

# Chronic Toxicity Testing Results for Wyckoff Eagle Harbor Groundwater Treatment Plant

# Monitoring Period: July 2019

Prepared for:

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Prepared by:

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Date Submitted: August 9, 2019

## **Data Quality Assurance:**

- Nautilus Environmental is accredited in accordance with NELAP by the State of Oregon Environmental Laboratory Accreditation Program (ORELAP ID 4053). It is also certified by the State of California Water Resources Control Board Environmental Laboratory Accreditation Program (Certificate No. 1802) and the State of Washington Department of Ecology (Lab ID C552). Specific fields of testing applicable to each accreditation are available upon request.
- All data have been reviewed and verified.
- All test results have met minimum test acceptability criteria under their respective US EPA protocols, unless otherwise noted in this report.
- All tests have met internal Quality Assurance Program requirements.

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Results verified by:

Eric Green, Project Manager

## Introduction

A toxicity test was performed using a groundwater composite sample collected on July 9, 2019 from the Wyckoff Eagle Harbor Groundwater Treatment Plant on Bainbridge Island in Washington. This test was performed to satisfy quarterly monitoring requirements according to the project Quality Assurance Project Plan (QAPP 2013). The chronic bioassay was conducted using the bivalve *Mytilus galloprovincialis* (Mediterranean mussel). Testing was performed at Enthalpy Analytical (formerly Nautilus Environmental) located in San Diego, California between July 10 and 12, 2019.

## Materials and Methods

The groundwater sample was collected into a low-density polyethylene cubitainer by Jacobs personnel, packed in a cooler containing ice, and shipped overnight to Enthalpy. Appropriate chain-of-custody (COC) procedures were employed during collection and transport. Upon arrival at the laboratory, the cooler was opened, the sample inspected, and the contents verified against information on the COC form. Standard water quality parameters were measured and recorded on a sample check-in form and are summarized in Table 1. The sample was stored at 4°C in the dark until used for testing.

Sample ID	070919				
Enthalpy Log-in Number	19-0742				
Collection Date; Time	7/9/2019; 1000h				
Receipt Date; Time	7/10/2019; 0945h				
Receipt Temperature (°C)	3.8				
Dissolved Oxygen (mg/L)	7.6				
рН	7.66				
Conductivity (µS/cm)	NM				
Salinity (ppt)	7.0				
Alkalinity (mg/L CaCO <sub>3</sub> )	395				
Total Chlorine (mg/L)	0.02				
Total Ammonia (mg/L as N)	1.3				

Table 1. Sample Information

NM = not measured

## **Test Methods**

Chronic toxicity testing was conducted according to the method set forth in USEPA (1995). This method is summarized in Table 2.

Test Period	7/10/2019, 1515h to 7/12/2019, 1535h
Test Organism	Mytilus galloprovincialis
Test Organism Source	Mission Bay (San Diego, CA)
Test Organism Age	4 hours post fertilization
Test Duration	48 ± 2 hours
Test Type	Static
Test Chamber, Test Solution Volume	30 mL glass vial, 10 mL
Test Temperature	15 ± 1°C
Dilution Water	Laboratory Seawater (Source: Scripps Institution of Oceanography [SIO] intake) diluted with de-ionized water
Additional Control	Brine Control (de-ionized water and hypersaline brine)
Test Salinity	30 ± 2 ppt
Source of Salinity	Hypersaline brine made by freezing seawater to a salinity of 90.3 ppt
Test Concentrations (% sample)	73.5 <sup>a</sup> , 35, 18, 9, 4, and 2%, lab and brine controls
Number of Replicates	5
Photoperiod	16 hours light/8 hours dark
Test Protocol	EPA/600/R-95/136
Test Acceptability Criteria for Controls	$\geq$ 50% mean survival, $\geq$ 90% mean development rate
Reference Toxicant	Copper chloride <sup>b</sup>
Statistical Software	CETIS™ 1.8.7.20

### Table 2. Summary of Methods for the Bivalve Larval Development Test

<sup>a</sup> Highest concentration tested due to the addition of hypersaline brine

<sup>b</sup> A deviation to the QAPP was approved by USEPA and Washington Department of Ecology to conduct reference toxicant testing with copper chloride. See QA section.

## Results

There were no statistically significant effects observed in any effluent concentration tested for the survival or development endpoint of the bivalve test. This results in a no observed effect concentration (NOEC) of 73.5 (the highest concentration tested) and a chronic toxic unit ( $TU_c$ ) of less than 1.36 for both endpoints.

Results for the chronic toxicity test are summarized in Tables 3 and 4. Individual statistical summaries for the test and copies of the laboratory bench sheets are provided in Appendix A. The sample check-in sheet and COC form are provided in Appendices B and C, respectively.

Table 3. Summary	of Statistical Results for the Chronic Toxicity	Tests
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Species	Species Endpoint		LOEC (% effluent)	Toxic Unit (TU <sub>c</sub> )	EC <sub>25</sub> (% effluent)	
Bivalve	Normal Development	73.5	> 73.5	< 1.36	> 73.5	
Divalve	Survival	73.5	> 73.5	< 1.36	> 73.5	

NOEC = No Observed Effect Concentration

LOEC = Lowest Observed Effect Concentration

Chronic Toxic Unit (TU<sub>c</sub>) = 100//NOEC. NOTE: Since 100% sample was not tested, the TU<sub>c</sub> value can only be calculated up to the highest concentration tested. If no toxicity is observed at this concentration, the TU<sub>c</sub> is reported as less than the calculated value. Effect Concentration 25 (IC<sub>25</sub>) = Concentration expected to cause an effect to 25% of the organisms

Concentration (% Effluent)	Mean Survival (%)	Mean Normal Development (%)								
0 (Brine Control)	98.6	96.3								
0 (Lab Control)	99.1	96.7								
2	97.0	95.0								
4	96.6	95.6								
9	100	96.3								
18	98.2	96.9								
35	97.8	96.0								
73.5 <sup>a</sup>	97.5	95.3								

## Table 4. Detailed Results for the Bivalve Development Chronic Toxicity Test

<sup>a</sup> Highest concentration tested due to the addition of hypersaline brine

## **Quality Assurance**

The sample was received within the required 36-hour holding time, in good condition, and within the appropriate temperature range of 0-6°C. All control acceptability criteria were met and water quality parameters remained within the appropriate ranges throughout the test. Statistical analyses followed standard USEPA flowchart selections. Dose-response relationships were reviewed to ensure the reliability of the results. Based on the dose response observed, the calculated effects concentrations were deemed reliable.

Results for the reference toxicant tests used to monitor laboratory performance and test organism sensitivity are summarized in Table 5. A deviation to the QAPP was approved by USEPA and Washington Department of Ecology to conduct reference toxicant testing with copper chloride rather than copper sulfate. The results for the concurrent reference toxicant test were within the acceptable range of the mean historical test results plus or minus two standard deviations. The reference toxicant statistical summaries and laboratory bench sheets are provided in Appendix D. Minor QA/QC issues that were unlikely to have any bearing on the final test results, such as slight temperature deviations, are noted on the data sheets and a list of qualifier codes used on bench data sheets is presented in Appendix E.

Species	Endpoint	EC₅₀ (μg/L copper)	Historical mean ± 2 SD (µg/L copper)	CV (%)
Bivalve	Normal Development	7.80	7.19 ± 1.70	11.9
Divalve	Survival Rate	29.7	28.3 ± 5.20	9.19

Table 5. Reference Toxicant Test Results

Effect Concentration 50 (EC $_{50}$ ) = Concentration expected to cause an effect to 50% of the organisms

CV = Coefficient of Variation.

## References

- CH2MHill. 2013. Quality Assurance Project Plan Groundwater Treatment Plant Operations, Maintenance, Bainbridge, Washington. Prepared for USEPA Region 10 June 5, 2013.
- Standard Guide for Conducting Static Acute Toxicity Tests with Embryos of Four Species of Saltwater Bivalve Molluscs. 1989. ASTM Standard E 724-89.
- Tidepool Scientific Software. 2000-2013. CETIS Comprehensive Environmental Toxicity Information System Software, Version 1.8.7.20.
- USEPA. 1995. Short-Term Method for Estimating the Chronic Toxicity of Effluents and Receiving Waters to the West Coast Marine and Estuarine Organisms. EPA/600/R-95/136. pp. 209-258 and 389-465.
- Washington State Department of Ecology. 2016. Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria. Publication No. WQ-R-95-80. Revised June 2016

Appendix A Statistical Summaries and Raw Bench Sheets **Bivalve Larval Development Test** 

Report Date: Test Code: 02 Aug-19 08:20 (p 1 of 2) 1907-S060 | 03-8657-3583

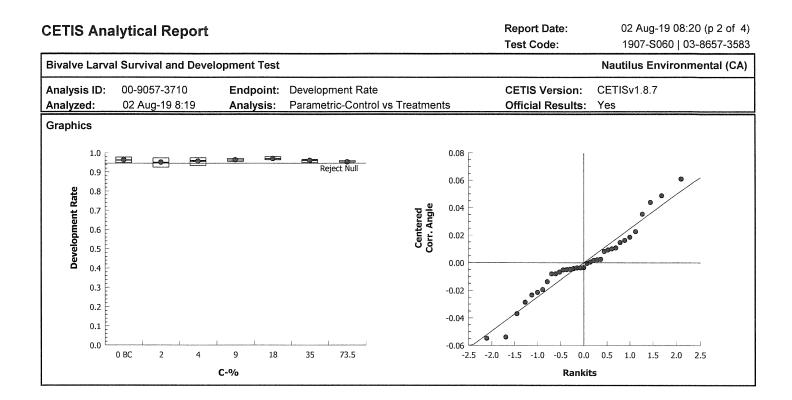
							16	est Code:	190	-3000   03	3-8657-3583		
Bivalve Larval	I Survival and D	evelop	oment Test						Nautilus	Environm	nental (CA)		
Batch ID: Start Date: Ending Date: Duration:	19-3820-1131 10 Jul-19 15:15 12 Jul-19 15:35 48h		Test Type: Protocol: Species: Source:	Species: Mytilus gallopro			D B		luent: Diluted Natural Seawater ine: Frozen Seawater				
Sample ID:	16-9245-2664		Code:					lient: Jaco	bs				
-	09 Jul-19 10:00		Material:	Effluent Sampl	е		Р	roject:					
Receive Date:	10 Jul-19 09:45		Source:	Jacobs									
Sample Age:	29h (3.8 °C)		Station:	Wyckoff									
Comparison S	Summary												
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method					
00-9057-3710	Development Rate 73.5		73.5	>73.5	NA	1.73%	1.361		ultiple Comp				
04-3000-7149	Survival Rate		73.5	>73.5	NA	4.69%	1.361	Steel Many	-One Rank	Sum Test			
Point Estimate	e Summary												
Analysis ID	Endpoint	and the store of the star	Level	%	95% LCL	95% UCL	TU	Method	14407-156-158 BIELEVILLE				
11-3898-7688	Development R	ate	EC25		N/A	N/A	<1.361		rpolation (IC	CPIN)			
			EC50	>73.5	N/A	N/A	<1.361						
10-1515-3673 Survival Rate			EC25		N/A	N/A	<1.361		Linear Interpolation (ICPIN)				
			EC50	>73.5	N/A	N/A	<1.361						
Test Acceptab	bility												
Analysis ID	Endpoint		Attrib	oute	Test Stat	TAC Lim	its	Overlap	Decision	*******			
00-9057-3710	Development R	ate	Contr	ol Resp	0.9626	0.9 - NL		Yes					
11-3898-7688	Development R	ate	Contr	ol Resp	0.9626	0.9 - NL		Yes	Yes Passes Acceptability Criteria				
04-3000-7149	Survival Rate			ol Resp	0.9862	0.5 - NL		Yes	Passes Ac				
10-1515-3673	Survival Rate		Contr	ol Resp	0.9862	0.5 - NL		Yes	Passes Ac	ceptability	Criteria		
Development	Rate Summary												
C-%	Control Type	Cour	nt Mear	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect		
0	Brine Control	5	0.962	6 0.9494	0.9757	0.948	0.9779	0.004749	0.01062	1.1%	0.0%		
	Lab Control	5	0.967	2 0.9486	0.9857	0.9504	0.991	0.006692	0.01496	1.55%	-0.48%		
2		5	0.949		0.9714	0.9245	0.9738		0.01751	1.84%	1.34%		
4		5	0.955		0.9758	0.9322	0.9744		0.01622	1.7%	0.72%		
9		5	0.963		0.9712	0.9542	0.9714		0.006451	0.67%	-0.07%		
18		5	0.969		0.9781	0.9628	0.9807		0.006948		-0.72%		
35 73.5		5 5	0.960 0.952		0.9692 0.9577	0.9486 0.9494	0.966 0.9595	0.003233 0.001777	0.007228 0.003974	0.75% 0.42%	0.24% 1.02%		
Survival Rate	Summary									0.1270			
C-%	Control Type	Cour	nt Mear	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect		
0	Brine Control	5	0.986		1	0.9569	1	0.008876	0.01985	2.01%	0.0%		
0	Lab Control	5	0.990		1	0.9526	1	0.009483	0.0212	2.14%	-0.44%		
2		5	0.969		1	0.9138	1	0.01536	0.03435	3.54%	1.66%		
4		5	0.966	0.9179	1	0.9052	1	0.01748	0.03908	4.04%	2.01%		
9		5	1	1	1	1	1	0	0	0.0%	-1.4%		
18		5	0.981	9 0.9425	1	0.9267	1	0.01419	0.03173	3.23%	0.44%		
35		5	0.977		1	0.9224	1	0.01532	0.03427	3.51%	0.87%		
73.5		5	0.975	0.9292	1	0.9181	1	0.01649	0.03688	3.78%	1.14%		

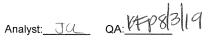
Bivalve Larval Survival and Development Test

			Report Date: Test Code:	02 Aug-19 08:20 (p 2 of 2) 1907-S060   03-8657-3583
				Nautilus Environmental (CA)
•	Den (	D 5		
3	Rep 4	Rep 5		
70	0.004	0.040		

Divalve L			it rest				
Developm	nent Rate Detail						
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	0.9615	0.9614	0.9779	0.964	0.948	
0	Lab Control	0.9679	0.9662	0.9603	0.9504	0.991	
2		0.9738	0.9507	0.952	0.9474	0.9245	
4		0.9744	0.9322	0.964	0.96	0.9476	
9		0.9615	0.9542	0.9668	0.962	0.9714	
18		0.9628	0.9685	0.9703	0.9807	0.9649	
35		0.9645	0.966	0.9643	0.9578	0.9486	
73.5		0.9494	0.9506	0.9513	0.9595	0.9531	
Survival F	Rate Detail						
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	1	1	0.9741	0.9569	1	
0	Lab Control	1	1	1	1	0.9526	
2		0.9871	0.9612	0.9871	1	0.9138	
4		1	1	0.9569	0.9698	0.9052	
9		1	1	1	1	1	
18		0.9267	1	1	1	0.9828	
35		1	1	0.9655	1	0.9224	
73.5		1	1	1	0.9569	0.9181	
Developm	nent Rate Binomials	5					
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	225/234	224/233	221/226	214/222	237/250	
0	Lab Control	241/249	229/237	242/252	249/262	219/221	
2		223/229	212/223	218/229	252/266	196/212	
4		228/234	220/236	214/222	216/225	199/210	
9		225/234	229/240	233/241	228/237	238/245	
18		207/215	246/254	229/236	254/259	220/228	
35		272/282	227/235	216/224	227/237	203/214	
73.5		225/237	231/243	254/267	213/222	203/213	
Survival I	Rate Binomials						
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	232/232	232/232	226/232	222/232	232/232	
0	Lab Control	232/232	232/232	232/232	232/232	221/232	
2		229/232	223/232	229/232	232/232	212/232	
4		232/232	232/232	222/232	225/232	210/232	
9		232/232	232/232	232/232	232/232	232/232	
18		215/232	232/232	232/232	232/232	228/232	
		232/232	232/232	224/232	232/232	214/232	
35		2321232	2021202	2271202	202/202		

CETIS Ana	alytical Rep	ort		Report Date: Test Code:		02 Aug-19 08:20 (p 1 of 4) 1907-S060   03-8657-3583						
Bivalve Larva	al Survival and I	Developm	ent Test						Nautilus	Environn	nental (CA)	
Analysis ID: Analyzed:	00-9057-3710 02 Aug-19 8:1		•					S Version: ial Results		CETISv1.8.7 Yes		
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU	
Angular (Corre	ected)	NA	C > T	NA	NA		1.73%	73.5	>73.5	NA	1.361	
Dunnett Mult	iple Compariso	n Test					and a stand of the second standing of the second standing of the second standing of the second standing of the					
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(α:5%)			
Brine Control	2		1.755	2.407	0.042 8	0.1665	CDF	Non-Signi	ficant Effect			
	4		0.9479	2.407	0.042 8	0.4763	CDF	Non-Signi	ficant Effect			
	9		-0.03727	2.407	0.042 8	0.8670	CDF	Non-Signi	ficant Effect			
	18		-1.066	2.407	0.042 8	0.9899	CDF	Non-Signi	ficant Effect			
l	35		0.4068	2.407	0.042 8	0.7190	CDF	Non-Significant Effect				
	73.5		1.494	2.407	0.042 8	0.2478	CDF	-	ficant Effect			
ANOVA Table	9											
Source	Sum Squ	ares	Mean Squ	lare	DF	F Stat	P-Value	Decision	(α:5%)			
Between	0.008611	49	0.0014352	248	6	1.921	0.1123	Non-Signi	ficant Effect			
Error	0.020919	07	0.0007471	096	28							
Total	0.029530	56			34							
Distributiona	I Tests											
Attribute	Test			Test Stat	,							
Variances	Bartlett I	Equality of	Variance	9.911	16.81	0.1284	Equal Variances					
Distribution	Shapiro-	Wilk W No	ormality	0.9578	0.9146	0.1960	Normal Distribution					
Development	t Rate Summary	,										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	Brine Control	5	0.9626	0.9494	0.9757	0.9615	0.948	0.9779	0.004749	1.1%	0.0%	
2		5	0.9497	0 0070	0.0744	0.0507	0.0045	0.9738			1 0 407	
			0.3437	0.9279	0.9714	0.9507	0.9245	0.9750	0.007829	1.84%	1.34%	
4		5	0.9556	0.9279 0.9355	0.9714 0.9758	0.9507 0.96	0.9245	0.9738	0.007829 0.007254	1.84% 1.7%	1.34% 0.72%	
4 9												
		5	0.9556	0.9355	0.9758	0.96	0.9322	0.9744	0.007254	1.7%	0.72%	
9		5 5	0.9556 0.9632	0.9355 0.9552	0.9758 0.9712	0.96 0.962	0.9322 0.9542	0.9744 0.9714	0.007254 0.002885	1.7% 0.67%	0.72% -0.07%	
9 18		5 5 5	0.9556 0.9632 0.9694	0.9355 0.9552 0.9608	0.9758 0.9712 0.9781	0.96 0.962 0.9685	0.9322 0.9542 0.9628	0.9744 0.9714 0.9807	0.007254 0.002885 0.003107	1.7% 0.67% 0.72%	0.72% -0.07% -0.72%	
9 18 35 73.5	rected) Transfo	5 5 5 5 5	0.9556 0.9632 0.9694 0.9602 0.9528	0.9355 0.9552 0.9608 0.9513	0.9758 0.9712 0.9781 0.9692	0.96 0.962 0.9685 0.9643	0.9322 0.9542 0.9628 0.9486	0.9744 0.9714 0.9807 0.966	0.007254 0.002885 0.003107 0.003232	1.7% 0.67% 0.72% 0.75%	0.72% -0.07% -0.72% 0.24%	
9 18 35 73.5	rected) Transfo Control Type	5 5 5 5 5	0.9556 0.9632 0.9694 0.9602 0.9528	0.9355 0.9552 0.9608 0.9513	0.9758 0.9712 0.9781 0.9692	0.96 0.962 0.9685 0.9643	0.9322 0.9542 0.9628 0.9486	0.9744 0.9714 0.9807 0.966	0.007254 0.002885 0.003107 0.003232	1.7% 0.67% 0.72% 0.75%	0.72% -0.07% -0.72% 0.24%	
9 18 35 73.5 Angular (Cor		5 5 5 5 5 <b>rmed Sun</b>	0.9556 0.9632 0.9694 0.9602 0.9528	0.9355 0.9552 0.9608 0.9513 0.9478	0.9758 0.9712 0.9781 0.9692 0.9577	0.96 0.962 0.9685 0.9643 0.9513	0.9322 0.9542 0.9628 0.9486 0.9494	0.9744 0.9714 0.9807 0.966 0.9595	0.007254 0.002885 0.003107 0.003232 0.001778	1.7% 0.67% 0.72% 0.75% 0.42%	0.72% -0.07% -0.72% 0.24% 1.02%	
9 18 35 73.5 Angular (Cor C-%	Control Type	5 5 5 5 rmed Sun Count	0.9556 0.9632 0.9694 0.9602 0.9528 mmary Mean	0.9355 0.9552 0.9608 0.9513 0.9478 95% LCL	0.9758 0.9712 0.9781 0.9692 0.9577 <b>95% UCL</b>	0.96 0.962 0.9685 0.9643 0.9513 Median	0.9322 0.9542 0.9628 0.9486 0.9494 Min	0.9744 0.9714 0.9807 0.966 0.9595 Max	0.007254 0.002885 0.003107 0.003232 0.001778 Std Err	1.7% 0.67% 0.72% 0.75% 0.42%	0.72% -0.07% -0.72% 0.24% 1.02% %Effect	
9 18 35 73.5 <b>Angular (Cor</b> <b>C-%</b> 0	Control Type	5 5 5 5 rmed Sun Count 5	0.9556 0.9632 0.9694 0.9602 0.9528 mmary <u>Mean</u> 1.378	0.9355 0.9552 0.9608 0.9513 0.9478 95% LCL 1.342	0.9758 0.9712 0.9781 0.9692 0.9577 <b>95% UCL</b> 1.414	0.96 0.962 0.9685 0.9643 0.9513 Median 1.373	0.9322 0.9542 0.9628 0.9486 0.9494 Min 1.341	0.9744 0.9714 0.9807 0.966 0.9595 Max 1.422	0.007254 0.002885 0.003107 0.003232 0.001778 Std Err 0.0129	1.7% 0.67% 0.72% 0.75% 0.42% CV% 2.09%	0.72% -0.07% -0.72% 0.24% 1.02% %Effect 0.0%	
9 18 35 73.5 Angular (Cor C-% 0 2	Control Type	5 5 5 5 <b>rmed Sun</b> <b>Count</b> 5 5	0.9556 0.9632 0.9694 0.9602 0.9528 mary Mean 1.378 1.347	0.9355 0.9552 0.9608 0.9513 0.9478 <b>95% LCL</b> 1.342 1.296	0.9758 0.9712 0.9781 0.9692 0.9577 <b>95% UCL</b> 1.414 1.398	0.96 0.962 0.9685 0.9643 0.9513 Median 1.373 1.347	0.9322 0.9542 0.9628 0.9486 0.9494 <b>Min</b> 1.341 1.292	0.9744 0.9714 0.9807 0.966 0.9595 Max 1.422 1.408	0.007254 0.002885 0.003107 0.003232 0.001778 <b>Std Err</b> 0.0129 0.01842	1.7% 0.67% 0.72% 0.75% 0.42% <b>CV%</b> 2.09% 3.06%	0.72% -0.07% -0.72% 0.24% 1.02% %Effect 0.0% 2.2%	
9 18 35 73.5 Angular (Cor C-% 0 2 4	Control Type	5 5 5 5 rrmed Sun Count 5 5 5 5	0.9556 0.9632 0.9694 0.9602 0.9528 mmary <u>Mean</u> 1.378 1.347 1.361	0.9355 0.9552 0.9608 0.9513 0.9478 <b>95% LCL</b> 1.342 1.296 1.313	0.9758 0.9712 0.9781 0.9692 0.9577 <b>95% UCL</b> 1.414 1.398 1.41	0.96 0.962 0.9685 0.9643 0.9513 Median 1.373 1.347 1.369	0.9322 0.9542 0.9628 0.9486 0.9494 Min 1.341 1.292 1.307	0.9744 0.9714 0.9807 0.966 0.9595 Max 1.422 1.408 1.41	0.007254 0.002885 0.003107 0.003232 0.001778 <b>Std Err</b> 0.0129 0.01842 0.01753	1.7% 0.67% 0.72% 0.75% 0.42% CV% 2.09% 3.06% 2.88%	0.72% -0.07% -0.72% 0.24% 1.02% %Effect 0.0% 2.2% 1.19%	
9 18 35 73.5 Angular (Cor C-% 0 2 4 9	Control Type	5 5 5 5 <b>rmed Sun</b> 5 5 5 5 5 5	0.9556 0.9632 0.9694 0.9528 mary Mean 1.378 1.347 1.361 1.378	0.9355 0.9552 0.9608 0.9513 0.9478 <b>95% LCL</b> 1.342 1.296 1.313 1.357	0.9758 0.9712 0.9781 0.9692 0.9577 <b>95% UCL</b> 1.414 1.398 1.41 1.4	0.96 0.962 0.9685 0.9643 0.9513 Median 1.373 1.347 1.369 1.375	0.9322 0.9542 0.9628 0.9486 0.9494 Min 1.341 1.292 1.307 1.355	0.9744 0.9714 0.9807 0.966 0.9595 <b>Max</b> 1.422 1.408 1.41 1.401	0.007254 0.002885 0.003107 0.003232 0.001778 <b>Std Err</b> 0.0129 0.01842 0.01753 0.007673	1.7% 0.67% 0.72% 0.75% 0.42% <b>CV%</b> 2.09% 3.06% 2.88% 1.25%	0.72% -0.07% -0.72% 0.24% 1.02% %Effect 0.0% 2.2% 1.19% -0.05%	





	· · · · · · · · · · · · · · · · · · ·	oort						Repo Test	Code:	1907	U U	20 (p 3 of 4) 3-8657-3583
Bivalve Larva	al Survival and	Developm	ent Test	1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 -						Nautilus	Environr	nental (CA)
Analysis ID: Analyzed:	04-3000-7149 02 Aug-19 8:		•	vival Rate parametric-	Control v	's T	reatments	CETIS Version: CETISv1.8.7 Official Results: Yes				
Data Transfor	rm	Zeta	Alt Hyp	Trials	Seed			PMSD	NOEL	LOEL	TOEL	TU
Angular (Corre	ected)	NA	C > T	NA	NA			4.69%	73.5	>73.5	NA	1.361
Steel Many-O	ne Rank Sum	Test										
Control	vs C-%		Test Stat	Critical	Ties	DF	P-Value	P-Type	Decision	(α:5%)		
Brine Control	2		23.5	16	1	8	0.5252	Asymp		ificant Effect		
	4		23.5	16		8	0.5252	Asymp	5	ificant Effect		
	9		32.5	16		8	0.9904	Asymp	-	ificant Effect		
	18		27.5	16		8	0.8571	Asymp	-	ificant Effect		
	35		26.5	16		8	0.7925	Asymp	-	ificant Effect		
	73.5		26	16	•	8	0.7547	Asymp	-	ificant Effect		
ANOVA Table	2											
Source	Sum Sq	uares	Mean Squ	are	DF		F Stat	P-Value	Decision	(a:5%)		
Between	0.04959		0.0082653		6		0.8102	0.5708		ificant Effect		
Error	0.28563		0.0102014		28			0.0100				
Total	0.33523											
Distributional	l Tests											
Attribute	Test			Test Stat	Critical		P-Value	Decision(	α:1%)			
Variances	Mod Le	vene Equa	lity of Variance		3.812		0.6720	Equal Variances				
Variances		Equality of	•	3.555	3.528		0.0096	•				
Distribution		-Wilk W No		0.9065	0.9146		0.0059	· · · · · · · · · · · · · · · · · · ·				
Survival Rate												
C-%	Control Type	Count	Mean	95% LCL	95% U	CL	Median	Min	Max	Std Err	CV%	%Effect
<b>C-%</b>	Control Type Brine Control	Count	<b>Mean</b>	95% LCL	95% U(	CL	Median	Min 0.9569	Max	<b>Std Err</b>	<b>CV%</b>	%Effect
0	Control Type Brine Control	5	0.9862	0.9616	1		1	0.9569	1	0.008876	2.01%	0.0%
0 2		5 5	0.9862 0.9698	0.9616 0.9272	1		1 0.9871	0.9569 0.9138	1 1	0.008876 0.01536	2.01% 3.54%	0.0% 1.66%
0 2 4		5 5 5	0.9862 0.9698 0.9664	0.9616 0.9272 0.9179	1 1 1		1 0.9871 0.9698	0.9569 0.9138 0.9052	1 1 1	0.008876 0.01536 0.01748	2.01% 3.54% 4.04%	0.0% 1.66% 2.01%
0 2 4 9		5 5 5 5	0.9862 0.9698 0.9664 1	0.9616 0.9272 0.9179 1	1	CL	1 0.9871 0.9698 1	0.9569 0.9138 0.9052 1	1 1 1 1	0.008876 0.01536 0.01748 0	2.01% 3.54% 4.04% 0.0%	0.0% 1.66% 2.01% -1.4%
0 2 4 9 18		5 5 5 5 5 5	0.9862 0.9698 0.9664 1 0.9819	0.9616 0.9272 0.9179 1 0.9425	1 1 1 1 1		1 0.9871 0.9698 1 1	0.9569 0.9138 0.9052 1 0.9267	1 1 1 1	0.008876 0.01536 0.01748 0 0.01419	2.01% 3.54% 4.04% 0.0% 3.23%	0.0% 1.66% 2.01% -1.4% 0.44%
0 2 4 9 18 35		5 5 5 5 5 5 5	0.9862 0.9698 0.9664 1 0.9819 0.9776	0.9616 0.9272 0.9179 1 0.9425 0.935	1 1 1 1 1 1	CL	1 0.9871 0.9698 1 1 1	0.9569 0.9138 0.9052 1 0.9267 0.9224	1 1 1 1 1	0.008876 0.01536 0.01748 0 0.01419 0.01532	2.01% 3.54% 4.04% 0.0% 3.23% 3.51%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87%
0 2 4 9 18 35 73.5	Brine Control	5 5 5 5 5 5 5 5 5	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975	0.9616 0.9272 0.9179 1 0.9425	1 1 1 1 1	CL	1 0.9871 0.9698 1 1	0.9569 0.9138 0.9052 1 0.9267	1 1 1 1	0.008876 0.01536 0.01748 0 0.01419	2.01% 3.54% 4.04% 0.0% 3.23%	0.0% 1.66% 2.01% -1.4% 0.44%
0 2 4 9 18 35 73.5 Angular (Corr	Brine Control	5 5 5 5 5 5 5 5	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975	0.9616 0.9272 0.9179 1 0.9425 0.935 0.9292	1 1 1 1 1 1		1 0.9871 0.9698 1 1 1 1	0.9569 0.9138 0.9052 1 0.9267 0.9224 0.9181	1 1 1 1 1 1	0.008876 0.01536 0.01748 0 0.01419 0.01532 0.01649	2.01% 3.54% 4.04% 0.0% 3.23% 3.51% 3.78%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87% 1.14%
0 2 4 9 18 35 73.5 Angular (Corr C-%	Brine Control rected) Transfo Control Type	5 5 5 5 5 5 5 5 0 0 0 0 0 0 0 0 0 0 0 0	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975 nmary Mean	0.9616 0.9272 0.9179 1 0.9425 0.935 0.9292 95% LCL	1 1 1 1 1 1 95% U(		1 0.9871 0.9698 1 1 1 1 1 <b>Median</b>	0.9569 0.9138 0.9052 1 0.9267 0.9224 0.9181 Min	1 1 1 1 1 1 1 Max	0.008876 0.01536 0.01748 0 0.01419 0.01532 0.01649 Std Err	2.01% 3.54% 4.04% 0.0% 3.23% 3.51% 3.78%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87% 1.14%
0 2 4 9 18 35 73.5 Angular (Corr C-% 0	Brine Control	5 5 5 5 5 5 5 5 5 5 5 0 7 med Sun 5	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975 mmary <u>Mean</u> 1.477	0.9616 0.9272 0.9179 1 0.9425 0.935 0.9292 95% LCL 1.371	1 1 1 1 1 1 1 <b>95% U</b> 1.583		1 0.9871 0.9698 1 1 1 1 1 <b>Median</b> 1.538	0.9569 0.9138 0.9052 1 0.9267 0.9224 0.9181 <b>Min</b> 1.362	1 1 1 1 1 1 1 1 <b>Max</b> 1.538	0.008876 0.01536 0.01748 0 0.01419 0.01532 0.01649 <b>Std Err</b> 0.0381	2.01% 3.54% 4.04% 0.0% 3.23% 3.51% 3.78% <b>CV%</b> 5.77%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87% 1.14% %Effect 0.0%
0 2 4 9 18 35 73.5 Angular (Corr C-% 0 2	Brine Control rected) Transfo Control Type	5 5 5 5 5 5 5 5 5 5 5 0 7 7 7 6 0 1 7 5 5 5	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975 mmary <u>Mean</u> 1.477 1.419	0.9616 0.9272 0.9179 1 0.9425 0.935 0.9292 95% LCL 1.371 1.294	1 1 1 1 1 1 1 <b>95% U(</b> 1.583 1.544		1 0.9871 0.9698 1 1 1 1 1 1 <b>Median</b> 1.538 1.457	0.9569 0.9138 0.9052 1 0.9267 0.9224 0.9181 <b>Min</b> 1.362 1.273	1 1 1 1 1 1 1 1 <b>Max</b> 1.538 1.538	0.008876 0.01536 0.01748 0 0.01419 0.01532 0.01649 <b>Std Err</b> 0.0381 0.04503	2.01% 3.54% 4.04% 0.0% 3.23% 3.51% 3.78% <b>CV%</b> 5.77% 7.09%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87% 1.14% %Effect 0.0% 3.9%
0 2 4 9 18 35 73.5 Angular (Corr C-% 0 2 4	Brine Control rected) Transfo Control Type	5 5 5 5 5 5 5 5 5 5 5 5 7 7 7 7 7 7 7 7	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975 mmary <u>Mean</u> 1.477 1.419 1.418	0.9616 0.9272 0.9179 1 0.9425 0.935 0.9292 <b>95% LCL</b> 1.371 1.294 1.269	1 1 1 1 1 1 1 <b>95% U(</b> 1.583 1.544 1.568		1 0.9871 0.9698 1 1 1 1 1 1 <b>Median</b> 1.538 1.457 1.396	0.9569 0.9138 0.9052 1 0.9267 0.9224 0.9181 <b>Min</b> 1.362 1.273 1.258	1 1 1 1 1 1 1 <b>Max</b> 1.538 1.538 1.538 1.538	0.008876 0.01536 0.01748 0 0.01419 0.01532 0.01649 <b>Std Err</b> 0.0381 0.04503 0.0539	2.01% 3.54% 4.04% 0.0% 3.23% 3.51% 3.78% <b>CV%</b> 5.77% 7.09% 8.5%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87% 1.14% %Effect 0.0% 3.9% 3.97%
0 2 4 9 18 35 73.5 Angular (Corr C-% 0 2 4 9	Brine Control rected) Transfo Control Type	5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975 <b>mary</b> Mean 1.477 1.419 1.418 1.538	0.9616 0.9272 0.9179 1 0.9425 0.935 0.9292 95% LCL 1.371 1.294 1.269 1.538	1 1 1 1 1 1 1 <b>95% U(</b> 1.583 1.544 1.568 1.538		1 0.9871 0.9698 1 1 1 1 1 1 <b>Median</b> 1.538 1.457 1.396 1.538	0.9569 0.9138 0.9052 1 0.9267 0.9224 0.9181 <b>Min</b> 1.362 1.273 1.258 1.538	1 1 1 1 1 1 1 1 <b>Max</b> 1.538 1.538 1.538 1.538 1.538	0.008876 0.01536 0.01748 0 0.01419 0.01532 0.01649 <b>Std Err</b> 0.0381 0.04503 0.0539 0	2.01% 3.54% 4.04% 0.0% 3.23% 3.51% 3.78% <b>CV%</b> 5.77% 7.09% 8.5% 0.0%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87% 1.14% %Effect 0.0% 3.9% 3.97% -4.13%
0 2 4 9 18 35 73.5 Angular (Corr C-% 0 2 4	Brine Control rected) Transfo Control Type	5 5 5 5 5 5 5 5 5 5 5 5 7 7 7 7 7 7 7 7	0.9862 0.9698 0.9664 1 0.9819 0.9776 0.975 mmary <u>Mean</u> 1.477 1.419 1.418	0.9616 0.9272 0.9179 1 0.9425 0.935 0.9292 <b>95% LCL</b> 1.371 1.294 1.269	1 1 1 1 1 1 1 <b>95% U(</b> 1.583 1.544 1.568		1 0.9871 0.9698 1 1 1 1 1 1 <b>Median</b> 1.538 1.457 1.396	0.9569 0.9138 0.9052 1 0.9267 0.9224 0.9181 <b>Min</b> 1.362 1.273 1.258	1 1 1 1 1 1 1 <b>Max</b> 1.538 1.538 1.538 1.538	0.008876 0.01536 0.01748 0 0.01419 0.01532 0.01649 <b>Std Err</b> 0.0381 0.04503 0.0539	2.01% 3.54% 4.04% 0.0% 3.23% 3.51% 3.78% <b>CV%</b> 5.77% 7.09% 8.5%	0.0% 1.66% 2.01% -1.4% 0.44% 0.87% 1.14% %Effect 0.0% 3.9% 3.97%

Analyst: Jac QA: 4798319

#### Report Date: 02 Aug-19 08:20 (p 4 of 4) **CETIS Analytical Report** 1907-S060 | 03-8657-3583 Test Code: Nautilus Environmental (CA) **Bivalve Larval Survival and Development Test** CETISv1.8.7 Analysis ID: 04-3000-7149 Endpoint: Survival Rate **CETIS Version:** Analyzed: 02 Aug-19 8:19 Analysis: Nonparametric-Control vs Treatments **Official Results:** Yes Graphics 0.15 1.0 202 **A**02 0.9 0.10 0.8 Survival Rate Centered Corr. Angle 0.05 0.7 0.6 0.00 0.5 -0.05 0.4 0.3 -0.10 0.2 -0.15 0.1

-0.20

-2.5 -2.0 -1.5 -1.0

-0.5 0.0 0.5 1.0 1.5 2.0 2.5

Rankits

0.0

0 BC

2

4

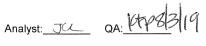
9

C-%

18

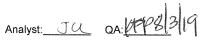
35

73.5

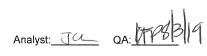


CETIS	S Anal	lytical Repo	ort					-	rt Date: Code:		0	20 (p 1 of 2 3-8657-358
Bivalve	e Larval	Survival and D	evelopmen	t Test				*****		Nautilus	Environ	mental (CA
Analys Analyz		11-3898-7688 02 Aug-19 8:19			Development R Linear Interpola				S Version: ial Results:	CETISv1 Yes	.8.7	
Linear	Interpo	lation Options						*****				
X Tran	sform	Y Transform	Seed	ł	Resamples	Exp 95%	CL Meth	od				
Linear		Linear	1716		1000	Yes		Point Interpo	lation			
Point E	stimate	es										
Level	%	95% LCL	95% UCL	τu	95% LCL	95% UCL						
EC25	>73.5		N/A	<1.361		NA						
EC50	>73.5	N/A	N/A	<1.361		NA						
Develo	pment l	Rate Summary				Calcul	ated Variat	te(A/B)			an a	
C-%	C	ontrol Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	А	в
0	В	rine Control	5	0.9626	0.948	0.9779	0.004749	0.01062	1.1%	0.0%	1121	1165
2			5	0.9497	0.9245	0.9738	0.007829	0.01751	1.84%	1.34%	1101	1159
4			5	0.9556	0.9322	0.9744	0.007254	0.01622	1.7%	0.72%	1077	1127
9			5	0.9632	0.9542	0.9714	0.002885	0.006451	0.67%	-0.07%	1153	1197
18			5	0.9694	0.9628	0.9807	0.003107	0.006948	0.72%	-0.72%	1156	1192
35			5	0.9602	0.9486	0.966	0.003232	0.007228	0.75%	0.24%	1145	1192
73.5			5	0.9528	0.9494	0.9595	0.001778	0.003975	0.42%	1.02%	1126	1182
Graphi	1.0 0.9	•••••	•••••		•							

C-%



CETIS	S Anal	ytical Repo	ort					-	ort Date: Code:		-	20 (p 2 of 2 3-8657-358
Bivalve	e Larval	Survival and D	evelopmen	t Test				**************************************		Nautilus	s Environi	mental (CA
Analys Analyz		10-1515-3673 02 Aug-19 8:20		point: lysis:	Survival Rate Linear Interpola	tion (ICPIN)			S Version: ial Results:	CETISv1 Yes	.8.7	
Linear	Interpol	ation Options						in soud-ser, Tarlanda an Annaise Lan	tara mangungi yang gingga antakisi iki kiti kiti kiti kata mang			Sofelineanar in an anns an an Air
X Trans	sform	Y Transform	See	d	Resamples	Exp 95%	CL Meth	od				
Linear		Linear	1423	3709	1000	Yes	Two-	Point Interp	olation			
Point E	Estimate	S										
Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL						
EC25	>73.5	N/A	N/A	<1.36	1 NA	NA	·····					
EC50	>73.5	N/A	N/A	<1.36	1 NA	NA						
Surviva	al Rate S	Summary				Calcul	lated Varia	te(A/B)				
C-%	C	ontrol Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	в
0	Br	rine Control	5	0.9862	2 0.9569	1	0.008876	0.01985	2.01%	0.0%	1144	1160
2			5	0.9698	8 0.9138	1	0.01536	0.03435	3.54%	1.66%	1125	1160
4			5	0.9664	4 0.9052	1	0.01748	0.03908	4.04%	2.01%	1121	1160
9			5	1	1	1	0	0	0.0%	-1.4%	1160	1160
18			5	0.9819	9 0.9267	1	0.01419	0.03173	3.23%	0.44%	1139	1160
35			5	0.9776	6 0.9224	1	0.01532	0.03427	3.51%	0.87%	1134	1160
73.5			5	0.975	0.9181	1	0.01649	0.03688	3.78%	1.14%	1131	1160
Graphi	1.0 0.9 0.9 0.8 0.7 0.5 0.5 0.4 0.3 0.2 0.1		•		•							
	0.0 <sup>E</sup> 0	10 20	30 40		60 70 80							
			C-%									



### **CETIS Test Data Worksheet**

### **Bivalve Larval Survival and Development Test**

Nautilus Environmental (CA)
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art Date: nd Date: ample Date:	12 .	lul-19 lul-19 lul-19			Mytilus galloprovi EPA/600/R-95/13 Effluent Sample			Sample Code: Sample Source: Sample Station:	
C-%	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal		Notes
			181			254	246	Ja	7/19/19
			182			213	203	<b>B</b>	
			183			249	241		
			184			267	254		
			185			266	252		
			186			225	216		
			187			252	242		
			188			212	196		
			189			240	229		
			190			221	219		
			191			215	207		
			192			282	272		
			193			224	216		
			194			228	220		
			195			262	249		
			196			236	229		
			197			237	229		
			198			222	213		
			199			214	203		
			200			223	212		
			201			210	199		
			202			222	214		
			203			229	223		
			204			250	237		
			205			222	214		4
			206			229	218	JU 7	126/19
			207			233	224		
			208			245	738		
			209			234	225		
			210			234	225		
			211			234	228		
			212			236	220		
			213			237	228		
			214			235	227		
			215			237	227		
			216			226	221		
			217			243	231		
			218			241	233		
			219			259	254		
			220			237	225		4

Analyst: JU QA: 1478/1/19

### **CETIS Test Data Worksheet**

Report Date: 06 Jul-19 14:59 (p 1 of 1) Test Code: 1907-5060 03-8657-3583/170AA50F

Start Date: End Date: Sample Date:	12 J	ul-19 ul-19 ul-19			Mytilus galloprovi EPA/600/R-95/13 Effluent Sample			Sample Code: 19- 0구식 <u>2</u> Sample Source: Jacobs Sample Station: Wyckoff
C-%	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal	Notes
0	BC	1	209			242	233	
0	BC	2	207			410	005	AC7/12/19
0	BC	3	216					
0	BC	4	202					
0	BC	5	204					
0	LC	1	183					
0	LC	2	197					
0	LC	3	187			251	242	-T1 7100 100
0	LC	4	195			001	040	JU 7/25719
0	LC	5	190					
2		1	203					
2		2	200					
2		-	206					
2		4	185					
2		5	188					
4		1	211					
4		2	212					
4		3	205					
4		4	186					
4		5	201					
9		1	210			010	20-	
9		2	189			240	232	JU 7/25/19
9		3	218					
9		4	213					
9		5	208					
18		1	191					
18		2	181					
18		3	196					
18		4	219					
18		5	194					
35		1	192					
35		2	214					
35		3	193					
35		4	215					
35		5	199					
a ad		1	220					
72.3		2	217					
72.3		3	184					
72.3		4	198			220	211	min aller lia
72.3		5	182			220	211	JU 7/25/19
			_	c= Eq			<u> </u>	

## Water Quality Measurements

Client:	Jacobs	т
Sample ID:	Wyckoff	Star
Sample Log No.:	19-0742	Enc
Test No.:	1907-5060	

 Test Species:
 M. galloprovincialis

 Start Date/Time:
 7/10/2019
 \5\5

 End Date/Time:
 7/12/2019
 \535

Concentration		Salinity		Т	emperatu	re	Diss	solved Ox	ygen		рН	
(% sample)		(ppt)			(°C)			(mg/L)			(pH units	·
	0	24	48	0	24	48	0	24	48	0	24	48
Lab Control	30.1	30.1	30.0	15.8	14.9	15.0	8.2	8.1	8,2	8.60	8.00	7.98
Brine Control	30.6	30.4	30.6	16.0	14.8	14.8	7.8	8.1	8.3	8.18	8.09	8.04
2	30,3	36.3	30.3	15.7	14.8	14.9	8.1	8,0	8.4	8.60	8.00	8.01
4	29.90	30.1	30.3	15.7	14.8	14.9	8.2	8.1	8.4	797	7.99	8.01
9	30,3	30.Z	36.3	15.7	14.7	14.9	8,2	8.1	8.3	7.91	7.98	8.05
18	303	30.3	30.3	15.6	14.9	15.0	8.3	8.0	ર.પ	7.83	8.01	8.10
35	30.3	30.3	30,3	15.F	14.8	15,1	8.6	8.1	S' 3	7.74	8.02	8.13
73.5	30,7	307	30.7	15.8	14.8	15.0	7.9	8.0	8.3	7,65	8.03	3.21
		·										

Technician Initials:	0 24 WQ Readings: کو کی کی Dilutions made by: کو ک			
Comments:	0 hrs: <u>ଜ୍</u> ଚାନ୍ତ ହାନ୍ତ ବ୍ରାଡୀଜ୍ୟ 24 hrs: 48 hrs:	 		
QC Check:	PTP S/1/19		Final Review:	EG 8/9/19

Nautilus Environmental. 4340 Vandever Avenue. San Diego, CA 92120.

### **Brine Dilution Worksheet**

Project:	JACOBS		_	Analyst:	EG	
Sample ID:	Wyckoff		_	Test Date:	7/10/2019	
Test No:	1907-S 660		-	Test Type:	Mussel Developm	ent
Salinity of Effl	uent	7.0	_			
Salinity of Brir	ie	93.8	_ Date	of Brine used:	6/11/2019	-
Target Salinity	,	30	Alkalinity of	f Brine Control:	(À)	_ mg/L as CaCO3
Test Dilution V	/olume	250	_		0	
		Effluent	Brine Control			
Salinity Adjust (TS - SE)/(SB - TS = target	TS) = salinity	0.36	0.47			
SE = Salinit	y of effluent					

SB = salinity of brine

Concentration %	Effluent Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
Control	NA	NA	NA	250
2	5.0	0.36	1.8	250
4	10.0	0.36	3.6	250
9	22.5	0.36	8.1	250
18	45.0	0.36	16.2	250
35	87.5	0.36	31.5	250
73.5	183.8	0.36	66.2	250

	DI Volume			
Brine Control	140.9	0.47	66.2	250
( ou lut out	Mar Attal	and the set		

(Alkalinky not meaned, technica emor.

Total Brine Volume Required (ml): 193.8

QC Check: 17 8/1/19

Final Review: E4 8 9 19

Nautilus Environmental. 4340 Vandever Avenue. San Diego, CA 92120.

### Larval Development Worksheet

)

Client:	Jacobs-Wyckoft
Test No.:	1907-5060
Test Species:	Mytilus galloprovincialis
Animal Source:	Missien Decer
Date Received:	5129/19 013A+B combined tank
Test Chambers:	30 mL glass shell vials
Sample Volume:	10 mL

N40

Start Date/Time:	7/10/2019	515	
End Date/Time:	7/12/2019	1535	
Technician Initials:	ACLEG		

#### Spawn Information

First Gamete Release Time:

#### **Gamete Selection**

Sex	Number Spawning
Male	6
Female	2 MB. ) (at

Sex	Beaker Number(s)	Condition (sperm motility, egg density, color, shape, etc.)
Male	1	good densities motile tim
Female 1	MBI	good density + color U
Female 2	mB2	Vight color, goozt demante churn
Female 3	Catl	Verywange, excellent dinst

Egg Fertilization Time: 1230

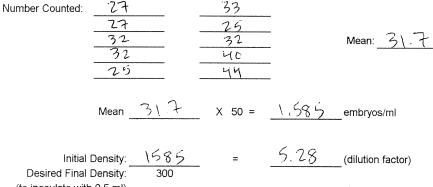
#### **Embryo Stock Selection**

Stock Number	% of embryos at 2-cell division stage
Female 1	100
Female 2	90
Female 3	80

### Stock(s) chosen for testing:

#### Embryo Inoculum Preparation

Target count on Sedgwick-Rafter slide for desired density is 6 embryos



(to inoculate with 0.5 ml)

When mean percent dividing is ≥ 90, prepare the embryo inoculum according to the calculated dilution factor. For example, if the dilution factor is 2.25, use 100 ml of existing stock (1 part) and 125 ml of dilution water (1.25 parts).

#### **Time Zero Control Counts**

Rep	No. Dividing	Total	% Dividing	Mean % Dividing
TØ A	019	219	100	
TØ B	261	261	(00)	
TØC	232	232	100	
TØ D	211	211	100	100
TØ E	233	233	100	
TØ F	238	238	100	
	x=232.	•		

48-h QC: 229/236 = 97 / 2

Comments:

QC Check:

Nautilus Environmental. 4340 Vandever Avenue. San Diego, CA 92120.

Final Review: 45/19

Appendix B Sample Check-In Information

Client:	Jacobs	
Sample ID:	WYCKoff	(070919)
Test ID No(s).:	1907-5060	( )

Nautilus Environmental

Sample Check-In Information

4340 Vandever Avenue	Sample ID:	WYCKO.	ff (0-	10919)		
San Diego, CA 92120	Test ID No(s).:	1907-5	6060		Sample Description:	
					A: no color, clear, no o	dor no debri
s	ample (A, B, C):					
	in No. (19-xxxx): 0742				3	
Sample Collection						
	ipt Date & Time: 71014 0945				COC Complete (Y/N)? A B C	
Number of Containers &	7 10 17 01 10				A B C	
Approx. Total Volun					Filtration? Y	
	nperature (°C) 3-8				0	
	nperature OK? <sup>1</sup> (Ŷ) N	YN	YN	Y N	Pore Size:	
	DO (mg/L)				Organisms or Deb	oris
	pH (units)				Salinity Adjustment	
Conduc	ctivity (µS/cm)					- Target ppt: 30
	Salinity (ppt) 7.0				Test: Source:	
A	Ikalinity (mg/L) <sup>2</sup> $395$					Target ppt:
	dness (mg/L) <sup>2, 3</sup>				Test: Source: pH Adjustment? Y N	Target ppt:
	hlorine (mg/L) 0.02					в с
	nician Initials KL	1			Initial pH:	
			1		Amount of HCl added:	
Test Performed: MUSSEl	Dave la conte					
Test Performed: 10554	Developmen Control/Dilution Wa	Hardness or Sa	V / Lab ART C	otner:	Final pH: Cl <sub>2</sub> Adjustment? Y N	
Additional Cont	rol? (V) N = Brine Catrol	Ikalinity:	Hardness or Sa	linity: 30 ent		в с
				<u> </u>	Initial Free Cl <sub>2</sub> :	
Test Performed:	Control/Dilution Wa	ater: 8:2 / Lab SV	V / LabART C	)ther:	STS added:	
		Hardness or Sal			Final Free Cl <sub>2</sub> :	
Additional Conti	rol? Y N =/					
					Sample Aeration? Y (N)	
Test Performed:	Control/Dilution Wa	iter: 8:2 / Lab SV	/ LabART C	other:	A	B C
	Alkalinity:	Hardness or Sal	inity:		Initial D.O.	
Additional Conti	rol? <u>Y N</u> = /	Alkalinity:	Hardness or Sa	linity:	Duration & Rate	
					Final D.O.	
_	of sample should be 0-6°C, if received			).		~
<sup>2</sup> mg/L as CaC	O3, <sup>3</sup> Measured for freshwater sample	s only, NA = Not Appli	cable		Subsamples for Additional Chemistry	Required? 🖤 N
Additional Comments: (A) Al Walind	at bar another I.	Mergin 1 1.1.	ah 04 A 4		NH3 Other	
	ig vi prime construit with		an evilor.		Tech Initials A <u>H</u> _ BC_	
			·		QC Check:	DEP 8/1/19
			·		Final Review:	OUCTIG

### **Total Ammonia Analysis** Freshwater

Clien	t: JACOBS				
Projec	t: Wyckoff				
Test Type	e: Mussel Deve	elopment			
	(: <u>0.</u> 0			Analyst:	
Test Start Date	e: <u>//10/2019</u>		Ar	nalysis Date:	
Sample ID	Nautilus ID	Sub-Sample Date	Test Day	NH3-N (mg/L)	N x 1.22 Ammonia (mg/L)
Blank Spike (10 mg/L NH <sub>3</sub> )		NA	NA	7,4	9.0
Wyckoff	19-0742	7/10/2019	Check In	1.3	1.6
Spike Check (10 mg/L NH3)		NA	NA		
Batch QA Sample	19-0773	7/16/19	NIA	36.0	43.9
Sample Duplicate <sup>a</sup>	Batch QA	NA	NA	36.3	44.3
Sample Duplicate + Spike <sup>a</sup>		NA	NA	43.5	53.1
Spike Check (10 mg/L NH3)		NA	NA	-	

<u>Relative Percent Difference (RPD) = [sample] (mg/L) - [sample duplicate] (mg/L)</u> x 100 [average ammonia] (mg/L)

Acceptable Range: 0-20%

<u>Percent Recovery = [spiked sample] (mg/L) - [sample] (mg/L)</u> x 100 nominal [spike] (mg/L)

Acceptable Range: 80-120%<sup>b</sup>

QC Sample ID	[NH <sub>3</sub> ]	[Sample Dup]	Measured [Spike]	Nominal [Spike]	RPD	% Recovery
Blank	0.0	NA	9.0	10	NA	90
Eatch QA (Maasdoom)	43.9	44.3	53.1	10	0.9	92
O18 208/1/10	1					

Comments:	
oonnento.	

Notes: "Unless otherwise noted, the last sample listed on the datasheet is used for duplicate and duplicate + spike QC check.

<sup>b</sup> Acceptable range for % recovery applies only to the blank spike. Spike recoveries in samples may vary based on sample matrix and are for information only.

<sup>c</sup> Calculation not performed due to one or both values below the method detection limit.

Method Detection Limit = 0.5 mg/L

AC 8/1/19 QC Check:

Final Review:

KTP 8/1/19

Appendix C Chain-of-Custody Form

#### Page 1 of 1

### Enthalpy Analytical (REGION COPY)

DateShipped: 7/9/2019

CarrierName: FedEx

AirbillNo: 7756 6640 3707

## CHAIN OF CUSTODY RECORD

### No: 10-070919-102015-0381

Project Code: WEH-029D Cooler #: 1 of 1

2019T10P000DD210W2LA00 Contact Name: Keith Allers Contact Phone: 206-780-1711

Sample Identifier	CLP Sample No.	Matrix/Sampler	Coll. Method	Analysis/Tur (Days	naround ;)	Tag/Prese	rvative/Bottles	Location		llection te/Time	Sample Type	
070919	• • • • • • • • • • • • • • • • • • •	Ground Water/ K.Allers	Composite	CHRTOX(8	Weeks)	(<	6 C) (1)	SP-11	07/09/	2019 10:00	Field Sample	3.8
<ul> <li>1 • • • • • • • • • • • • • • • • • • •</li></ul>	· · · · · · · · · · · · · · · · · · ·				· · · · · · · · · · · · · · · · · · ·							
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PERSON DEPENDENCE IN A DESIGNATION OF THE DESIGNATI	in the state of th	aan naamaan ahaa ahaa ahaa ahaa ahaa aha						Shipment for C	Case Com	lete? N		
Special Instructions	:							Samples Trans			Custody #	
Analysis Key: CHR	TOX=Chronic T	oxicity							······································	· · · · · ·	  - 	
Items/Reason	Relinquished b	y (Signature and O	rganization)	Date/Time	Received	by (Signature	and Organization)	) Date/Tim	ne Sam	nple Conditio	n Upon Receipt	
	1 /	the JAC	abs	7-9-2019 1025	1/2	serface	Noutitus	7/10/1 0945	9	good	<ul> <li>Projektovoste sette statovos</li> </ul>	
	and a share and an and an and a share and											
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Labt: 19-0742

Wyckoff Eagle Harbor GWTP 2019/WA

Appendix D Reference Toxicant Test Results **Bivalve Larval Development Test** 

30 Jul-19 11:41 (p 1 of 3) 190710msdv | 02-0190-4206

Bivalve Larval	l Survival and Developr	nent Test						Nautilus Environmental (CA
Batch ID: Start Date: Ending Date: Duration:	10 Jul-19 15:15	Fest Type: Protocol: Species: Source:	Development- EPA/600/R-95 Mytilus gallopr Mission Bay	/136 (1995)				iluted Natural Seawater lot Applicable
Sample ID: Sample Date: Receive Date: Sample Age:	10 Jul-19 I 10 Jul-19 S	Code: Material: Source: Station:	190710msdv Copper chlorid Reference Tox Copper Chlorid	ricant			Client: Ir Project:	nternal
Comparison S	Summary							
Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	τu	Metho	1
14-6066-2134 10-2312-9373 02-2288-3584	Combined Developmen Development Rate Survival Rate	t Ra 5 5 10	10 10 20	7.071 7.071 14.14	5.24% 1.95% 6.49%		Dunnet	t Multiple Comparison Test t Multiple Comparison Test t Multiple Comparison Test
Point Estimate	e Summarv							
Analysis ID	Endpoint	Level	µg/L	95% LCL	95% UCL	τu	Metho	ł
08-2094-6054	Combined Developmen	t Ra EC25 EC50	6.361	6.053 7.579	6.458 7.898		Linear	nterpolation (ICPIN)
02-8661-6124	Development Rate	EC25 EC50		6.239 7.692	6.452 7.885		Linear	nterpolation (ICPIN)
18-9463-9841	Survival Rate	EC25 EC50		19.48 27.42	25.51 31.18		Linear	nterpolation (ICPIN)
Test Acceptab	bility							
Analysis ID	Endpoint	Attrib	ute	Test Stat	TAC Limi	ts	Overla	p Decision
02-8661-6124 10-2312-9373	Development Rate Development Rate Survival Rate	Contro	ol Resp ol Resp ol Resp	0.9707 0.9707 0.9716	0.9 - NL 0.9 - NL 0.5 - NL		Yes Yes Yes	Passes Acceptability Criteria Passes Acceptability Criteria Passes Acceptability Criteria
02-2288-3584 18-9463-9841 14-6066-2134	Survival Rate Combined Developmen		ol Resp	0.9716	0.5 - NL		Yes	Passes Acceptability Criteria

Analyst: Ja QA: 1998/1119

C-µg/L

0

5

10

20

40

2.5

**Bivalve Larval Survival and Development Test** 

Count

5

5

5

5

5

5

Mean

0.9432

0.9532

0.9369

0.1057

0

0

95% LCL

0.8937

0.9255

0.8814

0

0

0.07905

95% UCL Min

0.8836

0.9224

0.8793

0.0819

0

0

0.9927

0.9924

0.1324

0

0

0.981

**Combined Development Rate Summary** 

Lab Control

**Control Type** 

### 000-089-187-4

Development Rate Summary											
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Мах	Std Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.9707	0.955	0.9865	0.9515	0.9872	0.005686	0.01271	1.31%	0.0%
2.5		5	0.9691	0.9587	0.9795	0.9607	0.9828	0.003752	0.008391	0.87%	0.17%
5		5	0.9598	0.9368	0.9828	0.9409	0.9835	0.00829	0.01854	1.93%	1.13%
10		5	0.1118	0.08853	0.135	0.09314	0.1357	0.008373	0.01872	16.75%	88.49%
20		5	0	0	0	0	0	0	0		100.0%
40		5	0	0	0	0	0	0	0		100.0%
Survival Rat	e Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.9716	0.9254	1	0.9095	1	0.01662	0.03715	3.82%	0.0%
2.5		5	0.9836	0.9593	1	0.9526	1	0.00877	0.01961	1.99%	-1.24%
5		5	0.9759	0.9333	1	0.9267	1	0.01534	0.03429	3.51%	-0.44%
10		5	0.9414	0.887	0.9958	0.8793	1	0.01959	0.04381	4.65%	3.11%
20		5	0.8509	0.7151	0.9867	0.7414	0.9741	0.04891	0.1094	12.85%	12.42%
40		5	0.1034	0.08784	0.1191	0.08621	0.1207	0.00562	0.01257	12.15%	89.35%
Combined D	evelopment Rate	Detail				-					
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
0	Lab Control	0.9701	0.944	0.931	0.8836	0.9872			· · · · · · · · · · · · · · · · · · ·		
2.5		0.9483	0.9224	0.9644	0.9828	0.9483					
5		0.8793	0.9835	0.9409	0.9052	0.9754					
10		0.09483	0.09483	0.0819	0.1293	0.1277					
20		0	0	0	0	0					
40		0	0	0	0	0					
Developmen	nt Rate Detail										

Developmer	Development Rate Detail										
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
0	Lab Control	0.9701	0.9733	0.9515	0.9716	0.9872					
2.5		0.9607	0.9683	0.9644	0.9828	0.9692					
5		0.9488	0.9835	0.9409	0.9502	0.9754					
10		0.1023	0.1	0.09314	0.1357	0.1277					
20		0	0	0	0	0					
40		0	0	0	0	0					
Survival Rate Detail											
Garvivarita	le Detail										
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
		<b>Rep 1</b>	<b>Rep 2</b> 0.9698	<b>Rep 3</b>	Rep 4	<b>Rep 5</b> 1					
C-µg/L	Control Type			•	· · ·	Rep 5 1 0.9784					
<b>С-µg/L</b> 0	Control Type	1	0.9698	•	· · ·	1					
<b>С-µg/L</b> 0 2.5	Control Type	1 0.9871	0.9698 0.9526	0.9784 1	0.9095 1	1 0.9784					
<mark>С-µg/L</mark> 0 2.5 5	Control Type	1 0.9871 0.9267	0.9698 0.9526 1	0.9784 1 1	0.9095 1 0.9526	1 0.9784 1					

Test Code:

Max

0.9872

0.9828

0.9835

0.1293

0

0

**Report Date:** 

Std Err

0.01783

0.009599

0.01

0.02

0

0

Std Dev

0.03986

0.02236

0.04472

0.02146

0

0

%Effect

0.0%

-1.07%

0.67%

88.79%

100.0%

100.0%

Nautilus Environmental (CA)

CV%

4.23%

2.35%

4.77%

20.31%

**Bivalve Larval Survival and Development Test** 

Report Date:	
Test Code:	

30 Jul-19 11:41 (p 3 of 3)

190710msdv | 02-0190-4206

Nautilus	Environmental (CA)

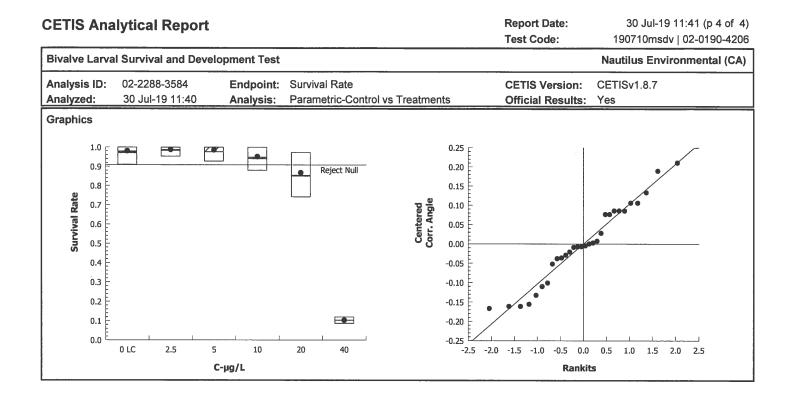
Combined	Development Rate	Binomials					
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Lab Control	227/234	219/232	216/232	205/232	231/234	
2.5		220/232	214/232	244/253	229/233	220/232	
5		204/232	239/243	239/254	210/232	238/244	
10		22/232	22/232	19/232	30/232	30/235	
20		0/232	0/232	0/232	0/232	0/232	
40		0/232	0/232	0/232	0/232	0/232	
Developme	ent Rate Binomials	;					
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Lab Control	227/234	219/225	216/227	205/211	231/234	
2.5		220/229	214/221	244/253	229/233	220/227	
5		204/215	239/243	239/254	210/221	238/244	
10		22/215	22/220	19/204	30/221	30/235	
20		0/198	0/226	0/172	0/172	0/219	
40		0/20	0/25	0/28	0/24	0/23	
Survival Ra	ate Binomials		4				
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Lab Control	232/232	225/232	227/232	211/232	232/232	
2.5		229/232	221/232	232/232	232/232	227/232	
5		215/232	232/232	232/232	221/232	232/232	
10		215/232	220/232	204/232	221/232	232/232	
20		198/232	226/232	172/232	172/232	219/232	
40		20/232	25/232	28/232	24/232	23/232	



CETIS Ar	nalytical Repo	ort						ort Date: Code:			1 (p 1 of 4) -0190-4206
Bivalve Lar	val Survival and [	Developme	ent Test						Nautilus	Environn	nental (CA)
Analysis ID Analyzed:	: 14-6066-2134 30 Jul-19 11:3		•	mbined Deve rametric-Cor	•			IS Version: ial Results:	CETISv1. Yes	8.7	
Data Transf	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Co	rrected)	NA	C > T	NA	NA		5.24%	5	10	7.071	
Dunnett Mu	Itiple Compariso	n Test									
Control	vs C-µg/L		Test Stat	Critical	MSD DF	P-Value	Р-Туре	Decision(	α:5%)		
Lab Control	2.5		-0.3336	2.227	0.104 8	0.8529	CDF	•	ficant Effect		
	5		0.2539	2.227	0.104 8	0.6511	CDF	-	ficant Effect		
	10*		21.7	2.227	0.104 8	<0.0001	CDF	Significant	t Effect		
ANOVA Tab											
Source	Sum Squ	lares	Mean Sq	uare	DF	F Stat	P-Value	Decision(			
Between Error	3.858434 0.087157	06	1.286145 0.0054473	316	3 16	236.1	<0.0001	Significant	tEffect		
Total	3.945591	00	0.000447	510	19						
Distributior	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Bartlett Equality of Variance			4.066	11.34	0.2544	Equal Var				
Distribution	Shapiro-Wilk W Normality			0.9628	0.866	0.6022	Normal D	istribution			
Combined I	Development Rate	e Summar	y								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.9432	0.8937	0.9927	0.944	0.8836	0.9872	0.01783	4.23%	0.0%
2.5		5	0.9532	0.9255	0.981	0.9483	0.9224	0.9828	0.01	2.35%	-1.07%
5 10		5 5	0.9369 0.1057	0.8814 0.07905	0.9924 0.1324	0.9409 0.09483	0.8793 0.0819	0.9835 0.1293	0.02	4.77%	0.67%
20		5	0.1057	0.07905	0.1324	0.09465	0.0819	0.1293	0.009599 0	20.31%	88.79% 100.0%
40		5	0	0	0	0	0	0	0		100.0%
Angular (Co	orrected) Transfor	rmed Sum	marv								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.343	1.232	1.454	1.332	1.223	1.457	0.04002	6.66%	0.0%
2.5		5	1.358	1.289	1.428	1.341	1.289	1.439	0.02504	4.12%	-1.16%
5		5	1.331	1.21	1.452	1.325	1.216	1.442	0.04345	7.3%	0.88%
10 20		5	0.3299	0.2867	0.3731	0.313	0.2902	0.3678	0.01556	10.54%	75.43%
40		5 5	0.03283 0.03283	0.03283 0.03283	0.03283 0.03283	0.03283 0.03283	0.03283 0.03283	0.03283 0.03283	0 0	0.0% 0.0%	97.55% 97.55%
Graphics											
	1.0										
		-				0.12 0.10				• •	
E.	0.9			Reject Null	_	0.08			•	•/	
udo	F					0.06 0.04					
l Devel Rate	0.7					0.04 0.02 0.02 0.00 0.00 0.00 0.00 0.00					
ad D Ra	0.6				ļ	0.00		0000000000			
bine	0.5							6000*			
Corr	0.4					-0.04		/			0.00
	0.3 <del> </del>					-0.08					
	0.2					-0.10					
	0.1					-0.12 -0.14					
	0.0 - 0.0 - 2.5		10		-	-0.16 <sup>E</sup>			Í Í		
	0 LC 2.5	5		20 40		-2.5	5 -2.0 -1.5	-1.0 -0.5 0.		1.5 2.0	2.5
		С-µд	/L					Rank			

CETIS Ana	alytical Repo	ort			Report Date:         30 Jul-19 11:41 (p           Test Code:         190710msdv   02-01						
Bivalve Larva	al Survival and D	evelopmen	t Test						Nautilus	Environn	nental (CA)
Analysis ID: Analyzed:	10-2312-9373 30 Jul-19 11:39		-	elopment R ametric-Cor	ate itrol vs Trea	tments		S Version: ial Results:	CETISv1. Yes	8.7	
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Corre	ected)	NA	C > T	NA	NA		1.95%	5	10	7.071	
Dunnett Mult	iple Comparison	Test									
Control	vs C-µg/L	Treat	Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(	a:5%)		
Lab Control	2.5		0.2821	2.227	0.053 8	0.6393	CDF	•	ficant Effect		
	5		1.198	2.227	0.053 8	0.2595	CDF	-	ficant Effect		
	10*		44.81	2.227	0.053 8	<0.0001	CDF	Significant			
NOVA Table	•										
Source	Sum Squa	ares	Mean Squ	are	DF	F Stat	P-Value	Decision(	α:5%)		
Between	4.140649		1.380216		3	982.3	<0.0001	Significant	Effect	`	
Error	0.0224815	56	0.0014050	98	16	_					
Total	4.163131				19						
Distributiona	l Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision(	α:1%)			
Variances	Bartlett E	11.34	0.5836	Equal Var							
Distribution	Shapiro-V	0.9494	0.866	0.3582	Normal Di	stribution					
Development	t Rate Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
)	Lab Control	5	0.9707	0.955	0.9865	0.9716	0.9515	0.9872	0.005686	1.31%	0.0%
2.5		5	0.9691	0.9587	0.9795	0.9683	0.9607	0.9828	0.003753	0.87%	0.17%
5		5	0.9598	0.9368	0.9828	0.9502	0.9409	0.9835	0.00829	1.93%	1.13%
10		5	0.1118	0.08853	0.135	0.1023	0.09314	0.1357	0.008373	16.75%	88.49%
20		5	0	0	0	0	0	0	0		100.0%
40		5	0	0	0	0	0	0	0		100.0%
	rected) Transfor	med Summ	ary								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.402	1.354	1.45	1.401	1.349	1.457	0.01723	2.75%	0.0%
2.5		5	1.396	1.363	1.428	1.392	1.371	1.439	0.0117	1.88%	0.48%
5		5	1.374	1.311	1.437	1.346	1.325	1.442	0.02274	3.7%	2.03%
10 20		5	0.34	0.3035	0.3766	0.3256	0.3101	0.3773	0.01317	8.66%	75.75%
40		5 5	0.03577 0.1027	0.0329 0.0948	0.03865 0.1106	0.03554 0.1022	0.03327 0.09463	0.03813	0.001035	6.47%	97.45%
		5	0.1027	0.0940	0.1100	0.1022	0.09463	0.112	0.002846	6.2%	92.68%
1.0 0.9 0.8 0.7 0.6 0.5 0.4 0.4 0.3 0.2 0.2 0.1 0.0	0 LC 2.5	та 5 <b>С-µg/L</b>	10 20	Reject Null		0.07 0.06 0.05 0.04 0.03 0.02 0.02 0.01 0.02 0.01 0.02 0.01 0.02 0.03 0.04 0.03 0.04 0.03 0.04 0.03 0.04 0.05 0.04 0.05 0.04 0.05 0.06 0.05 0.04 0.05 0.04 0.05 0.04 0.05 0.04 0.05 0.05	5 -2.0 -1.5	-1.0 -0.5 0. Rank	its		2.5
00-089-187-3	1			C	CETIS™ v1.	8.7.20			Analyst:]	<u>il</u> a	A: 10578/1

CETIS Ana	alytical Rep	ort			•				ıl-19 11:41 (p 3 of  4) nsdv   02-0190-4206			
Bivalve Larv	al Survival and I	Developme	nt Test						Nautilus	Environm	ental (CA)	
Analysis ID: Analyzed:	02-2288-3584 30 Jul-19 11:4			vival Rate ametric-Con	trol vs Treat	ments		S Version: ial Results		CETISv1.8.7 Yes		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU	
Angular (Corr	rected)	NA	C > T	NA	NA		6.49%	10	20	14.14		
Dunnett Mult	tiple Compariso	n Test										
Control	vs C-µg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(α:5%)			
Lab Control	2.5		-0.411	2.362	0.169 8	0.9267	CDF	Non-Signi	ficant Effect			
	5		-0.2831	2.362	0.169 8	0.9035	CDF	Non-Signi	ficant Effect			
	10		1.169	2.362	0.169 8	0.3469	CDF	Non-Signi	ficant Effect			
	20*		3.269	2.362	0.169 8	0.0069	CDF	Significan	t Effect			
	40*		15.48	2.362	0.169 8	<0.0001	CDF	Significan	t Effect			
ANOVA Tabl	le	· · · · · · · · · · · · · · · · · · ·										
Source	Sum Squ	Mean Squ	F Stat	P-Value	Decision	(α:5%)						
Between	4.850606	;	0.9701213		5	76.12	<0.0001	Significan	t Effect			
Error	0.305881	5	24			•						
Total	5.156487		29	-								
Distributiona	al Tests											
Attribute	Test			Test Stat	Critical	P-Value	Decision(	α:1%)				
11					45.00			,				
Variances	Bartlett Equality of Variance Shapiro-Wilk W Normality			11.19	15.09	0.0476	Equal Var	iances				
Variances Distribution				0.9592	15.09 0.9031	0.0476 0.2958	Equal Var Normal Di					
	Shapiro-						•					
Distribution	Shapiro-						•		Std Err	CV%	%Effect	
Distribution Survival Rat	Shapiro- te Summary	Wilk W Nor	mality	0.9592	0.9031	0.2958	Normal Di	stribution	<b>Std Err</b> 0.01662	<b>CV%</b> 3.82%	<b>%Effect</b> 0.0%	
Distribution Survival Rat C-µg/L	Shapiro- te Summary Control Type	Wilk W Nor	mality Mean	0.9592 95% LCL	0.9031 95% UCL	0.2958 Median	Normal Di Min	stribution Max				
Distribution Survival Rate C-µg/L 0	Shapiro- te Summary Control Type	Wilk W Nor Count	Mean 0.9716	0.9592 95% LCL 0.9254	0.9031 95% UCL 1	0.2958 Median 0.9784	Normal Di Min 0.9095	stribution Max 1	0.01662	3.82%	0.0%	
Distribution Survival Rate C-µg/L 0 2.5	Shapiro- te Summary Control Type	Count 5 5	Mean 0.9716 0.9836	0.9592 95% LCL 0.9254 0.9593	0.9031 95% UCL 1 1	0.2958 Median 0.9784 0.9871	Normal Di Min 0.9095 0.9526	Stribution Max 1 1	0.01662 0.00877	3.82% 1.99%	0.0% -1.24%	
Distribution Survival Rate C-µg/L 0 2.5 5	Shapiro- te Summary Control Type	Count 5 5 5 5	Mean 0.9716 0.9836 0.9759	0.9592 95% LCL 0.9254 0.9593 0.9333	0.9031 95% UCL 1 1 1	0.2958 Median 0.9784 0.9871 1	Normal Di Min 0.9095 0.9526 0.9267	stribution Max 1 1 1	0.01662 0.00877 0.01534	3.82% 1.99% 3.51%	0.0% -1.24% -0.44%	
Distribution Survival Rate C-µg/L 0 2.5 5 10	Shapiro- te Summary Control Type	Count 5 5 5 5 5 5	Mean 0.9716 0.9836 0.9759 0.9414	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887	0.9031 95% UCL 1 1 1 0.9958	0.2958 Median 0.9784 0.9871 1 0.9483	Normal Di Min 0.9095 0.9526 0.9267 0.8793	stribution Max 1 1 1 1	0.01662 0.00877 0.01534 0.01959	3.82% 1.99% 3.51% 4.65%	0.0% -1.24% -0.44% 3.11%	
Distribution Survival Rat C-µg/L 0 2.5 5 10 20 40	Shapiro- te Summary Control Type	Count 5 5 5 5 5 5 5 5 5 5	Mean 0.9716 0.9836 0.9759 0.9414 0.8509 0.1034	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887 0.7151	0.9031 <b>95% UCL</b> 1 1 0.9958 0.9867	0.2958 Median 0.9784 0.9871 1 0.9483 0.8534	Normal Di Min 0.9095 0.9526 0.9267 0.8793 0.7414	Stribution           Max           1           1           1           0.9741	0.01662 0.00877 0.01534 0.01959 0.04891	3.82% 1.99% 3.51% 4.65% 12.85%	0.0% -1.24% -0.44% 3.11% 12.42%	
Distribution Survival Rat C-µg/L 0 2.5 5 10 20 40	Shapiro- te Summary Control Type Lab Control rrected) Transfo Control Type	Count 5 5 5 5 5 5 5 5 7 med Sumr Count	Mean 0.9716 0.9836 0.9759 0.9414 0.8509 0.1034 mary Mean	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887 0.7151 0.08784 95% LCL	0.9031 95% UCL 1 1 1 0.9958 0.9867 0.1191 95% UCL	0.2958 Median 0.9784 0.9871 1 0.9483 0.8534 0.1034 Median	Normal Di Min 0.9095 0.9526 0.9267 0.8793 0.7414 0.08621 Min	Max           1           1           1           0.9741           0.1207	0.01662 0.00877 0.01534 0.01959 0.04891 0.00562 Std Err	3.82% 1.99% 3.51% 4.65% 12.85% 12.15%	0.0% -1.24% -0.44% 3.11% 12.42% 89.35%	
Distribution Survival Rate C-µg/L 0 2.5 5 10 20 40 Angular (Con C-µg/L 0	Shapiro- te Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 5 7 7 med Sum	Mean 0.9716 0.9836 0.9759 0.9414 0.8509 0.1034 mary	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887 0.7151 0.08784	0.9031 95% UCL 1 1 1 0.9958 0.9867 0.1191 95% UCL 1.573	0.2958 Median 0.9784 0.9871 1 0.9483 0.8534 0.1034	Normal Di Min 0.9095 0.9526 0.9267 0.8793 0.7414 0.08621	Max           1           1           1           0.9741           0.1207	0.01662 0.00877 0.01534 0.01959 0.04891 0.00562	3.82% 1.99% 3.51% 4.65% 12.85% 12.15%	0.0% -1.24% -0.44% 3.11% 12.42% 89.35%	
Distribution Survival Rate C-µg/L 0 2.5 5 10 20 40 Angular (Con C-µg/L	Shapiro- te Summary Control Type Lab Control rrected) Transfo Control Type	Count 5 5 5 5 5 5 5 5 7 med Sumr Count	Mean 0.9716 0.9836 0.9759 0.9414 0.8509 0.1034 mary Mean	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887 0.7151 0.08784 95% LCL	0.9031 95% UCL 1 1 1 0.9958 0.9867 0.1191 95% UCL 1.573 1.56	0.2958 Median 0.9784 0.9871 1 0.9483 0.8534 0.1034 Median	Normal Di Min 0.9095 0.9526 0.9267 0.8793 0.7414 0.08621 Min	Max           1           1           1           0.9741           0.1207	0.01662 0.00877 0.01534 0.01959 0.04891 0.00562 Std Err	3.82% 1.99% 3.51% 4.65% 12.85% 12.15%	0.0% -1.24% -0.44% 3.11% 12.42% 89.35%	
Distribution Survival Rate C-µg/L 0 2.5 5 10 20 40 Angular (Con C-µg/L 0	Shapiro- te Summary Control Type Lab Control rrected) Transfo Control Type	Count 5 5 5 5 5 5 5 5 5 7 rmed Sum Count 5	Mean           0.9716           0.9836           0.9759           0.9414           0.8509           0.1034           mary           Mean           1.432	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887 0.7151 0.08784 95% LCL 1.291	0.9031 95% UCL 1 1 1 0.9958 0.9867 0.1191 95% UCL 1.573	0.2958 Median 0.9784 0.9871 1 0.9483 0.8534 0.1034 Median 1.423	Normal Di Min 0.9095 0.9526 0.9267 0.8793 0.7414 0.08621 Min 1.265	Max           1           1           1           0.9741           0.1207           Max           1.538	0.01662 0.00877 0.01534 0.01959 0.04891 0.00562 Std Err 0.05081	3.82% 1.99% 3.51% 4.65% 12.85% 12.15% CV% 7.93%	0.0% -1.24% -0.44% 3.11% 12.42% 89.35% %Effect 0.0%	
Distribution Survival Rate C-µg/L 0 2.5 5 10 20 40 Angular (Con C-µg/L 0 2.5	Shapiro- te Summary Control Type Lab Control rrected) Transfo Control Type	Count           5	Mean           0.9716           0.9836           0.9759           0.9414           0.8509           0.1034           mary           Mean           1.432           1.462	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887 0.7151 0.08784 95% LCL 1.291 1.363	0.9031 95% UCL 1 1 1 0.9958 0.9867 0.1191 95% UCL 1.573 1.56	0.2958 Median 0.9784 0.9871 1 0.9483 0.8534 0.1034 Median 1.423 1.457	Normal Di Min 0.9095 0.9526 0.9267 0.8793 0.7414 0.08621 Min 1.265 1.351	Max           1           1           1           0.9741           0.1207           Max           1.538           1.538	0.01662 0.00877 0.01534 0.01959 0.04891 0.00562 <b>Std Err</b> 0.05081 0.03557	3.82% 1.99% 3.51% 4.65% 12.85% 12.15% <b>CV%</b> 7.93% 5.44%	0.0% -1.24% -0.44% 3.11% 12.42% 89.35% %Effect 0.0% -2.05%	
Distribution Survival Rate 0 2.5 5 10 20 40 Angular (Con C-µg/L 0 2.5 5 5	Shapiro- te Summary Control Type Lab Control rrected) Transfo Control Type	Count           5	Mean           0.9716           0.9836           0.9759           0.9414           0.8509           0.1034           mary           Mean           1.432           1.452	0.9592 95% LCL 0.9254 0.9593 0.9333 0.887 0.7151 0.08784 95% LCL 1.291 1.363 1.305	0.9031 95% UCL 1 1 1 0.9958 0.9867 0.1191 95% UCL 1.573 1.56 1.6	0.2958 Median 0.9784 0.9871 1 0.9483 0.8534 0.1034 Median 1.423 1.457 1.538	Normal Di Min 0.9095 0.9526 0.9267 0.8793 0.7414 0.08621 Min 1.265 1.351 1.297	Max           1           1           1           0.9741           0.1207           Max           1.538           1.538           1.538           1.538	0.01662 0.00877 0.01534 0.01959 0.04891 0.00562 <b>Std Err</b> 0.05081 0.03557 0.05312	3.82% 1.99% 3.51% 4.65% 12.85% 12.15% <b>CV%</b> 7.93% 5.44% 8.18%	0.0% -1.24% -0.44% 3.11% 12.42% 89.35% <b>%Effect</b> 0.0% -2.05% -1.41%	



CETIS	S Anal	ytical Repo	ort					-	ort Date: Code:			:41 (p 1 of 3 )2-0190-420
Bivalve	e Larval	Survival and D	evelopmen	t Test						Nautilu	s Environ	mental (CA
Analysis ID:         08-2094-6054           Analyzed:         30 Jul-19 11:40				point: lysis:	Combined Development Rate Linear Interpolation (ICPIN)				CETIS Version: C Official Results: Y		.8.7	
Linear	Interpol	ation Options										
X Trans	sform	Y Transform	See	d	Resamples	Exp 95%	6 CL Meth	nod				
Linear		Linear	4920	)75	1000	Yes		Point Interp	olation			<u> </u>
Point E	Estimate	S										
Level	µg/L	95% LCL	95% UCL									
EC25	6.361	6.053	6.458									
EC50	7.786	7.579	7.898									
Combined Development Rate Summary Calculated Variate(A/B)												
C-µg/L	C	ontrol Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	в
0	La	ab Control	5	0.9432		0.9872	0.01783	0.03986	4.23%	0.0%	1098	1164
2.5			5	0.9532		0.9828	0.01	0.02236	2.35%	-1.07%	1127	1182
5			5	0.9369		0.9835	0.02	0.04472	4.77%	0.67%	1130	1205
10			5	0.1057	7 0.0819	0.1293	0.009599	0.02146	20.31%	88.79%	122	1163
20			5	0	0	0	0	0		100.0%	0	1160
40			5	0	0	0	0	0		100.0%	0	1160
Graphi	cs											
otel Paradonado Paradonado	1.0 0.9 0.8 0.7 0.6 0.6 0.6 0.4 0.3 0.4 0.3 0.2											

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0.5 0.4 0.3 0.2 0.1

. . . . . . . . . .

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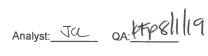
C-µg/L

25

30

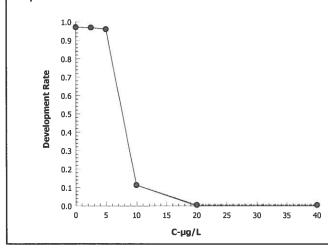
35

10



CETIS	Ana	lytical Repo	rt					•	ort Date: Code:			41 (p 2 of 3 2-0190-4206
Bivalve	Larva	Survival and D	evelopmen	t Test						Nautilu	s Environ	mental (CA)
Analysi Analyze		02-8661-6124 30 Jul-19 11:40		point: lysis:	Development F Linear Interpola		1)		IS Version: cial Results:	CETISv1 Yes	.8.7	
Linear I	nterpo	lation Options										
X Trans	sform	Y Transform	See	t	Resamples	Exp 95%	6 CL Met	nod				
Linear		Linear	1713	316	1000	Yes	Two	Point Interp	olation			
Point E Level EC25 EC50	stimate <u>µg/L</u> 6.368 7.8	95% LCL	<b>95% UCL</b> 6.452 7.885									
Develop	pment	Rate Summary				Calc	ulated Varia	te(A/B)				
C-µg/L	C	ontrol Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	в
0	L	ab Control	5	0.9707	7 0.9515	0.9872	0.005686	0.01271	1.31%	0.0%	1098	1131
2.5			5	0.969	1 0.9607	0.9828	0.003753	0.008392	0.87%	0.17%	1127	1163
5			5	0.959	B 0.9409	0.9835	0.00829	0.01854	1.93%	1.13%	1130	1177
10			5	0.1118		0.1357	0.008373	0.01872	16.75%	88.49%	123	1095
20			5	0	0	0	0	0		100.0%	0	987
40			5	0	0	0	0	0		100.0%	0	120

Graphics



CETIS	S Anal	ytical Repo	ort						-	ort Date: Code:			41 (p 3 of 3 2-0190-420
Bivalve	e Larval	Survival and D	evelopme	nt Test							Nautilu	s Environ	mental (CA
				dpoint: alysis:	Survival Rate Linear Interpola	ation (ICPI	N)			IS Version: cial Results	CETISv1 : Yes	.8.7	
Linear	Interpol	ation Options									<u></u>		
X Trans	sform	Y Transform	See	ed	Resamples	Exp 95%	% CL	Metho	d				
Linear		Linear	158	4768	1000	Yes		Two-P	oint Interp	olation			
Point F	Estimate	s											
Level		95% LCL	95% UCL										
EC25	μg/L 23.15	19.48	25.51	•	· · · · · · · ·								
EC25 EC50	29.69	27.42	25.51 31.18										
Surviva	al Rate S	ummary				Calc	ulated	Variate	e(A/B)				
C-µg/L	Co	ontrol Type	Count	Mean	Min	Max	Std	Err	Std Dev	CV%	%Effect	A	в
0	La	b Control	5	0.971	6 0.9095	1	0.01	662	0.03715	3.82%	0.0%	1127	1160
2.5			5	0.983	6 0.9526	1	0.00	0877	0.01961	1.99%	-1.24%	1141	1160
5			5	0.975	9 0.9267	1	0.01	534	0.03429	3.51%	-0.44%	1132	1160
10			5	0.9414		1	0.01	959	0.04381	4.65%	3.11%	1092	1160
20			5	0.850		0.9741	0.04	891	0.1094	12.85%	12.42%	987	1160
40			5	0.103	4 0.08621	0.1207	0.00	)562	0.01257	12.15%	89.35%	120	1160
Graphi	cs												
Survival Bate	0.9	0-0-0	e										

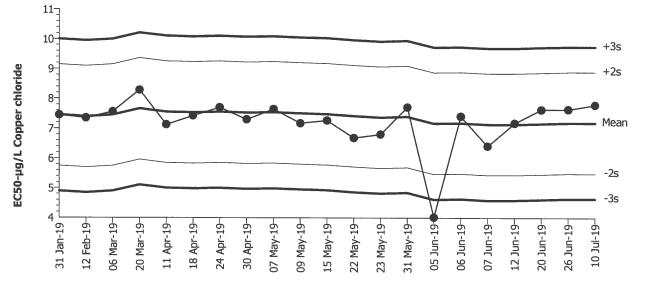
0.3 0.2 0.1

0.0 0 5 10 15 20 25 30 35 40 **C-μg/L** 

### **CETIS QC Plot**

Bivalve Larval Survival and Developmen	Nautilus Environmental (CA)		
Test Type: Development-Survival	Organism: Mytilus galloprovincialis (Bay Mussel	Material:	Copper chloride
Protocol: EPA/600/R-95/136 (1995)	Endpoint: Combined Development Rate	Source:	Reference Toxicant-REF





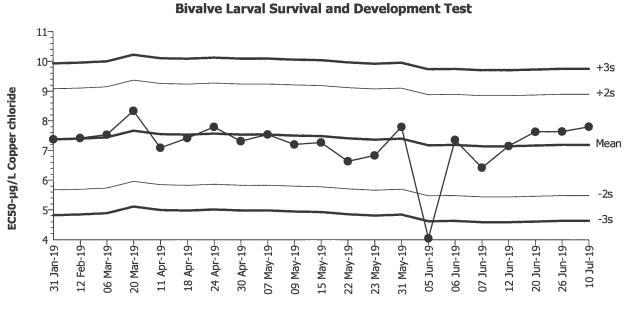
Mean:	7.186	Count:	20	-2s Warning Limit:	5.485	-3s Action Limit:	4.634
Sigma:	0.8505	CV:	11.80%	+2s Warning Limit:	8.887	+3s Action Limit:	9.737

**Quality Control Data** 

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2019	Jan	31	20:05	7.448	0.262	0.308			11-0188-3209	19-4158-0070
2		Feb	12	14:30	7.347	0.1613	0.1896			05-4773-4064	14-0529-6566
3		Mar	6	14:40	7.555	0.3693	0.4342			11-4050-7104	03-7884-0450
4			20	15:25	8.284	1.098	1.291			00-9922-7600	12-2384-4421
5		Apr	11	17:38	7.124	-0.06153	-0.07235			03-4197-9533	19-1101-0677
6			18	15:00	7.422	0.2363	0.2778			07-6552-9366	19-9120-3565
7			24	14:25	7.7	0.5144	0.6048			00-2566-7560	04-1354-8994
8			30	14:45	7.298	0.1119	0.1315			20-4872-1616	19-4433-9185
9		May	7	23:15	7.642	0.4555	0.5356			01-8514-7589	19-5887-7680
10			9	16:40	7.169	-0.01678	-0.01973			16-2457-2225	00-5727-2653
11			15	15:10	7.264	0.07801	0.09172			12-3612-1420	09-9400-1021
12			22	14:35	6.682	-0.5037	-0.5923			07-8876-2604	11-8156-7920
13			23	14:30	6.802	-0.3835	-0.451			04-4294-0564	00-1747-4708
14			31	14:45	7.705	0.5187	0.6099			03-7170-8467	07-0824-5140
15		Jun	5	14:30	4.018	-3.168	-3.725	(-)	(-)	14-9865-1579	13-4485-4153
16			6	15:45	7.404	0.2176	0.2558			20-7222-8009	13-0272-9420
17			7	13:35	6.408	-0.7782	-0.915			16-0229-2669	15-6829-1413
18			12	15:00	7.174	-0.01208	-0.0142			20-8735-2782	03-4367-9827
19			20	15:00	7.627	0.441	0.5186			00-4624-1892	07-7845-5970
20			26	15:55	7.636	0.4503	0.5295			00-8415-2643	12-3790-3484
21		Jul	10	15:15	7.786	0.5999	0.7053			02-0190-4206	08-2094-6054

### **CETIS QC Plot**

Bivalve Larval Survival and Development	Organism: Mytilus galloprovincialis (Bay Mussel Material: Copper chloride		Nautilus Environmental (CA)
Test Type: Development-Survival	Organism: Mytilus galloprovincialis (Bay Mussel	Material:	Copper chloride
Protocol: EPA/600/R-95/136 (1995)	Endpoint: Development Rate	Source:	Reference Toxicant-REF



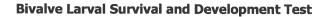
Mean:	7.188	Count:	20	-2s Warning Limit:	5.484	-3s Action Limit:	4.633
Sigma:	0.8519	CV:	11.90%	+2s Warning Limit:	8.892	+3s Action Limit:	9.744

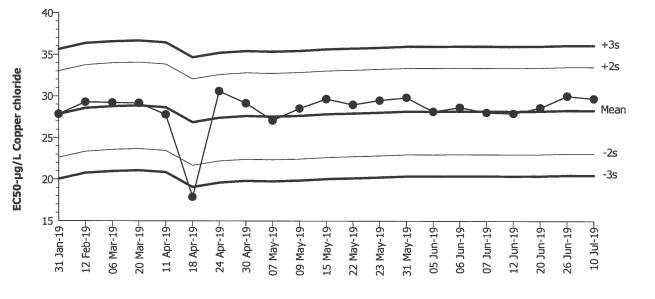
#### **Quality Control Data**

oint Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
2019	Jan	31	20:05	7.377	0.1885	0.2213			11-0188-3209	01-6713-0404
	Feb	12	14:30	7.421	0.2327	0.2731			05-4773-4064	11-5918-1928
	Mar	6	14:40	7.531	0.3435	0.4032			11-4050-7104	19-8242-5220
		20	15:25	8.334	1.146	1.345			00-9922-7600	07-3949-1564
	Apr	11	17:38	7.092	-0.09631	-0.1131			03-4197-9533	11-6047-7524
		18	15:00	7.416	0.2277	0.2673			07-6552-9366	15-9786-1055
		24	14:25	7.793	0.605	0.7101			00-2566-7560	21-1882-8302
		30	14:45	7.316	0.1277	0.1498			20-4872-1616	19-2600-3345
	May	7	23:15	7.538	0.3503	0.4112			01-8514-7589	17-7584-8304
0		9	16:40	7.2	0.01215	0.01426			16-2457-2225	15-1559-8997
1		15	15:10	7.264	0.07575	0.08892			12-3612-1420	11-8863-7865
2		22	14:35	6.635	-0.5528	-0.6489			07-8876-2604	17-1974-2634
3		23	14:30	6.832	-0.3556	-0.4175			04-4294-0564	17-8215-3713
4		31	14:45	7.79	0.6019	0.7065			03-7170-8467	14-6790-8476
5	Jun	5	14:30	4.043	-3.145	-3.691	(-)	(-)	14-9865-1579	11-4839-0192
6		6	15:45	7.353	0.1653	0.194			20-7222-8009	04-5983-4829
7		7	13:35	6.42	-0.7681	-0.9016			16-0229-2669	12-7601-7003
8		12	15:00	7.147	-0.04149	-0.0487			20-8735-2782	10-3598-4911
9		20	15:00	7.627	0.4393	0.5156			00-4624-1892	15-3942-3527
0		26	15:55	7.636	0.4482	0.5261			00-8415-2643	06-7718-5278
1	Jul	10	15:15	7.8	0.6115	0.7179			02-0190-4206	02-8661-6124

### **CETIS QC Plot**

Bivalve Larval Survival and Developmen	t Test		Nautilus Environmental (CA)
Test Type: Development-Survival	Organism: Mytilus galloprovincialis (Bay Mussel	Material:	Copper chloride
Protocol: EPA/600/R-95/136 (1995)	Endpoint: Survival Rate	Source:	Reference Toxicant-REF





Mean:	28.28	Count:	20	-2s Warning Limit:	23.08	-3s Action Limit:	20.48
Sigma:	2.6	CV:	9.19%	+2s Warning Limit:	33.48	+3s Action Limit:	36.08

### **Quality Control Data**

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2019	Jan	31	20:05	27.83	-0.4542	-0.1747			11-0188-3209	12-1004-4079
2		Feb	12	14:30	29.28	1.001	0.385			05-4773-4064	07-6241-6865
3		Mar	6	14:40	29.2	0.9189	0.3534			11-4050-7104	15-7196-8133
4			20	15:25	29.16	0.8836	0.3398			00-9922-7600	05-5691-6970
5		Apr	11	17:38	27.79	-0.495	-0.1904			03-4197-9533	08-6961-6395
6			18	15:00	17.87	-10.41	-4.002	(-)	(-)	07-6552-9366	08-9162-0556
7			24	14:25	30.58	2.3	0.8845			00-2566-7560	12-6072-8549
8			30	14:45	29.13	0.8524	0.3279			20-4872-1616	08-0824-0389
9		Мау	7	23:15	27.08	-1.205	-0.4633			01-8514-7589	14-6629-9437
10			9	16:40	28.51	0.2344	0.09017			16-2457-2225	07-4498-9014
11			15	15:10	29.64	1.359	0.5227			12-3612-1420	00-6090-1816
12			22	14:35	28.95	0.6734	0.259			07-8876-2604	08-9025-9816
13			23	14:30	29.48	1.2	0.4614			04-4294-0564	02-9635-2644
14			31	14:45	29.82	1.545	0.5941			03-7170-8467	20-0663-5874
15		Jun	5	14:30	28.14	-0.1374	-0.05284			14-9865-1579	15-6459-4753
16			6	15:45	28.65	0.3668	0.1411			20-7222-8009	14-8023-9336
17			7	13:35	28.03	-0.2521	-0.09697			16-0229-2669	01-4873-8064
18			12	15:00	27.92	-0.3649	-0.1404			20-8735-2782	03-5721-7619
19			20	15:00	28.59	0.3111	0.1197			00-4624-1892	18-2643-8450
20			26	15:55	30	1.72	0.6615			00-8415-2643	15-8975-2294
21		Jul	10	15:15	29.69	1.409	0.5418			02-0190-4206	18-9463-9841

### **CETIS Test Data Worksheet**

Report Date:0Test Code:02-

itart Date: ind Date: ample Date	10 J 12 J	lul-19 lul-19	14:48	Protocol:	: Mytilus galloprovi EPA/600/R-95/13 Copper chloride				Nautilus Environmental (CA) 190710msdv Reference Toxicant Copper Chloride			
C-µg/L	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal		Notes			
			1			225	219	Ja	7/17/19			
			2			B32022			1			
			3			244	238					
			4			721	214		1			
			5			226	Ô					
			6			233	229					
			7			219	Ø					
			8			221	210					
			9			215	204					
			10			229	220					
			11			198	Ø					
			12			235	30					
			13			254	2.39					
			14			234	231					
			15			243	239					
			16			220	22					
			17			211	205					
			18			227	216					
			19			172	Ø			r		
			20			172	Ø					
			21			24	Õ	TCL 7	/18/19	some cells 145		
	-		22			234	227	00-1	1	source cens 145		
			23			28	Ø		cells ly	520		
			24			25	$\tilde{O}$		2 Colls 14			
			25			20	(D)	Some	certs 145	ed.		
			26			. 204	i9	SOUVE	cerps 143			
			27			215	22					
			28		-	253	244					
			29			227	220					
			30			23	Ø		CEDEK 1450	d		

@ Q18 Ja 7/17/19

Analyst: JU QA: 178/1/19

CETIS	Test	Data	Works	heet
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Nautilus Environmental (CA)

Bivalve	Larval	Survival	and	Development Test
				a crophic root

Start Date: End Date: Sample Date:			14:48 2 7	@18 Species: C Protocol: /6/ <sub>19</sub> Material:	Mytilus galloprovi EPA/600/R-95/13 Copper chloride	ncialis 6 (1995)		Sample Code:190710msdvSample Source:Reference ToxicantSample Station:Copper Chloride
C-µg/L	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal	Notes
0	LC	1	22			238	229	Ac 7/12/19
0	LC	2	1					1.01
0	LC	3	18					
0	LC	4	17					
0	LC	5	14					
2.5		1	10			226	217	
2.5		2	4				4	
2.5		3	28					
2.5		4	6					
2.5		5	29			0	1.001	
5		1	9			713-	196	
5		2	15					
5		3	13				-	
5		4	8					
5		5	3 27			00.0	110	
10		2	27 16			222	42	
10		2	26					
10		4	20					
10		5	12					
20		1	11			203	<i>m</i>	
20		2	5			200	0	
20		3	19					
20		4	20				-	
20		5	7					
40	· · ·	1	25			0	$\cap$	cells lysed
40		2	24				$\sim$	uns you
40		3	23			1		
40		4	21					
40		5	30					

QC=AC

Analyst:\_<u>RT</u>\_QA:<u>KFP8/1</u>/19

Water Quality Measurements

Client: Internal

Sample ID: CuCl<sub>2</sub>

Test No.: 190710msdv

Test Species:M. galloprovincialisStart Date/Time:7/10/20191515End Date/Time:7/12/20191535

Concentration (µg/L)		Salinity (ppt)		Т	Temperature (°C)			Dissolved Oxygen (mg/L)			pH (pH units)		
(µg/L)	0	24	48	0	24	48	0	24	48	0	24	48	
Lab Control	31.7	31.7	31.6	16.0	14.8	15.2	8.5	8.0	8.2	8.00	7.98	296	
2.5	31.8	32,0	31.9	15.7	14.8	15,0	8.4	8,0	٤.)	8.01	7.98	798	
5	31.8	32.0	31.9	15.7	14.7	15.0	8,5	8.0	8.3	8,01	7.98	798	
10	31.9	32.4	31.9	15.8	14.7	14.9	8.4	8.0	8.6	\$ .68	7.97	799	
20	31.8	31.9	31.9	15.7	14.7	14,9	8.4	8.0	8.3	799	7.97	799	
40	318	31.8	31.8	15.8	14.7	15.0	84	79	8.3	8.01	7.92	8.00	
Technician Initials:		Readings: made by:		24 RT	48 P.~		Vol. Cu	conc. mac stock ado Final Volu	ded (mL): me (mL):	۱.º 50	0		

Comments:	0 hrs:	
	24 hrs:	
	48 hrs:	
QC Check:	PH 7/31/19	Final Review: 1959 8/1/19

Nautilus Environmental. 4340 Vandever Avenue. San Diego, CA 92120.

#### Larval Development Worksheet

Client:	Internal/Cucl2
Test No.:	190710 msdy
Test Species:	Mytilus galloprovincialis
Animal Source:	Missign bacy
Date Received:	5129/19 013A+B combined tank
Test Chambers:	30 mL glass shell vials
Sample Volume:	10 mL

140

Start Date/Time:	7/10/2019	515	
End Date/Time:	7/12/2019	1535	
Technician Initials:	ACLEG		

#### Spawn Information

First Gamete Release Time:

#### **Gamete Selection**

Sex	Number Spawning
Male	6
Female	2MB, 1 (at

Sex	Beaker Number(s)	Condition (sperm motility, egg density, color, shape, etc.)
Male	1	good demetrie metric to
Female 1	mbi	gued density - color
Female 2	INB2	Vight COLOY, OCTOR DEMANT. Chil
Female 3	Catl	Verygrange, exailent dins

Egg Fertilization Time: 1230

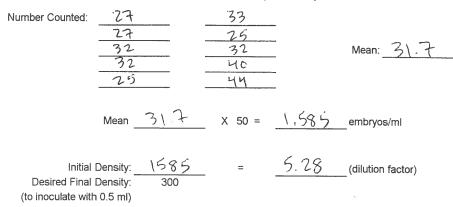
#### Embryo Stock Selection

Stock Number	% of embryos at 2-cell division stage	
Female 1	100	
Female 2	90	
Female 3	80	

Stock(s) chosen for testing:

#### Embryo Inoculum Preparation

Target count on Sedgwick-Rafter slide for desired density is 6 embryos



When mean percent dividing is  $\geq$  90, prepare the embryo inoculum according to the calculated dilution factor. For example, if the dilution factor is 2.25, use 100 ml of existing stock (1 part) and 125 ml of dilution water (1.25 parts).

**Time Zero Control Counts** 

Rep	No. Dividing	Total	% Dividing	Mean % Dividing
TØ A	219	219	100	
TØ B	261	261	(00)	
TØC	232	232	100	
TØ D	211	211	100	100
TØ E	233	233	100	
TØ F	238	.238	100	
	x=232			

48-h QC: 229/236 = 97 / 2

Comments:

QC Check:

RH 7/31/19

Nautilus Environmental. 4340 Vandever Avenue. San Diego, CA 92120.

Final Review: KTP 8///19

Appendix E List of Qualifier Codes



### **Glossary of Qualifier Codes:**

- Q1 Temperatures out of recommended range; corrective action taken and recorded in Test Temperature Correction Log
- Q2 Temperatures out of recommended range; no action taken, test terminated same day
- Q3 Sample aerated prior to initiation or renewal due to dissolved oxygen (D.O.) levels below 6.0 mg/L
- Q4 Test aerated; D.O. levels dropped below 4.0 mg/L
- Q5 Test initiated with aeration due to an anticipated drop in D.O.
- Q6 Airline obstructed or fell out of replicate and replaced; drop in D.O. occurred
- Q7 Salinity out of recommended range
- Q8 Spilled test chamber/ Unable to recover test organism(s)
- Q9 Inadequate sample volume remaining, 50% renewal performed
- Q10 Inadequate sample volume remaining, no renewal performed
- Q11 Sample out of holding time; refer to QA section of report
- Q12 Replicate(s) not initiated; excluded from data analysis
- Q13 Survival counts not recorded due to poor visibility or heavy debris
- Q14 D.O. percent saturation was checked and was  $\leq 110\%$
- Q15 Did not meet minimum test acceptability criteria. Refer to QA section of report.
- Q16 Percent minimum significant difference (PMSD) was <u>below</u> the lower bound limit for acceptability. This indicates that statistics may be over-sensitive in detecting a difference from the control due to low variability in the data set.
- Q17 Percent minimum significant difference (PMSD) was <u>above</u> the upper bound limit for acceptability. This indicates that statistics may be under-sensitive in detecting a difference from the control due to high variability in the data set.
- Q18 Incorrect Entry
- Q19 Illegible Entry
- Q20 Miscalculation
- Q21 Other (provide reason in comments section)
- Q22 Greater than 10% mortality observed upon receipt and/or in holding prior to test initiation. Organisms acclimated to test conditions at Nautilus and ultimately deemed fit to use for testing.
- Q23 Test organisms received at a <u>temperature</u> greater than 3°C outside the recommended test temperature range. However, due to age-specific protocol requirements and/or sample holding time constraints, the organisms were used to initiate tests upon the day of arrival. Organisms were acclimated to the appropriate test conditions upon receipt and prior to test initiation.
- Q24 Test organisms received at <u>salinity</u> greater than 3 ppt outside of the recommended test salinity range. However, due to age-specific protocol requirements and/or sample holding time constraints, the organisms were used to initiate tests upon the day of arrival. Organisms were acclimated to the appropriate test conditions upon receipt and prior to test initiation.