

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

## Monitoring Period: February 2020

Prepared for:

Jacobs 1100 112<sup>th</sup> Ave NE Suite 500 Bellevue, WA, 98004

Prepared by:

Enthalpy Analytical (formerly Nautilus Environmental) 4340 Vandever Avenue San Diego, CA 92120 (858) 587-7333

Date Submitted: March 18, 2020

## Data Quality Assurance:

- Enthalpy Analytical 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.

California 4340 Vandever Avenue San Diego, California 92120 858.587.7333 fax: 619.279.5919

Results verified by:

Eric Green, Project Manager

## Introduction

A toxicity test was performed using a groundwater composite sample collected on February 4, 2020 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 located in San Diego, California.

## **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	020420
Enthalpy Log-in Number	20-0143
Collection Date; Time	2/4/2020; 0926h
Receipt Date; Time	2/5/2020; 0950h
Receipt Temperature (°C)	2.8
Dissolved Oxygen (mg/L)	9.4
рН	7.42
Conductivity (µS/cm)	10,710
Salinity (ppt)	6.6
Alkalinity (mg/L CaCO <sub>3</sub> )	409
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.

Table 2. Summary of Methous for u	
Test Period	2/5/2020, 1310h to 2/7/2020, 1230h
Test Organism	Mytilus galloprovincialis
Test Organism Source	Taylor Shellfish (Shelton, WA)
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 94.1 ppt
Test Concentrations (% sample)	73.3 <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 73.3 (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	Endpoint	NOEC (% effluent)	LOEC (% effluent)	Toxic Unit (TU <sub>c</sub> )	EC <sub>25</sub> (% effluent)
Biyalya	Normal Development	73.3	> 73.5	< 1.36	> 73.3
Bivalve Survival		73.3	> 73.5	< 1.36	> 73.3

NOEC = No Observed Effect Concentration

LOEC = Lowest Observed Effect Concentration

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

Concentration (% Effluent)	Mean Survival (%)	Mean Normal Development (%)
0 (Brine Control)	92.4	99.0
0 (Lab Control)	93.5	98.4
2	94.3	98.6
4	99.7	98.8
9	96.9	98.7
18	96.1	98.0
35	93.7	99.0
73.3 <sup>a</sup>	97.7	98.6

## 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. 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 D.

## **Reference Toxicant**

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 E.

Species	Endpoint	EC₅₀ (μg/L copper)	Historical mean ± 2 SD (µg/L copper)	CV (%)
Bivalve	Normal Development	7.13	8.96 ± 6.03	33.7
Divalve	Survival Rate	29.8	29.8 ± 4.88	8.19

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

## **CETIS Summary Report**

26 Feb-20 15:22 (p 1 of 2) 2002-S016 | 11-4010-6265

Bivalve Larva	Survival and D	evelopm	ent Test						Nautilus	Environn	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	11-1242-0616 05 Feb-20 13:1 07 Feb-20 12:3 47h	0 P 0 S	est Type: rotocol: pecies: ource:	Development-S EPA/600/R-95/ Mytilus gallopro Taylor Shellfish	136 (1995) wincialis		Anal Dilu Brin Age	ent: Dilut e: Froz	ed Natural S en Seawate		
•	15-3935-9903 04 Feb-20 09:2 05 Feb-20 09:5 28h (2.8 °C)	6 N 0 S	ode: laterial: ource: tation:	20-0143 Effluent Sample Jacobs Wyckoff G7 W		Hvent	Clie Proj		obs		
Comparison S	Summary										
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method			
07-1815-1513 04-4290-4136	Development R Survival Rate	late	73.3 73.3	>73.3 >73.3	NA NA	1.72% 12.8%	<1.364 <1.364		ultiple Com		
Test Acceptat	oility								÷		
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Lin	nits	Overlap	Decision		
07-1815-1513	Development R	late	Contr	ol Resp	0.9895	0.9 - NL		Yes Passes Acceptability Criteria			Criteria
04-4290-4136	Survival Rate		Contr	ol Resp	0.9236	0.5 - NL		Yes		ceptability	
Development	Rate Summary										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Brine Control	5	0.989	5 0.9807	0.9983	0.982	1	0.003161	0.007069	0.71%	0.0%
0	Lab Control	5	0.983	8 0.9667	1	0.9608	0.9926	0.006158	0.01377	1.4%	0.58%
2		5	0.986	2 0.979	0.9934	0.9792	0.9926	0.002595	0.005804	0.59%	0.34%
4		5	0.988	2 0.9742	1	0.9752	1	0.00505	0.01129	1.14%	0.13%
9		5	0.987	2 0.9818	0.9926	0.9828	0.9924	0.001934	0.004323	0.44%	0.23%
18		5	0.979	7 0.9577	1	0.9576	1	0.007949	0.01777	1.81%	0.99%
35		5	0.989	9 0.979	1	0.9758	1	0.003927	0.008781	0.89%	-0.04%
73.3		5	0.985	8 0.9732	0.9984	0.9741	1	0.004546	0.01016	1.03%	0.37%
Survival Rate	Summary										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Brine Control	5	0.923	6 0.8306	1	0.8049	1	0.03348	0.07487	8.11%	0.0%
0	Lab Control	5	0.935	0.8284	1	0.8293	1	0.03839	0.08585	9.18%	-1.23%
2		5	0.943	1 0.8275	1	0.7805	1	0.04161	0.09305	9.87%	-2.11%
4		5	0.996		1	0.9837	1	0.003252	0.007272	0.73%	-7.92%
9		5	0.969		1	0.9024	1	0.01998	0.04468	4.61%	-4.93%
18		5	0.961		1	0.9106	1	0.0177	0.03958	4.12%	-4.05%
35		5	0.936	6 0.844	1	0.8211	1	0.03336	0.0746	7.97%	-1.41%

Analyst: EG JCL QA: EG 2/27/20

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Bivalve	Larval	Survival	and	Development	Test

26 Feb-20 1	5:22 (p 2 of	2)
2002 2016	44 4040 00	005

Report Date:

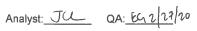
Test Code:

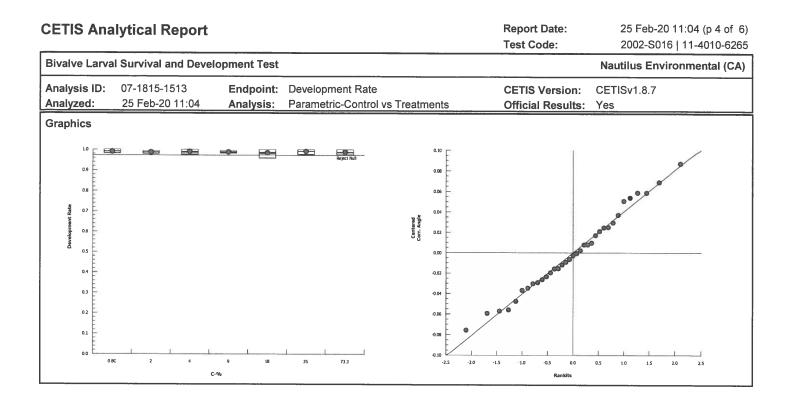
2002-S016 | 11-4010-6265

Nautilus Environmental (CA)
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	t Rate Detail					
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Brine Control	0.982	0.9841	0.9899	0.9915	1
0	Lab Control	0.9926	0.9608	0.9918	0.981	0.9926
2		0.9792	0.9926	0.9917	0.9829	0.9845
4		0.9752	0.9846	1	1	0.9811
9		0.9828	0.9924	0.9837	0.986	0.991
18		0.9576	0.9911	0.9847	1	0.9652
35		0.9911	1	0.9924	0.9901	0.9758
73.3		0.9793	1	0.9914	0.9843	0.9741
Survival Rate	e Detail					
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Brine Control	0.9024	1	0.8049	0.9593	0.9512
0	Lab Control	1	0.8293	0.9919	0.8537	1
2		0.7805	1	0.9837	0.9512	1
4		0.9837	1	1	1	1
9		0.9431	1	1	1	0.9024
18		0.9593	0.9106	1	1	0.935
35		0.9106	0.9512	1	0.8211	1
73.3		1	1	0.9431	1	0.9431
Development	t Rate Binomials					in a second s
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Brine Control	109/111	124/126	98/99	117/118	117/117
0	Lab Control	135/136	98/102	121/122	103/105	135/136
2		94/96	134/135	120/121	115/117	127/129
4			10 11 100			12//120
		118/121	128/130	140/140	140/140	156/159
9					140/140 141/143	
9 18		118/121	128/130	140/140		156/159
1		118/121 114/116	128/130 131/132	140/140 121/123	141/143	156/159 110/111
18		118/121 114/116 113/118	128/130 131/132 111/112	140/140 121/123 129/131	141/143 125/125	156/159 110/111 111/115
18 35	e Binomials	118/121 114/116 113/118 111/112	128/130 131/132 111/112 117/117	140/140 121/123 129/131 131/132	141/143 125/125 100/101	156/159 110/111 111/115 121/124
18 35 73.3	e Binomials Control Type	118/121 114/116 113/118 111/112	128/130 131/132 111/112 117/117	140/140 121/123 129/131 131/132	141/143 125/125 100/101	156/159 110/111 111/115 121/124
18 35 73.3 Survival Rate		118/121 114/116 113/118 111/112 142/145	128/130 131/132 111/112 117/117 123/123	140/140 121/123 129/131 131/132 115/116	141/143 125/125 100/101 125/127	156/159 110/111 111/115 121/124 113/116
18 35 73.3 Survival Rate C-%	Control Type	118/121 114/116 113/118 111/112 142/145 Rep 1	128/130 131/132 111/112 117/117 123/123 Rep 2	140/140 121/123 129/131 131/132 115/116 Rep 3	141/143 125/125 100/101 125/127 Rep 4	156/159 110/111 111/115 121/124 113/116 Rep 5
18 35 73.3 Survival Rate C-% 0	Control Type Brine Control	118/121 114/116 113/118 111/112 142/145 <b>Rep 1</b> 111/123	128/130 131/132 111/112 117/117 123/123 <b>Rep 2</b> 123/123	140/140 121/123 129/131 131/132 115/116 <b>Rep 3</b> 99/123	141/143 125/125 100/101 125/127 <b>Rep 4</b> 118/123	156/159 110/111 111/115 121/124 113/116 Rep 5 117/123
18 35 73.3 Survival Rate C-% 0 0	Control Type Brine Control	118/121 114/116 113/118 111/112 142/145 <b>Rep 1</b> 111/123 123/123	128/130 131/132 111/112 117/117 123/123 <b>Rep 2</b> 123/123 102/123	140/140 121/123 129/131 131/132 115/116 <b>Rep 3</b> 99/123 122/123	141/143 125/125 100/101 125/127 <b>Rep 4</b> 118/123 105/123	156/159 110/111 111/115 121/124 113/116 Rep 5 117/123 123/123
18 35 73.3 Survival Rate C-% 0 0 2	Control Type Brine Control	118/121 114/116 113/118 111/112 142/145 <b>Rep 1</b> 111/123 123/123 96/123	128/130 131/132 111/112 117/117 123/123 <b>Rep 2</b> 123/123 102/123 123/123	140/140 121/123 129/131 131/132 115/116 <b>Rep 3</b> 99/123 122/123 121/123	141/143 125/125 100/101 125/127 <b>Rep 4</b> 118/123 105/123 117/123	156/159 110/111 111/115 121/124 113/116 Rep 5 117/123 123/123 123/123
18 35 73.3 Survival Rate C-% 0 0 2 4	Control Type Brine Control	118/121 114/116 113/118 111/112 142/145 <b>Rep 1</b> 111/123 123/123 96/123 121/123	128/130 131/132 111/112 117/117 123/123 <b>Rep 2</b> 123/123 102/123 123/123 123/123	140/140 121/123 129/131 131/132 115/116 <b>Rep 3</b> 99/123 122/123 121/123 123/123	141/143 125/125 100/101 125/127 <b>Rep 4</b> 118/123 105/123 117/123 123/123	156/159 110/111 111/115 121/124 113/116 Rep 5 117/123 123/123 123/123 123/123
18 35 73.3 Survival Rate C-% 0 0 2 4 9	Control Type Brine Control	118/121 114/116 113/118 111/112 142/145 <b>Rep 1</b> 111/123 123/123 96/123 121/123 116/123	128/130 131/132 111/112 117/117 123/123 <b>Rep 2</b> 123/123 123/123 123/123 123/123	140/140 121/123 129/131 131/132 115/116 <b>Rep 3</b> 99/123 122/123 121/123 123/123	141/143 125/125 100/101 125/127 <b>Rep 4</b> 118/123 105/123 117/123 123/123 123/123	156/159 110/111 111/115 121/124 113/116 Rep 5 117/123 123/123 123/123 123/123 123/123 111/123

	ETIS Analytical Report								2002	25 Feb-20 11:04 (p 3 of 6) 2002-S016   11-4010-6265		
Bivalve Larva	al Survival and	Developn	nent Test						Nautilus	Environr	nental (CA)	
Analysis ID: Analyzed:	07-1815-1513 25 Feb-20 11:			velopment R ametric-Con		iments		S Version: ial Results:	CETISv1. Yes	8.7		
Data Transfor	rm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU	
Angular (Corre	ected)	NA	C > T	NA	NA		1.72%	73.3	>73.3	NA	1.364	
Dunnett Mult	iple Compariso	n Test										
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(	x:5%)			
Brine Control 2			0.5629	2.407	0.067 8	0.6531	CDF		icant Effect		-,	
	4		0.03426	2.407	0.067 8	0.8477	CDF	Non-Significant Effect Non-Significant Effect				
	9		0.435	2.407	0.067 8	0.7075	CDF					
	18		1.142	2.407	0.067 8	0.3883	CDF	-	icant Effect			
	35		-0.117	2.407	0.067 8	0.8864	CDF	-	icant Effect			
	73.3		0.5034	2.407	0.067 8	0.6789	CDF	-	icant Effect			
ANOVA Table	)											
Source	Sum Sqi	uares	Mean Squ	iare	DF	F Stat	P-Value	Decision(α:5%)				
Between	0.004377	651	0.0007296	085	6	0.3806	0.8851	Non-Signif	icant Effect			
Error	0.05367923		0.0019171	15	28							
Total	0.058056	88			34	_						
Distributiona	l Tests											
Attribute	Test			Test Stat	Critical	P-Value	Decision(	α:1%)				
Variances	Bartlett	Equality of	Variance	6.978	16.81	0.3229	Equal Var	iances				
Distribution	Shapiro	Wilk W N	ormality	0.984	0.9146	0.8798	Normal Di	nal Distribution				
Development	Doto Summon											
	. Rate Summary	1										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
<b>C-%</b>	-		<b>Mean</b> 0.9895	<b>95% LCL</b>	<b>95% UCL</b>	<b>Median</b> 0.9899	<b>Min</b> 0.982	Max 1	<b>Std Err</b> 0.003162	<b>CV%</b> 0.71%	%Effect 0.0%	
	Control Type	Count										
0	Control Type	Count 5	0.9895	0.9807	0.9983	0.9899	0.982	1	0.003162	0.71%	0.0%	
0 2	Control Type	Count 5 5	0.9895 0.9862	0.9807 0.979	0.9983 0.9934	0.9899 0.9845	0.982 0.9792	1 0.9926	0.003162 0.002596	0.71% 0.59%	0.0% 0.34%	
0 2 4	Control Type	<b>Count</b> 5 5 5	0.9895 0.9862 0.9882	0.9807 0.979 0.9742	0.9983 0.9934 1	0.9899 0.9845 0.9846	0.982 0.9792 0.9752	1 0.9926 1	0.003162 0.002596 0.00505	0.71% 0.59% 1.14%	0.0% 0.34% 0.13%	
0 2 4 9	Control Type	<b>Count</b> 5 5 5 5 5	0.9895 0.9862 0.9882 0.9872	0.9807 0.979 0.9742 0.9818	0.9983 0.9934 1 0.9926	0.9899 0.9845 0.9846 0.986	0.982 0.9792 0.9752 0.9828	1 0.9926 1 0.9924	0.003162 0.002596 0.00505 0.001934	0.71% 0.59% 1.14% 0.44%	0.0% 0.34% 0.13% 0.23%	
0 2 4 9 18	Control Type	<b>Count</b> 5 5 5 5 5 5	0.9895 0.9862 0.9882 0.9872 0.9797	0.9807 0.979 0.9742 0.9818 0.9577	0.9983 0.9934 1 0.9926 1	0.9899 0.9845 0.9846 0.986 0.9847	0.982 0.9792 0.9752 0.9828 0.9576	1 0.9926 1 0.9924 1	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927	0.71% 0.59% 1.14% 0.44% 1.81%	0.0% 0.34% 0.13% 0.23% 0.99%	
0 2 4 9 18 35 73.3	Control Type	Count 5 5 5 5 5 5 5 5 5 5 5	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858	0.9807 0.979 0.9742 0.9818 0.9577 0.979	0.9983 0.9934 1 0.9926 1 1	0.9899 0.9845 0.9846 0.986 0.9847 0.9911	0.982 0.9792 0.9752 0.9828 0.9576 0.9758	1 0.9926 1 0.9924 1 1	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927	0.71% 0.59% 1.14% 0.44% 1.81% 0.89%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04%	
0 2 4 9 18 35 73.3	Control Type Brine Control	Count 5 5 5 5 5 5 5 5 5 5 5	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858	0.9807 0.979 0.9742 0.9818 0.9577 0.979	0.9983 0.9934 1 0.9926 1 1 0.9984	0.9899 0.9845 0.9846 0.986 0.9847 0.9911	0.982 0.9792 0.9752 0.9828 0.9576 0.9758	1 0.9926 1 0.9924 1 1	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927	0.71% 0.59% 1.14% 0.44% 1.81% 0.89%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04%	
0 2 4 9 18 35 73.3 Angular (Corr	Control Type Brine Control rected) Transfo	Count 5 5 5 5 5 5 5 7 7 med Sun	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858	0.9807 0.979 0.9742 0.9818 0.9577 0.979 0.9732	0.9983 0.9934 1 0.9926 1 1 0.9984 <b>95% UCL</b>	0.9899 0.9845 0.9846 0.986 0.9847 0.9911 0.9843 Median	0.982 0.9792 0.9752 0.9828 0.9576 0.9758 0.9741 Min	1 0.9926 1 0.9924 1 1 1 <b>Max</b>	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927 0.004546 Std Err	0.71% 0.59% 1.14% 0.44% 1.81% 0.89% 1.03%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04% 0.37%	
0 2 4 9 18 35 73.3 Angular (Corr C-%	Control Type Brine Control rected) Transfo Control Type	Count 5 5 5 5 5 5 7 7 med Sun Count	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858 mmary Mean	0.9807 0.979 0.9742 0.9818 0.9577 0.979 0.9732 95% LCL	0.9983 0.9934 1 0.9926 1 1 0.9984	0.9899 0.9845 0.9846 0.986 0.9847 0.9911 0.9843	0.982 0.9792 0.9752 0.9828 0.9576 0.9758 0.9741 <b>Min</b> 1.436	1 0.9926 1 0.9924 1 1 1 1 <b>Max</b> 1.525	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927 0.004546 Std Err 0.01556	0.71% 0.59% 1.14% 0.44% 1.81% 0.89% 1.03% <b>CV%</b> 2.37%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04% 0.37% <b>%Effect</b> 0.0%	
0 2 4 9 18 35 73.3 Angular (Corr C-% 0 2	Control Type Brine Control rected) Transfo Control Type	Count 5 5 5 5 5 5 5 7 rmed Sun Count 5	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858 mmary Mean 1.471	0.9807 0.979 0.9742 0.9818 0.9577 0.979 0.9732 95% LCL 1.428	0.9983 0.9934 1 0.9926 1 1 0.9984 <b>95% UCL</b> 1.514	0.9899 0.9845 0.9846 0.986 0.9847 0.9911 0.9843 <b>Median</b> 1.47	0.982 0.9792 0.9752 0.9828 0.9576 0.9758 0.9741 <b>Min</b> 1.436 1.426	1 0.9926 1 0.9924 1 1 1 1 <b>Max</b> 1.525 1.485	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927 0.004546 Std Err	0.71% 0.59% 1.14% 0.44% 1.81% 0.89% 1.03% <b>CV%</b> 2.37% 1.77%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04% 0.37% <b>%Effect</b> 0.0% 1.06%	
0 2 4 9 18 35 73.3 Angular (Corr C-% 0 2	Control Type Brine Control rected) Transfo Control Type	Count 5 5 5 5 5 5 5 5 rmed Sun Count 5 5	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858 mmary Mean 1.471 1.455	0.9807 0.979 0.9742 0.9818 0.9577 0.979 0.9732 <b>95% LCL</b> 1.428 1.423	0.9983 0.9934 1 0.9926 1 1 0.9984 <b>95% UCL</b> 1.514 1.487	0.9899 0.9845 0.9846 0.986 0.9847 0.9911 0.9843 <b>Median</b> 1.47 1.446	0.982 0.9792 0.9752 0.9828 0.9576 0.9758 0.9741 <b>Min</b> 1.436	1 0.9926 1 0.9924 1 1 1 1 <b>Max</b> 1.525	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927 0.004546 <b>Std Err</b> 0.01556 0.01151 0.02456	0.71% 0.59% 1.14% 0.44% 1.81% 0.89% 1.03% <b>CV%</b> 2.37% 1.77% 3.74%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04% 0.37% <b>%Effect</b> 0.0% 1.06% 0.06%	
0 2 4 9 18 35 73.3 Angular (Corr C-% 0 2 4 9	Control Type Brine Control rected) Transfo Control Type	Count 5 5 5 5 5 5 5 5 rmed Sun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858 mmary Mean 1.471 1.455 1.47	0.9807 0.979 0.9742 0.9818 0.9577 0.979 0.9732 <b>95% LCL</b> 1.428 1.423 1.402 1.434	0.9983 0.9934 1 0.9926 1 1 0.9984 <b>95% UCL</b> 1.514 1.487 1.538 1.483	0.9899 0.9845 0.9846 0.986 0.9847 0.9911 0.9843 <b>Median</b> 1.47 1.446 1.446 1.452	0.982 0.9792 0.9752 0.9828 0.9576 0.9758 0.9741 <b>Min</b> 1.436 1.426 1.413 1.439	1 0.9926 1 0.9924 1 1 1 1 <b>Max</b> 1.525 1.485 1.529 1.484	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927 0.004546 <b>Std Err</b> 0.01556 0.01151	0.71% 0.59% 1.14% 0.44% 1.81% 0.89% 1.03% <b>CV%</b> 2.37% 1.77% 3.74% 1.37%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04% 0.37% <b>%Effect</b> 0.0% 1.06% 0.06% 0.82%	
0 2 4 9 18 35 73.3 Angular (Corr C-% 0 2 4	Control Type Brine Control rected) Transfo Control Type	Count 5 5 5 5 5 5 5 7 med Sun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	0.9895 0.9862 0.9882 0.9872 0.9797 0.9899 0.9858 <b>mmary</b> <b>Mean</b> 1.471 1.455 1.47 1.459	0.9807 0.979 0.9742 0.9818 0.9577 0.979 0.9732 <b>95% LCL</b> 1.428 1.423 1.402	0.9983 0.9934 1 0.9926 1 1 0.9984 <b>95% UCL</b> 1.514 1.487 1.538	0.9899 0.9845 0.9846 0.986 0.9847 0.9911 0.9843 <b>Median</b> 1.47 1.446 1.446	0.982 0.9792 0.9752 0.9828 0.9576 0.9758 0.9741 <b>Min</b> 1.436 1.426 1.413	1 0.9926 1 0.9924 1 1 1 1 <b>Max</b> 1.525 1.485 1.529	0.003162 0.002596 0.00505 0.001934 0.007949 0.003927 0.004546 <b>Std Err</b> 0.01556 0.01151 0.02456 0.008907	0.71% 0.59% 1.14% 0.44% 1.81% 0.89% 1.03% <b>CV%</b> 2.37% 1.77% 3.74%	0.0% 0.34% 0.13% 0.23% 0.99% -0.04% 0.37% <b>%Effect</b> 0.0% 1.06% 0.06%	

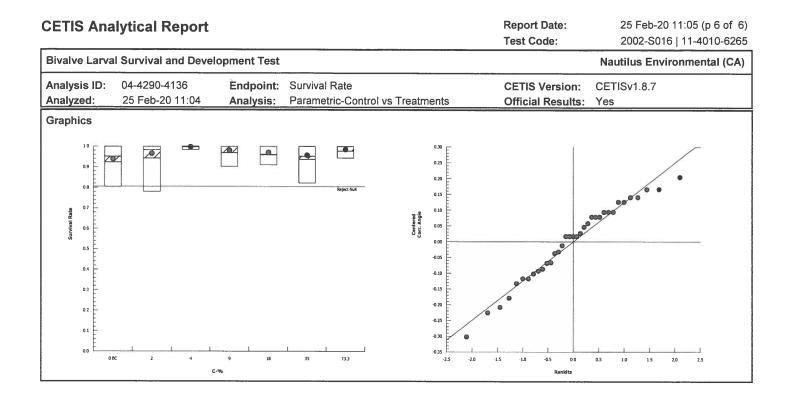




CETIS Ana	ETIS Analytical Report								25 Feb-20 11:04 (p 5 of 6) 2002-S016   11-4010-6265		
Bivalve Larv	al Survival and [	Develop	oment Test						Nautilus	Environn	nental (CA)
Analysis ID: Analyzed:	D:         04-4290-4136         Endpoint:         Survival Rate           25 Feb-20 11:04         Analysis:         Parametric-Control vs Treatme		tments	CETIS Version: CETISv1.8.7 Official Results: Yes							
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Corrected) NA C > T		NA	NA		12.8%	73.3	>73.3	NA	1.364		
Dunnett Mult	tiple Compariso	n Test									
Control	vs C-%		Test Sta	at Critical	MSD DF	P-Value	Р-Туре	Decision(	a:5%)		
Brine Control	2		-0.7365	2.407	0.208 8	0.9740	CDF		icant Effect		
	4		-2.174	2.407	0.208 8	0.9998	CDF	-	icant Effect		
	9		-1.28	2.407	0.208 8	0.9948	CDF	-	icant Effect		
	18		-0.9071	2.407	0.208 8	0.9838	CDF	-	icant Effect		
	35		-0.4469	2.407	0.208 8	0.9454	CDF	-	icant Effect		
	73.3		-1.458	2.407	0.208 8	0.9971	CDF	-	icant Effect		
ANOVA Table	e							· · · · ·			-
Source	Sum Squ	ares	Mean S	quare	DF	F Stat	P-Value	Decision(	a:5%)		
Between	0.113521		0.01892	017	6	1.017	0.4344	Non-Significant Effect			
Error	0.5207969 0.01859		989	28							
Total	0.634317	9			34						
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	α:1%)			
Variances	Bartlett E	Equality	of Variance	7.921	16.81	0.2439	Equal Var	iances			
Distribution	Shapiro-	Wilk W	Normality	0.9664	0.9146	0.3519	Normal Di	Distribution			
Survival Rate	e Summary										
C-%	Control Type	Cour	nt Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Brine Control	5	0.9236	0.8306	1	0.9512	0.8049	1	0.03348	8.11%	0.0%
2		5	0.9431	0.8275	1	0.9837	0.7805	1	0.04161	9.87%	-2.11%
4		5	0.9967	0.9877	1	1	0.9837	1	0.003252	0.73%	-7.92%
9		5	0.9691	0.9136	1	1	0.9024	1	0.01998	4.61%	-4.93%
18		5	0.961	0.9118	1	0.9593	0.9106	1	0.0177	4.12%	-4.05%
35		5	0.9366	0.844	1	0.9512	0.8211	1	0.03336	7.97%	-1.41%
					1	1	0.9431	1	0.01394	3.19%	-5.81%
73.3		5	0.9772	0.9385							
73.3 Angular (Cor	rected) Transfor	med S	ummary		. <u></u>						
73.3 Angular (Cor C-%	Control Type	med Si Cour	ummary nt Mean	95% LCL	95% UCL		Min	Max	Std Err	CV%	%Effect
73.3 Angular (Cor C-% 0		med Si Cour	ummary nt Mean 1.322	<b>95% LCL</b> 1.133	<b>95% UCL</b> 1.51	1.348	<b>Min</b> 1.113	1.526	<b>Std Err</b> 0.06804	<b>CV%</b> 11.51%	<b>%Effect</b> 0.0%
73.3 Angular (Cor C-% 0 2	Control Type	rmed So Cour 5 5	ummary nt Mean 1.322 1.385	<b>95% LCL</b> 1.133 1.157	<b>95% UCL</b> 1.51 1.614	1.348 1.443	<b>Min</b> 1.113 1.083	1.526 1.526			
73.3 Angular (Cor C-% 0 2 4	Control Type	rmed Si Cour 5 5 5 5	ummary nt Mean 1.322 1.385 1.509	<b>95% LCL</b> 1.133 1.157 1.463	<b>95% UCL</b> 1.51 1.614 1.555	1.348 1.443 1.526	<b>Min</b> 1.113	1.526 1.526 1.526	0.06804	11.51%	0.0%
73.3 Angular (Cor C-% 0 2 4 9	Control Type	<b>Cour</b> 5 5 5 5 5	ummary nt Mean 1.322 1.385 1.509 1.432	<b>95% LCL</b> 1.133 1.157 1.463 1.269	<b>95% UCL</b> 1.51 1.614 1.555 1.595	1.348 1.443 1.526 1.526	Min 1.113 1.083 1.443 1.253	1.526 1.526 1.526 1.526	0.06804 0.08228	11.51% 13.28%	0.0% -4.81%
73.3 Angular (Cor C-% 0 2 4 9 18	Control Type	<b>med S</b> <b>Cour</b> 5 5 5 5 5 5 5	ummary nt Mean 1.322 1.385 1.509 1.432 1.4	<b>95% LCL</b> 1.133 1.157 1.463 1.269 1.25	<b>95% UCL</b> 1.51 1.614 1.555 1.595 1.549	1.348 1.443 1.526 1.526 1.368	Min 1.113 1.083 1.443 1.253 1.267	1.526 1.526 1.526 1.526 1.526	0.06804 0.08228 0.01655	11.51% 13.28% 2.45%	0.0% -4.81% -14.19%
73.3 Angular (Cor C-% 0 2 4 9	Control Type	<b>Cour</b> 5 5 5 5 5	ummary nt Mean 1.322 1.385 1.509 1.432	<b>95% LCL</b> 1.133 1.157 1.463 1.269	<b>95% UCL</b> 1.51 1.614 1.555 1.595	1.348 1.443 1.526 1.526	Min 1.113 1.083 1.443 1.253	1.526 1.526 1.526 1.526	0.06804 0.08228 0.01655 0.05863	11.51% 13.28% 2.45% 9.16%	0.0% -4.81% -14.19% -8.36%

- 20

Analyst: JU QA: EU 2/27/20



Analyst: JU QA: EL 2/22/26

## **CETIS Test Data Worksheet**

 Report Date:
 02 Feb-20 12:41 (p 1 of 1)

 Test Code:
 2002 - 50% 11-4010-6265/43F4A419

Nautilus Environmental (CA)

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

		95 Feb-20 97 Feb-20 94 Feb-20		7 Feb-20 4 Feb-20		Species: Protocol: Material:	Mytilus galloprovincialis EPA/600/R-95/136 (1995) Effluent Sample			Sample Code: 20- 0143 Sample Source: Jacobs Sample Station: Wyckoff G WTP Effluent
C-%	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal	Notes		
			256			123	121	RT 2/19/20		
			257			121	118	1		
			258			127	125			
			259			112	111			
			260			143	141			
			261			145	142			
			262			111	109			
			263			132	131			
			264			123	123			
			265			116	113			
			266			101	100			
			267			129	127			
			268			135	134 98			
			269			99	98			
			270			116	115			
			271			117	117			
			272			102	98			
			273			112	111			
			274			105	103			
			275			[2]	120			
			276			117	117			
			277			125	125			
			278			124	121			
			279			130	128			
			280			136	135			
			281			116	114			
			282			115	111			
			283			96	94			
			284			118	113			
			285			126	124	$\checkmark$		
			286			131	129	RT 2/21/20		
			287			hi	110			
			288			140	140	RT Z/21/20 J A(S 2/24/2020		
			289			159	156	Λ		
			290			132	131			
			291			136	135			
			292			117	115			
			293			1(8	117	1		
			294			140	140			
			295			122	121			

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## **CETIS Test Data Worksheet**

Report Date: 02 Feb-20 12:40 (p 1 of 1) Test Code: 2002 - SOL6 11-4010-6265/43F4A419

Bivalve Larv	valve Larval Survival and Development Test							Nautilus Environmental (CA)			
Start Date: End Date: Sample Date	-1 3				Sample Code: 20-0143 Sample Source: Jacobs Sample Station: Wyckoff GWTP FIFWent						
C-%	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal	Notes			
0	BC	1	262								
0	BC	2	285								
0	BC	3	269			106	106	NM 217/20			
0	BC	4	293					· · · · · ·			
0	BC	5	271								
0	LC	1	291								
0	LC	2	272								
0	LC	3	295			123	122	NM			
0	LC	4	274								
0	LC	5	280								
2		1	283								
2		2	268								
2		3	275			117	117	NM			
2		4	292								
2		5	267								
4		1	257								
4		2	279								
4		3	294			140	139	NM			
4		4	288								
4		5	289								
9		1	281								
9		2	263								
9		3	256			118	116	NM			
9		4	260								
9		5	287								
18		1	284								
18		2	273								
18		3	286			130	128	NM			
18		4	277								
18		5	282								
35		1	259								
35		2	276								
35		3	290			137	135	NM			
35		4	266	-							
35		5	278								
74.1		1	261								
74.1 -73 74.1	.3	2	264								
		3	270		-	117	117	NM			
4.1		4	258								
₱		5	265								

Ats QC= \$4

Analyst: AC ] QA: EG 2/27/20

# Marine Chronic Bioassay DM-014

## Water Quality Measurements

Client: Jacobs

Sample ID: Wyckoff GUTP EFFluent Sample Log No.: 20-0143

Test No.: 2002-Sol 0

 Test Species:
 M. galloprovincialis

 Start Date/Time:
 2/5/2020
 1310

 End Date/Time:
 2/7/2020
 1330

Concentration (% sample)	Salinity (ppt)			Т	emperatu (°C)	ire	Dissolved Oxygen (mg/L)			pH (pH units)		
	0	24	48	0	24	48	0	24	48	0	24	48
Lab Control	30,3	30,0	29.9	14.3	14.5	14.7	8.7	8.5	8.4	8.08	8.04	7.97
Brine Control	30.5	30.2	30.4	14.9	14,2	14.5	83	8.6	8.4	8.24	8.09	8.01
2	30.3	30.0	30.2	14.8	14.7	14.8	8.4	8.5	8.3	8,07	8.06	8.00
4	30.3	30.0	30.1	14.5	14.5	14.6	8.5	3.6	8.4	8.04	8.06	8.03
9	30.3	3.0.1	30.1	14.3	14,4	14.5	8.4	3.6	8.4	7.94	8.06	8.07
18	30,3	30.1	30.2	14,4	14.7	14.9	8,4	8.5	8.3	7.84	8.06	8.12
35	30,3	30.0	30.2	14.1	14.6	14.8	8.3	8.5	8.3	7.69	8.07	8-19
73.3	30,5	30,3	30.4	14.0	14.6	14.7	8.4	3.5	8.4	7,60	8.09	8.Z7

Technician Initials:	WQ Readings: Dilutions made by:	0 EG EG	24 DM -	48  P++ 080 ///-	Environmental Chamber:	D
Comments:	0 hrs: 24 hrs: 48 hrs:					
QC Check:	EG 2/27/20				Final Revie	w: <u>PH 313/20 // AZ 3/ 6/20</u>

Enthalpy Analytical. 4340 Vandever Avenue. San Diego, CA 92120.

# Marine Chronic Bioassay

## **Brine Dilution Worksheet**

Project:	JACOBS			Analyst:	EG		
Sample ID:	Wyckoff GWTY	effluent		Test Date:	2/5/2020		
Test No:	<b>o:</b> <u>2002-S016</u>			Test Type:	Mussel Development		
Salinity of Ef	fluent	6.6					
Salinity of Br	ine	94.1	Date	of Brine used:	12/20/2019	_	
Target Salini	ty	30	Alkalinity of	Brine Control:	83	_ mg/L as CaCO3	
Test Dilution	Volume	250			,		
		Effluent	Brine Control				
(TS - SE)/(SB TS = targe	•	0.37	0.47				

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	0.37	1.8	250
4	10.0	0.37	3.7	250
9	22.5	0.37	8.2	250
18	45.0	0.37	16.4	250
35	87.5	0.37	31.9	250
73.3	183.1	0.37	66.9	250

	DI Volume			
Brine Control	142.9	0.47	66.9	250

Total Brine Volume Required (ml): 195.8

QC Check: Eq 2/27/20

Final Review: <u>p# 3/3/2071 AC, 3</u>/6/20

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

#### Marine Chronic Bioassay DM-013

Client/Sample:	Jacobs/Wyckoff GUTP EAGluent
Test No.:	2002-5016
Test Species:	Mytilus galloprovincialis
Animal Source/Ba	ich Tank: Taylor / 19B
Date Received:	11/20/19
Test Chambers:	30 mL glass shell vials
Sample Volume:	10 mL

0935

#### Larval Development Worksheet

Start Date/Time:	2/5/2020	1310	
End Date/Time:	2/7/2020	1230	
Technician Initials:	EG		

#### Spawn Information

Sex	Beaker Number(s)	Condition (sperm motility, egg density, color, shape, etc.)
Male	1,2,3	good denotby + motility
Female 1	1	Ok shape, good density pink color
Female 2	2	great shake, OK density, pale color
Female 3		

Sex	Number Spawning
Male	3+
Female	2

Embryo	Stock	Selection	

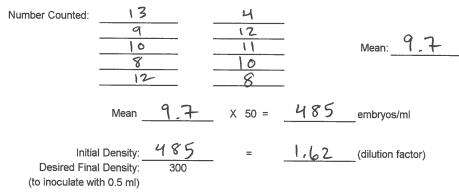
Stock Number	% of embryos at 2-cell division stage
Female 1	
Female 2	100
Female 3	

# Egg Fertilization Time: 1025

Stock(s) chosen for testing:

#### Embryo Inoculum Preparation

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



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).

Gamete Selection

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

TØ Vial No.	No. Dividing	Dividing Total % Dividing		Mean % Dividing
TØ A	119	119 119 100		
TØ B	144	144	100	
тøс	119	119	100	100
TØ D	127	127	100	(00
TØ E	121	121	100	
TØ F	107	107	100	
<u>x</u> =	123			

48-h QC: 116/118 = 98.3%

Comments:

QC Check:

EG 2/27/20



Final Review: <u>R41 3/3/20 AC 3/6</u>/2.

Appendix B Sample Check-In Information

340 Vandever Avenue	Sample ID:	LLINC D	off Gulin	Peffluent	(120470)		formation DC-005
an Diego, CA 92120	Test ID No(s).:		-5016		Sample Description:		
an Diego, CA 32120	Test ID NO(S)	2002	3016		A: no colar, clear, no	dec	- a debas
					n. n= cons, clear, no	000 , 1	1000000
Sample (A	A, B, C):						
Log-in No. (20							
Sample Collection Date a	& Time: 2/4/20 0926				COC Complete (Y/N)?		
Sample Receipt Date a	& Time: 2/5/20 0450				ABC		
Number of Containers & Containe	er Type:    L cubi						
Approx. Total Volume Receiv					Filtration? Y (N)		
Check-in Temperatur	re (°C) 2.8				Initials: A) B) (	)	
Temperature		YN	Y N	Y N	Pore Size:		
DO (I	(mg/L) 9.4				Organisms or	Debris	
pH (	units) 7.42				Salinity Adjustment?		
Conductivity (µ	<b>IS/cm)</b> 10,710				Test: Mussel Source: F	Targe	et ppt: 30
Salini	ity (ppt) 6,6				Test: Source:		et ppt:
Alkalinity (					Test: Source:	Targe	et ppt:
Hardness (m	ng/L) <sup>2, 3</sup>				pH Adjustment? Y (N)		
Total Chlorine (	mg/L) 0.02				A	В	С
Technician In	nitials RT/ER/HS				Initial pH:		
0 0	4				Amount of HCI added:		
Test Performed: MUSSel Develop	Men Control/Dilution War	ter: 8:2 / <u>Lat</u>	SW_/ Lab ART	Other:	Final pH:		
Test Performed: MUSSEL Develop	Other Control/Dilution Wat Alkalinity: 97	ter: 8:2 / Lab Hardness or	Salinity: 3 Salinity:	Other:	Final pH: Cl <sub>2</sub> Adjustment? Y (N)		
Test Performed: MV35l DWClop Additional Control? (Y	<u>Diver</u> Control/Dilution War Alkalinity: <u>97</u> DN = <u>Blive Control</u> A	ter: 8:2 / Lat Hardness or Ikalinity:3	Salinity: <u>3</u> Sa	Other:		В	c
Test Performed: MV35ll DW10f Additional Control? (Y	$\frac{\text{Other A}  \text{Control/Dilution Wat}}{\text{Alkalinity:}  97}}{\text{ON} =  \text{Blive Curtual A}}$	ter: 8:2 / Lat Hardness or Ikalinity:{3	SW_/ Lab ART Salinity: Hardness or S	Other:	Cl <sub>2</sub> Adjustment? Y	В	с
Test Performed: MV35ll DW106 Additional Control? (Y	<u>N</u> = <u>Blive Control</u> A	Ikalinity: <u>02</u> ter: 8:2 / Lab	SW / Lab ART	alinity: <u>50pp (</u> Other:	Cl <sub>2</sub> Adjustment? Y N	B	с 
Additional Control? (Y	<u>N</u> = <u>Blive Cuntrol</u> A <u>Control/Dilution Wat</u> Alkalinity:	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or	Hardness or S     SW / Lab ART     Salinity:	alinity: <u>50pp (</u> Other:	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> :	B	с 
Additional Control? (Y	<u>N</u> = <u>Blive Control</u> A	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or	Hardness or S     SW / Lab ART     Salinity:	alinity: <u>50pp (</u> Other:	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> :	В	с 
Additional Control? (Y	<u>Control/Dilution Wat</u> <u>Control/Dilution Wat</u> <u>Alkalinity:</u> <u>N</u> =A	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or Ikalinity:	Hardness or S     SW / Lab ART     Salinity: Hardness or S	alinity: <u>50 pp (</u> Other: alinity:	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> : STS added: Final Free Cl <sub>2</sub> : Sample Aeration? Y N		
Additional Control? (Y	<u>N</u> = <u>Blive Cuntual</u> A           Control/Dilution Wat           Alkalinity:           N           N           Control/Dilution Wat           Control/Dilution Wat	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or Ikalinity: ter: 8:2 / Lab	SW / Lab ART ( Salinity:	alinity: <u>50 pp (</u> Other: alinity:	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> : STS added: Final Free Cl <sub>2</sub> : Sample Aeration? Y N A	B	C C
Additional Control?Y Test Performed: Additional Control?Y Test Performed:	<u>N</u> = <u>Bive Cuntual</u> A           Control/Dilution Wat           Alkalinity:           N           N           Control/Dilution Wat           Alkalinity:           Alkalinity:	Ikalinity: <u>02</u> ter: 8:2 / Lab — Hardness or Ikalinity: ter: 8:2 / Lab — Hardness or	Hardness or S     SW / Lab ART     Salinity:     Hardness or S     SW / Lab ART     Salinity:	alinity: <u>30 pp (</u>	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> : STS added: Final Free Cl <sub>2</sub> : Sample Aeration? Y N A Initial D.O.		
Additional Control?Y Test Performed: Additional Control?Y Test Performed:	<u>N</u> = <u>Bive Cuntual</u> A           Control/Dilution Wat           Alkalinity:           N           N           Control/Dilution Wat           Alkalinity:           Alkalinity:	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or Ikalinity: ter: 8:2 / Lab	Hardness or S     SW / Lab ART     Salinity:     Hardness or S     SW / Lab ART     Salinity:	alinity: <u>30 pp (</u>	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> : STS added: Final Free Cl <sub>2</sub> : Sample Aeration? Y N A Initial D.O. Duration & Rate		
Additional Control? (Y <u>Test Performed:</u> Additional Control? Y <u>Test Performed:</u> Additional Control? Y	Control/Dilution Wat          Control/Dilution Wat         Alkalinity:         N       =         Control/Dilution Wat         Control/Dilution Wat         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or Ikalinity: ter: 8:2 / Lab Hardness or Ikalinity:	Hardness or S     SW / Lab ART     Salinity: Hardness or S     SW / Lab ART     Salinity: Hardness or S	alinity: <u>50 pp (</u>	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> : STS added: Final Free Cl <sub>2</sub> : Sample Aeration? Y N A Initial D.O.		
Additional Control?Y Test Performed: Additional Control?Y Test Performed: Additional Control?Y Notes:Temperature of sample	Control/Dilution Wat          Control/Dilution Wat         Alkalinity:         N       =         Control/Dilution Wat         Control/Dilution Wat         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or Ikalinity: ter: 8:2 / Lab Hardness or Ikalinity: more than 24 hou	Hardness or S     SW / Lab ART     Salinity:     Hardness or S     SW / Lab ART     Salinity:     Hardness or S     Sinity:     Hardness or S     Salinity:	alinity: <u>50 pp (</u>	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> : STS added: Final Free Cl <sub>2</sub> : Sample Aeration? Y N A Initial D.O. Duration & Rate	B	C
Additional Control?Y Test Performed: Additional Control?Y Test Performed: Additional Control?Y Notes:Temperature of sample	<u>Control/Dilution Wat</u> Control/Dilution Wat         Alkalinity:         N       =         Control/Dilution Wat         Alkalinity:       Alkalinity:         Alkalinity:       Alkalinity:         N       =       Alkalinity:         Alkalinity:       Alkalinity:       Alkalinity:         N	Ikalinity: <u>02</u> ter: 8:2 / Lab Hardness or Ikalinity: ter: 8:2 / Lab Hardness or Ikalinity: more than 24 hou	Hardness or S     SW / Lab ART     Salinity:     Hardness or S     SW / Lab ART     Salinity:     Hardness or S     Sinity:     Hardness or S     Salinity:	alinity: <u>50 pp (</u>	Cl <sub>2</sub> Adjustment? Y N A Initial Free Cl <sub>2</sub> : STS added: Final Free Cl <sub>2</sub> : Sample Aeration? Y N A Initial D.O. Duration & Rate Final D.O.	B nistry Require	C

519401171

#### **Overlying Water**

#### Total Ammonia Analysis Freshwater

DC-001

DI BI Test Start D	Analyst: DM Analysis Date: 2/13/20				
	Nautilus	Sub-Sample	Test	NH3-N	N x 1.22 Ammonia
Sample ID	ID	Date	Day	(mg/L)	(mg/L)
ank Spike (10 mg/L NH3)		NA	NA	7.4	9.0
yckoff	20-0143	2/5/2020	Check In	1,3	1.6
					1
				2.0	
bike Check (10 mg/L NH <sub>3</sub> )		NA	NA	7.4	9,0
·······					
Imple Duplicate <sup>a</sup>		NA	NA	1,2	1.5
mple Duplicate + Spike <sup>a</sup>		NA	NA	8,7	10.6
ike Check (10 mg/L NH <sub>3</sub> )		NA	NA	7.4	9.0

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

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

Measured Nominal [NH<sub>3</sub>] [Sample Dup] [Spike] [Spike] % Recovery QC Sample ID RPD 0.0 9.0 90 Blank 10 NA NA 10.6 Wychoff 1.6 1.5 90 10 6.5

Comments:

Notes: <sup>a</sup>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 samp e 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

QC Check: BO Z/28/20

EG 2 28 20 Final Review:

Enthalpy Analytical. 4340 Vandever Avenue. San Diego, CA 92120.

Appendix C Chain-of-Custody Form Page 1 of 1

Enthalpy Analyyical (REGION COPY)

DateShipped: 2/4/2020 CarrierName: FedEx AirbillNo: 7776 8157 0320

#### CHAIN OF CUSTODY RECORD Wyckoff Eagle Harbor GWTP 2019/WA Project Code: WEH-029K Cooler #: 1 of 1

#### No: 10-020420-095301-0439

2020T10P000DD210W2LA00 Contact Name: Keith Allers Contact Phone: 206-780-1711

Sample Identifier	CLP Sample No.	Matrix/Sampler	Coll. Method	Analysis/Turnaround (Days)	Tag/Preservative/Bottles	Location	Collection Date/Time	Sample Type
020420		Ground Water/ K.Allers	Composite	CHRTOX(8 Weeks)	(< 6 C) (1)	SP-11	02/04/2020 09:26	Field Sample

	Shipment for Case Complete? N
Special Instructions:	Samples Transferred From Chain of Custody #
Analysis Key: CHRTOX=Chronic Toxicity	

Items/Reason	Relinquished by (Signature and Organization)			ure and Organization)	Date/Time	Sample Cond	lition Upon Receipt	in#:
	Kuth aller JAGOBS	2-4-2020 1000	Elizabet Dugse	Enthalpy	2/5/20 0950	GOOD	2.8°C	20-014
	<i>P</i> *							

Appendix D 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.

Appendix E Reference Toxicant Test Results

## **CETIS Summary Report**

Report Date: Test Code: 11 Mar-20 15:07 (p 1 of 3) 200205msdv | 06-6849-2235

Bivalve Larva	l Survival and Develop	ment Test						Nautilus Environmental (CA
Batch ID: Start Date: Ending Date: Duration:	05 Feb-20 13:10 07 Feb-20 12:30	Test Type: Protocol: Species: Source:	Development- EPA/600/R-95 Mytilus gallopr Taylor Shellfis	/136 (1995) ovincialis			Analyst: Diluent: Brine: Age:	Diluted Natural Seawater Not Applicable
Sample ID: Sample Date: Receive Date: Sample Age:	05 Feb-20 05 Feb-20 9	Code: Material: Source: Station:	200205msdv Copper chlorid Reference Tox Copper Chlorid	ticant			Client: Project:	Internal
Comparison S								
Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	τu	Meth	od
18-0959-6796	Combined Developmer	nt Ra 2.5	5	3.536	11.2%		Dunn	ett Multiple Comparison Test
06-6130-0163	Development Rate	2.5	5	3.536	2.67%			ett Multiple Comparison Test
12-4039-9276	Survival Rate	20	40	28.28	18.1%			ett Multiple Comparison Test
Point Estimate	e Summary							
	e Summary Endpoint	Level	μg/L	95% LCL	95% UCL	ти	Meth	od
Analysis ID	•		μ <mark>g/L</mark> 5.654 7.103	<b>95% LCL</b> 4.978 6.652	<b>95% UCL</b> 6.09 7.393	TU		od Ir Interpolation (ICPIN)
Point Estimate Analysis ID 04-8167-3886 20-3119-3253	Endpoint	nt Ra EC25 EC50 EC25	5.654 7.103 5.698	4.978 6.652 5.291	6.09 7.393 6.115	TU	Linea	
Analysis ID 04-8167-3886	Endpoint Combined Developmen Development Rate	nt Ra EC25 EC50	5.654 7.103	4.978 6.652	6.09 7.393	TU	Linea	r Interpolation (ICPIN)
Analysis ID 04-8167-3886 20-3119-3253	Endpoint Combined Developmen Development Rate Survival Rate	nt Ra EC25 EC50 EC25 EC50 EC50	5.654 7.103 5.698 7.132 24.74	4.978 6.652 5.291 6.86 22.76	6.09 7.393 6.115 7.41 25.13	TU	Linea	ar Interpolation (ICPIN) ar Interpolation (ICPIN)
Analysis ID 04-8167-3886 20-3119-3253 07-0404-6516 Test Acceptat	Endpoint Combined Developmen Development Rate Survival Rate	nt Ra EC25 EC50 EC25 EC50 EC50	5.654 7.103 5.698 7.132 24.74 29.83	4.978 6.652 5.291 6.86 22.76	6.09 7.393 6.115 7.41 25.13 30.09		Linea	ar Interpolation (ICPIN) ar Interpolation (ICPIN) ar Interpolation (ICPIN)
Analysis ID 04-8167-3886 20-3119-3253 07-0404-6516 Test Acceptak Analysis ID	Endpoint Combined Developmen Development Rate Survival Rate	nt Ra EC25 EC50 EC25 EC50 EC25 EC50 Attrib	5.654 7.103 5.698 7.132 24.74 29.83	4.978 6.652 5.291 6.86 22.76 28.51	6.09 7.393 6.115 7.41 25.13 30.09		Linea Linea Linea	ar Interpolation (ICPIN) ar Interpolation (ICPIN) ar Interpolation (ICPIN)
Analysis ID 04-8167-3886 20-3119-3253 07-0404-6516 Test Acceptak Analysis ID 06-6130-0163	Endpoint Combined Developmen Development Rate Survival Rate ility Endpoint	nt Ra EC25 EC50 EC25 EC50 EC25 EC50 Attrib Contro	5.654 7.103 5.698 7.132 24.74 29.83 ute	4.978 6.652 5.291 6.86 22.76 28.51 <b>Test Stat</b>	6.09 7.393 6.115 7.41 25.13 30.09 TAC Limi		Linea Linea Linea <b>Over</b>	ar Interpolation (ICPIN) ar Interpolation (ICPIN) ar Interpolation (ICPIN)
Analysis ID 04-8167-3886 20-3119-3253 07-0404-6516 Test Acceptat Analysis ID 06-6130-0163 20-3119-3253	Endpoint Combined Developmen Development Rate Survival Rate ility Endpoint Development Rate	nt Ra EC25 EC50 EC25 EC50 EC25 EC50 Attrib Contro Contro	5.654 7.103 5.698 7.132 24.74 29.83 ute	4.978 6.652 5.291 6.86 22.76 28.51 <b>Test Stat</b> 0.9893	6.09 7.393 6.115 7.41 25.13 30.09 TAC Limi 0.9 - NL		Linea Linea Linea <b>Over</b> Yes	ar Interpolation (ICPIN) ar Interpolation (ICPIN) ar Interpolation (ICPIN) Iap Decision Passes Acceptability Criteria
Analysis ID 04-8167-3886 20-3119-3253 07-0404-6516	Endpoint Combined Developmen Development Rate Survival Rate ility Endpoint Development Rate Development Rate	nt Ra EC25 EC50 EC25 EC50 EC25 EC50 Attrib Contro Contro Contro	5.654 7.103 5.698 7.132 24.74 29.83 ute DI Resp DI Resp	4.978 6.652 5.291 6.86 22.76 28.51 <b>Test Stat</b> 0.9893 0.9893	6.09 7.393 6.115 7.41 25.13 30.09 TAC Limi 0.9 - NL 0.9 - NL		Linea Linea Linea <b>Over</b> Yes Yes	ar Interpolation (ICPIN) ar Interpolation (ICPIN) ar Interpolation (ICPIN) ar Interpolation (ICPIN) Iap Decision Passes Acceptability Criteria Passes Acceptability Criteria

000-089-187-4

## **CETIS Summary Report**

C-µg/L

0

2.5

**Bivalve Larval Survival and Development Test** 

Count

5

5

Mean

0.8927

0.8944

95% LCL 95% UCL

0.9656

0.9873

0.8197

0.8014

Min

0.7886

0.8049

Max

0.9268

0.9922

**Combined Development Rate Summary** 

Lab Control

**Control Type** 

#### 000-089-187-4

5		5	0.7715	0.6788	0.8641	0.6423	0.8293	0.03338	0.07464	9.68%	13.58%
10		5	0	0	0	0	0	0	0		100.0%
20		5	0	0	0	0	0	0	0		100.0%
40		5	0	0	0	0	0	0	0		100.0%
Developme	ent Rate Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.9893	0.9804	0.9982	0.9828	1	0.003192	0.007137	0.72%	0.0%
2.5		5	0.9858	0.9683	1	0.963	1	0.006318	0.01413	1.43%	0.35%
5		5	0.8669	0.7847	0.9491	0.776	0.9519	0.02962	0.06623	7.64%	12.37%
10		5	0	0	0	0	0	0	0		100.0%
20		5	0	0	0	0	0	0	0		100.0%
40		5	0	0	0	0	0	0	0		100.0%
Survival Ra	ate Summary							******			
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.9024	0.8269	0.978	0.7967	0.9431	0.02721	0.06084	6.74%	0.0%
2.5		5	0.9073	0.8135	1	0.8049	1	0.03378	0.07553	8.32%	-0.54%
5		5	0.8943	0.7559	1	0.7236	1	0.04985	0.1115	12.46%	0.9%
10		5	0.8829	0.7738	0.992	0.7642	0.9919	0.03929	0.08787	9.95%	2.16%
20		5	0.8959	0.8087	0.9832	0.8293	1	0.03142	0.07027	7.84%	0.72%
40		5	0	0	0	0	0	0	0		100.0%
Combined	Development Rate	e Detail									
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
0	Lab Control	0.9106	0.9268	0.9106	0.9268	0.7886					
2.5		0.9431	0.8455	0.8862	0.8049	0.9922					
5		0.8049	0.8049	0.8293	0.776	0.6423					
10		0	0	0	0	0					
20		0	0	0	0	0					
40		0	0	0	0	0					
Developme	ent Rate Detail										
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
		0.00/0									

C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Lab Control	0.9912	0.9828	1	0.9828	0.9898
2.5		0.9831	0.963	0.9909	1	0.9922
5		0.8319	0.9519	0.887	0.776	0.8876
10		0	0	0	0	0
20		0	0	0	0	0
40		0	0	0	0	0
Survival Ra	te Detail					an a
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Lab Control	0.9187	0.9431	0.9106	0.9431	0.7967
2.5		0.9593	0.878	0.8943	0.8049	1
5		0.9675	0.8455	0.935	1	0.7236
10		0.9919	0.7642	0.9187	0.9106	0.8293
20		0.935	1	0.8618	0.8537	0.8293
40		0	0	0	0	0

**Report Date:** Test Code:

Std Err

0.02627

0.03348

Std Dev

0.05874

0.07486

11 Mar-20 15:07 (p 2 of 3) 200205msdv | 06-6849-2235

Nautilus Environmental (CA)

CV%

6.58%

8.37%

%Effect

0.0%

-0.19%



0/122

0/115

Rep 1

113/123

118/123

119/123

122/123

115/123

0/123

0/1

0/94

0/129

Rep 2

116/123

108/123

104/123

94/123

123/123

0/123

0/1

0/113

0/106

Rep 3

112/123

110/123

115/123

113/123

106/123

0/123

0/1

10

20

40

0

2.5

5

10

20

40

C-µg/L

**Survival Rate Binomials** 

**Control Type** 

Lab Control

Report Date:	11
Test Code:	200

11 Mar-20 15:07 (p 3 of 3) 200205msdv | 06-6849-2235

							Test Code.	20020011507   00-0649-2200
Bivalve La	arval Survival and I	Developme	nt Test					Nautilus Environmental (CA)
Combined	d Development Rat	e Binomials	5					
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
0	Lab Control	112/123	114/123	112/123	114/123	97/123		
2.5		116/123	104/123	109/123	99/123	128/129		
5		99/123	99/123	102/123	97/125	79/123		
10		0/123	0/123	0/123	0/123	0/123		
20		0/123	0/129	0/123	0/123	0/123		
40		0/123	0/123	0/123	0/123	0/123		
Developm	ent Rate Binomials	6						
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
0	Lab Control	112/113	114/116	112/112	114/116	97/98	1999-1899-1997-1997-1997-1997-1997-1997-	na y synandar analys ar yn yn yn gymra dy ffyddiad yn faf far fa'r far yn
2.5		116/118	104/108	109/110	99/99	128/129		
5		99/119	99/104	102/115	97/125	79/89		

0/112

0/105

Rep 4

116/123

99/123

123/123

112/123

105/123

0/123

0/1

0/102

0/102

Rep 5

98/123

89/123

123/123

102/123

102/123

0/123

0/1

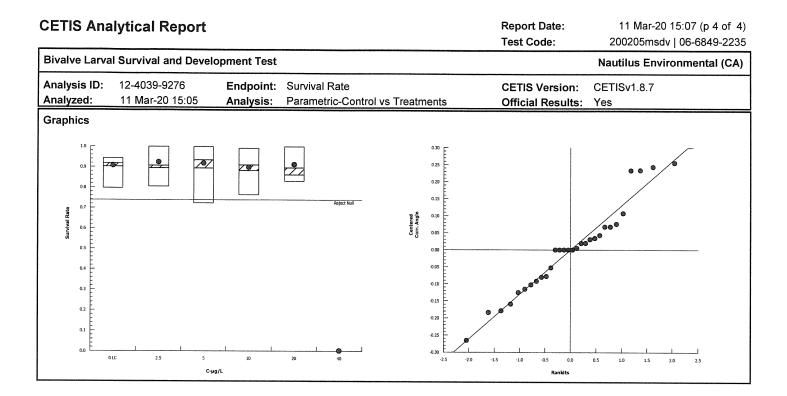
Analyst: Jac QA: 13/12/20

CETIS An	alytical Rep	ort						ort Date: Code:			06 (p 1 of 4) 6-6849-2235
Bivalve Larv	al Survival and I	Developm	ent Test		an a	ana ana ina ina ina ina ina ina ina ina		N MARANA MANYA MANA MANYA KATA KATA KATA KATA KATA KATA KATA KA	Nautilus	Environr	nental (CA)
Analysis ID: Analyzed:	18-0959-6796 11 Mar-20 15:0			mbined Deve rametric-Cor				IS Version: al Results:	CETISv1 Yes	8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C > T	NA	NĂ		11.2%	2.5	5	3.536	****
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-µg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(	a:5%)		
Lab Control	2.5		-0.2838	2.108	0.146 8	0.7688	CDF	Non-Signif	icant Effect		
	5*		2.431	2.108	0.146 8	0.0284	CDF	Significant	Effect		
ANOVA Tabl	le										
Source	Sum Squ	ares	Mean Sq	uare	DF	F Stat	P-Value	Decision(	a:5%)		
Between	0.106906		0.053452		2 4.452			Significant	Effect		
Error	0.144065		0.012005	46	12						
Total	0.250971	0			14						
Distribution	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	α:1%)			
Variances		Variance	1.501	9.21	0.4722	Equal Var	riances				
Distribution	Shapiro-	0.8999	0.8328	0.0948	Normal D	stribution					
Combined D	evelopment Rate	e Summar	У								
C-µg/L	Control Type	Count	Mean	95% LCL			Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.8927	0.8197	0.9656	0.9106	0.7886	0.9268	0.02627	6.58%	0.0%
2.5 5		5 5	0.8944 0.7715	0.8014 0.6788	0.9873 0.8641	0.8862 0.8049	0.8049	0.9922	0.03348	8.37%	-0.19%
10		5	0.7715	0.0788	0.0041	0.8049 0	0.6423 0	0.8293 0	0.03338 0	9.68%	13.58% 100.0%
20		5	0	0	0	0	0	0	0		100.0%
40		5	0	0	0	0	0	0	0		100.0%
Angular (Co	rrected) Transfor	med Sum	mary								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.244	1.138	1.351	1.267	1.093	1.297	0.03837	6.9%	0.0%
2.5		5	1.264	1.082	1.446	1.227	1.113	1.483	0.06546	11.58%	-1.58%
5 10		5 5	1.076 0.0451	0.9702 0.04509	1.181 0.04511	1.113 0.0451	0.9297	1.145	0.03803	7.91%	13.54%
20		5	0.04489	0.04309	0.04511	0.0451	0.0451 0.04404	0.0451 0.0451	0 0.000212	0.0% 1.06%	96.38% 96.39%
40		5	0.0451	0.04509	0.04511	0.0451	0.0451	0.0451	0	0.0%	96.38%
Graphics		98.940.922421.041.1722.441.44									
1.0											
						0.25 E				٠	
0.9						0.20					
		Z		Reject Null	-	0.15					/
0.7 L					g	0.10					
Combined Development Rate					Centered	L 0.05				9 8	
alitica e e e e e e e e e e e e e e e e e e e						0.00			<u> </u>		
0.4						-0.05	•				
0.3						-0.10					
0.2											
						-0.15					
0.1						-0.20					
0.0 E	0 LC 2.5	5	10 20	40	1	-0.25 -2.5 -2	2.0 -1.5 -1.0	-0.5 0.0	0.5 1.0	1.5 2.0	J 2.5
		C-µg/L						Rankits			

CETIS™ v1.8.7.20

	alytical Repo	ort						ort Date: Code:			07 (p 2 of 4 6-6849-223
Bivalve Larv	al Survival and I	Developme	nt Test		nin di Anna anna anna anna anna anna anna ann				Nautilus	Environ	mental (CA
Analysis ID: Analyzed:	06-6130-0163 11 Mar-20 15:0		-	velopment R rametric-Con		tments		S Version:		8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	rected)	NA	C > T	NA	NA		2.67%	2.5	5	3.536	-
Dunnett Mul	tiple Comparisor	n Test									
Control	vs C-µg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(α:5%)		
Lab Control	2.5		0.2372	2.108	0.093 8	0.5698	CDF		ficant Effect		
	5*		5.987	2.108	0.093 8	<0.0001	CDF	Significan	t Effect		
ANOVA Tabl	e										
Source	Sum Squ	iares	Mean Squ	Jare	DF	F Stat	P-Value	Decision	(α:5%)		
Between	0.221848	2	0.1109241		2	22.99	<0.0001	Significan	t Effect		
Error	0.0579058		0.0048254	187	12						
Total	0.279754				14	*					
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision(	α:1%)			
Variances	Bartlett E	Equality of V	ariance	4.061	9.21	0.1312	Equal Var				
Distribution	Shapiro-		0.9697	0.8328	0.8539	Normal Di			_		
Developmen	nt Rate Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.9893	0.9804	0.9982	0.9898	0.9828	1	0.003192	0.72%	0.0%
2.5		5	0.9858	0.9683	1	0.9909	0.963	1	0.006318	1.43%	0.35%
5		5	0.8669	0.7847	0.9491	0.887	0.776	0.9519	0.02962	7.64%	12.37%
10		5	0	0	0	0	0	0	0		100.0%
20		5	0	0	0	0	0	0	0		100.0%
40		5	0	0	0	0	0	0	0		100.0%
Angular (Cor	rrected) Transfor	rmed Sumn	nary							11 - I	
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.47	1.427	1.513	1.47	1.439	1.524	0.01552	2.36%	0.0%
2.5		5	1.459	1.392	1.526	1.475	1.377	1.521	0.02415	3.7%	0.71%
5		5	1.207	1.08	1.333	1.228	1.078	1.35	0.04551	8.44%	17.9%
10		5	0.04814	0.04511	0.05118	0.04726	0.04528	0.05159	0.001095	5.08%	96.72%
20		5	0.04752	0.04476	0.05028	0.04858	0.04404	0.04953	0.000994	4.68%	96.77%
20 40							0.04404 0.5236	0.04953 0.5236	0.000994 0	4.68% 0.0%	96.77% 64.37%
10 20 40 <b>Graphics</b>		5	0.04752	0.04476	0.05028	0.04858					
20 40 Graphics		5	0.04752	0.04476	0.05028	0.04858					
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028	0.04858 0.5236					
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028	0.04858 0.5236					
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236					
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236			0		
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028	0.04858 0.5236		0.5236	0		
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236	0.5236	0.5236	0		
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236		0.5236	0		
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236	0.5236	0.5236	0		
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236	0.5236	0.5236	0		
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236	0.5236	0.5236	0		
20 40 Graphics		5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236	0.5236	0.5236	0		
20 40 Graphics	οιc 2.5	5	0.04752	0.04476 0.5234	0.05028 0.5238	0.04858 0.5236	0.5236	0.5236	0		

CETIS An	alytical Rep	ort			Report Date:         11 Mar-20 15:07 (p           Test Code:         200205msdv   06-684						
Bivalve Larv	al Survival and I	Developm	ent Test	den den en en de de la referir de la referir y de la arte			****	16 (16 y - 16) / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201 / 201	Nautilus	Environn	nental (CA)
Analysis ID: Analyzed:	12-4039-9276 11 Mar-20 15:(		n <b>dpoint</b> : Su n <b>alysis:</b> Pa	vival Rate ametric-Cor	itrol vs Trea	tments		S Version:	CETISv1. Yes	8.7	÷
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C > T	NA	NA		18.1%	20	40	28.28	
Dunnett Mul	Itiple Compariso	n Test									
Control	vs C-µg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(	α:5%)		
Lab Control	2.5		-0.3009	2.305	0.228 8	0.8829	CDF		ficant Effect		
	5		-0.2032	2.305	0.228 8	0.8593	CDF		ficant Effect		
	10		0.1492	2.305	0.228 8	0.7481	CDF	-	ficant Effect		
	20		-0.08069	2.305	0.228 8	0.8252	CDF	-	ficant Effect		
ANOVA Tabl	le										
Source	Sum Squ	ares	Mean Squ	lare	DF	F Stat	P-Value	Decision(	α:5%)		
Between	0.006000	22	0.0015000	)55	4	0.06134	0.9925		ficant Effect		
Error	0.489131		0.024456	55	20						
Total	0.495131	2			24						
Distribution	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	α:1%)			
Variances	Bartlett E	Equality of	Variance	1.868	13.28	0.7600	Equal Var	iances			
Distribution	Shapiro-	Wilk W No	ormality	0.9544	0.8877	0.3148	Normal Di	stribution			
Survival Rat	e Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	8.8.				
0						Meulan	Min	Max	Std Err	CV%	%Effect
	Lab Control	5	0.9024	0.8269	0.978	0.9187	0.7967	0.9431	0.02721	<b>CV%</b>	%Effect 0.0%
2.5	Lab Control	5 5		0.8269 0.8135							
2.5 5	Lab Control		0.9024		0.978	0.9187	0.7967	0.9431	0.02721	6.74%	0.0%
	Lab Control	5	0.9024 0.9073	0.8135	0.978 1	0.9187 0.8943	0.7967 0.8049	0.9431 1	0.02721 0.03378	6.74% 8.32%	0.0% -0.54%
5	Lab Control	5 5	0.9024 0.9073 0.8943	0.8135 0.7559	0.978 1 1	0.9187 0.8943 0.935	0.7967 0.8049 0.7236	0.9431 1 1	0.02721 0.03378 0.04985	6.74% 8.32% 12.46%	0.0% -0.54% 0.9%
5 10	Lab Control	5 5 5	0.9024 0.9073 0.8943 0.8829	0.8135 0.7559 0.7738	0.978 1 1 0.992	0.9187 0.8943 0.935 0.9106	0.7967 0.8049 0.7236 0.7642	0.9431 1 1 0.9919	0.02721 0.03378 0.04985 0.03929	6.74% 8.32% 12.46% 9.95%	0.0% -0.54% 0.9% 2.16%
5 10 20 40	Lab Control	5 5 5 5 5	0.9024 0.9073 0.8943 0.8829 0.8959 0	0.8135 0.7559 0.7738 0.8087	0.978 1 1 0.992 0.9832	0.9187 0.8943 0.935 0.9106 0.8618	0.7967 0.8049 0.7236 0.7642 0.8293	0.9431 1 1 0.9919 1	0.02721 0.03378 0.04985 0.03929 0.03142	6.74% 8.32% 12.46% 9.95%	0.0% -0.54% 0.9% 2.16% 0.72%
5 10 20 40		5 5 5 5 5	0.9024 0.9073 0.8943 0.8829 0.8959 0	0.8135 0.7559 0.7738 0.8087	0.978 1 1 0.992 0.9832	0.9187 0.8943 0.935 0.9106 0.8618	0.7967 0.8049 0.7236 0.7642 0.8293	0.9431 1 1 0.9919 1	0.02721 0.03378 0.04985 0.03929 0.03142	6.74% 8.32% 12.46% 9.95%	0.0% -0.54% 0.9% 2.16% 0.72%
5 10 20 40 Angular (Co	rrected) Transfor	5 5 5 5 5 <b>med Sum</b>	0.9024 0.9073 0.8943 0.8829 0.8959 0	0.8135 0.7559 0.7738 0.8087 0	0.978 1 1 0.992 0.9832 0	0.9187 0.8943 0.935 0.9106 0.8618 0	0.7967 0.8049 0.7236 0.7642 0.8293 0	0.9431 1 0.9919 1 0	0.02721 0.03378 0.04985 0.03929 0.03142 0	6.74% 8.32% 12.46% 9.95% 7.84%	0.0% -0.54% 0.9% 2.16% 0.72% 100.0%
5 10 20 40 Аngular (Со С-µg/L	rrected) Transfor Control Type	5 5 5 5 med Sum Count	0.9024 0.9073 0.8943 0.8829 0.8959 0 mary Mean	0.8135 0.7559 0.7738 0.8087 0 <b>95% LCL</b>	0.978 1 1 0.992 0.9832 0 <b>95% UCL</b>	0.9187 0.8943 0.935 0.9106 0.8618 0 Median	0.7967 0.8049 0.7236 0.7642 0.8293 0 Min	0.9431 1 0.9919 1 0 <b>Max</b>	0.02721 0.03378 0.04985 0.03929 0.03142 0 Std Err	6.74% 8.32% 12.46% 9.95% 7.84%	0.0% -0.54% 0.9% 2.16% 0.72% 100.0%
5 10 20 40 Аngular (Сол С-µg/L 0	rrected) Transfor Control Type	5 5 5 5 <b>med Sum</b> 5	0.9024 0.9073 0.8943 0.8829 0.8959 0 mary Mean 1.262	0.8135 0.7559 0.7738 0.8087 0 <b>95% LCL</b> 1.146	0.978 1 1 0.992 0.9832 0 95% UCL 1.378	0.9187 0.8943 0.935 0.9106 0.8618 0 Median 1.282	0.7967 0.8049 0.7236 0.7642 0.8293 0 <b>Min</b> 1.103	0.9431 1 0.9919 1 0 Max 1.33	0.02721 0.03378 0.04985 0.03929 0.03142 0 <b>Std Err</b> 0.04177	6.74% 8.32% 12.46% 9.95% 7.84% <b>CV%</b> 7.4% 12.3%	0.0% -0.54% 0.9% 2.16% 0.72% 100.0% <b>%Effect</b> 0.0% -2.36%
5 10 20 40 Аngular (Сол С-µg/L 0 2.5	rrected) Transfor Control Type	5 5 5 5 <b>med Sum</b> 5 5 5	0.9024 0.9073 0.8943 0.8829 0.8959 0 mary Mean 1.262 1.292	0.8135 0.7559 0.7738 0.8087 0 <b>95% LCL</b> 1.146 1.095	0.978 1 1 0.992 0.9832 0 <b>95% UCL</b> 1.378 1.49	0.9187 0.8943 0.935 0.9106 0.8618 0 <b>Median</b> 1.282 1.24	0.7967 0.8049 0.7236 0.7642 0.8293 0 <b>Min</b> 1.103 1.113	0.9431 1 0.9919 1 0 Max 1.33 1.526	0.02721 0.03378 0.04985 0.03929 0.03142 0 <b>Std Err</b> 0.04177 0.0711	6.74% 8.32% 12.46% 9.95% 7.84% CV% 7.4%	0.0% -0.54% 0.9% 2.16% 0.72% 100.0% %Effect 0.0%
5 10 20 40 <b>Angular (Co</b> <b>C-μg/L</b> 0 2.5 5	rrected) Transfor Control Type	5 5 5 5 <b>med Sum</b> 5 5 5 5 5	0.9024 0.9073 0.8943 0.8829 0.8959 0 mary Mean 1.262 1.292 1.282	0.8135 0.7559 0.7738 0.8087 0 <b>95% LCL</b> 1.146 1.095 1.038	0.978 1 1 0.992 0.9832 0 <b>95% UCL</b> 1.378 1.49 1.527	0.9187 0.8943 0.935 0.9106 0.8618 0 <b>Median</b> 1.282 1.24 1.313	0.7967 0.8049 0.7236 0.7642 0.8293 0 <b>Min</b> 1.103 1.113 1.017	0.9431 1 0.9919 1 0 Max 1.33 1.526 1.526	0.02721 0.03378 0.04985 0.03929 0.03142 0 <b>Std Err</b> 0.04177 0.0711 0.08811	6.74% 8.32% 12.46% 9.95% 7.84% <b>CV%</b> 7.4% 12.3% 15.36%	0.0% -0.54% 0.9% 2.16% 0.72% 100.0% <b>%Effect</b> 0.0% -2.36% -1.59%



CETIS	S Anal	ytical Repo	ort						port Date: st Code:			:07 (p 1 of 3 06-6849-223
Bivalve	e Larval	Survival and D	evelopme	nt Test						Nautilu	s Enviror	mental (CA)
Analys	is ID:	04-8167-3886	End	dpoint:	Combined Dev	velopment R	late	CE	TIS Version:	CETISv1	.8.7	
Analyz	ed:	11 Mar-20 15:0	5 <b>An</b> a	alysis:	Linear Interpo	lation (ICPIN	I)	Of	ficial Results	: Yes		
Linear	Interpol	ation Options			an an an Air an Air Maer Is ann a tùmhaidh an ta			INTERPRETATION CANCELON AND A CANDING STATE		Talanda kapa-akan tiké di Kabutatan Apira pan	****	99999999999999999999999999999999999999
X Tran	sform	Y Transform	n See	ed	Resamples	Exp 95%	GCL M	lethod				
Linear		Linear	361	23	1000	Yes	T١	wo-Point Inte	rpolation			
Point E	Estimate	s										
Level	µg/L	95% LCL	95% UCL									
EC25	5.654	4.978	6.09						·····	a Manufa Andrea a Roy, Nora Ana an Ana an		
EC50	7.103	6.652	7.393									
Combi	ned Dev	elopment Rate	Summary			Calc	ulated Va	riate(A/B)		unzer (za juga za politika)		
C-µg/L	Co	ontrol Type	Count	Mean	Min	Max	Std Er	r Std Dev	/ CV%	%Effect	А	в
0	La	b Control	5	0.8927	7 0.7886	0.9268	0.0262	7 0.05874	6.58%	0.0%	549	615
2.5			5	0.8944	0.8049	0.9922	0.0334	8 0.07486	8.37%	-0.19%	556	621
5			5	0.7715	5 0.6423	0.8293	0.0333	8 0.07464	9.68%	13.58%	476	617
10			5	0	0	0	0	0		100.0%	0	615
20			5	0	0	0	0	0		100.0%	0	621
40			5	0	0	0	0	0		100.0%	0	615
Graphi are completed beneformer Fate												

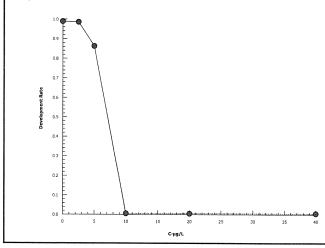
0.2

C-µg/L

25 30 35 40

CETIS	Ana	lytical Rep	ort			•	ort Date: Code:	11 Mar-20 15:07 (p 2 of 200205msdv   06-6849-22				
Bivalve	Larval	Survival and [	Developme	ent Test						Nautilu	s Enviror	mental (CA
-	•					Development Rate Linear Interpolation (ICPIN)				CETISv1 Yes	.8.7	ga 2012 d 4 14 d 4 1 4 19 19 19 19 19 19 19 19 19 19 19 19 19
Linear I	nterpo	lation Options										
X Trans	form	Y Transform	n Se	ed	Resamples	Exp 95%	% CL Met	hod				
Linear		Linear	37	2019	1000	Yes	Two	-Point Interp	olation			
Point E	stimate	s										
Level	µg/L	95% LCL	95% UC	L								
EC25	5.698	5.291	6.115					· · · · · · · · · · · · · · · · · · ·				
EC50	7.132	6.86	7.41									
Develop	oment l	Rate Summary				Calc	ulated Varia	ate(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	0.989	3 0.9828	1	0.003192	0.007136	0.72%	0.0%	549	555
2.5			5	0.985	8 0.963	1	0.006318	0.01413	1.43%	0.35%	556	564
5			5	0.866	9 0.776	0.9519	0.02962	0.06623	7.64%	12.37%	476	552
10			5	0	0	0	0	0		100.0%	0	543
20			5	0	0	0	0	0		100.0%	0	557
40			5	0	0	0	0	0		100.0%	0	5

Graphics



CETIS	S Analytical Repo	ort					-	ort Date: Code:			:07 (p 3 of 3) 06-6849-2235
Bivalve	e Larval Survival and D	evelopment	Test						Nautilus	s Environ	mental (CA)
Analysi Analyzo				Survival Rate Linear Interpola	tion (ICPIN)			IS Version: cial Results:	CETISv1 Yes	.8.7	
Linear	Interpolation Options					ter Antonio (CATOMAG)				****	
X Trans	sform Y Transform	n Seed		Resamples	Exp 95% CL	Met	hod				
Linear	Linear	66426	53	1000	Yes	Two	-Point Interp	olation			
Point E	Estimates										
Level	µg/L 95% LCL	95% UCL									
EC25	24.74 22.76	25.13								2910 CMANA ( 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1	
EC50	29.83 28.51	30.09									
Surviva	al Rate Summary				Calculate	d Varia	ate(A/B)				
C-µg/L	Control Type	Count	Mean	Min	Max S	td Err	Std Dev	CV%	%Effect	А	в
0	Lab Control	5	0.9024	0.7967	0.9431 0	02721	0.06084	6.74%	0.0%	555	615
2.5		5	0.9073	0.8049	1 0	03378	0.07553	8.32%	-0.54%	558	615
5			0.8943		1 0	04985	0.1115	12.46%	0.9%	550	615
10			0.8829		0.9919 0	03929	0.08787	9.95%	2.16%	543	615
20			0.8959			03142	0.07027	7.84%	0.72%	551	615
40		5	0	0	0 0		0		100.0%	0	615
Graphic		5 20 2								7	

Analyst: Ja QA: 45 3/120

Test Type: Protocol:	Development-Surviv EPA/600/R-95/136 (		Organism: Endpoint:		alloprovincia d Developm			aterial: ource:		chloride ice Toxicant	-REF
			Biva	ilve Larval Sur	vival and Develo	pment Test					
	25										
	20-										+3s
	15-										+2s
er chloride	10-		×	9					•		$\backslash$
EC50-µg/L Copper chloride											Mean
ECSO	5-							•			-25
											-3s
	-5	19				2 0		-6]	0 g		
	17 11/10 12	07 Aug-19- 14 Aug-19-	1/ Aug-19- 20 Aug-19- 24 Aug-19-	28 Aug-19-	11 John 19	18 Sep-19- 17 Oct-19-	30 Oct-19 27 Nov-19	05 Dec-19	11 Dec-19- 08 Jan-20-	22 Jan-20- 04 Feb-20-	05 Feb-20

(	CE	TIS	QC	Plot	

**Bivalve Larval Survival and Development Test** 

Nautilus Environmental (CA)

Mean:	8.915	Count:	20	-2s Warning Limit:	2.837	-3s Action Limit:	-0.2019
Sigma:	3.039	CV:	34.10%	+2s Warning Limit:	14.99	+3s Action Limit:	18.03

#### **Quality Control Data**

Point `	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1 2	2019	Jul	17	14:50	8.497	-0.4178	-0.1375			04-5072-3133	00-7236-3161
2			23	14:30	13.76	4.846	1.595			07-6771-8781	18-1893-5656
3			30	15:30	7.313	-1.602	-0.5272			15-3542-8276	10-4430-8659
4		Aug	7	15:30	7.395	-1.52	-0.5003			01-2834-9487	15-5629-3220
5			14	14:15	7.255	-1.66	-0.5462			18-5609-6564	17-5885-5207
6			17	14:00	7.582	-1.333	-0.4386			15-9584-4385	11-8998-1524
7			20	14:15	10.86	1.947	0.6405			14-8361-1578	03-1832-9380
8			24	16:00	7.414	-1.501	-0.4938			19-4374-5817	01-6546-9581
9			28	14:30	7.348	-1.567	-0.5156			01-0546-0046	21-3090-7111
10		Sep	11	14:30	11.93	3.02	0.9936			09-2717-2159	04-2480-9094
11			12	14:25	8.444	-0.4706	-0.1548			19-6218-6352	07-5188-6358
12			18	13:20	7.4	-1.515	-0.4985			10-9359-1611	21-3838-7021
13		Oct	17	12:30	4.368	-4.547	-1.496			01-8239-7270	07-0806-0577
14			30	12:30	7.518	-1.397	-0.4597			07-8198-2858	11-8079-0492
15		Nov	27	20:00	7.249	-1.666	-0.5484			12-9914-0499	16-0529-7707
16		Dec	5	13:15	4.982	-3.933	-1.294			04-7411-4445	13-6587-0425
17			11	13:35	7.245	-1.67	-0.5495			10-8800-1613	10-7929-5811
18 2	2020	Jan	8	13:40	12.34	3.425	1.127			07-8444-5322	01-1422-4896
19			22	13:25	14.72	5.805	1.91			02-1152-2212	07-1224-7163
20		Feb	4	16:30	14.68	5.761	1.896			19-9078-6483	21-0369-4045
21			5	13:10	7.103	-1.812	-0.5963			06-6849-2235	04-8167-3886

## **CETIS QC Plot**

Bivalve La	rval Survival and Developmer	nt Test			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
		Biva	lve Larval Survival and Development Test		
	20-				
					+35
	15				+2s
ide	10-		$\frown$		
EC50-µg/L Copper chloride				-0	Mean
J/L Copp	5-				•
EC50-µ					-25
	-				-35
	- 17-but 71 - 23-but 72 - 23-but - 23-but - 23-but - 24-but - 24-b	17 Aug-19	28 Aug-19 11 Sep-19 12 Sep-19 13 Sep-19 17 Oct-19 30 Oct-19	04-19	
	17. 23. 23. 23. 23. 23. 17. 14. 14. 14. 14. 14. 14. 14. 14. 14. 14	17 Ai 20 Ai 24 Ai	28 A 11 Sc 12 Sc 18 Sc 17 O 30 O	27 Nov-19	11 Dec-19- 08 Jan-20- 22 Jan-20- 04 Feb-20- 05 Feb-20-

Mean:	8.961	Count:	20	-2s Warning Limit:	2.929	-3s Action Limit:	-0.0867
Sigma:	3.016	CV:	33.70%	+2s Warning Limit:	14.99	+3s Action Limit:	18.01

Quality	Control	Data
---------	---------	------

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2019	Jul	17	14:50	8.504	-0.4571	-0.1516			04-5072-3133	09-0911-7730
2			23	14:30	13.38	4.42	1.466			07-6771-8781	07-7153-3575
3			30	15:30	7.388	-1.573	-0.5215			15-3542-8276	07-3589-9194
4		Aug	7	15:30	7.473	-1.488	-0.4934			01-2834-9487	19-8086-2685
5			14	14:15	7.466	-1.495	-0.4957			18-5609-6564	14-6389-5644
6			17	14:00	7.563	-1.398	-0.4634			15-9584-4385	19-0402-2449
7			20	14:15	10.76	1.803	0.5976			14-8361-1578	12-0750-5104
В			24	16:00	7.521	-1.44	-0.4775			19-4374-5817	04-6745-5945
9			28	14:30	7.351	-1.61	-0.5339			01-0546-0046	10-3410-8075
10		Sep	11	14:30	11.98	3.022	1.002			09-2717-2159	17-4622-9429
11			12	14:25	8.608	-0.3534	-0.1172			19-6218-6352	06-5225-4823
12			18	13:20	7.546	-1.415	-0.4693			10-9359-1611	16-7089-5314
13		Oct	17	12:30	4.375	-4.586	-1.521			01-8239-7270	19-1864-9270
14			30	12:30	7.481	-1.48	-0.4906			07-8198-2858	15-7183-3565
15		Νον	27	20:00	7.297	-1.664	-0.5517			12-9914-0499	01-7534-7240
16		Dec	5	13:15	5.087	-3.874	-1.284			04-7411-4445	10-0471-4567
17			11	13:35	7.32	-1.641	-0.544			10-8800-1613	20-9346-8800
18	2020	Jan	8	13:40	12.43	3.468	1.15			07-8444-5322	06-2499-4329
19			22	13:25	14.68	5.72	1.897			02-1152-2212	04-4145-0874
20		Feb	4	16:30	15.01	6.047	2.005	(+)		19-9078-6483	06-3219-7963
21			5	13:10	7.132	-1.829	-0.6064			06-6849-2235	20-3119-3253



Bivalve Lar	val Survival and Developmer	nt Test	Nautilus Er	vironmental (CA
	Development-Survival EPA/600/R-95/136 (1995)	Organism: Mytilus galloprovincialis (Bay Mussel Endpoint: Survival Rate	Material:Copper chlorideSource:Reference Toxical	nt-REF
		Bivalve Larval Survival and Development Test		
	38-			
				+3s
				+2s
	34 - -			
ide	32			
ECSO-µg/L Copper chloride	30-			®Mann
ig/L Copi	28-			- near
EC50-µ			-	
	26			
	24-			-25
	22			-3s
	16 Jul-19 17 Jul-19- 23 Jul-19- 30 Jul-19- 07 Aug-19-	14 Aug-19- 17 Aug-19- 24 Aug-19- 28 Aug-19- 11 Sep-19- 12 Sep-19- 13 Sep-19- 13 Cot+19- 17 Oct+19-	30 Oct-19 27 Nov-19 05 Dec-19 11 Dec-19 08 Jan-20 22 Jan-20	05 Feb-20
		11 12 11 12 11 12 11 12 11 12 11 12 11	37 37 37 37 37 37 37 37 37 37 37 37 37 3	8

Mean:	29.78	Count:	20	-2s Warning Limit:	24.9	-3s Action Limit:	22.47
Sigma:	2.438	CV:	8.19%	+2s Warning Limit:	34.66	+3s Action Limit:	37.09

**Quality Control Data** 

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2019	Jul	16	14:35	29.46	-0.3165	-0.1298			04-6285-8375	17-5419-9497
2			17	14:50	30.25	0.4679	0.1919			04-5072-3133	00-3161-9686
3			23	14:30	29.36	-0.4224	-0.1733			07-6771-8781	15-4437-4125
4			30	15:30	29.67	-0.107	-0.04388			15-3542-8276	20-8453-4017
5		Aug	7	15:30	29.85	0.07493	0.03073			01-2834-9487	07-4855-2818
6			14	14:15	28.85	-0.9345	-0.3833			18-5609-6564	13-1367-1354
7			17	14:00	29.6	-0.1763	-0.07233			15-9584-4385	20-0172-5237
8			20	14:15	37.92	8.139	3.338	(+)	(+)	14-8361-1578	02-5800-6574
9			24	16:00	30.04	0.2574	0.1056			19-4374-5817	17-7461-0750
10			28	14:30	28.66	-1.115	-0.4575			01-0546-0046	13-4512-6481
11		Sep	11	14:30	33.71	3.935	1.614			09-2717-2159	01-1883-2964
12			12	14:25	29.16	-0.6159	-0.2526			19-6218-6352	02-6393-7831
13			18	13:20	29.04	-0.741	-0.304			10-9359-1611	04-3365-2341
14		Oct	17	12:30	24.88	-4.898	-2.009	(-)		01-8239-7270	13-2801-3685
15			30	12:30	29.32	-0.4571	-0.1875			07-8198-2858	20-5233-5110
16		Nov	27	20:00	29.07	-0.7133	-0.2926			12-9914-0499	00-1104-7300
17		Dec	5	13:15	28.21	-1.566	-0.6422			04-7411-4445	20-5035-4724
18			11	13:35	29.18	-0.6007	-0.2464			10-8800-1613	02-9848-3585
19	2020	Jan	8	13:40	29.6	-0.1789	-0.0734			07-8444-5322	01-5655-1706
20			22	13:25	29.76	-0.02439	-0.01			02-1152-2212	19-4150-8988
21		Feb	5	13:10	29.83	0.04632	0.019			06-6849-2235	07-0404-6516

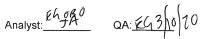
## **CETIS Test Data Worksheet**

Report Date: Test Code:

02 Feb-20 12:41 (p 1 of 1) 06-6849-2235/200205msdv

Start Date: End Date: Sample Date	07 F	<sup>=</sup> eb-20 <sup>=</sup> eb-20 <sup>=</sup> eb-20	)		Mytilus galloprovi EPA/600/R-95/13 Copper chloride			Sample Code: 200205msdv Sample Source: Reference Toxicant Sample Station: Copper Chloride
C-µg/L	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal	Notes
			226			115	0	JA 2/21/20
			227			0	0	1
			228			113	112	
			229			105	0	
			230			118	116	
			231			125	97	JA 2/28/20
			232			102	0	
			233			106	0	
			234			110	109	
			235			116	114	
			236			112	112	
			237			122	0	
			238			108	104	
			239			0	0	
			240			ALZE 1	10-90	9 JA 3/6/20 EG QC:23/10/20 VS QC:23/11/20 0/102. EG QC:23/10/20 0/102.
			241			902	0	*5 QCid 3/11/20 0/129
			242			A. @ 102	0	EL QC'd 3/10/20 0/102.
			243			0	$\circ$	
			244			94	0	
			245			104	99	
			246			0	0	
			247			112	D	
			248			113	0	
			249			116	114	
			250			98	97	
			251			115	102	
			252			129	128	
			253			89	79	
			254			0	0	
			255			99	99	

(B) KS Q18 3/10/20 (B) KS Q18 3/11/20



000-089-187-4

### **CETIS Test Data Worksheet**

 Report Date:
 02 Feb-20 1

 Test Code:
 06-6849-223

02 Feb-20 12:41 (p 1 of 1) 06-6849-2235/200205msdv

				velopment Test				Nautilus Environmental (CA
Start Date: End Date: Sample Date:	05 Feb-20 07 Feb-20 : 05 Feb-20			Protocol:	Mytilus galloprovincialis EPA/600/R-95/136 (1995) Copper chloride			Sample Code:200205msdvSample Source:Reference ToxicantSample Station:Copper Chloride
C-µg/L	Code	Rep	Pos	Initial Density	Final Density	# Counted	# Normal	Notes
0	LC	1	228					
0	LC	2	249					
0	LC	3	236			110	109	NM 2/7/20
0	LC	4	235				1 - 1	101 C 21112-
0	LC	5	250					
2.5		1	230					
2.5		2	238					
2.5		3	234	· · · · · · · · · · · · · · · · · · ·		114	114	NM
2.5		4	255					
2.5		5	252					
5		1	240					
5		2	245					
5		3	251			116	103	NM
5		4	231			110	100	
5		5	253		-			
10		1	237	····				
10		2	244					
10		3	248			112	0	NM
10		4	247			11 ~~	Ű	
10		5	232		-			
20		1	226			-		
20		2	241					
20		3	233			105	0	NM
20		4	229					
20		5	242					
40		1	254					
40		2	227					
40		3	243			0	0	Cells lysed NM
40		4	246					
40		5	239		+			

QC= Eh

Analyst: #() QA: EG 3/10/20

## Marine Chronic Bioassay

DM-014

Client: Internal

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

Test No.: 200205msdv

## Water Quality Measurements

Test Species:	M. gallopro	vincialis
Start Date/Time:	2/5/2020	1310
End Date/Time:	2/7/2020	1230

Concentration (µg/L)		Salinity (ppt)		Т	emperatu (°C)	ire	Dis	solved Ox (mg/L)	xygen		pH (pH units	3)
(+-37	0	24	48	0	24	48	0	24	48	0	24	48
Lab Control	31.7	31.5	31.6	14.6	15,2	14.9	89	3.8	8.5	8.04	8.00	7.97
2.5	31.9	31.6	31.7	14.6	14.7	14.7	8.9	8.8	8.5	8.01	8.01	7.98
5	31.9	31.6	31.7	14.6	14.7	14.6	8.6	8.7	8.5	7.99	2.01	7.98
10	31.9	31.8	31.9	14.7	15.1	15.2	8.5	8.6	8.3	7.98	8.01	7.98
20	32.0	31.6	31.7	14.8	15.0	15.0	8.5	8.6	8.4	8,02	8.02	7.99
40	31,9	31.6	31.6	14.7	15.0	14.9	8.5	8.6	8.4	8.04	8.03	07.9
									-			
								1				1
and an analysis and a substantial machine and an advantage of a machine solar distance and an and a substance a			0	0.4	40				d	L	4.0	-
echnician Initials:	WO F	Readings:	0 €4	24 Dm	48 HOV	]		conc. mai	de (µg/L): ded (mL):		40	-
	Dilutions	-			00000				ime (mL):			1
		·	L		<b>1</b>	Cι			on (µg/L):			1
nvironmental Chaml	oer:	D								,		
omments:	0 hrs:											
	24 hrs:						1979 in			51001157, 21-170-911-101-101-1		
	48 hrs:	MAR	KL217/20							*-**		

QC Check:

RH03/10/20

Final Review: <u>EG 3/10/20</u>

# Marine Chronic Bioassay DM-013

Client/Sample:	Internal / Cullz		
Test No.:	200205 msdv		
Test Species:	Mytilus galloprovincialis		
Animal Source/Batch Tank: Taylor / 19B			
Date Received:	11/20/19		
Test Chambers:	30 mL glass shell vials		
Sample Volume:	10 mL		

0935

#### Larval Development Worksheet

Start Date/Time:	2/5/2020	1310	
End Date/Time:	2/7/2020	1233	
Technician Initials:	EG		

#### Spawn Information

First Gamete	Release	Time:
--------------	---------	-------

Gamete Selection

Sex	Number Spawning	
Male	3+	
Female	2	

Sex	Beaker Number(s)	Condition (sperm motility, egg density, color, shape, etc.)		
Male	1,2,3	good density + motility		
Female 1	1	Ok shape, good density, pink color		
Female 2	2	great shake, OK density, pale color		
Female 3				

Egg Fertilization Time: 1025

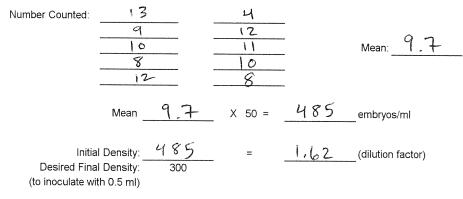
Stock(s) chosen for testing:

#### Embryo Stock Selection

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

#### Embryo Inoculum Preparation

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



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 Ze	ro Control Cou	unts		
TØ Vial No.	No. Dividing	Total	% Dividing	Mean % Dividing
TØ A	119	119	100	
TØ B	144	144	100	
TØC	119	119	100	100
TØ D	127	127	100	
TØ E	121	121	100	
TØ F	107	107	100	
<u>X</u> =	123			

48-h QC: 116/118 = 98.3%

Comments:

QC Check:

Rt03/10/20

Enthalpy Analytical. 4340 Vandever Avenue. San Diego, CA 92120.

Final Review: EG 3/10/20