3	5	0	11	14	3
2500		0	0	0	0	0	0	0	0	0	0
Survival Detail											
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	LW	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
500		1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
1000		1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
1500		1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	0.000
2000		1.000	1.000	1.000	0.000	1.000	1.000	0.000	1.000	1.000	1.000
2500		0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
Survival Binomials											
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	LW	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
500		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
1000		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
1500		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	0/1
2000		1/1	1/1	1/1	0/1	1/1	1/1	0/1	1/1	1/1	1/1
2500		0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1

## Ceriodaphnia Survival and Reproduction Test

Pacific EcoRisk

Test Type: Reproduction-Survival (7d)

Organism: Ceriodaphnia dubia (Water Flea)

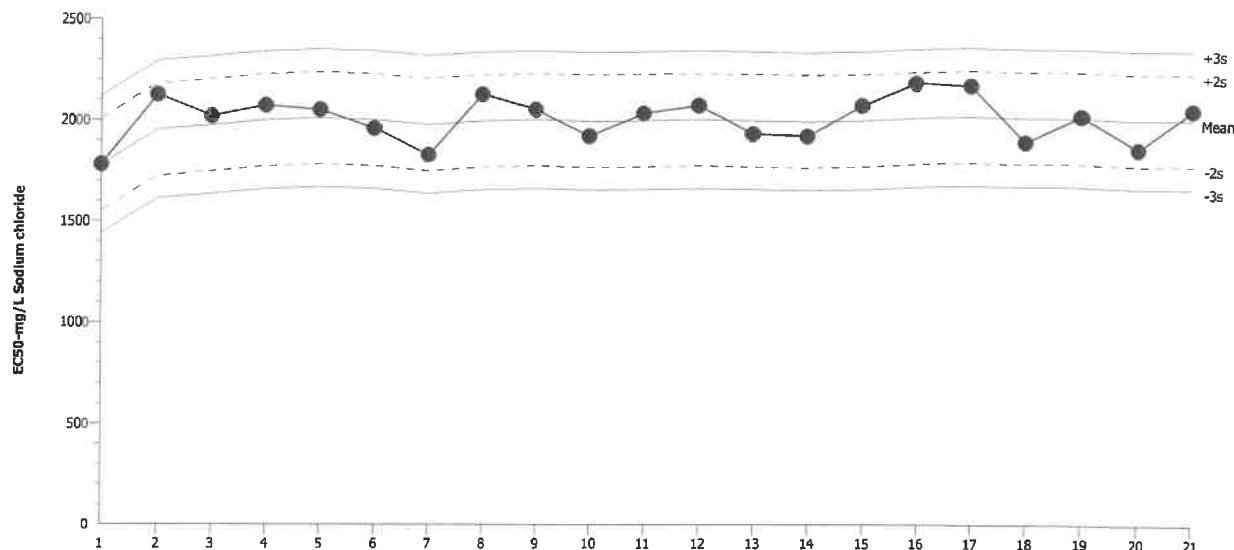
Material: Sodium chloride

Protocol: EPA-821-R-02-013 (2002)

Endpoint: Survival

Source: Reference Toxicant-REF

Ceriodaphnia Survival and Reproduction Test



Mean: 2003  
Sigma: 113.8

Count: 20  
CV: 5.68%

-2s Warning Limit: 1775  
+2s Warning Limit: 2230

-3s Action Limit: 1661  
+3s Action Limit: 2344

## Quality Control Data

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2018	Aug	29	14:50	1780	-222.7	-1.957			03-9264-7933	02-8153-7063
2		Sep	11	14:40	2125	121.7	1.07			17-7763-6788	12-2919-7286
3			12	14:04	2019	15.9	0.1397			17-4569-5270	18-9812-2558
4			18	12:26	2071	68.13	0.5987			20-2968-4066	17-1744-5971
5			19	11:34	2050	46.88	0.412			16-4284-4765	03-9142-8586
6			25	17:25	1957	-45.58	-0.4005			14-3900-9954	21-1313-3142
7		Oct	3	15:35	1825	-178	-1.565			07-6007-9059	16-4049-1493
8			9	16:46	2125	121.7	1.07			04-5469-0891	20-3055-9291
9			11	14:50	2050	46.88	0.412			20-2439-9413	10-4540-0750
10			16	13:11	1918	-84.64	-0.7438			03-5850-8111	20-6659-7771
11			18	15:16	2032	28.64	0.2517			05-8033-5759	02-3631-3458
12			19	15:05	2071	68.13	0.5987			02-1441-2791	17-1340-7957
13			23	15:40	1930	-72.54	-0.6374			10-7048-8617	14-7553-0745
14			30	10:35	1918	-84.64	-0.7438			05-8645-6876	01-6608-5367
15			31	14:47	2071	68.13	0.5987			15-6701-8818	10-0650-6684
16		Nov	6	15:55	2180	176.7	1.553			06-4622-5066	07-3608-9199
17			8	16:11	2170	166.8	1.465			07-3988-3316	08-3419-0126
18			13	16:18	1890	-112.9	-0.9922			10-7032-1533	04-1396-8369
19			27	13:39	2019	15.9	0.1397			01-2067-8558	07-2924-3826
20			30	14:32	1855	-147.9	-1.3			06-8813-8043	14-0037-6886
21		Dec	4	15:51	2050	46.88	0.412			08-4916-6117	20-2985-9813

## Ceriodaphnia Survival and Reproduction Test

Pacific EcoRisk

Test Type: Reproduction-Survival (7d)

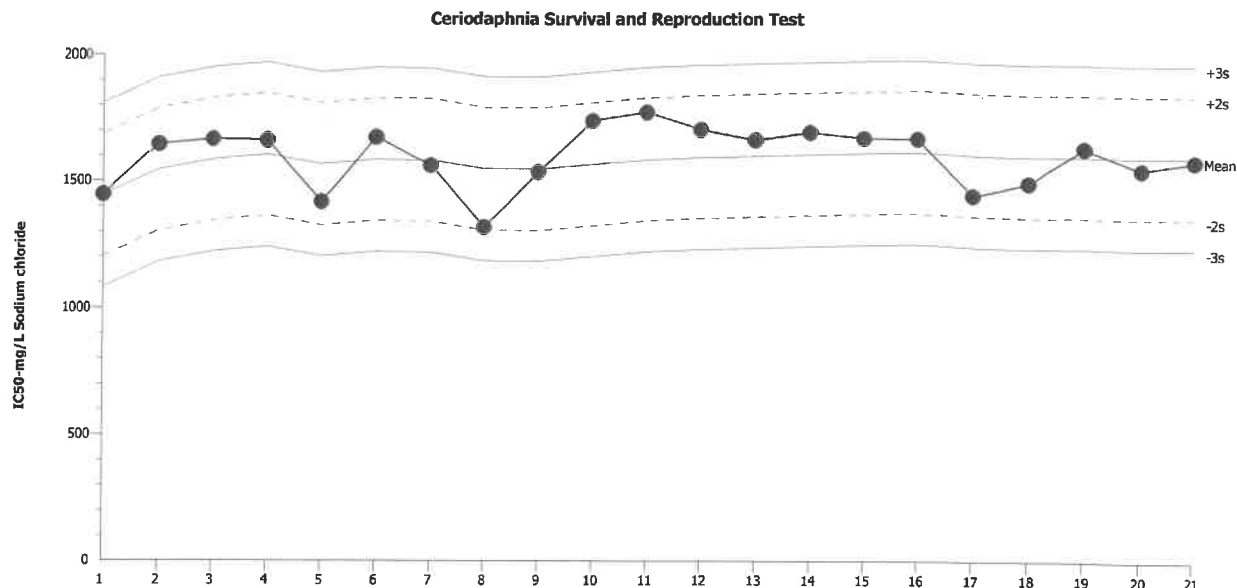
Organism: Ceriodaphnia dubia (Water Flea)

Material: Sodium chloride

Protocol: EPA-821-R-02-013 (2002)

Endpoint: Reproduction

Source: Reference Toxicant-REF



Mean: 1598

Count: 20

-2s Warning Limit: 1355

-3s Action Limit: 1234

Sigma: 121.2

CV: 7.58%

+2s Warning Limit: 1840

+3s Action Limit: 1961

## Quality Control Data

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2018	Aug	29	14:50	1447	-151.1	-1.247			03-9264-7933	16-8090-8266
2		Sep	11	14:40	1646	48.43	0.3996			17-7763-6788	12-3840-6964
3			12	14:04	1666	67.78	0.5592			17-4569-5270	09-7553-7941
4			18	12:26	1660	62.12	0.5126			20-2968-4066	11-3715-5377
5			19	11:34	1418	-179.9	-1.485			16-4284-4765	11-9866-6961
6			25	17:25	1673	75.42	0.6222			14-3900-9954	18-5535-4978
7		Oct	3	15:35	1561	-37.21	-0.307			07-6007-9059	08-5057-3824
8			9	16:46	1317	-281.2	-2.32	(-)		04-5469-0891	07-2283-5254
9			11	14:50	1535	-62.65	-0.5169			20-2439-9413	04-4179-5524
10			16	13:11	1738	140.1	1.156			03-5850-8111	05-4684-8364
11			18	15:16	1772	173.7	1.433			05-8033-5759	10-5626-5735
12			19	15:05	1704	105.9	0.8742			02-1441-2791	18-9658-3991
13			23	15:40	1663	65.4	0.5396			10-7048-8617	19-2272-0008
14			30	10:35	1694	95.9	0.7912			05-8645-6876	20-8136-4320
15			31	14:47	1670	72.45	0.5978			15-6701-8818	09-4862-8045
16		Nov	6	15:55	1669	70.63	0.5828			06-4622-5066	01-6239-3016
17			8	16:11	1445	-153.4	-1.265			07-3988-3316	04-7517-9392
18			13	16:18	1493	-105.4	-0.8698			10-7032-1533	07-8371-8990
19			27	13:39	1633	34.89	0.2879			01-2067-8558	17-6140-1063
20			30	14:32	1548	-49.52	-0.4085			06-8813-8043	05-8989-8676
21		Dec	4	15:51	1581	-16.75	-0.1382			08-4916-6117	16-9668-1931

Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: \_\_\_\_\_ Reference Toxicant: \_\_\_\_\_ Material: Sodium Chloride Test Date: 12/4/18  
 Project #: 29680 Test ID: 81022 Randomization: 10.7.6 Control Water: \_\_\_\_\_ Mod EPAMH

	Day	pH		D.O.		Cond. (µS/cm)		Temp (°C)	Survival / Reproduction										SIGN-OFF	
		New	Old	New	Old	New	Old		A	B	C	D	E	F	G	H	I	J		
Lab Water Control	0	7.87		9.1		360		24.5	0	0	0	0	0	0	0	0	0	0	Date: 12/4/18 New WQ: KL Test Init.: TF	Time: 1551
	1	7.89	7.68	7.2	8.2	359	406	24.3	0	0	0	0	0	0	0	0	0	0	Date: 12/5/18 New WQ: KL Counts: TF	Time: 1355
	2	7.69	7.72	8.4	6.9	359	431	24.0	0	0	0	0	0	0	0	0	0	0	Date: 12/6/18 New WQ: KL Counts: JB	Time: 1301
	3	8.03	7.65	7.1	5.0	418	427	24.0	0	0	0	0	0	0	0	0	0	0	Date: 12/7/18 New WQ: KL Counts: JB	Time: 1534
	4	7.71	7.69	7.9	4.4	370	400	25.3	6	6	5	4	6	6	6	7	6	6	Date: 12/8/18 New WQ: KL Counts: TF	Time: 1326
	5	7.83	7.61	7.7	7.4	357	406	24.0	0	0	11	10	12	11	11	10	10	0	Date: 12/9/18 New WQ: KL Counts: KL	Time: 1430
	6	7.76	7.76	9.0	7.8	371	416	24.6	12	10	13	0	0	0	12	0	0	13	Date: 12/10/18 New WQ: KL Counts: KL	Time: 1430
	7	-	8.07	-	8.0	-	399	24.7	15	10	0	13	12	14	0	14	12	14	Date: 12/11/18 New WQ: - Counts: -	Time: 1340
	8																		Date: Old WQ: Counts: Time:	
Total=									33	32	29	27	30	31	29	31	28	33	Mean Neonates/Female = 30.3	

	Day	pH		D.O.		Cond. (µS/cm)		Temp (°C)	Survival / Reproduction										RT BATCH NUMBER	
		New	Old	New	Old	New	Old		A	B	C	D	E	F	G	H	I	J		
500 mg/L	0	7.85		8.9		1351		24.5	0	0	0	0	0	0	0	0	0	0	288	
	1	7.85	7.69	7.3	8.3	1327	1570	25.0	0	0	0	0	0	0	0	0	0	0	288	
	2	7.69	7.86	8.5	7.0	1326	1477	24.0	0	0	0	0	0	0	0	0	0	0	288	
	3	7.93	7.66	7.4	4.9	1306	1505	24.7	0	0	0	0	0	0	0	0	0	0	288	
	4	7.69	7.70	8.3	5.5	1362	1494	24.0	6	6	6	6	5	6	6	6	6	6	288	
	5	7.81	7.58	8.2	7.3	1380	1448	25.1	12	12	10	11	12	12	12	11	0	11	288	
	6	7.75	7.81	9.0	8.0	1333	1565	24.7	0	0	0	0	12	13	11	0	9	0	288	
	7	-	8.00	-	8.0	-	1406	25.2	12	12	13	15	0	0	0	14	13	12	-	
	8																			
Total=									30	30	29	32	29	31	29	31	28	29	Mean Neonates/Female = 29.8	

Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: \_\_\_\_\_ Reference Toxicant: \_\_\_\_\_ Material: Sodium Chloride Test Date: 12/4/16  
 Project #: 29680 Test ID: 81022 Randomization: 10.7-6 Control Water: Mod EPAMH

	Day	pH		D.O.		Cond. (µS/cm)		Temp (°C)	Survival / Reproduction										SIGN-OFF
		New	Old	New	Old	New	Old		A	B	C	D	E	F	G	H	I	J	
1000 mg/L	0	7.84		9.0		2333		24.6	0	0	0	0	0	0	0	0	0	0	
	1	7.84	7.70	7.5	8.3	2285	2518	24.3	0	0	0	0	0	0	0	0	0	0	
	2	7.68	7.94	8.7	7.5	2287	2538	24.0	0	0	0	0	0	0	0	0	0	0	
	3	7.88	7.67	7.7	5.2	2320	2647	24.2	0	0	0	0	0	0	0	0	0	0	
	4	7.68	7.70	8.9	4.8	2325	2473	25.8	5	6	5	4	4	6	5	6	5	5	
	5	7.76	7.61	8.5	7.2	2285	2522	25.2	10	11	10	10	11	11	12	10	10	11	
	6	7.73	7.81	9.1	8.1	2312	2634	24.8	0	0	0	13	11	12	0	10	0	0	
	7	-	7.95	-	8.6	-	2441	24.4	13	12	12	0	0	0	13	0	12	14	
	8																		
Total=									28	29	27	27	26	29	30	26	27	30	Mean Neonates/Female = 27.9
1500 mg/L	0	7.85		9.3		3255		24.4	0	0	0	0	0	0	0	0	0	0	
	1	7.83	7.68	7.6	8.3	3191	3852	24.7	0	0	0	0	0	0	0	0	0	0	
	2	7.66	7.83	9.0	7.6	3231	3586	25.9	0	0	0	0	0	0	0	0	0	0	
	3	7.84	7.65	7.9	5.4	3300	3518	24.5	0	0	0	0	0	0	0	0	0	0	
	4	7.63	7.68	9.4	5.7	3175	3508	25.0	3	4	4	4	5	3	3	3	3	8/3	
	5	7.76	7.68	8.6	7.8	3267	3496	25.1	0	9	0	7	0	4	9	10	8	-	
	6	7.72	7.84	9.3	8.1	3207	3860	24.9	6	0	8	9	11	12	0	10	0	-	
	7	-	7.93	-	8.1	-	3604	24.9	0	9	10	10	10	7	9	3	6/3	-	
	8																		
Total=									9	22	22	21	26	14	21	16	17	8/3	Mean Neonates/Female = 17.1



Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: \_\_\_\_\_ Reference Toxicant: \_\_\_\_\_ Material: Sodium Chloride Test Date: 12/4/10  
 Project #: 29680 Test ID: 81022 Randomization: 10-7-6 Control Water: \_\_\_\_\_ Mod EPAMH

	Day	pH		D.O.		Cond. (µS/cm)		Temp (°C)	Survival / Reproduction										SIGN-OFF
		New	Old	New	Old	New	Old		A	B	C	D	E	F	G	H	I	J	
2000 mg/L	0	7.81		9.3		4231		24.4	0	0	0	0	0	0	0	0	0	0	
	1	7.76	7.67	8.0	8.1	4148	4443	24.5	0	0	0	0	0	0	0	0	0	0	
	2	7.65	7.51	9.1	7.5	4172	4500	24.8	0	0	0	0	0	0	1/0	0	0	0	
	3	7.74	7.62	8.0	5.8	4169	4740	24.0	0	0	0	0	0	0	-	0	0	0	
	4	7.63	7.69	9.7	4.7	4213	5430	25.6	0	0	0	0	0	0	-	0	0	0	
	5	7.73	7.74	9.3	8.0	4161	4415	25.1	3	0	0	1	0	3	-	3	5	0	
	6	7.71	7.82	9.5	8.0	4155	4576	25.0	0	0	0	1/0	0	0	-	0	0	3	
	7	-	7.92	-	8.3	-	4445	25.1	6	0	5	-	3	2	-	8	9	0	
	8											-			-				
Total=									9	0	5	1/1	3	5	1/0	11	14	3	Mean Neonates/Female = 5.1
2500 mg/L	0	7.79		9.5		5136		24.5	0	0	0	0	0	0	0	0	0	0	
	1	7.75	7.69	8.0	8.2	4904	5714	24.9	0	0	0	1/0	0	1/0	0	1/0	0	1/0	
	2	7.64	7.78	9.4	7.6	5140	5326	23.4	1/0	1/0	1/0	-	1/0	-	1/0	-	1/0	-	
	3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
	4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
	5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
	6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
	7								-	-	-	-	-	-	-	-	-	-	
	8								-	-	-	-	-	-	-	-	-	-	
Total=									1/0	1/0	1/0	1/0	1/0	1/0	1/0	1/0	1/0	1/0	Mean Neonates/Female = 0.0