

Appendix H Chronic Toxicity Study

Renewal of NPDES CA0109223
Carlsbad Desalination Project



Poseidon Salinity Tolerance Study Interim Report - Chronic Toxicity Test Results October 2014 through July 2015

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Data Quality Assurance:

- Nautilus Environmental is accredited in accordance with NELAP by the State of Oregon Environmental Laboratory Accreditation Program (Certificate No. 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 EPA protocols, unless otherwise noted in this report.
- o All test results have met internal Quality Assurance Program requirements.

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INTRODUCTION

Chronic bioassay testing was conducted to determine the sensitivity of several marine organisms to salinity increases above ambient seawater levels. Testing was conducted in response to the amendments made to the Ocean Plan (OP) by the California State Water Quality Control Board (SWQCB); amendments to the Ocean Plan state that at the end of the zone of initial dilution (ZID), receiving water salinity must be within 2,000 mg/L of ambient salinity levels. This limit was based on salinity tolerance studies performed by Phillips et al. (2012) and in a literature review reported by Roberts et al. (2012). Poseidon Water (Poseidon) has contracted with Nautilus Environmental (Nautilus) to conduct salinity studies with the aim of applying for a facility-specific alternative receiving water limit for the Carlsbad Desalination Plant brine discharge.

This study includes a suite of salinity tolerance bioassays using the species and endpoints listed in the OP amendment. These include: purple sea urchin (*Strongylocentrotus purpuratus*) fertilization and development, sand dollar (*Dendraster excentricus*) fertilization and development, red abalone (*Haliotis rufescens*) development, Pacific topsmelt (*Atherinops affinis*) survival and growth, and giant kelp (*Macrocystis pyrifera*) germination and germ-tube growth. The goal of this study is to characterize individual species sensitivity with regards to increased salinity using standard whole effluent toxicity (WET) methods. The approach for this study includes conducting each test three times over the course of approximately one year to address possible seasonal effects and potential differences in batch sensitivities. Results for tests conducted between October 2014 and July 2015 are included in this report. Further testing is scheduled to be completed by February 2016 and will be reported separately.

Tests were spaced several months apart in order to account for potential differences in sensitivity due to seasonal effects. Three tests per year were schedule to generally cover organism spawning, non-spawning, and recruitment periods. Table 1 outlines the testing schedule; including the tests which have already been completed (shaded gray) with results reported herein, and tests scheduled to be completed by February 2016.

Table 1. Salinity Tolerance Test Schedule

Species and Endpoint	Winter	Spring/Summer	Fall
Abalone Development	Dec. 2014	May 2015	Sept Oct. 2015
Kelp Germination and Growth	Dec. 2015- Feb. 2016	May 2015	Sept Oct. 2015
Topsmelt Survival and Growth	Dec. 2015- Feb. 2016	May 2015	Sept Oct. 2015
Urchin Fertilization	Dec. 2015- Feb. 2016	July 2015	Oct. 2014
Sand Dollar Fertilization	Dec. 2015- Feb. 2016	July 2015	Oct. 2014
Urchin Development	Dec. 2015- Feb. 2016	July 2015	Oct. 2014
Sand Dollar Development	Dec. 2015- Feb. 2016	July 2015	Oct. 2014

Dates shaded gray = test already completed and results reported herein

MATERIALS AND METHODS

Test Material

All bioassays were performed in accordance with the United States Environmental Protection Agency (USEPA) protocol "Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to West Coast Marine and Estuarine Organisms" (EPA/600/R-95/136, August 1995). All organisms were acclimated in the laboratory to, or collected directly from ambient seawater. Species-specific test methods are summarized in Tables 2 through 6.

All testing was conducted using natural seawater collected from the intake at Scripps Institution of Oceanography (SIO) in La Jolla, California. After collection, the seawater is held at Nautilus in a 5,000-gallon storage tank, and is continuously cycled through an in-line 20-µm fiber filter system and a chiller unit. The ambient salinity of SIO seawater is approximately 33.5 parts per thousand (ppt). Salinity of each test concentration was increased with the addition of hypersaline seawater (brine) prepared at Nautilus using methods described in USEPA protocols. Briefly, filtered seawater is partially frozen in a -20 °C freezer overnight. The liquid that remains after the freezing period (now concentrated in salinity) is decanted from the ice, which is composed mainly of fresh water. The brine used for this study was approximately two to three times the salinity of ambient seawater. A brine control was also tested at ambient salinity by mixing brine with deionized water and seawater. The brine control incorporated the highest percentage brine used in the dilution series and was tested to ensure that any effects observed in the test concentrations were not attributable to the brine itself.

All definitive tests were conducted using the same salinity concentrations. Nominal test concentrations consisted of 35.0, 35.5, 36.0, 36.5, 37.0, 37.5, 38.0, and 38.5 ppt, as well as an ambient seawater control (lab control), and a brine control.

After dilutions were mixed, minor salinity adjustments were made by addition of brine or seawater so that the measured salinity levels at test initiation matched the nominal salinity to the nearest 0.1 ppt. A Hach SensION5 meter with 0.1 ppt accuracy, calibrated daily to a certified 35 ppt standard was used for all salinity measurements in this study. Water quality parameters (i.e., pH, dissolved oxygen (DO), salinity, and temperature) were measured in all test concentrations prior to test initiation, and once daily depending on the duration of the test.

Table 2. Red Abalone Chronic Test Specifications

Endpoint: 48-hour larval development Test Organism, Age: Haliotis rufescens (red abalone), newly fertilized embryos (within one hour of fertilization) Test Organism Source: American Abalone (Davenport, California) 5 replicates, 150-200 embryos per replicate Number of Replicates, Organisms per Replicate: Test Chamber Type, Volume Glass shell vial containing 20 mL of test solution per Replicate: Feeding: none Test Acceptability: Mean normal development must be 80 percent or greater in the control, must have statistically significant effect at 56 micrograms per liter (µg/L) zinc in the reference toxicant; must achieve a percent

minimum significant difference (PMSD) of less than 20.

Table 3. Echinoderm Larval Development Chronic Test Specifications

Endpoint:	72-hour larval development
Test Organism, Age:	Strongylocentrotus purpuratus (purple urchin) and Dendraster excentricus (sand dollar), newly fertilized embryos (within one hour of fertilization)
Test Organism Source:	Field Collected (San Diego, California)
Number of Replicates, Organisms per Replicate:	5 replicates, 150-200 embryos per replicate
Test Chamber Type, Volume per Replicate:	Glass shell vial containing 10 mL of test solution
Feeding:	none
Test Acceptability:	Mean normal development must be 80 percent or greater in the control; must achieve a PMSD of less than 25.

Table 4. Echinoderm Fertilization Chronic Test Specifications

Endpoint: Egg fertilization (20-min sperm exposure, followed by 20-min egg

fertilization period)

Test Organism: Strongylocentrotus purpuratus (purple urchin) and Dendraster

excentricus (sand dollar)

Test Organism Source: Field Collected (San Diego, California)

Number of Replicates, Organisms per Replicate: 5 replicates, 2000 eggs per replicate. Sperm to egg ratio determined

before each test with a preliminary rangefinding test.

Test Chamber Type, Volume

per Replicate:

Glass scintillation vial containing 10 mL of test solution

Feeding: none

Test Acceptability: Mean fertilization must be 70 percent or greater in the control; must

achieve a PMSD of less than 25.

Table 5. Giant Kelp Chronic Test Specifications

Endpoints: 48-hour spore germination and growth

Test Organism: Macrocystis pyrifera (giant kelp)

Test Organism Source: Field Collected (La Jolla, California)

Number of Replicates, Organisms per Replicate: 5 replicates, ~250,000 spores per replicate

Test Chamber Type, Volume

per Replicate:

Glass petri dish containing 30 mL of test solution

Light Intensity; Photoperiod: 200 ± 40 foot-candles (ft-c); 16 h light, 8 h darkness

Feeding: none

Test Acceptability: Mean germination in the controls must be equal to or greater than 70

percent, and mean length equal to or greater than 10 micrometers (µm). The no observed effect concentration (NOEC) for mean length in the reference toxicant must be equal to or less than 35 $\mu g/L$

copper. The PMSD for both endpoints must be less than 20.

Endpoints:

Table 6. Pacific Topsmelt Chronic Test Specifications

Test Organism, Age: Atherinops affinis (Pacific topsmelt), 12-15 days old at initiation

Test Organism Source: Aguatic Biosystems, Inc. (Fort Collins, Colorado)

Number of Replicates, Organisms per Replicate: 5 replicates, 5 animals per replicate

7-day survival and growth (as dry biomass)

Test Chamber Type, Volume

per Replicate:

500 mL plastic cup containing 250 mL of test solution

Feeding: Artemia nauplii, twice daily

Test Acceptability: Mean survival must be 80 percent or greater in the control, 0.85 mg

average weight of control larvae; the median lethal effect

concentration (LC₅₀) with copper must be less than 205 µg/L in the reference toxicant; must achieve a PMSD less than 25 for survival

and less than 50 for growth.

Data Analysis

Toxicity test responses were statistically evaluated using the Comprehensive Environmental Toxicity Information System™ (CETIS) version 1.8.7.20 program by Tidepool Scientific Software. All comparisons were made according to flowchart specifications provided in method quidance (USEPA 1995). Results were used to calculate the No Observed Effect Concentration (NOEC), Lowest Observed Effect Concentration (LOEC), and point estimates including estimated median lethal concentration (LC₅₀) or effect values (EC₂₅ and EC₅₀; the concentration calculated by the dose-response in the test where 25 or 50 percent of the organisms were adversely effected). All point estimates were calculated using linear interpolation to ensure comparability of statistical method between tests regardless of minor data variations. Since we are interested in effects relative to ambient seawater salinity, organism performance in the test concentrations was compared to that in the lab control rather than the brine control. In cases where the brine control result was less than lab control, a statistical comparison was made between the two controls to ensure no adverse effects were produced due to the addition of brine.

Concurrent reference toxicant tests were conducted to evaluate the sensitivity of each batch of organisms. Copper chloride was used as the reference toxicant for all species, except for abalone for which zinc sulfate was used. The NOEC, LOEC, and LC50/EC50 value for each reference toxicant test is reported in the Quality Assurance section. The mean LC₅₀/EC₅₀ value plus or minus two standard deviations is also reported for up to the last twenty tests performed at Nautilus.

RESULTS

In tests conducted to date, there were no statistically significant effects observed in any test concentration of the Pacific topsmelt, giant kelp, purple urchin or sand dollar fertilization, or the sand dollar larval development tests. The larval development endpoint for purple urchins and abalone appear to be the most sensitive of this suite of tests to increased salinity, with the abalone being more sensitive than purple urchins. These findings are similar to those reported by Phillips et al. in 2012.

A summary of the statistical analysis results for all tests conducted to date is provided in Table 7. Detailed summaries of all test results are presented in Appendix A, and raw data and statistical analysis worksheets are provided in Appendix B.

Table 7. Summary of Statistical Results for all Chronic Salinity Tolerance Tests to Date (October 2014 through July 2015)

Species	Endpoint	Test Start Date	Measured Salinity at Test Initiation (ppt)	NOEC (ppt)	LOEC (ppt)	EC ₂₅ (ppt)	95% CI	EC ₅₀ (ppt)	95% CI
Abalone	Development Rate	12/10/14	34.3 (BC), 33.0 (LC), 35.0, 35.5, 36.0, 36.5, 37.0, 37.5, 38.0, 38.5	36.0	36.5	36.4	36.3 - 36.5	36.7	36.6 - 36.8
Abalone	Development Rate	05/20/15	33.3 (BC), 33.2 (LC), 35.0, 35.5, 36.0, 36.5, 37.0, 37.5, 38.0, 38.5	36.0	36.5	37.1	36.8 - 37.2	37.3	37.3 - 37.4
Purple Urchin	Development Rate	10/30/14	33.7 (BC), 33.3 (LC), 34.9, 35.5, 36.0, 36.5, 37.0, 37.5, 38.0, 38.5	36.5	37.0	38.1	37.8 - 38.2	> 38.5	N/A
Purple Urchin	Development Rate	7/22/15	33.4 (BC), 33.5 (LC), 34.9, 35.4, 35.9, 36.4, 36.9, 37.4, 37.9, 38.5	36.9	37.4	37.7	37.5 – 37.9	38.1	38.0 – 38.3
Sand Dollar	Development Rate	10/30/14	33.7 (BC), 33.3 (LC), 34.9, 35.5, 36.0, 36.5, 37.0, 37.5, 38.0, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Sand Dollar	Development Rate	7/22/15	33.4 (BC), 33.5 (LC), 34.9, 35.4, 35.9, 36.4, 36.9, 37.4, 37.9, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Purple Urchin	Fertilization Rate	10/30/14	33.7 (BC), 33.3 (LC), 34.9, 35.5, 36.0, 36.5, 37.0, 37.5, 38.0, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Purple Urchin	Fertilization Rate	7/22/15	33.4 (BC), 33.5 (LC), 34.9, 35.4, 35.9, 36.4, 36.9, 37.4, 37.9, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Sand Dollar	Fertilization Rate	10/30/14	33.7 (BC), 33.3 (LC), 34.9, 35.5, 36.0, 36.5, 37.0, 37.5, 38.0, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Sand Dollar	Fertilization Rate	7/22/15	33.4 (BC), 33.5 (LC), 34.9, 35.4, 35.9, 36.4, 36.9, 37.4, 37.9, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Giant Kelp	Germination	05/12/15	34.0 (BC), 33.0 (LC), 35.0, 35.5,	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Giant Keip	Growth	03/12/13	36.0, 36.6, 37.0, 37.5, 38.1, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A
Pacific	Survival	05/05/15	33.3 (BC), 33.3 (LC), 35.0, 35.5,	38.5	>38.5	>38.5	N/A	> 38.5	N/A
Topsmelt	Growth	05/05/15	36.0, 36.5, 37.0, 37.5, 38.0, 38.5	38.5	> 38.5	> 38.5	N/A	> 38.5	N/A

ppt = parts per thousand

BC = Brine Control, LC = Lab Control (ambient seawater). The lab control (ambient seawater) was used for all statistical comparisons.

NOEC = No Observed Effect Level; the lowest level or concentration resulting in no observed effect

LOEC = Lowest Observed Effect Level; the lowest level or concentration resulting in an observed effect

EC25/ EC50 = the concentration at which an adverse effect is observed in 25 or 50 percent of the organisms

95% CI = 95 percent confidence interval; lower and upper confidence limits

N/A = Not applicable; confidence limits cannot be calculated

QUALITY ASSURANCE

All laboratory and brine controls met the minimum test acceptability criteria. Variability among replicates was within the appropriate limits for all tests as described in EPA 1995, and the ability to detect a statistical difference was deemed appropriate. Minor deviations are noted on datasheets and a glossary of qualifier codes is available in Appendix C.

Final salinity readings in most tests were within 0.2 ppt of the initial measurement with two exceptions. The final salinity measured in the kelp test was 0.5 to 0.7 ppt higher than the initial reading, likely due to some evaporation because this test is conducted under a specified light intensity. Final readings in the topsmelt test were a maximum of 0.4 ppt higher than the initial readings. Neither of these two tests resulted in any adverse effects to the test organisms in any of the salinity concentrations tested.

Reference Toxicant Test

All reference toxicant tests included in this study met test acceptability criteria. Additionally, the LC_{50} or EC_{50} values calculated for all endpoints were within two standard deviations of the internal control chart means, indicating typical test organism sensitivity to copper (zinc for abalone). Reference toxicant results are summarized in Table 8, and provided in full in Appendix D.

Table 8. Summary of Reference Toxicant Statistical Results

Test Endpoint	Test Start Date	NOEC (µg/L)	LOEC (µg/L)	LC ₅₀ /EC ₅₀ value (µg/L)	Mean LC ₅₀ /EC ₅₀ ± 2 SD (μg/L)
Abalone	12/10/14	18	32	40.8	56.7 ± 23.4
Development	5/20/15	32	56	55.5	56.5 ± 21.8
Purple Urchin	10/30/14	<2.5	2.5	6.16	8.97 ± 5.17
Development	7/22/15	<2.5	2.5	10.4	9.85 ± 4.93
Sand Dollar	10/30/14	10	>10	14.0	20.8 ± 10.5
Development	7/22/15	10	20	14.6	18.5 ± 10.9
Purple Urchin	10/30/14	<10	10	19.0	40.8 ± 24.5
Fertilization	7/22/15	<10	10	35.9	22.9 ± 16.9
Sand Dollar	10/30/14	10	20	26.2	23.1 ± 34.6
Fertilization	7/22/15	<10	10	16.8	24.1 ± 24.7
Kelp Germination Growth	5/13/15	10 10	32 32	118 280	110 ± 63.1 196 ± 157
Pacific Topsmelt Survival Growth	5/5/15	37.5 37.5	75 >37.5	62.7 63.5	83.1 ± 38.5 85.3 ± 41.7

The reference toxicant for abalone is zinc sulfate; for all other species copper chloride was used. Values reported are in micrograms per liter zinc or copper.

NOEC = No Observed Effect Level; the lowest level or concentration resulting in no observed effect

LOEC = Lowest Observed Effect Level; the lowest level or concentration resulting in an observed effect

LC₅₀ = Lethal concentration 50, concentration expected to cause mortality to 50 percent of test organisms.

Mean $LC_{50} \pm 2$ SD = Historical mean of LC_{50} data for previous tests conducted at Nautilus, plus or minus two standard deviations.

REFERENCES

- California State Water Resources Control Board (SWRCB) 2014. Draft Staff Report Including the Draft Substitute Environmental Documentation Amendment to the Water Quality Control Plan For Ocean Waters of California Addressing Desalination Facility Intakes, Brine Discharges, and the Incorporation of other Non-substantive Changes. Sacramento, CA, July 2014.
- Phillips, B.M., B.S. Anderson, K. Siegler, J.P. Voorhees, S. Katz, L. Jennings and R.S. Tjeerdema. 2012. Hyper-Saline Toxicity Thresholds for Nine California Ocean Plan Toxicity Test Protocols. Final Report. University of California, Davis, Department of Environmental Toxicology at Grand Canyon.
- Roberts, P., S. Jenkins, J. Paduan, D. Schlenk and J. Weis. 2012. Management of Brine Discharges to Coastal Waters: Recommendations of a Science Advisory Panel. Environmental Review Panel (ERP). Southern California Coastal Water Research Project (SCCWRP). Costa Mesa, CA. Technical Report 694. http://www.swrcb.ca.gov/water_issues/programs/ocean/desalination/docs/dpr.pdf
- Tidepool Scientific Software. 2000-2013. CETIS Comprehensive Environmental Toxicity Information System Software, Version 1.8.7.20.
- USEPA 1995. Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to West Coast Marine and Estuarine Organisms. EPA/600/R-95/136, August 1995.

APPENDIX A

Chronic Test Data Summaries

Appendix Table A-1. Summary of Salinity Tolerance Chronic Test Results Red Abalone (*Haliotis rufescens*) 48-hour Development

Nominal Test Concentration	December 10, 2014	Test Date	May 20, 2015 Tes	0, 2015 Test Date	
(ppt)	Mean Percent Normal Development	Standard Deviation	Mean Percent Normal Development	Standard Deviation	
Lab Control (LC)	94.0	2.6	96.6	2.7	
Brine Control (BC)	94.6	1.8	97.0	2.6	
35.0	95.0	2.6	98.4	1.7	
35.5	94.6	2.3	96.8	2.2	
36.0	90.6	1.8	96.4	1.7	
36.5	63.4	7.7	83.4	7.7	
37.0	22.6	7.4	78.6	10	
37.5	1.20	2.7	34.2	5.8	
38.0	0.00	0.0	7.20	6.4	
38.5	0.00	0.0	1.40	1.1	
NOEC	36.0		36.0		
LOEC	36.5		36.5		
EC ₂₅ /EC ₅₀	36.4/36.7		37.1/37.3		

The measured test concentrations were within 0.1 ppt of each nominal concentration at test initiation. All statistical endpoints (i.e., NOEC, LOEC, EC_{25}/EC_{50}) were calculated based on initial measured test concentrations.

Values in **bold** indicate a statistically significant decrease compared to the lab control using Dunnett's Multiple Comparison Test.

NOEC = No Observed Effect Concentration, or the level at which there is no adverse effect.

LOEC = Lowest Observed Effect Concentration, or the lowest concentration at which there is a statistically significant adverse effect.

Appendix Table A-2. Summary of Salinity Tolerance Chronic Test Results Purple Urchin (*Strongylocentrotus purpuratus*) 72-hour Development

Nominal Test Concentration	October 30, 2014 T	est Date	July 22, 2015 Tes	t Date
(ppt)	Mean Percent Normal Development	Standard Deviation	Mean Percent Normal Development	Standard Deviation
Lab Control (LC)	95.6	2.1	96.8	1.9
Brine Control (BC)	96.2	1.1	96.4	3.6
35.0	95.6	1.8	95.4	1.8
35.5	95.0	1.9	95.4	2.2
36.0	95.8	2.3	95.0	2.1
36.5	94.0	3.8	94.6	3.9
37.0	87.4	4.5	92.2	5.4
37.5	82.6	7.7	81.6	5.2
38.0	74.4	8.0	63.8	10
38.5	52.4	8.8	26.0	10
NOEC	36.5		36.9	
LOEC	37.0		37.4	
EC ₂₅ /EC ₅₀	38.1/>38.5		37.7/38.1	

The measured test concentrations were within 0.1 ppt of each nominal concentration at test initiation. All statistical endpoints (i.e., NOEC, LOEC, EC $_{25}$ /EC $_{50}$) were calculated based on initial measured test concentrations.

Values in **bold** indicate a statistically significant decrease compared to the lab control using Dunnett's Multiple Comparison Test.

NOEC = No Observed Effect Concentration, or the level at which there is no adverse effect.

LOEC = Lowest Observed Effect Concentration, or the lowest concentration at which there is a statistically significant adverse effect.

Appendix Table A-3. Summary of Salinity Tolerance Chronic Test Results Sand Dollar (*Dendraster excentricus*) 72-hour Development

Nominal Test Concentration	October 30, 2014 T	est Date	July 22, 2015 Tes	t Date
(ppt)	Mean Percent Normal Development	Standard Deviation	Mean Percent Normal Development	Standard Deviation
Lab Control (LC)	97.2	0.84	96.4	1.5
Brine Control (BC)	96.8	2.7	96.0	3.0
35.0	98.4	1.7	97.6	1.7
35.5	98.4	1.1	96.2	1.8
36.0	98.8	1.6	96.2	1.9
36.5	96.8	1.3	94.4	4.0
37.0	97.8	1.3	96.2	3.1
37.5	98.0	1.2	96.6	0.89
38.0	97.4	3.0	93.0	2.2
38.5	97.0	1.9	94.2	2.5
NOEC	38.5		38.5	
LOEC	>38.5		>38.5	
EC ₂₅ /EC ₅₀	>38.5/>38.5	5	>38.5/>38.5	5

The measured test concentrations were within 0.1 ppt of each nominal concentration at test initiation. All statistical endpoints (i.e., NOEC, LOEC, EC_{25}/EC_{50}) were calculated based on initial measured test concentrations.

Values in **bold** indicate a statistically significant decrease compared to the lab control using Dunnett's Multiple Comparison Test.

NOEC = No Observed Effect Concentration, or the level at which there is no adverse effect.

LOEC = Lowest Observed Effect Concentration, or the lowest concentration at which there is a statistically significant adverse effect.

Appendix Table A-4. Summary of Salinity Tolerance Chronic Test Results Purple Urchin (*Strongylocentrotus purpuratus*) Fertilization

Nominal Test Concentration	October 30, 2014 1	Test Date	July 22, 2015 Tes	st Date
(ppt)	Mean Percent Fertilization	Standard Deviation	Mean Percent Fertilization	Standard Deviation
Lab Control (LC)	80.8	6.8	93.6	2.4
Brine Control (BC)	83.6	3.6	93.4	3.1
35.0	86.8	6.3	94.6	3.2
35.5	82.8	4.3	93.4	2.1
36.0	83.2	2.5	95.2	2.8
36.5	84.8	6.3	96.4	1.5
37.0	84.2	4.1	94.0	2.1
37.5	82.2	4.0	95.6	1.5
38.0	82.2	2.5	92.4	2.5
38.5	81.6	3.8	91.8	4.0
NOEC	38.5		38.5	
LOEC	>38.5		>38.5	
EC ₂₅ /EC ₅₀	>38.5/>38.	5	>38.5/>38.	5

The measured test concentrations were within 0.1 ppt of each nominal concentration at test initiation. All statistical endpoints (i.e., NOEC, LOEC, EC $_{25}$ /EC $_{50}$) were calculated based on initial measured test concentrations.

Values in **bold** indicate a statistically significant decrease compared to the lab control using Dunnett's Multiple Comparison Test.

NOEC = No Observed Effect Concentration, or the level at which there is no adverse effect.

LOEC = Lowest Observed Effect Concentration, or the lowest concentration at which there is a statistically significant adverse effect.

Appendix Table A-5. Summary of Salinity Tolerance Chronic Test Results Sand Dollar (*Dendraster excentricus*) Fertilization

Nominal Test Concentration	October 30, 2014	Test Date	July 22, 2015 Te	st Date
(ppt)	Mean Percent Fertilization	Standard Deviation	Mean Percent Fertilization	Standard Deviation
Lab Control (LC)	97.0	1.7	88.6	3.1
Brine Control (BC)	96.2	1.6	90.4	3.3
35.0	96.8	1.8	90.6	2.8
35.5	95.2	1.1	91.0	6.1
36.0	94.8	2.4	89.6	3.4
36.5	96.0	1.9	92.6	2.8
37.0	96.4	1.3	92.0	1.4
37.5	93.6	3.4	93.6	1.9
38.0	93.8	4.4	88.6	4.2
38.5	95.6	3.1	91.2	3.3
NOEC	38.5		38.5	
LOEC	>38.5		>38.5	
EC ₂₅ /EC ₅₀	>38.5/>38.	5	>38.5/>38.5	

The measured test concentrations were within 0.1 ppt of each nominal concentration at test initiation. All statistical endpoints (i.e., NOEC, LOEC, EC_{25}/EC_{50}) were calculated based on initial measured test concentrations.

Values in **bold** indicate a statistically significant decrease compared to the lab control using Dunnett's Multiple Comparison Test.

NOEC = No Observed Effect Concentration, or the level at which there is no adverse effect.

LOEC = Lowest Observed Effect Concentration, or the lowest concentration at which there is a statistically significant adverse effect.

 EC_{25}/EC_{50} = The calculated test concentration at which 25 or 50 percent of the test organisms are adversely affected.

Appendix Table A-6. Summary of Salinity Tolerance Chronic Test Results Giant Kelp (*Macrocystis pyrifera*) 48-hour Germination and Growth

Nominal Test Concentration –	May 20, 2015 Test Date			
(ppt)	Mean Percent Germination	Standard Deviation	Mean Growth (μm)	Standard Deviation
Lab Control (LC)	89.4	4.2	14.6	0.3
Brine Control (BC)	92.2	3.3	14.4	1.1
35.0	92.4	2.8	15.3	1.5
35.5	91.2	3.8	15.3	1.5
36.0	91.0	5.6	15.9	0.68
36.5	94.2	2.8	16.0	0.61
37.0	90.4	2.1	16.0	0.95
37.5	91.8	1.9	16.4	0.52
38.0	93.8	3.3	15.8	0.53
38.5	92.6	5.4	15.3	1.1
NOEC	38.5 38.5			
LOEC	>38.5		>38.5	
EC ₂₅ /EC ₅₀	>38.5/>38.5	5	>38.5/>38.5	

The measured test concentrations were within 0.1 ppt of each nominal concentration at test initiation. All statistical endpoints (i.e., NOEC, LOEC, EC_{25}/EC_{50}) were calculated based on initial measured test concentrations.

Values in **bold** indicate a statistically significant decrease compared to the lab control using Dunnett's Multiple Comparison Test.

NOEC = No Observed Effect Concentration, or the level at which there is no adverse effect.

LOEC = Lowest Observed Effect Concentration, or the lowest concentration at which there is a statistically significant adverse effect.

Appendix Table A-7. Summary of Salinity Tolerance Chronic Test Results Pacific Topsmelt (*Atherinops affinis*) 7-day Survival and Growth

Nominal Test Concentration	May 5, 2015 Test Date				
(ppt)	Mean Percent Survival	Standard Deviation	Mean Growth (mg/org.)	Standard Deviation	
Lab Control (LC)	88.0	18	1.16	0.14	
Brine Control (BC)	96.0	8.9	1.08	0.10	
35.0	100	0.0	1.04	0.05	
35.5	100	0.0	1.11	0.05	
36.0	96.0	8.9	1.14	0.21	
36.5	100	0.0	1.20	0.23	
37.0	92.0	18	0.90	0.14	
37.5	92.0	11	1.08	0.12	
38.0	96.0	8.9	1.10	0.15	
38.5	92.0	11.0	1.07	0.13	
NOEC	38.5		38.5		
LOEC	>38.5		>38.5		
EC ₂₅ /EC ₅₀	>38.5/>38.5		>38.5/>38.5		

The measured test concentrations were within 0.1 ppt of each nominal concentration at test initiation. All statistical endpoints (i.e., NOEC, LOEC, EC_{25}/EC_{50}) were calculated based on initial measured test concentrations.

Values in **bold** indicate a statistically significant decrease compared to the lab control using Dunnett's Multiple Comparison Test.

NOEC = No Observed Effect Concentration, or the level at which there is no adverse effect.

LOEC = Lowest Observed Effect Concentration, or the lowest concentration at which there is a statistically significant adverse effect.

APPENDIX B

Chronic Test Data Raw Data and Statistical Analyses Red Abalone 48-hour Larval Development

Test Date: December 10, 2014

CETIS Summary Report

Ending Date: 12 Dec-14 17:00

Report Date:

07 Aug-15 11:15 (p 1 of 1) 1/12-S285 | 17-5757-1101

rest Code:	1412-5265	17-3737-1101
	Nautilus Enviro	nmental (CA)

Red Abalone Larval Development Test Batch ID:

14-4277-1885 Test Type: Development 10 Dec-14 15:30

Protocol: EPA/600/R-95/136 (1995) Species: Haliotis rufescens

Analyst: Diluent:

Natural Seawater

Brine:

Not Applicable

Age:

Duration: 50h

05-4416-6489 Sample Date: 10 Dec-14

Noutilus Brine Code: 14-817(15 Brined seawater

American Abalone

Client:

Poseidon

Receive Date: 10 Dec-14

Material: Source:

Source:

Poseidon

Project:

Sample Age: 16h

Station:

Batch Note:

Start Date:

Sample ID:

Linear interpolation is reported for consistency among statistical point estimate methods over all tests conducted. Linear

regression also analyzed for comparison.

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
00-3414-5689	Development Rate	36	36.5	36.25	5.83%		Dunnett Multiple Comparison Test

Point Estimate	Summary
----------------	---------

Analysis ID	Endpoint	Level	ppt	95% LCL	95% UCL TU	Method
15-5698-7246	Development Rate	EC25	36.42	36.36	36.48	Linear Regression (MLE)
		EC50	36.69	36.64	36.73	
02-9593-9815	Development Rate	EC25	36.36	36.26	36.51	Linear Interpolation (ICPIN)
	,	EC50	36.7	36.62	36.78	•

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits	Overlap	Decision
00-3414-5689	Development Rate	Control Resp	0.94	0.8 - NL	Yes	Passes Acceptability Criteria
02-9593-9815	Development Rate	Control Resp	0.94	0.8 - NL	Yes	Passes Acceptability Criteria
15-5698-7246	Development Rate	Control Resp	0.94	0.8 - NL	Yes	Passes Acceptability Criteria
00-3414-5689	Development Rate	PMSD	0.05833	NL - 0.2	No	Passes Acceptability Criteria

Development Rate Summary

C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Brine Control	5	0.946	0.9234	0.9686	0.92	0.97	0.008124	0.01817	1.92%	0.0%
0	Lab Control	5	0.94	0.9071	0.9729	0.91	0.98	0.01183	0.02646	2.82%	0.63%
35		5	0.95	0.9183	0.9817	0.91	0.98	0.0114	0.0255	2.68%	-0.42%
35.5		5	0.946	0.9174	0.9746	0.91	0.97	0.0103	0.02302	2.43%	0.0%
36		5	0.906	0.8834	0.9286	0.88	0.93	0.008124	0.01817	2.01%	4.23%
36.5		5	0.634	0.5388	0.7292	0.57	0.75	0.03429	0.07668	12.09%	32.98%
37		5	0.226	0.1345	0.3175	0.16	0.32	0.03295	0.07369	32.61%	76.11%
37.5		5	0.012	0	0.04532	0	0.06	0.012	0.02683	223.6%	98.73%
38		5	0	0	0	0	0	0	0		100.0%
38.5		5	0	0	0	0	0	0	0		100.0%

Development Rate Detail

C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Brine Control	0.97	0.92	0.95	0.95	0.94
0	Lab Control	0.98	0.93	0.95	0.93	0.91
35		0.91	0.95	0.98	0.96	0.95
35.5		0.97	0.96	0.94	0.95	0.91
36		0.88	0.93	0.91	0.91	0.9
36.5		0.67	0.57	0.61	0.57	0.75
37		0.16	0.19	0.32	0.17	0.29
37.5		0.06	0	0	0	0
38		0	0	0	0	0
38.5		0	0	0	0	0

Report Date: Test Code: 30 Jun-15 13:57 (p 1 of 2) 1412-S285 | 17-5757-1101

Red Abalone Larval Development Test Nautilus Environmental (CA) Endpoint: Development Rate **CETIS Version:** CETISv1.8.7 Analysis ID: 00-3414-5689 Parametric-Control vs Treatments Official Results: Yes Analysis: Analyzed: 30 Jun-15 13:56 **PMSD** NOEL LOEL **TOEL** TU **Data Transform** Trials Seed Alt Hyp Zeta 5.83% 36.25 Angular (Corrected) NA C > T NA NA 36 36.5

Dunnett Multi	ple C	omparison Test							
Control	vs	C-ppt	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(α:5%)
Lab Control		35	-0.5021	2.407	0.104	8	0.9522	CDF	Non-Significant Effect
		35.5	-0.2519	2.407	0.104	8	0.9145	CDF	Non-Significant Effect
		36	1.594	2.407	0.104	8	0.2140	CDF	Non-Significant Effect
		36.5*	9.393	2.407	0.104	8	<0.0001	CDF	Significant Effect
		37*	19.36	2.407	0.104	8	<0.0001	CDF	Significant Effect
		37.5*	28.65	2.407	0.104	8	<0.0001	CDF	Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)
Between	7.50991	1.251652	6	267.3	<0.0001	Significant Effect
Error	0.1311219	0.004682924	28			
Total	7.641032		34			

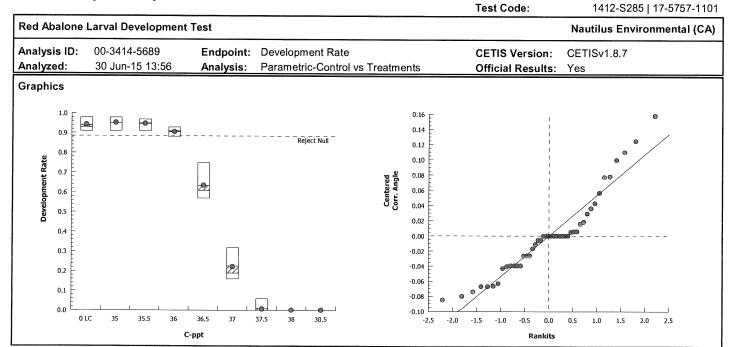
Distributional To	ests				
Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance	5.01	16.81	0.5425	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9331	0.9146	0.0346	Normal Distribution

Developm	ent Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.94	0.9071	0.9729	0.93	0.91	0.98	0.01183	2.82%	0.0%
35		5	0.95	0.9183	0.9817	0.95	0.91	0.98	0.0114	2.68%	-1.06%
35.5		5	0.946	0.9174	0.9746	0.95	0.91	0.97	0.0103	2.43%	-0.64%
36		5	0.906	0.8834	0.9286	0.91	0.88	0.93	0.008124	2.01%	3.62%
36.5		5	0.634	0.5388	0.7292	0.61	0.57	0.75	0.03429	12.09%	32.55%
37		5	0.226	0.1345	0.3175	0.19	0.16	0.32	0.03295	32.61%	75.96%
37.5		5	0.012	0	0.04532	0	0	0.06	0.012	223.6%	98.72%
38		5	0	0	0	0	0	0	0		100.0%
38.5		5	0	0	0	0	0	0	0		100.0%

Angular (Co	rrected) Transfor	med Sumn	nary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.329	1.252	1.407	1.303	1.266	1.429	0.02788	4.69%	0.0%
35		5	1.351	1.278	1.424	1.345	1.266	1.429	0.02615	4.33%	-1.64%
35.5		5	1.34	1.279	1.402	1.345	1.266	1.397	0.02219	3.7%	-0.82%
36		5	1.26	1.222	1.299	1.266	1.217	1.303	0.01395	2.48%	5.19%
36.5		5	0.9227	0.8217	1.024	0.8963	0.8556	1.047	0.0364	8.82%	30.58%
37		5	0.4915	0.3831	0.5999	0.451	0.4115	0.6013	0.03903	17.76%	63.03%
37.5		5	0.08951	-0.02013	0.1991	0.05002	0.05002	0.2475	0.03949	98.65%	93.27%
38		5	0.05002	0.05001	0.05003	0.05002	0.05002	0.05002	0	0.0%	96.24%
38.5		5	0.05002	0.05001	0.05003	0.05002	0.05002	0.05002	0	0.0%	96.24%

Report Date:

30 Jun-15 13:57 (p 2 of 2)



Report Date: Test Code: 30 Jun-15 13:57 (p 1 of 2)

1412-S285 | 17-5757-1101

								Test	Code:	141	2-S285 1	7-5757-1101
Red Ab	alone La	rval Developn	nent Test							Nautilus	s Environi	mental (CA)
Analysi Analyze		5-5698-7246 30 Jun-15 13:5		-	Development F inear Regress				IS Version: ial Results:	CETISv1 Yes	.8.7	
Linear I	Regressi	on Options										
Model F	unction			Thresh	old Option	Threshold	Optimized	Pooled	Het Corr	Weighted	i	
Log-Nor	mal [NE	D=A+B*log(X)]		Control	Threshold	0.06	Yes	No	Yes	Yes		
Regres	sion Sun	nmary										
Iters	LL	AlCc	BIC	Mu	Sigma	Adj R2	F Stat	Critical	P-Value	Decision((a:5%)	
7	-1104	2215	2220	1.565	0.004658	0.9807	-0.3747	2.364	1.0000	Non-Signi	ficant Lack	k of Fit
Point E	stimates											
Level	ppt	95% LCL	95% UCL									
EC25	36.42	36.36	36.48									
EC50	36.69	36.64	36.73									
Regress	sion Para	ameters										
Parame	ter	Estimate	Std Error	95% LC	L 95% UCL	t Stat	P-Value	Decision((a:5%)			
Thresho	ld	0.05502	0.006813	0.04127	0.06877	8.076	<0.0001	Significan	t Parameter			
Slope		214.7	11.12	192.2	237.1	19.3	<0.0001	Significan	t Parameter			
Intercep	t	-335.9	17.41	-371	-300.7	-19.29	<0.0001	Significan	t Parameter			
ANOVA	ANOVA Table											
Source		Sum Squa	ıres Mea	n Square	DF	F Stat	P-Value	Decision((a:5%)			
Model		3144.959		4.959	1	2237	<0.0001	Significant				
Lack of		-3.93311		5552	6	-0.3747	1.0000	Non-Signi	ficant			
Pure Err		62.98253		9515	36							
Residua		59.04942	1.40 	5939	42							
Residua	al Analys	is										
Attribut		Method			Test Stat		P-Value	Decision(
Goodne	ss-of-Fit	Pearson C	•		59.05	58.12	0.0422	-	t Heterogenit	-		
\		Likelihood			63.56	58.12	0.0174	_	t Heterogenit	ty		
Variance			e Equality		e 1.323 0.9177	2.305	0.2746	Equal Vari		_		
Distribut	ЮП	· · · · · · · · · · · · · · · · · · ·	ilk W Norm Darling A2∃	-	1.152	0.9498 2.492	0.0035 0.0052		al Distributio al Distributio			
			Daning A2	Normanty	1,102		B-101110 - 11 - 11 - 11		ai Distributio	II		
-		te Summary	0 1				ated Variate		C) (0)	0/ 55 4	Α.	
C-ppt		Control	Count 5	Mean 0.94	Min	Max 0.98	Std Err	Std Dev	CV% 2.82%	%Effect 0.0%	470	B 500
Λ	Lab	Control	5	0.95	0.91 0.91	0.98	0.01183 0.0114	0.02646 0.0255	2.82% 2.68%	-1.06%	470 475	500
-			5	0.946	0.91	0.98	0.0114	0.02302	2.43%	-0.64%	473	500
35			9		0.88	0.93	0.0103	0.02302	2.43%	3.62%	453	500
35 35.5			5	() unk			J.UUU 124	0.01017	£.UI/U	J.UZ /0	700	500
35 35.5 36			5 5	0.906 0.634				0.07668	12.09%	32.55%	317	500
35 35.5 36 36.5			5	0.634	0.57	0.75	0.03429	0.07668 0.07369	12.09% 32.61%	32.55% 75.96%	317 113	500 500
35.5 36 36.5 37			5 5	0.634 0.226	0.57 0.16	0.75 0.32	0.03429 0.03295	0.07369	32.61%	75.96%	113	500
35 35.5 36 36.5			5	0.634	0.57	0.75	0.03429					

Report Date: Test Code:

Development Rate

30 Jun-15 13:57 (p 2 of 2) 1412-S285 | 17-5757-1101

Red Abalone Larval Development Test Nautilus Environmental (CA) Analysis ID: 15-5698-7246 Endpoint: Development Rate **CETIS Version:** CETISv1.8.7 Analyzed: 30 Jun-15 13:56 Analysis: Linear Regression (MLE) Official Results: Yes Graphics Log-Normal [NED=A+B*log(X)] 1.01 3.0 F 0.91 0.81 2.0 0.71 0.61 0.50 0,40 0.30 0.20 0.10 -1.5 0.00 C-ppt Rankits 3,0 3.0 2.5 2.5 2.0 2.0 1.5 Weighted Residual Weighted Residual 1.0 1.0 0.5 0.0 -0.5 -0.5 -1.5

C-ppt

Report Date: Test Code:

07 Aug-15 11:14 (p 1 of 1)

1412-S285 | 17-5757-1101

Red Abalone Larval Development Test

Nautilus Environmental (CA)

Analysis ID: 02-9593-9815 Analyzed: 07 Aug-15 11:13

Endpoint: Development Rate

Analysis: Linear Interpolation (ICPIN) **CETIS Version:**

CETISv1.8.7

Batch Note: Linear interpolation is reported for consistency among statistical point estimate methods over all tests conducted. Linear

Official Results: Yes

regression also analyzed for comparison.

Linear Interpolation Options

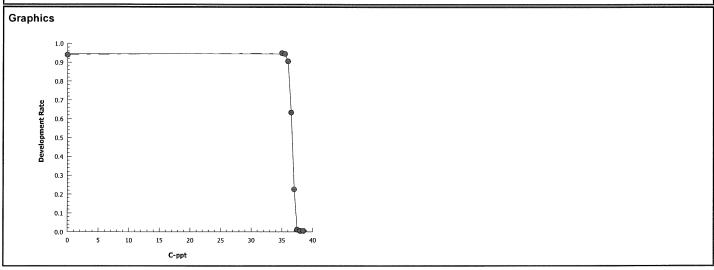
X Transform Y Transform Seed Resamples Exp 95% CL Method Linear Linear 1835851 1000 Yes Two-Point Interpolation

Point Estimates

Level ppt 95% LCL 95% UCL EC25 36.36 36.26 36.51 EC50 36.7 36.62 36.78

Development Rate Summary			Calculated Variate(A/B)								
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.94	0.91	0.98	0.01183	0.02646	2.82%	0.0%	470	500
35		5	0.95	0.91	0.98	0.0114	0.0255	2.68%	-1.06%	475	500
35.5		5	0.946	0.91	0.97	0.0103	0.02302	2.43%	-0.64%	473	500
36		5	0.906	0.88	0.93	0.008124	0.01817	2.01%	3.62%	453	500
36.5		5	0.634	0.57	0.75	0.03429	0.07668	12.09%	32.55%	317	500
37		5	0.226	0.16	0.32	0.03295	0.07369	32.61%	75.96%	113	500
37.5		5	0.012	0	0.06	0.012	0.02683	223.6%	98.72%	6	500
38		5	0	0	0	0	0		100.0%	0	500
38.5		5	0	0	0	0	0		100.0%	0	500

Developn	nent Rate Detail					
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Lab Control	0.98	0.93	0.95	0.93	0.91
35		0.91	0.95	0.98	0.96	0.95
35.5		0.97	0.96	0.94	0.95	0.91
36		0.88	0.93	0.91	0.91	0.9
36.5		0.67	0.57	0.61	0.57	0.75
37		0.16	0.19	0.32	0.17	0.29
37.5		0.06	0	0	0	0
38		0	0	0	0	0
38.5		0	0	0	0	0



Report Date:

09 Dec-14 16:04 (p 1 of 2)

Test Code: 14/2-5285 17-5757-1101/1412-S

Red Abalone Larval Development Test Nautilus Environmental (CA) Start Date: 10 Dec-14 Species: Haliotis rufescens Sample Code: 14-12 Dec-14 End Date: Protocol: EPA/600/R-95/136 (1995) Sample Source: Poseidon Sample Date: 10 Dec-14 Material: Brined seawater Sample Station: C-ppt Code Rep Pos # Counted # Normal Notes 31 DD AD 32 0 Õ 33 34 35 95 a lot of cellular debis A (-715" 36 37 95 38 39 allular debris 107 95 39 40 cellulas debris 41 42 94 43 90 44 little to no cell division 45 0 46 0 47 Little to no cell division 0 48 a cellular debis 49 50 cell war debins 51 91 52 cellulardebris 53 91 54 55 91 56 93 57 58 little to no cell division 59 0 60 little to no cell division 61 cellular debris 62 01 63 Little to no cell dinsion 64 0 65 92 66 93 67 cellular debis 68 16 69 70 71 94 72 little to no cell division 0 73 Little to no cell division 74 75 98/2/312/16/14 76 PA > 80

77

CETIS Test Data Worksheet

Report Date: Test Code:

09 Dec-14 16:04 (p 2 of 2) 17-5757-1101/1412-S

78 0 3 100 79 100 C-ppt Code Rep # Normal Notes Very few surving eggs/ommyos, lots of cell delins Little to no cell dersion, some cell delives 0 100 95 100

Analyst: QA: Sk Vz/17

CETIS Test Data Worksheet

Report Date:

09 Dec-14 16:04 (p 1 of 2)

Test Code: 1412-5285 17-5757-1101/1412-S

Red Abalone Larval Development Test Nautilus Environmental (CA) Start Date: 10 Dec-14 Species: Haliotis rufescens Sample Code: 14-**End Date:** 12 Dec-14 Protocol: EPA/600/R-95/136 (1995) Sample Source: Poseidon Sample Date: 10 Dec-14 Brined seawater Material: Sample Station: C-ppt Code Rep Pos # Counted # Normal Notes AC1412/14 BC вс вс BC ВС AC 12/12/14 LC LC LC LC LC AC 12/12/14 AC12/12/14 35.5 35.5 35.5 35.5 35.5 XC1412/14 36.5 & MIZ/14 Some cellulum debris present 36.5 36.5 36.5 36.5 Lots of cellular defins U 6 12/12/14 & 12/12/14 Very With dendopment 37.5 37.5 37.5 37.5 37.5 tox ounder Little/No development 812/12/14 38.5 38.5

CETIS Test Data Worksheet

Report Date: Test Code: 09 Dec-14 16:04 (p 2 of 2)

17-5757-1101/1412-S

C-ppt	Code		Pos	# Counted	# Normal	Notes
38.5		3	60			
38.5		4	47			
38.5		5	46			

OC=VCR

Analyst: VA QA: AULIGIN

Marine Chronic Bioassay

Water Quality Measurements

Client/Project: Poseidon/Salinity Tolerance Study

Sample ID: Brine (frozen seawater)

Tes

Test No.: 1412-S285

Test Species: H. rufescens

Start Date/Time: 12/10/2014 \530

End Date/Time: 12/12/2014 1700

Concentration (ppt)		Salinity (ppt)		T	emperatu (°C)	re	Diss	solved Ox (mg/L)	ygen		pH (pH units)
	0	24	48	0	24	48	0	24	48	0	24	48
Lab Control	33.0	33.0	53.O	15.0	15.6	155	82	4.O	7.9	8.08	7.98	7.96
Brine Control	34.3	34.3	34.2	15.6	15.3	15.2	8.0	8.0	7.9	8.06	7.98	7.90
35. 🗸	35.0	35.0	34,9	15.2	15.0	15,1	8.0	8.6	7.9	8.05	7.98	7.96
35.5	35.5	35.5	355	15.2	15,0	15,0	8.0	7.9	7.9	8.04	7.98	7.96
36. <i>O</i>	36.0	36.0	36.0	15.3	15.1	15.2	8-0	7.9	7.9	8.03	7.98	7,97
36.5	36,5	36.6	36.6	15.4	14.9	15.0	8-0	3.0	7.9	8.04	7.99	7.98
37, 🖒	37,0	37.0	37.1	15.1	17.9	14,7	8.1	8.0	7.9	8.03	8,00	7.99
37.5	37,5	37.6	37.6	15.0	15.0	15.0	8.0	8.0	7.9	8,02	7,99	7.99
38.0	38,0	39.0	38.1	14.5	14.9	15.0	B. D	4.0	7.9	8.02	7.99	7.99
38.5	38.5	38.5	388.6	14.6	15,1	150	8.1	8.0	7.9	8.02	7.99	7.99
			15115116									

		0	24	48
Technician Initials:	WQ Readings:			NH
	Dilutions made by:	VERIAC	Services.	

Comments:	Ohrs: Hach Sensions	3 meter used to measure	2 salinity
	24 hrs:)
	48 hrs:		
QC Check:	AC 12/16/14	Final Review:	€ 1-22-15

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

Marine Chronic Bioassay

Brine Dilution Worksheet

Project:	Poseidon		Analyst: PA
Sample ID:	frozen seawater		Test Date: 12/10/2014 1530
Test No: 1412-528-5			Test Type: Abalone Development
Salinity of Se	awater	32.9	ACRIS Bring
Salinity of Bri	ine .	91.2	Date of Brine used: 11/5 to 11/25; mixed 12/8/14
Test Dilution	Volume	400	Alkalinity of Brine Control: mg/L as CaCO3

TS = target salinity SE = salinity of effluent SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
34.0	100.0	250	NA	NA	400
35.0	96.4	385.6	0.04	14.4	400
35.5	95.5	382.2	0.05	17.8	400
36.0	94.7	378.7	0.06	21.3	400
36.5	93.8	375.3	0.07	24.7	400
37.0	93.0	371.9	0.08	28.1	400
37.5	92.1	368.4	0.09	31.6	400
38.0	91.3	365.0	0.10	35.0	400
38.5	90.4	361.6	0.11	38.4	400

DI Volume Brine Control 52.6 300 0.73 38.4 400 CS. 1 0.56 Total Brine Volume Required (ml): 76.8

QC Check: AC 12/16/14	Final Review: Sc 1/22/15
a) The brine control was	originally calculated incorrectly Additi
DI water was added +	originally calculated incorrectly Addition achieve the appropriate test salinity

meretore, the percent of brine controlled for all test concentrations except the 38.0 and 38.5 ppt.

Marine Chronic Bioassay

Abalone Embryo-Larval Development

Client:	Poseidon,	Salinity Study		Test Species:	Haliotis rufes	cens
Sample ID:	Brine (From	Salinity Study cen seawater	St.	art Date/Time:	12/10/2014	15
Test No.:	1412-5	285	_ E	nd Date/Time:	12/12/2014	17
Animal Source/Dat	e Received:	American Abalone 12/09/14				ewww.ewedow.com/ststn
Number of abalone	e and condition upo	on receipt/holding:				
Males:	4/6001	ion de term	_			
Females:	41600d	medition	-			
					Females:	1
	Tris & peroxide ad	dition time	- 1	Males:	Temales:	
	Spawn time			1330	1400	
	Number of spawne	ers		4		
	Condition of spaw	n (light, moderate, heavy)		Heavy	moderate	
	Fertilization time			143	5	
	ī			ı		
		Embryo counts (per 0	l			
		1	121			
		2	131			
		3	145			
	l	Mean	8 16. 1			
Time of test Initiation: Technician Initials:	1530			48 hr. QC	97°/s	
Comments:						
QC Check:	AC 12/16/1	f	Final Review:	Se 1-2	22-15	

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

Red Abalone 48-hour Larval Development

Test Date: May 20, 2015

Report Date: Test Code:

23 Jun-15 10:01 (p 1 of 1)

1505-S270 | 17-5102-4942

								Test Co	de:	150	5-S270 17	-5102-494
Red Abalone	Larval Developi	ment Test								Nautilus	s Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	10-1456-6728 20 May-15 14: 22 May-15 15: 50h	15 Pro t 55 Spe	tocol: cies:	Development EPA/600/R-95/ Haliotis rufesce American Abalo	Analyst Diluent: Brine: Age:	: Natu	ral Seawate en Seawate					
Sample ID: Sample Date: Receive Date: Sample Age:		Sou	erial: rce:	010BD3A2D- Notural Seawat Poseidon Nautilus Brine		s rine	2000	Client: Project:	Pose :	eidon		
Comparison S	Summary											
Analysis ID	Endpoint	•		LOEL	TOEL	PMSD	TU	IV	lethod			
00-3926-0772	Development F	Rate	36	36.5	36.25	6.17%		D	unnett M	ultiple Com	parison Tes	t
Point Estimat	e Summary											
Analysis ID	Endpoint		Level ppt 95% LCL 95% UCL			TU	IV	lethod				
13-6034-8183	Development F	Rate	EC25 EC50	37.06 37.34	36.8 37.26	37.15 37.39		Linear Interpolation (ICPIN)			CPIN)	
Test Acceptat	oility							,				
Analysis ID	Endpoint		Attribu	ıte	Test Stat	TAC Limi	ts	0	verlap	Decision		
00-3926-0772	Development F	Rate	Contro	l Resp	0.966	0.8 - NL		Y	'es		cceptability	
13-6034-8183	Development F		Control Resp		0.97	0.8 - NL			'es		cceptability	
00-3926-0772	Development F	Rate	PMSD		0.06168	NL - 0.2		N	lo ————	Passes A	cceptability	Criteria
Development	Rate Summary											
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max		td Err	Std Dev	CV%	%Effect
0	Brine Control	5	0.97	0.9371	1	0.93	1		.01183	0.02646	2.73%	0.0%
0	Lab Control	5	0.966	0.9325	0.9995	0.94	1		.01208	0.02702	2.8%	0.41%
35		5	0.984	0.9632	1	0.96	1		.007483	0.01673 0.02168	1.7% 2.24%	-1.44% 0.21%
35.5 36		5 5	0.968 0.964	0.9411 0.9432	0.9949 0.9848	0.94 0.94	0.99 0.98		.009695	0.02166	2.24% 1.74%	0.62%
36.5		5	0.834	0.9432	0.9296	0.94	0.90		.03444	0.07701	9.23%	14.02%
37		5	0.786	0.6577	0.9290	0.71	0.94		.04622	0.07701	13.15%	18.97%
37.5		5	0.342	0.2699	0.4141	0.25	0.4		.02596	0.05805	16.97%	64.74%
38		5	0.072	0	0.1512	0.01	0.16		.02853	0.0638	88.61%	92.58%
38.5		5	0.014	0	0.02816	0	0.03		.005099		81.44%	98.56%
Development	Rate Detail											
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Brine Control	0.96	0.98	1	0.93	0.98						
0	Lab Control	0.95	1	0.94	0.95	0.99						
35		1	0.96	0.98	1	0.98						
35.5		0.94	0.98	0.98	0.99	0.95						
36		0.98	0.96	0.94	0.96	0.98						
36.5		0.71	0.87	0.88	0.81	0.9						
37		0.82	0.66	0.76	0.75	0.94						
37 37.5		0.82 0.38	0.66 0.25	0.76 0.35	0.75 0.4	0.94 0.33						

@ Nautilus Brine - QISVOR 6/30/15

Analyst: AC QA: 4 6/23/19

CETIS™ v1.8.7.20 000-089-180-4

Report Date:

23 Jun-15 10:00 (p 1 of 2) 1505-S270 | 17-5102-4942

Test Code: **Red Abalone Larval Development Test** Nautilus Environmental (CA) 00-3926-0772 **CETIS Version:** CETISv1.8.7 Analysis ID: Endpoint: Development Rate Analyzed: Parametric-Control vs Treatments Official Results: Yes 22 Jun-15 16:38 Analysis: **Data Transform** Zeta Alt Hyp **Trials** Seed **PMSD** NOEL LOEL TOEL TU NΑ C > T NΑ NA 6.17% 36 36.5 36.25 Angular (Corrected) **Dunnett Multiple Comparison Test** P-Type Decision(a:5%) Control vs C-ppt Test Stat Critical MSD DF P-Value Lab Control -0.925 2.478 0.141 8 0.9903 CDF Non-Significant Effect 35 2.478 8 Non-Significant Effect 35.5 0.029 0.141 0.8820 CDF 36 0.2994 2.478 0.141 8 0.8036 CDF Non-Significant Effect 36.5* 4.267 2.478 0.141 8 0.0005 CDF Significant Effect 0.141 8 < 0.0001 2.478 CDF Significant Effect 37* 5.248 37.5* 13.65 2.478 0.141 8 < 0.0001 CDF Significant Effect 38* 20.31 2.478 0.141 8 < 0.0001 CDF Significant Effect Significant Effect 38.5* 22.6 2.478 0.141 8 < 0.0001 CDF **ANOVA Table** Mean Square DF F Stat P-Value Decision(a:5%) Source Sum Squares 8 < 0.0001 1.379343 170 Significant Effect Between 11.03475 0.2921088 0.008114133 36 Error 11.32686 44 Total **Distributional Tests** Attribute Test Stat Critical P-Value Decision(α:1%) 10.49 20.09 0.2325 Variances Bartlett Equality of Variance **Equal Variances** Distribution Shapiro-Wilk W Normality 0.9729 0.9308 0.3681 Normal Distribution **Development Rate Summary** 95% UCL CV% C-ppt **Control Type** Count Mean 95% LCL Median Min Max Std Err %Effect 0 Lab Control 5 0.966 0.9325 0.9995 0.95 0.94 1 0.01208 2.8% 0.0% 5 0.984 0.9632 0.98 0.96 0.007483 1.7% -1.86% 35 1 1 35.5 5 0.968 0.9411 0.9949 0.98 0.94 0.99 0.009695 2.24% -0.21% 36 5 0.964 0.9432 0.9848 0.96 0.94 0.98 0.007484 1.74% 0.21% 36.5 5 0.834 0.7384 0.9296 0.87 0.71 0.9 0.03444 9.23% 13.66% 5 0.786 0.6577 0.9143 0.76 0.66 0.94 0.04622 13.15% 18.63% 37 37.5 5 0.342 0.2699 0.4141 0.35 0.25 0.4 0.02596 16.97% 64.6% 38 5 0.072 0 0.1512 0.08 0.01 0.16 0.02853 88.61% 92.55% 38.5 5 0.014 0 0.02816 0.01 0 0.03 0.005099 81.44% 98.55% Angular (Corrected) Transformed Summary C-ppt **Control Type** Count Mean 95% LCL 95% UCL Median Min Max Std Err CV% %Effect 6.33% 0.0% Lab Control 5 1.401 1.291 1.511 1.345 1.323 1.521 0.03965 0 35 5 1.454 1.372 1.535 1.429 1.369 1.521 0.02944 4.53% -3.76% 35.5 5 1.399 1.322 1 477 1.429 1.323 1.471 0.02787 4.45% 0.12% 1.429 0.02017 3.26% 1.22% 5 1.369 1.323 36 1.384 1.328 1.44 36.5 5 1.035 1.281 1.202 1.002 1.249 0.04442 8.58% 17.35% 1.158 0.06262 12.71% 21.34% 37 5 1.102 0.9282 1.276 1.059 0.9483 1.323 5 0.5236 0.6847 0.02794 10.02% 55.5% 37.5 0.5459 0.7011 0.6331 0.6235 0.06217 56.96% 82.58% 5 0.1002 0.4115 38 0.2441 0.07145 0.4167 0.2868

Analyst: AC QA: 46173/15

41.55%

91.92%

0.1717

0.1002

0.05002

0.1741

0.02105

0.05482

0.1133

38.5

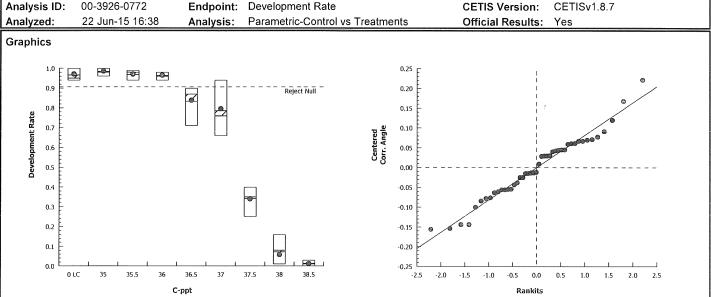
5

Report Date: Test Code: 23 Jun-15 10:01 (p 2 of 2) 1505-S270 | 17-5102-4942

Red Abalone Larval Development Test

Analysis ID: 00-3926-0772 Endpoint: Development Rate

CETIS Version: CETISv1.8.7



Report Date: Test Code: 23 Jun-15 10:01 (p 1 of 1)

1505-S270 | 17-5102-4942

Red Abalone Larval Development Test	Nautilus Environmental (CA)
-------------------------------------	-----------------------------

Analysis ID: 13-6034-8183 Endpoint: Development Rate CETIS Version: CETISv1.8.7

Analyzed: 23 Jun-15 10:00 Analysis: Linear Interpolation (ICPIN) Official Results: Yes

Linear Interpola	Linear Interpolation Options											
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method							
Linear	Linear	742320	1000	Yes	Two-Point Interpolation							

Point Estimates

Graphics

0.2

Level	ppt	95% LCL	95% UCL
EC25	37.06	36.8	37.15
EC50	37.34	37.26	37.39

Developn	nent Rate Summary	/			Cal	culated Variat	e(A/B)				
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Brine Control	5	0.97	0.93	1	0.01183	0.02646	2.73%	0.0%	485	500
35		5	0.984	0.96	1	0.007483	0.01673	1.7%	-1.44%	492	500
35.5		5	0.968	0.94	0.99	0.009695	0.02168	2.24%	0.21%	484	500
36		5	0.964	0.94	0.98	0.007484	0.01673	1.74%	0.62%	482	500
36.5		5	0.834	0.71	0.9	0.03444	0.07701	9.23%	14.02%	417	500
37		5	0.786	0.66	0.94	0.04622	0.1033	13.15%	18.97%	393	500
37.5		5	0.342	0.25	0.4	0.02596	0.05805	16.97%	64.74%	171	500
38		5	0.072	0.01	0.16	0.02853	0.0638	88.61%	92.58%	36	500
38.5		5	0.014	0	0.03	0.005099	0.0114	81.44%	98.56%	7	500

20

Report Date:

22 Jun-15 16:29 (p 1 of 2) 17-5102-4942/1505-S270

Test Code:

Red Abalone	Larval Developmen	t Test			Nautilus Environmental (CA)
Start Date:	20 May-15 14:15	Species:	Haliotis rufescens	Sample Code:	10BD3A2D
End Date:	22 May-15 15:55	Protocoli	EDA/600/D 05/126 (1005)	Commis Courses	Possidon

End Date:			5 15:5 -		:ol: EPA/600/R-95/136 (1995)	Sample Source: Poseidon
Sample Dat					al: Natural Seawater	Sample Station: Nautilus Brine
C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
			31	00	96	
			32	100	94	
			33	100	(0)	
			34	100	100	
			35	100	96	
			36	100	3	
			37	100	98	
			38	100	100	
			39	100	33	
			40	100	100	
· · · · · · · · · · · · · · · · · · ·			41	100	8-2	
			42	100	94	
			43	100	98	
			44	100	98 95	
			45	(00)	1	
			46	100	93	
			47	100	9.9	
1000 materia			48	100	71	
			49	100	87	
			50	100	16	
			51	100	90	
			52	100	2	
			53	100	40	
			54	100	35	
			55	100	95	
			56	100	<u> </u>	
			57	100	76	
			58	100	94	
			59	100	98	
			60	100	98	
			61	100	96	
			62	100		
			63	100	38	
			64	100	100	
			65	100	98	
			66	100	0	
			67	100	81	
			68	100	66	
			69	100	88,	
			70	100	98	
			71	100	94	
			72	(00)		
			73	100	25	
			74	100	98	
			75	100	99	
			76	100	75	
			77	100	96	

Report Date: Test Code: 22 Jun-15 16:29 (p 2 of 2) 17-5102-4942/1505-S270

C-ppt	Code Rep Pos #Counted #Normal				# Normal	Notes
			78	100	8	
			79	100	95	
			80	100	98	

Analyst: CH QA: 36/23/15

Report Date: 19 May-15 15:21 (p 1 of 2)

Test Code: \\$05-\$27017-5102-4942/685E852E

Red Abalone	Larva	i Dev	elopn	ent Test				Nautilus Environmental (CA)
Start Date: End Date: Sample Date:	22 N	Лау-1 Лау-1 Лау-1 Лау-1	5	Protoc	es: Haliotis ol: EPA/60 al: Natura	00/R-95/136 (1995)	Sample Code: Sample Source: Sample Station:	
C-ppt	Code	Rep	Pos	# Counted	# Normal		Notes	
0	вс	1	31			2000.00	A00000	
0	ВС	2	59					
0	ВС	3	34					
0	вс	4	46					
0	ВС	5	80				ATTENDED TO THE PARTY OF THE PA	
0	LC	1	79					
0	LC	2	64					
0	LC	3	71				A 100 Miles	
0	LC	4	55					
0	LC	5	75					
35		1	40					
35		2	35					
35		3	65					
35		4	38					
35		5	70					
35.5		1	58					
35.5		2	37				1 100	
35,5		3	43					
35.5		4	47					
35.5		5	44					
36		1	60					
36		2	61					
36		3	42					
36		4	77					
36		5	74					
36.5		1	48					
36.5		2	49					
36.5		3	69					
36.5		4	67					
36.5		5	51					
37		1	41					
37		2	68					
37		3	57					
37	+	4	76					

37.5 37.5

37.5

37.5

37.5

38.5

38.5

Report Date: Test Code: 19 May-15 15:21 (p 2 of 2) 17-5102-4942/685E852E

	C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
	38.5		3	62			
İ	38.5		4	52			
1	38.5		5	56			



Analyst: QA: ACC/22/15

Water Quality Measurements

Client: Poseidon	Test Species: H. rufescens
Sample ID: bone (frozen Seawater)	Start Date/Time: 5/20/2015 1415
Sample Log No.:	End Date/Time: 5/22/2015 しょう
Test No.: 1505-5270	

Concentration		Salinity (ppt)		T	Temperature (°C)			Dissolved Oxygen (mg/L)			pH (pH units)		
@18 6/23/15	0	24	48	0	24	48	0	24	48	0	24	48	
Brine Control	33.3	33.2	33 1	15.2	15.6	15.7	8.3	8.3	8.2	8.08	804	802	
Lab Control	33.2	33.3	33.3	14.8	15.5	15.3	8.5	8.3	8.4	8-10	8.05	8.00	
35	35.0	34.9	34.9	14.8	15.3	15.2	8.5	8.3	8:4	8.07	8.06	8.00	
35.5	35.5	35.5	35.5	14.7	15.2	153	8.5	8.3	8:4	8.07	8.06	807	
36	0.95	36.0	36.0	14.7	153	15.1	8.5	8.4	8.4	8.06	806	8.07	
36.5	365	36.5	36.5	14.8	15.1	15.0	8.5	8.3	8.4	8.06	8.06	808	
37	37.0	37.0	37-0	143	15.1	151	8.6	8.4	8.4	8.07	806	8.08	
37.5	37.5	37.5	37.5	14.4	15.1	15:2	8.5	8.4	8.4	8.06	20.8	8.08	
38	380	38.1	380	15.0	15.2	152	8.4	8.3	85	8,05	8.06	8.09	
38.5	38.5	38.5	38.6	14.9	15.1	15.1	8.5	8.3	8:4	8,05	8.05	8.09	

		- 99-1										
Technician Initials:	WQ R Dilutions i	leadings: made by:		24 A6	48 AD							
Comments:	0 hrs: 24 hrs: 48 hrs:		ty me	lasiren	ents u	ith F	tach S	ienslow	5 mete			
QC Check:		5/29/	15	-				Fina	l Review:	G 617	3/15	

Brine Dilution Worksheet

Project:

Poseidon

Analyst: ______

Sample ID:

frozen seawater

Test Date: 5/20 /\S

Test No:

1505-5270

Test Type: How de / Ababre Development & RYS/22/15

Salinity of Seawater

33.2

Salinity of Brine

84.3

Date of Brine used: 4/28/15

Test Dilution Volume

300

Alkalinity of Brine Control: 1 1 2 mg/L as CaCO3

TS = target salinity

SE = salinity of effluent

SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)	
34.0	100.0	250	NA	NA	300	
35.0	96.5	289.4	0.04	10.6	300	0.026418787
35.5	95.5	286.5	0.05	13.5	300	0.033757339
36.0	94.5	283.6	0.06	16.4	300	0.04109589
36.5	93.5	280.6	0.07	19.4	300	0.048434442
37.0	92.6	277.7	0.08	22.3	300	0.055772994
37.5	91.6	274.8	0.09	25.2	300	0.063111546
38.0	90.6	271.8	0.10	28.2	300	0.070450098
38.5	89.6	268.9	0.12	31.1	300	0.07778865

DI Volume

Brine Control	47.9	0.65	31.1	300

Total Brine Volume Required (ml):

197.8

QC Check: AC 5/29/15

Final Review: 4 6/73/15

Abalone Embryo-Larval Development

Final Review: 4 6/23/15

Client:	Poseidun		-	Test Species:	Haliotis rufe	
Sample ID:	Nautilus 1	onne	_ Sta	art Date/Time:	5/20/2015	14
Test No.:	1505-82	10	_ Er	nd Date/Time:	5/22/2015	15
Animal Source/D	Date Received: A	merican Abalone/ 「ラーリー	US			
Number of abalo	one and condition upon	receipt/holding:				
Males:	4, spawned	s/19/15 in holding	_			
Females:	4	<i>0</i>	_			
				Males:	Females:	
	Tris & peroxide addi	tion time		1035	1030	_
	Spawn time			1312	1314	
	Number of spawners	3		3	2-4	_
		(light, moderate, heavy)		moderale	heavy	-
	Fertilization time			132	5	
		Embryo counts (per 1 2 3 Mean	0.5 ml) 124 · 160 292 92			
Time of test Initiation				48 hr. Q0	: 97%	_

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

AC 5/29/15

Comments:

QC Check:

Purple Urchin 72-hour Larval Development

Test Date: October 30, 2014

CETIS Summary Report

Report Date:

30 Jun-15 09:47 (p 1 of 1) 1410-S149 | 21-4399-6332

Test Code:	1410-S149 21-4

	·····							Test Code:	14	10-S149 21	-4399-63
Echinoid Emi	bryo-Larval Dev	elopment	t Test						Nautilu	s Environm	ental (CA
Batch ID: Start Date: Ending Date: Duration:	04-0332-3382 30 Oct-14 15:0 02 Nov-14 17:0 74h	00 P 00 S	est Type: rotocol: pecies: ource:	Development EPA/600/R-95, Strongylocentr Pt. Loma	` ,	tus			tural Seawat zen Seawat		
Sample ID: Sample Date: Receive Date: Sample Age:	: 30 Oct-14	M S	ode: aterial: ource: tation:	Brine Brined seawate Poseidon Nauhus	ete.			Client: Po Project:	seidon		
Sample Note:	Frozen seawat	ter prepare	ed at Nauti	lus was used as	s brine.						
Comparison S	Summary										
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method			
18-9905-4562		Rate	36.5	37	36.75	5.72%			Multiple Com	parison Tes	st .
Point Estimat	e Summary				**************************************						
Analysis ID	Endpoint		Level	ppt	95% LCL	95% UCL	TU	Method			
09-1602-6129	Development F	Rate	EC25 EC50	38.06 >38.5	37.77 N/A	38.22 N/A		Linear In	terpolation (I	CPIN)	
Test Acceptat	oility										
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	its	Overlap	Decision		
09-1602-6129			ol Resp	0.956	0.8 - NL		Yes		cceptability	Criteria	
18-9905-4562	Development F	Rate		l Resp	0.956	0.8 - NL		Yes		cceptability	
18-9905-4562	Development F	Rate	PMSD	\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\	0.05724	NL - 0.25		No	Passes A	cceptability	Criteria
Development	Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effec
0	Brine Control	5	0.962	0.9484	0.9756	0.95	0.98	0.004899	0.01095	1.14%	0.0%
0	Lab Control	5	0.956	0.9303	0.9817	0.94	0.99			2.17%	0.62%
35		5	0.956	0.9334	0.9786	0.93	0.98			1.9%	0.62%
35.5		5	0.95	0.9268	0.9732	0.93	0.97	0.008367	0.01871	1.97%	1.25%
36		5	0.958	0.9297	0.9863	0.92	0.98	0.0102	0.0228	2.38%	0.42%
36.5		5	0.94	0.8927	0.9873	0.89	0.98	0.01703	0.03808	4.05%	2.29%
37		5	0.874	0.8181	0.9299	0.82	0.93	0.02015	0.04506	5.16%	9.15%
37.5		5	0.826	0.7308	0.9212	0.71	0.89	0.03429	0.07668	9.28%	14.14%
38		5	0.744	0.6448	0.8432	0.65	0.85		0.07987	10.74%	22.66%
38.5		5	0.524	0.4141	0.6339	0.39	0.6	0.03957	0.08849	16.89%	45.53%
Development											
	Control Type	Rep 1	Rep 2		Rep 4	Rep 5					
	Brine Control	0.96	0.95	0.96	0.98	0.96					
	Lab Control	0.94	0.94	0.99	0.96	0.95			•		
35		0.98	0.95	0.96	0.96	0.93					
35.5		0.93	0.96	0.93	0.96	0.97					
36		0.98	0.97	0.96	0.92	0.96					
36.5		0.96	0.91	0.96	0.89	0.98					
37		0.82	0.85	0.86	0.91	0.93					
37.5		0.85	0.89	0.71	0.89	0.79					
38		0.85	0.71	0.8	0.65	0.71					
38.5		0.6	0.48	0.59	0.39	0.56					
JU.U		0.0	0.40	0.03	0.00	0.00					

Report Date:

30 Jun-15 09:46 (p 1 of 2)

Test Code:

 Code:
 1410-S149 | 21-4399-6332

Echinoid Embryo-Larval Development TestNautilus Environmental (CA)Analysis ID:18-9905-4562Endpoint:Development RateCETIS Version:CETISv1.8.7Analyzed:30 Jun-15 9:46Analysis:Parametric-Control vs TreatmentsOfficial Results:Yes

Sample Note: Frozen seawater prepared at Nautilus was used as brine.

Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corrected)	NA	C > T	NA	NA	5.72%	36.5	37	36.75	

Dunnett Multip	ole Co	omparison Test			10000000000				
Control	vs	C-ppt	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(a:5%)
Lab Control		35	0.06846	2.478	0.115	8	0.8722	CDF	Non-Significant Effect
		35.5	0.3886	2.478	0.115	8	0.7720	CDF	Non-Significant Effect
		36	-0.07106	2.478	0.115	8	0.9045	CDF	Non-Significant Effect
		36.5	0.7114	2.478	0.115	8	0.6382	CDF	Non-Significant Effect
		37*	3.311	2.478	0.115	8	0.0069	CDF	Significant Effect
		37.5*	4.717	2.478	0.115	8	0.0001	CDF	Significant Effect
		38*	6.924	2.478	0.115	8	<0.0001	CDF	Significant Effect
		38.5*	11.98	2.478	0.115	8	<0.0001	CDF	Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)
Between	1.491822	0.1864777	8	34.5	<0.0001	Significant Effect
Error	0.1945804	0.005405012	36			
Total	1.686402	The state of the s	44	<u></u>		

Distributional To	ests				
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)
Variances	Bartlett Equality of Variance	5.305	20.09	0.7246	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9797	0.9308	0.6077	Normal Distribution

Developm	ent Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.956	0.9303	0.9817	0.95	0.94	0.99	0.009274	2.17%	0.0%
35		5	0.956	0.9334	0.9786	0.96	0.93	0.98	0.008124	1.9%	0.0%
35.5		5	0.95	0.9268	0.9732	0.96	0.93	0.97	0.008367	1.97%	0.63%
36		5	0.958	0.9297	0.9863	0.96	0.92	0.98	0.0102	2.38%	-0.21%
36.5		5	0.94	0.8927	0.9873	0.96	0.89	0.98	0.01703	4.05%	1.67%
37		5	0.874	0.8181	0.9299	0.86	0.82	0.93	0.02015	5.16%	8.58%
37.5		5	0.826	0.7308	0.9212	0.85	0.71	0.89	0.03429	9.28%	13.6%
38		5	0.744	0.6448	0.8432	0.71	0.65	0.85	0.03572	10.74%	22.18%
38.5		5	0.524	0.4141	0.6339	0.56	0.39	0.6	0.03957	16.89%	45.19%

Angular (C	Angular (Corrected) Transformed Summary											
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	Lab Control	5	1.366	1.29	1.443	1.345	1.323	1.471	0.02741	4.49%	0.0%	
35		5	1.363	1.307	1.42	1.369	1.303	1.429	0.02041	3.35%	0.23%	
35.5		5	1.348	1.295	1.402	1.369	1.303	1.397	0.01915	3.18%	1.32%	
36		5	1.37	1.303	1.436	1.369	1.284	1.429	0.02405	3.93%	-0.24%	
36.5		5	1.333	1.232	1.434	1.369	1.233	1.429	0.03632	6.09%	2.42%	
37		5	1.212	1.125	1.299	1.187	1.133	1.303	0.03133	5.78%	11.27%	
37.5		5	1.147	1.024	1.27	1.173	1.002	1.233	0.04422	8.62%	16.05%	
38		5	1.044	0.9276	1.161	1.002	0.9377	1.173	0.0421	9.01%	23.56%	
38.5		5	0.8095	0.6989	0.9201	0.8455	0.6745	0.8861	0.03984	11.01%	40.76%	

Report Date: Test Code: 30 Jun-15 09:47 (p 2 of 2) 1410-S149 | 21-4399-6332

Echinoid Embryo-Larval Development Test Nautilus Environmental (CA) Analysis ID: 18-9905-4562 Endpoint: Development Rate **CETIS Version:** CETISv1.8.7 Analyzed: 30 Jun-15 9:46 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 1.0 0.16 0.14 Reject Null 0.12 0.10 8.0 0.08 0.7 0.06 0.04 0.6 0.02 0.5 0.00 -0.02 -0.04 -0.06 -0.08 -0.10 -0.12 0.1 -0.14 0.0 -0.16 0 LC 37.5 38.5 -1.5 -0.5 1.0 2.0 C-ppt Rankits

Report Date:

30 Jun-15 09:47 (p 1 of 1)

Test Code:

1410-S149 | 21-4399-6332

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Analysis ID: Analyzed:

09-1602-6129 30 Jun-15 9:46 Endpoint: Development Rate Analysis: Linear Interpolation (ICPIN) **CETIS Version:**

CETISv1.8.7 Official Results: Yes

Sample Note: Frozen seawater prepared at Nautilus was used as brine.

Linear	Interpo	lation O	ptions
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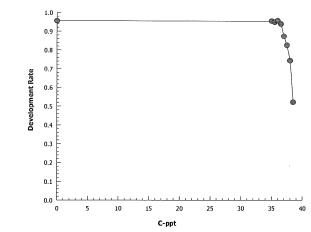
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	1731366	1000	Yes	Two-Point Interpolation

Point Estimates

Level	ppt	95% LCL	95% UC
EC25	38.06	37.77	38.22
EC50	>38.5	N/A	N/A

Developm	nent Rate Summary	у			Cal	culated Variat	te(A/B)				
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.956	0.94	0.99	0.009274	0.02074	2.17%	0.0%	478	500
35		5	0.956	0.93	0.98	0.008124	0.01817	1.9%	0.0%	478	500
35.5		5	0.95	0.93	0.97	0.008367	0.01871	1.97%	0.63%	475	500
36		5	0.958	0.92	0.98	0.0102	0.0228	2.38%	-0.21%	479	500
36.5		5	0.94	0.89	0.98	0.01703	0.03808	4.05%	1.67%	470	500
37		5	0.874	0.82	0.93	0.02015	0.04506	5.16%	8.58%	437	500
37.5		5	0.826	0.71	0.89	0.03429	0.07668	9.28%	13.6%	413	500
38		5	0.744	0.65	0.85	0.03572	0.07987	10.74%	22.18%	372	500
38.5		5	0.524	0.39	0.6	0.03957	0.08849	16.89%	45.19%	261	500

Graphics



Report Date:

28 Oct-14 16:36 (p 1 of 2)

Test Code: (410-5)44 21-4399-6332/1410-S149

				elopment Te				Nautilus Environmental (C
Start Date:	30 (Oct-14	ļ			ylocentrotus purpuratus	Sample Code:	Brine
End Date: Sample Date	1 80 .	Nov-1	4			00/R-95/136 (1995)	Sample Source:	Poseidon
Sample Date	ē: 30 (Oct-14	('1	Materi	al: Brined	seawater	Sample Station:	
C-ppt	Code	Rep	Pos	# Counted	# Normal		Notes	
		-	1	5/2/00 100	8883 65		SESTHULLY	
			2	1 1000	18× H		30,0	
			3	160,000				
			4	1 100,00	17796			
			5	100	93			
			6	100	96	TO COMMANDE TO THE PARTY OF THE		
			7	100	98			
			8	100	71	ed intship		
			9	100	2891	Q21 Recounted QC	check	
	+		10	100	82	acins 86/100		
			11	100	92	06/100		
			12					
			13	160	39 95			***************************************
			14		48			740.41
			15	100		The state of the s		
			16	100	86			
			17		98			
			18	160	79			
			19	160				
			20	160	96			
***************************************			21	100	96			
			22	100	97			
				(00)	71	20 C C C C C C C C C C C C C C C C C C C		
			23	100	91	QC: 5 88/100		
			24	100	56			
			25	100	85			
			26	100	96			
			27	100	60			
			28	(00	59		NN 277	
			29	100	93			
			30	100	96 96			
			31	100	96			
			32	100	96			
			33	too	95			
			34	100	95			
			35	160	93			
			36	100	89			
			37	100	96			
			38	100	93			
			39	100	85			
			40	100	96			
			41	100	98	1/		,
			42	100	97			11 11 12 12 12 12 12 12 12 12 12 12 12 1
			43	100	99	•		V
THE STATE OF THE S			44	100	89	Qcing 83/100		
			45	100	96	V 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0		
			46	100	80			110000000000000000000000000000000000000
			47	100	89			

Report Date: Test Code:

28 Oct-14 16:36 (p 2 of 2) 21-4399-6332/1410-S149

C-ppt	Code	Pos	# Counted	# Normal	Notes
		48	100	85	
		49	100	94	
		50	100	98	

Analyst: EG QACIDS/14

Report Date:

28 Oct-14 16:36 (p 1 of 2)

Test Code: 1410-\$149 21-4399-6332/1410-\$149

Nautilus Environmental (CA)

Start Date: End Date:

30 Oct-14

Echinoid Embryo-Larval Development Test

Species: Strongylocentrotus purpuratus Protocol: EPA/600/R-95/136 (1995)

Sample Code: Brine Sample Source

End Date: Sample Date	169 KE	Noy-14	114			S00/R-95/136 (1995) Sample Source: Poseidon
					al: Brined	seawater Sample Station:
C-ppt		Rep		# Counted	1	Notes
0	BC	1	37	100	98	
0	BC	2	13			
0	BC	3	45			
0	BC	4	7			
0	BC	5	19			
0	LC	1	49			
0	LC	2	18	100	94	
0	LC	3	43	•		
0	LC	4	31			
0	LC	5	33			
35		1	41			
35		2	34			
35		3	26			
35		4	40			
35		5	5			
35.5		1	29			
35.5		2	3			
35.5		3	38			
35.5		4	6			
35.5		5	42			
36		1	16			
36		2	21			
36		3	20			
36		4	11			
36		5	30			
36.5		1	4			
36.5		2	9			
36.5		3	32			
36.5		4	47			
36.5		5	50			
37		1	10			
37		2	25			
37		3	15	100	93	
37		4	23	17	• •	
37		5	35	***************************************		
37.5		1	48			
37.5		2	44			
37.5		3	2			
37.5		4	36			/
37.5		5	17			
38		1	39			
38		2	22			,
38		3	46			
38		4	1			
38		5	8			
38.5		1	27			
	-					
38.5		2	14			



C-ppt 38.5

38.5

38.5

Code Rep Pos

3 28

4

5

Report Date: Test Code

28 Oct-14 16:36 (p 2 of 2) 21-4399-6332/1410-\$149

		rest code.	21-4399-0332/1410-3149
# Counted	# Normal	Notes	

OCEAC

12

24

Water Quality Measurements

Client/Project: Poseidon/Salinity Tolerance Study

Sample ID: Brine (frozen seawater)

Test No.: 1410-S15148 1410-S149

Test Species: S.purpuratus

Start Date/Time: 10/30/2014 15 : 00

End Date/Time: 11/3/2014 | 7.00

11/2/14 Q18

Concentration (ppt)		(p	inity pt)				erature C)		D	issolve (m	d Oxyg g/L)	en		-	H units)	
	0	24	48	72	0	24	48	72	0	24	48	72	0	24	48	72
Lab Control	33,3	33.3	33.4	33.6	15.6	15.7	15.3	15.1	8.7	7.9	7.5	7.8	8.04	7.98	8.07	7.93
Brine Control	33, 7	33,7	33.8	34.3	15.4	157	15.1	14.9	8.8	7.8	7.8	78	8.06	8.00	8.11	7,94
35	34.9	34.9	35.0	35.0	14,8	15.8	15.0	16.0	88	7.7	7.7	7.9	8.07	7.99	8.11	790
35.5	35.5	355	35.5	35.5	15,0	157	15.0	14.9	8.8	8.0	7.7	7.8	8,07	8.01	8.09	7.98
36	36.0	36.0	36.1	36.1	14.7	15.7	14.8	15.0	6.7	8.1	7.8	7.9	8,13	8.15	8.11	7-98
36.5	36.5	36.5	36.5	36.5	14.7	15.8	14.7	15.0	000	7.8	7.8	7.7	8.10	8.05	8.10	7-98
37	37.0	37.0	31.1	37-1	14.8	159	14.8	15.0	8.8	7.7	7.9	7.9	8.09	7.99	8.11	7.98
37.5	37.5	87.5	37.4	37.4	14.9	15.9	4.7	15.0	84	7.8	7.8	7.9	8.08	8.00	8.19	7.99
38	36.0	37,9	38.0	38.0	15.4	15.9	14.6	15.1	8.7	7.8	7.9	7.9	8.06	8.00	8.17	7.99
38.5	38.5	38.4	38.5	38.5	15.6	16.0	4.7	16.1	8 8	7.8	7.9	7.9	8.05	8.00	8.17	8.00
												-				

Technician Initials: WQ Readings: 6 A6 FF AV Dilutions made by: A4

Comments:

0 hrs:

24 hrs:

48 hrs:

72 hrs:

QC Check: <u>AC 12-15-14</u>

Final Review: 47 12/5/14

Echinoderm Larval Development Worksheet

Client:	Possiden Salinty Tolerance Stud	Start Date/Time: 10/30/2014 / 75.00
Sample ID: Test No.:	Brine (for frozen seawater)	End Date/Time: 11/3/2014 114 17 00
rest No	1410-5149	Species: S. purpuratus
Tech initials:	P4	Animal Source: Point Loma
Injection Time:	14:30	Date Collected: 10(6/14
	0.0 - 0	
Sperm Absorbance at	400 nm: 0.823 (target range of 0.8 - 1.0 for de	ensity of 4x10 ⁶ sperm/ml)
		·
	10	
Eggs Counted:	<u>1</u>	<i>lℓ0</i> eggs/ml
	24	
	(target counts of 26 eggs per vertical pa	ass on Sedgwick-Rafter
	slide for a final density of 1000 eggs/ml	,
	7.8	
	<u>~~</u>	
Initial density:	$\underline{(110)}$ eggs/ml = (1) dilution factor	765
Final density:	$\frac{\{\{l\}\} \text{ eggs/ml}}{1000 \text{ eggs/ml}} = \frac{\{l,l\}}{1.0} \text{ dilution factor}$ $\frac{1}{1.0} \text{ part egg stock}$	egg stock 00 ml
Third dorlony.	(0, 11 parts seawater	seawater 422 z ml
	o, i parto sodwater	•
Prepare the egg stock a	according to the calculated dilution factor. For example, if the	e dilution factor is 2.25, use 100 ml of existing stock (1
part) and 125 ml of dilut	tion water (1.25 parts).	· ·
Volume of Sperm stock	needed to fertilize eggs:	
	735	
Egg Stock (mL) =	= 605	
Sperm Stock (μL) =	225	
Egg/Sperm Ratio =	Iml! tal	
E (11) () T	1446	
Fertilization Time:	1 (()	
Embryo Stock Fertilizati	on Checks: No. No.	
•		·⁄
10 minutes (1st fert.)	14:55 100 0	100
20 minutes (2nd fert. If r	needed)	
	16.0	
Test Initiation Time:	Embryo Stock Added:	0.25 ml
Test Termination:		
	No. No. %	
	Normal Abnormal Normal	
72-hour QC check ^a	97 3 97	•
End of test QC check		
	3.00	
Comments:	a If the embryo development does not meet the mean test	acceptability criterion of 80% normally
	developed, continue the test up to 96-hrs (ASTM 1999).	tier .
	·	1 - 121 - 1
QC Check:	K8 1124119	Final Review: AC12(5)14
	40 Vandavar Avanya - San Diana - CA 20400	THE TOYIOW.
ivautiius Erivironmental. 43	40 Vandever Avenue. San Diego, CA 92120.	

Purple Urchin 72-hour Larval Development

Test Date: July 22, 2015

CETIS Summary Report

Report Date:

04 Aug-15 14:41 (p 1 of 1)

Test Code:

1507-S081 | 09-7618-8627

Batch ID:	00-7317-1895		Test Type:	Development				Analyst:		V-V-2000-200-200-00-00-00-00-00-00-00-00-00	•
Start Date:	22 Jul-15 16:1	0	Protocol:	EPA/600/R-95/	136 (1995)			·=	atural Seawat	ter	
	25 Jul-15 16:1		Species:	Strongylocentro	. ,	itus			ozen Seawat		
Duration:	72h		Source:	Pt. Loma	parpara			Age:	Jouwal	. 	
Sample ID:	13-8856-1048		Code:	1507-S081				Client: Po	seidon	· · · · · · · · · · · · · · · · · · ·	
Sample Date:	22 Jul-15		Material:	Brined seawate	er			Project:			
Receive Date:			Source:	Poseidon							
Sample Age:	16h		Station:								
Comparison S	Summary		47,000,000				There exists and suffere)			
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method			
16-2651-6908	Development F	Rate	36.9	37.4	37.15	5.54%		Dunnett	Multiple Com	nparison Tes	st
Point Estimate	e Summary										
Analysis ID	Endpoint		Level	ppt	95% LCL	95% UCL	TU	Method			
18-0652-3476	Development F	Rate	EC25	37.69	37.58	37.77		Linear R	egression (M	1LE)	
	_		EC50	38.13	38.05	38.21				·	
01-1762-9710	Development F	Rate	EC25	37.65	37.49	37.88		Linear In	iterpolation (I	CPIN)	
			EC50	38.14	37.98	38.28					
Test Acceptab	-		•								
Analysis ID	Endpoint Dayslanmant F	2-4-	Attrib			TAC Limi	ts	Overlap			O '' :
01-1762-9710 16-2651-6908	Development F Development F			ol Resp	0.968	0.8 - NL		Yes		cceptability	
18-0652-3476	Development F			ol Resp ol Resp	0.968 0.968	0.8 - NL 0.8 - NL		Yes Yes		cceptability	
16-2651-6908	•		PMSD		0.966	0.6 - NL NL - 0.25		res No		cceptability	
	Rate Summary		55		3.0000	.,_ 0.20		. 10	. 43500 /		Jinona
•	Control Type	Coun	t Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effec
	Brine Control	5	0.964	0.9187	1	0.92	1	0.01631	0.03647	3.78%	0.0%
JU. T			0.968	0.9441	0.9919	0.95	1	0.008602		1.99%	-0.41%
	Lab Control	5				0.93	0.98			1.9%	1.04%
33.5	Lab Control	5 5	0.954	0.9314	0.9766	0.93					4.040/
33.5 34.9	Lab Control			0.9314 0.9268	0.9766	0.93	0.98	0.009798	3 0.02191	2.3%	1.04%
33.5 34.9 35.4	Lab Control	5	0.954				0.98 0.97			2.3% 2.23%	1.45%
33.5 34.9 35.4 35.9	Lab Control	5 5	0.954 0.954	0.9268	0.9812	0.92		0.00948			
33.5 34.9 35.4 35.9 36.4 36.9	Lab Control	5 5 5	0.954 0.954 0.95 0.946 0.922	0.9268 0.9237 0.8974 0.8555	0.9812 0.9763	0.92 0.92	0.97	0.00948 [°] 0.01749	7 0.02121 0.03912 0.05357	2.23%	1.45%
33.5 34.9 35.4 35.9 36.4 36.9 37.4	Lab Control	5 5 5 5 5	0.954 0.954 0.95 0.946 0.922 0.816	0.9268 0.9237 0.8974 0.8555 0.7517	0.9812 0.9763 0.9946 0.9885 0.8803	0.92 0.92 0.88 0.85 0.77	0.97 0.98 0.98 0.89	0.00948' 0.01749 0.02396 0.02315	7 0.02121 0.03912 0.05357 0.05177	2.23% 4.14% 5.81% 6.34%	1.45% 1.87% 4.36% 15.35%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9	Lab Control	5 5 5 5 5 5	0.954 0.954 0.95 0.946 0.922 0.816 0.638	0.9268 0.9237 0.8974 0.8555 0.7517	0.9812 0.9763 0.9946 0.9885 0.8803 0.766	0.92 0.92 0.88 0.85	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9	Lab Control	5 5 5 5 5	0.954 0.954 0.95 0.946 0.922 0.816	0.9268 0.9237 0.8974 0.8555 0.7517	0.9812 0.9763 0.9946 0.9885 0.8803	0.92 0.92 0.88 0.85 0.77	0.97 0.98 0.98 0.89	0.00948' 0.01749 0.02396 0.02315	7 0.02121 0.03912 0.05357 0.05177	2.23% 4.14% 5.81% 6.34%	1.45% 1.87% 4.36%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5	Rate Detail	5 5 5 5 5 5 5	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869	0.92 0.92 0.88 0.85 0.77 0.47	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Development	Rate Detail Control Type	5 5 5 5 5 5 5 7	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869	0.92 0.92 0.88 0.85 0.77 0.47 0.19	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Development	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 5 7 7 8 8 8 9 9 9	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869	0.92 0.92 0.88 0.85 0.77 0.47 0.19	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Development C-ppt	Rate Detail Control Type	5 5 5 5 5 5 5 5 5 7 7 8 8 8 9 9 0.99 0.96	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Development C-ppt 33.4 33.5 34.9	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 7 7 8 8 8 9 9 0.99 0.96 0.95	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93 0.95	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4 1 0.97	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92 0.96 0.96	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Development C-ppt 33.4 33.5 34.9 35.4	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 7 7 8 8 8 9 0.99 0.96 0.95 0.96	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93 0.95 0.95	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98 1 0.98 0.92	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4 1 0.97 0.93 0.96	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92 0.96 0.96 0.95	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 37.9 38.5 Development C-ppt 33.4 33.5 34.9 35.9	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 7 7 8 8 8 9 1 9 1 9 1 9 1 9 1 9 1 9 1 9 1 9	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93 0.95 0.95 0.98 0.97	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98 1 0.98 0.92 0.95	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4 1 0.97 0.93 0.96 0.97	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92 0.96 0.96 0.95 0.94	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 37.9 38.5 Development C-ppt 33.4 33.5 34.9 35.4 35.9	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 5 7 7 8 8 9 9 0.99 0.96 0.95 0.92 0.98	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93 0.95 0.95 0.98 0.97 0.95	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98 1 0.98 0.92 0.95 0.95	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4 1 0.97 0.93 0.96 0.97 0.88	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92 0.96 0.96 0.95 0.94 0.97	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Development C-ppt 33.4 33.5 34.9 35.4 36.4 36.9	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 5 7 7 8 8 9 9 0.99 0.96 0.95 0.99 0.98 0.99	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93 0.95 0.95 0.98 0.97 0.95 0.98	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98 1 0.98 0.92 0.95 0.95 0.85	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4 1 0.97 0.93 0.96 0.97 0.88 0.91	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92 0.96 0.96 0.95 0.94 0.97 0.9	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Development 10 C-ppt 33.4 33.5 34.9 35.4 35.9 36.4 36.9 37.4	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 5 7 7 0.99 0.96 0.95 0.96 0.92 0.98 0.97 0.89	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93 0.95 0.95 0.98 0.97 0.95 0.98 0.77	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98 1 0.98 0.92 0.95 0.95 0.85 0.79	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4 1 0.97 0.93 0.96 0.97 0.88	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92 0.96 0.96 0.95 0.94 0.97 0.9 0.78	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%
33.5 34.9 35.4 35.9 36.4 37.9 38.5 Development C-ppt 33.4 33.5 34.9 35.4 35.9 36.4 36.9	Rate Detail Control Type Brine Control	5 5 5 5 5 5 5 5 5 7 7 8 8 9 9 0.99 0.96 0.95 0.99 0.98 0.99	0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 Rep 2 0.93 0.95 0.95 0.98 0.97 0.95 0.98	0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 Rep 3 0.98 1 0.98 0.92 0.95 0.95 0.85	0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 Rep 4 1 0.97 0.93 0.96 0.97 0.88 0.91	0.92 0.92 0.88 0.85 0.77 0.47 0.19 Rep 5 0.92 0.96 0.96 0.95 0.94 0.97 0.9	0.97 0.98 0.98 0.89 0.74	0.00948' 0.01749 0.02396 0.02315 0.04609	7 0.02121 0.03912 0.05357 0.05177 0.1031	2.23% 4.14% 5.81% 6.34% 16.15%	1.45% 1.87% 4.36% 15.35% 33.82%

Analyst: AC QA: 46M/15

Report Date: Test Code: 04 Aug-15 14:41 (p 1 of 2) 1507-S081 | 09-7618-8627

		***************************************					Test			7-S081 09	
Echinoid Em	nbryo-Larval Dev	elopme	nt Test						Nautilus	Environm	nental (CA
Analysis ID: Analyzed:	16-2651-6908 04 Aug-15 14:		•	evelopment R arametric-Cor		tments		IS Version: ial Results:	CETISv1. Yes	.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	ti ili viiki kali kali ka ka ka palang vii vysi ken peda	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	rected)	NA	C > T	NA	NA		5.54%	36.9	37.4	37.15	
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-ppt		Test Sta	t Critical	MSD DF	P-Value	P-Type	Decision(α:5%)		
33.5	34.9		0.822	2.478	0.126 8	0.5873	CDF	Non-Signif	icant Effect		
33.5	35.4		0.8018	2.478	0.126 8	0.5967	CDF	Non-Signif	icant Effect		
33.5	35.9		1.002	2.478	0.126 8	0.5023	CDF	-	icant Effect		
33.5	36.4		1.052	2.478	0.126 8	0.4784	CDF	-	icant Effect		
33.5	36.9		1.912	2.478	0.126 8	0.1492	CDF	_	icant Effect		
33.5	37.4*		5.283	2.478	0.126 8	<0.0001	CDF	Significant			
33.5	37.9*		9.269	2.478	0.126 8	<0.0001	CDF	Significant			
33.5	38.5*		17.05	2.478	0.126 8	<0.0001	CDF	Significant			
ANOVA Table	e							A STATE OF THE STA			
Source	Sum Squ	ares	Mean Sc	quare	DF	F Stat	P-Value	Decision(a:5%)		
Between	3.356625		0.419578	31	8	64.46	<0.0001	Significant	Effect		
Error	0.234323	4	0.006508	3983	36						
Total	3.590948				44	_					
Distributiona	al Tests		710777777						DOLLOW DOLLAR AND HOUSE AND ADDRESS OF THE PARTY OF THE P		
Attribute	Test			Test Stat	Critical	P-Value	Decision((a:1%)			
		Equality o	of Variance	Test Stat 6.686	Critical 20.09	P-Value 0.5709	Decision Equal Var	<u> </u>			
Attribute Variances Distribution								iances			
Variances Distribution	Bartlett E	Wilk W N		6.686	20.09	0.5709	Equal Var	iances			
Variances Distribution Developmen	Bartlett E Shapiro-\	Wilk W N	Normality	6.686	20.09 0.9308	0.5709	Equal Var	iances	Std Err	CV%	%Effec
Variances Distribution	Bartlett E Shapiro-\ t Rate Summary	Wilk W N	Normality	6.686 0.9785	20.09 0.9308	0.5709 0.5599	Equal Var Normal Di	iances stribution	Std Err 0.008602	CV% 1.99%	%Effec
Variances Distribution Development C-ppt 33.5	Bartlett E Shapiro-¹ t Rate Summary Control Type	Wilk W N	Normality t Mean	6.686 0.9785 95% LCL	20.09 0.9308 95% UCL 0.9919 0.9766	0.5709 0.5599 Median	Equal Var Normal Di	iances stribution Max			
Variances Distribution Developmen C-ppt 33.5 34.9	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count	Mean 0.968	6.686 0.9785 95% LCL 0.9441	20.09 0.9308 95% UCL 0.9919	0.5709 0.5599 Median 0.96	Equal Var Normal Di Min 0.95	iances istribution Max 1	0.008602	1.99%	0.0%
Variances Distribution Developmen C-ppt 33.5 34.9 35.4	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count 5 5	Mean 0.968 0.954	6.686 0.9785 95% LCL 0.9441 0.9314	20.09 0.9308 95% UCL 0.9919 0.9766	0.5709 0.5599 Median 0.96 0.95	Equal Var Normal Di Min 0.95 0.93	Max 1 0.98	0.008602 0.008124	1.99% 1.9%	0.0% 1.45% 1.45% 1.86%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count 5 5 5	Mean 0.968 0.954 0.954	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812	0.5709 0.5599 Median 0.96 0.95 0.96	Min 0.95 0.93 0.92	Max 1 0.98 0.98	0.008602 0.008124 0.009798	1.99% 1.9% 2.3%	0.0% 1.45% 1.45%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count 5 5 5 5	Mean 0.968 0.954 0.95	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763	0.5709 0.5599 Median 0.96 0.95 0.96 0.95	Equal Var Normal Di Min 0.95 0.93 0.92 0.92	Max 1 0.98 0.98 0.97	0.008602 0.008124 0.009798 0.009487	1.99% 1.9% 2.3% 2.23%	0.0% 1.45% 1.45% 1.86%
Variances Distribution Developmen C-ppt	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count 5 5 5 5	Mean 0.968 0.954 0.954 0.95 0.946	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.95	Equal Var Normal Di Min 0.95 0.93 0.92 0.92 0.88	Max 1 0.98 0.98 0.97 0.98	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4 36.9	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count 5 5 5 5 5 5	0.968 0.954 0.95 0.95 0.946 0.922	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.95 0.95	Min 0.95 0.93 0.92 0.92 0.88 0.85	Max 1 0.98 0.98 0.97 0.98 0.98	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396	1.99% 1.9% 2.3% 2.23% 4.14% 5.81%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7%
Variances Distribution Developmen C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816	95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.95 0.91 0.79	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77	Max 1 0.98 0.98 0.97 0.98 0.98 0.98	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75%
Variances Distribution Developmen C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5	Bartlett E Shapiro-¹ t Rate Summary Control Type	Count 5 5 5 5 5 5 5 5 5 5	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26	95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.95 0.91 0.79 0.64	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47	Max 1 0.98 0.98 0.97 0.98 0.98 0.98 0.89 0.74	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09%
Variances Distribution Developmen C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5	Bartlett E Shapiro-\ t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 5 5 5	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 mmary Mean	95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.95 0.91 0.79 0.64	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47	Max 1 0.98 0.98 0.97 0.98 0.98 0.98 0.89 0.74	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Angular (Cor	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 5 6 7 7 7 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.95 0.91 0.79 0.64 0.22	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19	Max 1 0.98 0.98 0.97 0.98 0.97 0.98 0.74 0.44	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Angular (Cor	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 med Su Count	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 mmary Mean	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.95 0.91 0.79 0.64 0.22	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19	iances stribution Max 1 0.98 0.98 0.97 0.98 0.98 0.98 0.44 0.44	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Angular (Cor C-ppt 33.5	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 med Su Count 5	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 mmary Mean 1.4	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 95% LCL 1.314	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 95% UCL 1.487	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.91 0.79 0.64 0.22 Median 1.369	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19 Min 1.345	iances istribution Max 1 0.98 0.98 0.97 0.98 0.98 0.74 0.44 Max 1.521	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572 Std Err 0.03119	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32% CV% 4.98%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14%
Variances Distribution Developmen C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Angular (Cor C-ppt 33.5 34.9 35.4	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 6 7 7 7 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 mmary Mean 1.4 1.358	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 95% LCL 1.314 1.301	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 95% UCL 1.487 1.416	0.5709 0.5599 Median 0.96 0.95 0.95 0.95 0.91 0.79 0.64 0.22 Median 1.369 1.345	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19 Min 1.345 1.303	iances istribution Max 1 0.98 0.98 0.97 0.98 0.89 0.74 0.44 Max 1.521 1.429	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572 Std Err 0.03119 0.02062	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32% CV% 4.98% 3.39%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14% %Effect 0.0% 3.0%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Angular (Cor C-ppt 33.5 34.9 35.4 35.9	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 6 med Su Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 mmary Mean 1.4 1.358 1.359	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 95% LCL 1.314 1.301 1.295	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 95% UCL 1.487 1.416 1.424	0.5709 0.5599 Median 0.96 0.95 0.95 0.95 0.91 0.79 0.64 0.22 Median 1.369 1.345 1.369	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19 Min 1.345 1.303 1.284	iances istribution Max 1 0.98 0.98 0.97 0.98 0.89 0.74 0.44 Max 1.521 1.429 1.429	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572 Std Err 0.03119 0.02062 0.02336	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32% CV% 4.98% 3.39% 3.84%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14% %Effect 0.0% 3.0% 2.92%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Angular (Cor C-ppt 33.5 34.9 35.4 35.9 36.4 35.9 36.4	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 6 med Su Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 mmary Mean 1.4 1.358 1.359 1.349	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 95% LCL 1.314 1.301 1.295 1.289	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 95% UCL 1.487 1.416 1.424 1.41	0.5709 0.5599 Median 0.96 0.95 0.95 0.95 0.91 0.79 0.64 0.22 Median 1.369 1.345 1.369 1.345	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19 Min 1.345 1.303 1.284 1.284	max 1 0.98 0.98 0.97 0.98 0.98 0.74 0.44 Max 1.521 1.429 1.429 1.397	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572 Std Err 0.03119 0.02062 0.02336 0.02173	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32% CV% 4.98% 3.39% 3.84% 3.6%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14% %Effect 0.0% 3.0% 2.92% 3.65%
Variances Distribution Development C-ppt 33.5 34.9 35.4 35.9 36.4 37.4 37.9 38.5 Angular (Cor C-ppt 33.5 34.9 35.4 35.9 36.4 36.9 36.4 36.9	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.968 0.954 0.954 0.95 0.946 0.922 0.816 0.638 0.26 mmary Mean 1.4 1.358 1.359 1.349 1.347	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 95% LCL 1.314 1.301 1.295 1.289 1.246	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9946 0.9885 0.8803 0.766 0.3869 95% UCL 1.487 1.416 1.424 1.41	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.91 0.79 0.64 0.22 Median 1.369 1.345 1.345 1.345	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19 Min 1.345 1.303 1.284 1.217	Max 1 0.98 0.98 0.98 0.98 0.98 0.98 0.44 0.44 Max 1.521 1.429 1.397 1.429	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572 Std Err 0.03119 0.02062 0.02336 0.02173 0.0361	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32% CV% 4.98% 3.39% 3.84% 3.6% 6.0%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14% %Effec 0.0% 3.0% 2.92% 3.65% 3.83% 6.97%
Variances Distribution Development 33.5 34.9 35.4 35.9 36.4 36.9 37.4 37.9 38.5 Angular (Cor C-ppt 33.5 34.9	Bartlett E Shapiro-\t t Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.968 0.954 0.954 0.955 0.946 0.922 0.816 0.638 0.26 mmary 1.4 1.358 1.359 1.349 1.347 1.303	6.686 0.9785 95% LCL 0.9441 0.9314 0.9268 0.9237 0.8974 0.8555 0.7517 0.51 0.1331 95% LCL 1.314 1.301 1.295 1.289 1.246 1.17	20.09 0.9308 95% UCL 0.9919 0.9766 0.9812 0.9763 0.9885 0.8803 0.766 0.3869 95% UCL 1.487 1.416 1.424 1.41 1.447	0.5709 0.5599 Median 0.96 0.95 0.96 0.95 0.91 0.79 0.64 0.22 Median 1.369 1.345 1.345 1.366	Min 0.95 0.93 0.92 0.92 0.88 0.85 0.77 0.47 0.19 Min 1.345 1.303 1.284 1.284 1.217 1.173	Max 1 0.98 0.98 0.98 0.98 0.98 0.98 0.44 Max 1.521 1.429 1.429 1.429 1.429 1.429	0.008602 0.008124 0.009798 0.009487 0.01749 0.02396 0.02315 0.04609 0.04572 Std Err 0.03119 0.02062 0.02336 0.02173 0.0361 0.04784	1.99% 1.9% 2.3% 2.23% 4.14% 5.81% 6.34% 16.15% 39.32% CV% 4.98% 3.39% 3.84% 3.6% 6.0% 8.21%	0.0% 1.45% 1.45% 1.86% 2.27% 4.75% 15.7% 34.09% 73.14% %Effect 0.0% 3.0% 2.92% 3.65% 3.83%

0.89

0.47

0.21

0.77

0.64

0.44

0.79

0.64

0.24

37.4

37.9

38.5

Report Date: Test Code: 04 Aug-15 14:41 (p 2 of 2) 1507-S081 | 09-7618-8627

Echinoid Embryo-Larval Development Test Nautilus Environmental (CA) Analysis ID: 16-2651-6908 CETISv1.8.7 Endpoint: Development Rate **CETIS Version:** Analyzed: 04 Aug-15 14:39 Parametric-Control vs Treatments Analysis: Official Results: Yes **Development Rate Detail** C-ppt **Control Type** Rep 1 Rep 2 Rep 3 Rep 4 Rep 5 33.5 Lab Control 0.96 0.95 0.97 0.96 34.9 0.95 0.95 0.98 0.93 0.96 35.4 0.96 0.98 0.92 0.96 0.95 35.9 0.92 0.97 0.95 0.97 0.94 36.4 0.98 0.95 0.95 0.88 0.97 36.9 0.97 0.98 0.85 0.91 0.9

0.85

0.74

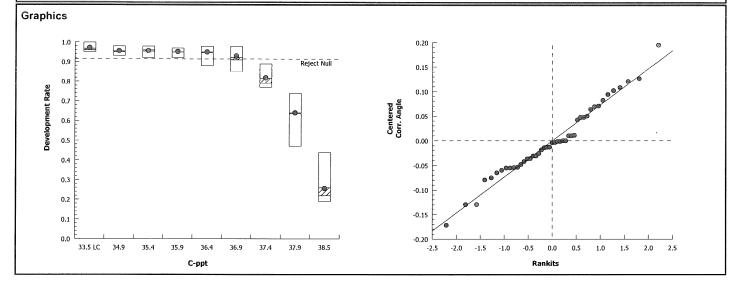
0.19

0.78

0.7

0.22

Angular (C	ngular (Corrected) Transformed Detail									
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5				
33.5	Lab Control	1.369	1.345	1.521	1.397	1.369				
34.9		1.345	1.345	1.429	1.303	1.369				
35.4		1.369	1.429	1.284	1.369	1.345				
35.9		1.284	1.397	1.345	1.397	1.323				
36.4		1.429	1.345	1.345	1.217	1.397				
36.9		1.397	1.429	1.173	1.266	1.249				
37.4		1.233	1.071	1.095	1.173	1.083				
37.9		0.7554	0.9273	0.9273	1.036	0.9912				
38.5		0.476	0.7253	0.512	0.451	0.4882				



Report Date:

04 Aug-15 14:41 (p 1 of 2)

Test Code: 1507-S081 | 09-7618-8627

								Test	Code:	150	17-S081 0	9-7618-86
Echino	id Emb	ryo-Larval Deve	elopment T	est						Nautilu	s Environi	mental (CA
Analysi	s ID:	18-0652-3476	End	point: De	evelopment F	Rate		CETI	S Version:	CETISv1	.8.7	
			•	•				ial Results:				
Linear	Ragrasi	sion Options	***************************************									
											_	
					ld Option		Optimized		Het Corr	Weighted	1	· · · · · · · · · · · · · · · · · · ·
Log-Nor	rmal [NE	ED=A+B*log(X)]		Control T	hreshold	0.032	Yes	No	Yes	Yes		
Regres	sion Su	ımmary										
Iters	LL	AlCc	BIC	Mu	Sigma	Adj R2	F Stat	Critical	P-Value	Decision	(a:5%)	
8	-1454	2915	2920	1.581	0.007532	0.9372	0.392	2.364	0.8793	Non-Sign	ficant Lack	k of Fit
Point E	stimate	S										
Level	ppt	95% LCL	95% UCL									
EC25	37.69	37.58	37.77				· · · · · · · · · · · · · · · · · · ·					
EC50	38.13	38.05	38.21									
Regres	sion Pa	rameters									***************************************	
Parame	ter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision(a:5%)			
Thresho	old	0.04539	0.006405	0.03247	0.05832	7.087	<0.0001	Significant	Parameter	***************************************		
Slope		132.8	10.96	110.6	154.9	12.11	<0.0001	-	Parameter			
Intercep	it	-209.9	17.32	-244.9	-175	-12.12	<0.0001	Significant	Parameter			
ANOVA	Table											
Source		Sum Squa	ires Mea	n Square	DF	F Stat	P-Value	Decision(a:5%)			
Model		1521.97	1521	1.97	1	658.3	<0.0001	Significant				
Lack of	Fit	5.955364	0.99	2561	6	0.392	0.8793	Non-Signif	icant			
Pure Err	ror	91.1523	2.53	2008	36							
Residua	ı	97.10767	2.31	2087	42						anno misso de la companya de la comp	
Residua	al Analy	sis										
Attribut	е	Method			Test Stat	Critical	P-Value	Decision(a:5%)			
Goodne	ss-of-Fi	Pearson C	hi-Sq GOF		97.11	58.12	<0.0001	Significant	Heterogeni	ty		
		Likelihood	Ratio GOF		99.08	58.12	<0.0001	Significant	Heterogeni	ty		
Variance	es	Bartlett Eq	uality of Var	iance	8.286	15.51	0.4060	Equal Vari	ances			
		Mod Lever	ne Equality o	of Variance	0.5334	2.305	0.8209	Equal Vari	ances			
Distribut	ion	Shapiro-W	ilk W Norma	ality	0.9912	0.9498	0.9790	Normal Dis	stribution			
		Anderson-l	Darling A2 N	Normality	0.1858	2.492	0.9613	Normal Dis	stribution		·	
Develop	ment F	late Summary				Calcul	ated Variate	e(A/B)				
C-ppt	Co	ontrol Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
33.5	La	b Control	5	0.968	0.95	1	0.008602	0.01924	1.99%	0.0%	484	500
34.9			5	0.954	0.93	0.98	0.008124	0.01817	1.9%	1.45%	477	500
35.4			5	0.954	0.92	0.98	0.009798	0.02191	2.3%	1.45%	477	500
35.9			5	0.95	0.92	0.97	0.009487	0.02121	2.23%	1.86%	475	500
36.4			5	0.946	0.88	0.98	0.01749	0.03912	4.14%	2.27%	473	500
36.9			5	0.922	0.85	0.98	0.02396	0.05357	5.81%	4.75%	461	500
37.4			5	0.816	0.77	0.89	0.02315	0.05177	6.34%	15.7%	408	500
37.9			5	0.638	0.47	0.74	0.04609	0.1031	16.15%	34.09%	319	500
28.5			E	0.26	Λ 10	0.44	0.04572	n 1022	20.220/	72 1/10/	120	500

38.5

5

0.26

0.19

0.44

0.04572

0.1022

39.32%

73.14%

130

500

Report Date:

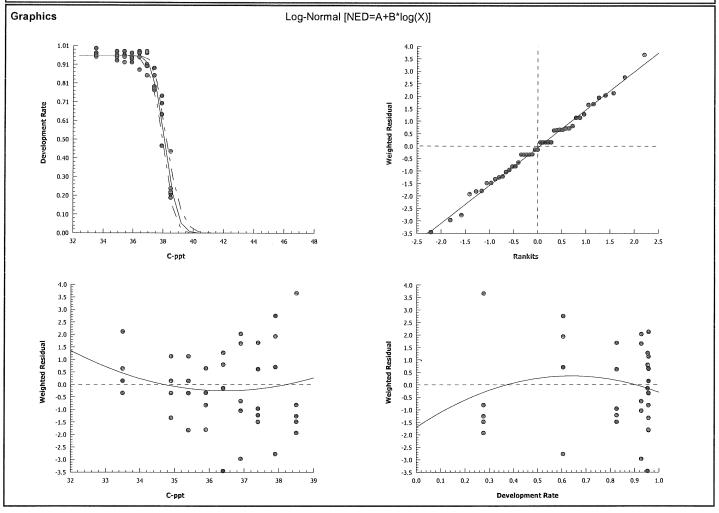
04 Aug-15 14:41 (p 2 of 2) 1507-S081 | 09-7618-8627

Test Code:

Echinoid Embryo-Larval Development Test	Nautilus Environmental (CA)

Analysis ID:	18-0652-3476	Endpoint:	Development Rate	CETIS Version:	CETISv1.8.7
Analyzed:	04 Aug-15 14:41	Analysis:	Linear Regression (MLF)	Official Results:	Ves

Develop	Development Rate Detail									
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5				
33.5	Lab Control	0.96	0.95	1	0.97	0.96				
34.9		0.95	0.95	0.98	0.93	0.96				
35.4		0.96	0.98	0.92	0.96	0.95				
35.9		0.92	0.97	0.95	0.97	0.94				
36.4		0.98	0.95	0.95	0.88	0.97				
36.9		0.97	0.98	0.85	0.91	0.9				
37.4		0.89	0.77	0.79	0.85	0.78				
37.9		0.47	0.64	0.64	0.74	0.7				
38.5		0.21	0.44	0.24	0.19	0.22				



Report Date: **Test Code:**

04 Aug-15 14:41 (p 1 of 1)

1507-S081 | 09-7618-8627

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

01-1762-9710 Analysis ID:

Linear Interpolation Options

Endpoint: Development Rate

CETIS Version:

CETISv1.8.7

Analyzed: 04 Aug-15 14:41 Analysis:

Linear Interpolation (ICPIN)

Official Results: Yes

X Transform	Y Transf

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	1146216	1000	Yes	Two-Point Interpolation

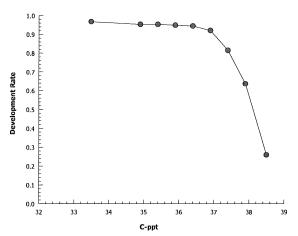
Point Estimates

Level	ppt	95% LCL	95% UCI
EC25	37.65	37.49	37.88
EC50	38.14	37.98	38.28

Developn	nent Rate Summary	Calculated Variate(A/B)									
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
33.5	Lab Control	5	0.968	0.95	1	0.008602	0.01924	1.99%	0.0%	484	500
34.9		5	0.954	0.93	0.98	0.008124	0.01817	1.9%	1.45%	477	500
35.4		5	0.954	0.92	0.98	0.009798	0.02191	2.3%	1.45%	477	500
35.9		5	0.95	0.92	0.97	0.009487	0.02121	2.23%	1.86%	475	500
36.4		5	0.946	0.88	0.98	0.01749	0.03912	4.14%	2.27%	473	500
36.9		5	0.922	0.85	0.98	0.02396	0.05357	5.81%	4.75%	461	500
37.4		5	0.816	0.77	0.89	0.02315	0.05177	6.34%	15.7%	408	500
37.9		5	0.638	0.47	0.74	0.04609	0.1031	16.15%	34.09%	319	500
38.5		5	0.26	0.19	0.44	0.04572	0.1022	39.32%	73.14%	130	500

Developn	Development Rate Detail									
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5				
33.5	Lab Control	0.96	0.95	1	0.97	0.96				
34.9		0.95	0.95	0.98	0.93	0.96				
35.4		0.96	0.98	0.92	0.96	0.95				
35.9		0.92	0.97	0.95	0.97	0.94				
36.4		0.98	0.95	0.95	0.88	0.97				
36.9		0.97	0.98	0.85	0.91	0.9				
37.4		0.89	0.77	0.79	0.85	0.78				
37.9		0.47	0.64	0.64	0.74	0.7				
38.5		0.21	0.44	0.24	0.19	0.22				

Graphics



Report Date:

21 Jul-15 17:17 (p 1 of 2)

Echinoid Embryo-Larval Development Test Nautilus Environmental (CA) Start Date: 22 Jul-15 Species: Strongylocentrotus purpuratus Sample Code: 1507-S∂� End Date: 25 Jul-15

Protocol: EPA/600/R-95/136 (1995)

Sample Source: Poseidon

d Date: mple Date		Jul-15 Jul-15			ol: EPA/60 al: Brined	20/R-95/136 (1995) Sample Source: Poseidon seawater Sample Station:	
C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes	
			1	100	95	organism present in vial (copeped)	7/30/15
			2	100	47	Organism Stesovii (11 Vivil Ooke 1000)	1190113
			3	100			
			4	100	97		
			5	100	22		
***************************************			6	100	85		
			7	100	74	CONTROL CONTRO	
			8	100	93		
			9	100	95		
			10	100	85		
			11	100	98		
			12	100	150		
			13	100	97		
			14	100	100		
			15	100	99		
			16	100	94		
			17	100	95		
			18	100	92		
			19	100	79		
			20	100	95		
			21	100	95		
			22	100	96		
			23	100	90		
			24	100	86		
			25	100	21		
			26	100	90		
			27		97		
			28	100	64	0	14/16
		-	29	100		Z)	14/10
			30	100	70		
			31	100	19		
			32	(00	95		
			33	100	91		
			34		44		
			35	100	92		
			36	(00)	90		
			37	100	92		
	-		38		00		
			39	100	98		
			40	100			
			41		97		
			42	100	96		
	-	-	43	100	95		
-				001	77		
			44	000	44		
			45	100	98		
			46	100	78		
			47	lõÕ	98		V

Report Date: 21 Jul-15 17:17 (p 2 of 2)

Test Code: 1507-508| 09-7618-8627/3A2F74D3

C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
			48	100	24	814/16
			49	100	ace	
			50	100	89	

Report Date:

21 Jul-15 17:17 (p 1 of 2)

Test Code: /507-508/ 09-7618-8627/3A2F74D3

Nautilus Environmental (CA)

Echinoid Embryo-Larval Development	Test	
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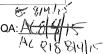
22 Jul-15 25 Jul-15

Start Date:

Species: Strongylocentrotus purpuratus Protocol: EPA/600/R-95/136 (1995)

Sample Code: 1507-S0%

Start Date: End Date: Sample Date	25 .	Jul-15 Jul-15 Jul-15		Protoc		gylocentrotus purpuratus 900/R-95/136 (1995) I seawater	Sample Code: Sample Source: Sample Station:	
C-ppt	Code	Rep	Pos	# Counted	# Normal		Notes	
0	ВС	1	15	100	99	A 7/28/15		
0	ВС	2	4					
0	ВС	3	39					
0	BC	4	14				1000	
0	ВС	5	18				The state of the s	
0	LC	1	49	100	100	AC 7/28/15		
0	LC	2	21			1		
0	LC	3	12					
0	LC	4	3	A COLOR			· · · · · · · · · · · · · · · · · · ·	
0	LC	5	22					
35		1	42					
35		2	1			100.14		
35		3	47					
35		4	8					
35		5	26					
35.5		1	41	-				
35.5		2	38					
35.5		3	37					
35.5	Ì	4	36					
35.5		5	17					
36		1	35					
36		2	30					
36		3	9					
36		4	27					
36		5	16					
36.5		1	45					
36.5		2	20					
36.5		3	32					
36.5		4	24					
36.5		5	13					
37		1	40					
37		2	11					
37		3	10					
37		4	33				****	
37		5	23				A 140 10 AAAAA	
37.5		1	50					
37.5		2	43					
37.5		3	19					
37.5		4	6					
37.5		5	46					
38		1	2					
38		2	28				****	
38		3	44					
38		4	7					
38		5	29					
38.5		1	25					
38.5		2	34					



Report Date: 21 Jul-15 17:17 (p 2 of 2)

C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
38.5		3	48			
38.5		4	31			
38.5		5	5			

Que

Water Quality Measurements

Client: Poseidon

Test Species: S. purpuratus

Sample ID: Nautilus brine (frozen seawater)

Start Date/Time: 7/21/2015 7/22/15

Sample Log No.: →

End Date/Time: 7/24/2015 7 75/15 1010

Test No.: 1507-5081

Concentration (ppt)			inity pt)		Temperature (°C)				Dissolved Oxygen (mg/L)				pH (pH units)			
	0	24	48	72	0	24	48	72	0	24	48	72	0	24	48	72
Lab Control	33.5	33.4	33.5	33.3	14.0	15.5	15.2	15.4	8.4	7.9	7.9	8:0	809	8.65	8.08	1
Brine Control	33.4	33.3	33.4	33.3	160	15.2	15.0	15.2	8.4	8.0	7.8	8.2	810	8,07	8.08	806
35.0	34.9	34.8	34.9	34.8	160	15.2	14.9	153	8.6	8.0	8.0	8.1	8.09	8.07	8.08	8.06
35.5	35.4	35.4	35.4	35.3	16.0	15.2	14.8	52	8.7	8-1	8.0	8.2	8.09	30.8	8.06	801
36.0	35.9	35.9	35.9	35.9	15.8	15.2	14.8	15.4	8.8	8.1	8.1	8.2	8.09	8.08	8.07	8.67
36.5	36.4	36.4	36,4	36.3	15.9	15.1	14.8	15.4	8.8	8.1	8.0	8.2	8.09	8.09	8.07	807
37.0	36.9	36.9	36.9	36.8	15.9	15.1	14.7	154	8.8	8.0	8.0	8.2	8.09	8.09	8.08	8.07
37.5	37.4	37.4	37.4	37.3	15.8	15.1	14.8	15.4	8.8	8.1	8.0	8.2	8.09	8.09	8.08	8.07
38.0	37.9	37.9	37.9	37,8	15.6	15.2	14.7	15.6	8.8	8.0	8,0	8.2	809	8.09	8.09	8.07
38.5	38.5	38,5	38.4	38:3	15.6	15.1	14.7	15·Le	8.8	8.1	8.0	8-2	8.09	8.09	8.09	8.07
															J	

		0	24	48	72
Technician Initials:	WQ Readings:	AO	EG	CH	Q4
	Dilutions made by:	AC			

Comments:	0 hrs:	@ AC 0187/21/15	Hach Sension 5 Salinity meter
	24 hrs:	(
	48 hrs:		
	72 hrs:		
QC Che	ck :Дс{	74/15	Final Review: 6/4/15

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

Echinoderm Larval Development Worksheet

Client:	Poseidon	Start Date/Time: 12415/ 1610
Sample ID:	Naufly oxine (Frozen Seawater)	End Date/Time: 1251157 610
Test No.:	1507-308)	Species: S. purpuratus
Tech initials: Injection Time:	PA/A/ 1525	Date Collected: 1/9/15
Sperm Absorbance at 4	400 nm: 4525 At 91,527 (target range of 0.8 - 1.0 for density of 4x	10 ⁶ sperm/ml)
Eggs Counted:	Mean: 3 X 50 = 560 ((target counts of 20 eggs per vertical pass on Sedgy slide for a final density of 1000 eggs/ml)	eggs/ml vick-Rafter
Initial density: Final density:		egg stock 50 ml
	according to the calculated dilution factor. For example, if the dilution filution water (1.25 parts).	factor is 2.25, use 100 ml of existing stock
Add 100 μL sperm stoc	k per 100mL of egg stock. For example, if you have 60mL of egg stoc	ck, add 60μL sperm stock.
Embryo Stock Fertilizati	on Checks (Initiate test only when fertilization is ≥90%):	
10 minutes (1st fert.) 20 minutes (2nd fert. If r	No. No. Time Fert. Unfert. %	
Fertilization Time:	15 55	
Test Initiation Time:	Embryo Stock Added: 0.25 ml	
Test Termination: 72-hour QC check ^a	No. No. % Normal Abnormal Normal $O(O)$ O	
End of test QC check	gener gan anger.	
Comments:	^a If the embryo development does not meet the mean test acceptable developed, continue the test to 96-hrs (ASTM 1999).	lity criterion of 80% normally
QC Check:	AC8 4 15	Final Review: 5/4/13

Sand Dollar 72-hour Larval Development

Test Date: October 30, 2014

CETIS Summary Report

Report Date: Te

30 Jun-15 09:48 (p 1 of 1) 40

toport bate.	00 0011 10 00.10 (p 1 0)
est Code:	1410-S151 00-7545-094

					***************************************				171	0 0 10 1 10	0-7040-004
Echinoid Em	bryo-Larval Dev	elopmen	nt Test			1800.000			Nautilus	s Environr	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	20-9369-4926 30 Oct-14 16:2 02 Nov-14 17:2 73h	20 F	Test Type: Protocol: Species: Source:	Development EPA/600/R-95/ Dendraster exc Mission Bay	` '				tural Seawat ozen Seawate		
Sample ID:	15-8340-2497	C	Code:	Brine		4		Client: Po	seidon		
Sample Date:		N	Material:	Brined seawate	er			Project:			
Receive Date			Source:	Poseidon	•						
Sample Age:	16h	S	Station:	Noutilus E	Brine						
Sample Note:	: Frozen seawat	er prepar	ed at Nauti	lus was used as	brine.						
Comparison	Summary										
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method			
11-3618-3366	Development F	Rate	38.5	>38.5	NA	3.76%		Dunnett	Multiple Com	parison Te	st
Point Estimat	te Summary										
Analysis ID	Endpoint		Level	ppt	95% LCL	95% UCL	TU	Method			
04-0439-9908	Development F	Rate	EC25	>38.5	N/A	N/A		Linear In	terpolation (I	CPIN)	
		- Charles Company	EC50	>38.5	N/A	N/A					
Test Acceptal	bility										
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Overlap	Decision		
04-0439-9908	Development F	Rate	Contro	ol Resp	0.972	0.8 - NL		Yes	Passes A	cceptability	Criteria
11-3618-3366	Development F	Rate	Contro	ol Resp	0.972	0.8 - NL		Yes	Passes A	cceptability	Criteria
11-3618-3366	Development F	Rate	PMSD		0.03765	NL - 0.25		No	Passes A	cceptability	Criteria
Development	Rate Summary						Section Control Communication				
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Brine Control	5	0.968	0.9347	1	0.92	0.98	0.012	0.02683	2.77%	0.0%
0	Lab Control	5	0.972	0.9616	0.9824	0.96	0.98	0.003742	0.008367	0.86%	-0.41%
35		5	0.984	0.9632	1	0.96	1	0.007483	0.01673	1.7%	-1.65%
35.5		5	0.984	0.9698	0.9982	0.97	1	0.005099	0.0114	1.16%	-1.65%
36		5	0.988	0.9676	1	0.96	1	0.007348	0.01643	1.66%	-2.07%
36.5		5	0.968	0.9518	0.9842	0.95	0.98	0.005831	0.01304	1.35%	0.0%
37		5	0.978	0.9618	0.9942	0.97	1	0.005831		1.33%	-1.03%
37.5		5	0.98	0.9648	0.9952	0.97	1	0.005477	0.01225	1.25%	-1.24%
38		5	0.974	0.9372	1	0.93	1	0.01327	0.02966	3.05%	-0.62%
38.5		5	0.97	0.9468	0.9932	0.94	0.99	0.008367	0.01871	1.93%	-0.21%
Development		D. 1	. -	D 2	D 1	D -					
C-ppt	Control Type	Rep 1	Rep 2		Rep 4	Rep 5					*************
0	Brine Control	0.98	0.98	0.98	0.92	0.98					
						0.96					
	Lab Control	0.97	0.98	0.97	0.98						
35	Lab Control	0.98	. 1	1	0.98	0.96					
35 35.5	Lab Control	0.98 0.98	. 1	1 0.97	0.98 0.98	0.96 0.99					
35 35.5 36	Lab Control	0.98 0.98 0.96	. 1 1 1	1 0.97 0.99	0.98 0.98 1	0.96 0.99 0.99					
35 35.5 36 36.5	Lab Control	0.98 0.98	. 1	1 0.97	0.98 0.98	0.96 0.99					
35 35.5 36 36.5	Lab Control	0.98 0.98 0.96	. 1 1 1	1 0.97 0.99	0.98 0.98 1	0.96 0.99 0.99					
35 35.5 36 36.5 37	Lab Control	0.98 0.98 0.96 0.96	. 1 1 1 0.97	1 0.97 0.99 0.98	0.98 0.98 1 0.95	0.96 0.99 0.99 0.98					
0 35 35.5 36 36.5 37 37.5	Lab Control	0.98 0.98 0.96 0.96 0.98	. 1 1 1 0.97 0.97	1 0.97 0.99 0.98 0.97	0.98 0.98 1 0.95 0.97	0.96 0.99 0.99 0.98					

Report Date: Test Code: 30 Jun-15 09:48 (p 1 of 2)

e: 1410-S151 | 00-7545-0940

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Analysis ID:11-3618-3366Endpoint:Development RateCETIS Version:CETISv1.8.7Analyzed:30 Jun-15 9:47Analysis:Parametric-Control vs TreatmentsOfficial Results:Yes

Sample Note: Frozen seawater prepared at Nautilus was used as brine.

Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corrected)	NA	C > T	NA	NA	3.76%	38.5	>38.5	NA	

Dunnett Multi	ple C	omparison Test							
Control	vs	C-ppt	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(α:5%)
Lab Control		35	-1.362	2.478	0.090	8	0.9978	CDF	Non-Significant Effect
		35.5	-1.236	2.478	0.090	8	0.9966	CDF	Non-Significant Effect
		36	-1.82	2.478	0.090	8	0.9996	CDF	Non-Significant Effect
		36.5	0.2822	2.478	0.090	8	0.8093	CDF	Non-Significant Effect
		37	-0.6538	2.478	0.090	8	0.9781	CDF	Non-Significant Effect
		37.5	-0.8304	2.478	0.090	8	0.9870	CDF	Non-Significant Effect
		38	-0.6709	2.478	0.090	8	0.9791	CDF	Non-Significant Effect
		38.5	0.02403	2.478	0.090	8	0.8832	CDF	Non-Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.02641647	0.003302059	8	0.9943	0.4570	Non-Significant Effect
Error	0.1195557	0.003320992	36			
Total	0.1459722		44	Administration		

Distributional Tes	ts				
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)
Variances	Bartlett Equality of Variance	7.437	20.09	0.4903	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9715	0.9308	0.3283	Normal Distribution

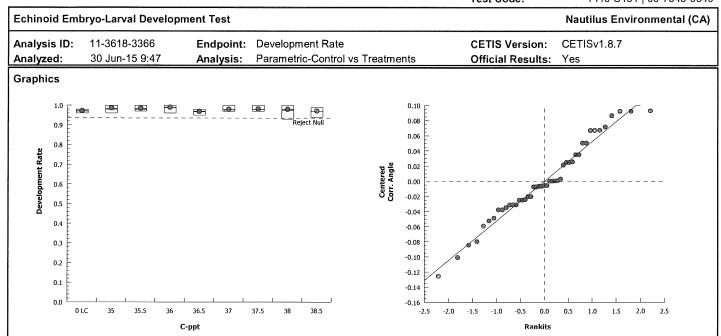
Developm	ent Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.972	0.9616	0.9824	0.97	0.96	0.98	0.003742	0.86%	0.0%
35		5	0.984	0.9632	1	0.98	0.96	1	0.007483	1.7%	-1.24%
35.5		5	0.984	0.9698	0.9982	0.98	0.97	1	0.005099	1.16%	-1.24%
36		5	0.988	0.9676	1	0.99	0.96	1	0.007349	1.66%	-1.65%
36.5		5	0.968	0.9518	0.9842	0.97	0.95	0.98	0.005831	1.35%	0.41%
37		5	0.978	0.9618	0.9942	0.97	0.97	1	0.005831	1.33%	-0.62%
37.5		5	0.98	0.9648	0.9952	0.98	0.97	1	0.005477	1.25%	-0.82%
38		5	0.974	0.9372	1	0.98	0.93	1	0.01327	3.05%	-0.21%
38.5		5	0.97	0.9468	0.9932	0.97	0.94	0.99	0.008367	1.93%	0.21%

Control Type	Count									
	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
ab Control	5	1.404	1.373	1.435	1.397	1.369	1.429	0.01127	1.8%	0.0%
	5	1.454	1.372	1.535	1.429	1.369	1.521	0.02944	4.53%	-3.53%
	5	1.449	1.39	1.509	1.429	1.397	1.521	0.0214	3.3%	-3.21%
	5	1.47	1.394	1.547	1.471	1.369	1.521	0.02763	4.2%	-4.72%
	5	1.394	1.348	1.44	1.397	1.345	1.429	0.01646	2.64%	0.73%
	5	1.428	1.361	1.495	1.397	1.397	1.521	0.02403	3.76%	-1.7%
	5	1.434	1.371	1.498	1.429	1.397	1.521	0.02276	3.55%	-2.16%
	5	1.429	1.31	1.547	1.429	1.303	1.521	0.04258	6.67%	-1.74%
	5	1.403	1.336	1.47	1.397	1.323	1.471	0.02416	3.85%	0.06%
	ab Control	5 5 5 5 5 5 5	5 1.454 5 1.449 5 1.47 5 1.394 5 1.428 5 1.434 5 1.429	5 1.454 1.372 5 1.449 1.39 5 1.47 1.394 5 1.394 1.348 5 1.428 1.361 5 1.434 1.371 5 1.429 1.31	5 1.454 1.372 1.535 5 1.449 1.39 1.509 5 1.47 1.394 1.547 5 1.394 1.348 1.44 5 1.428 1.361 1.495 5 1.434 1.371 1.498 5 1.429 1.31 1.547	5 1.454 1.372 1.535 1.429 5 1.449 1.39 1.509 1.429 5 1.47 1.394 1.547 1.471 5 1.394 1.348 1.44 1.397 5 1.428 1.361 1.495 1.397 5 1.434 1.371 1.498 1.429 5 1.429 1.31 1.547 1.429	5 1.454 1.372 1.535 1.429 1.369 5 1.449 1.39 1.509 1.429 1.397 5 1.47 1.394 1.547 1.471 1.369 5 1.394 1.348 1.44 1.397 1.345 5 1.428 1.361 1.495 1.397 1.397 5 1.434 1.371 1.498 1.429 1.397 5 1.429 1.31 1.547 1.429 1.303	5 1.454 1.372 1.535 1.429 1.369 1.521 5 1.449 1.39 1.509 1.429 1.397 1.521 5 1.47 1.394 1.547 1.471 1.369 1.521 5 1.394 1.348 1.44 1.397 1.345 1.429 5 1.428 1.361 1.495 1.397 1.397 1.521 5 1.434 1.371 1.498 1.429 1.307 1.521 5 1.429 1.31 1.547 1.429 1.303 1.521	5 1.454 1.372 1.535 1.429 1.369 1.521 0.02944 5 1.449 1.39 1.509 1.429 1.397 1.521 0.0214 5 1.47 1.394 1.547 1.471 1.369 1.521 0.02763 5 1.394 1.348 1.44 1.397 1.345 1.429 0.01646 5 1.428 1.361 1.495 1.397 1.397 1.521 0.02403 5 1.434 1.371 1.498 1.429 1.307 1.521 0.02276 5 1.429 1.31 1.547 1.429 1.303 1.521 0.04258	5 1.454 1.372 1.535 1.429 1.369 1.521 0.02944 4.53% 5 1.449 1.39 1.509 1.429 1.397 1.521 0.0214 3.3% 5 1.47 1.394 1.547 1.471 1.369 1.521 0.02763 4.2% 5 1.394 1.348 1.44 1.397 1.345 1.429 0.01646 2.64% 5 1.428 1.361 1.495 1.397 1.521 0.02403 3.76% 5 1.434 1.371 1.498 1.429 1.303 1.521 0.04258 6.67% 5 1.429 1.31 1.547 1.429 1.303 1.521 0.04258 6.67%

Analyst: A QA: 6/30/19

000-089-170-2 CETIS™ v1.8.7.20

Report Date: Test Code: 30 Jun-15 09:48 (p 2 of 2) 1410-S151 | 00-7545-0940



Report Date:

30 Jun-15 09:48 (p 1 of 1)

Test Code:

1410-S151 | 00-7545-0940

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Analysis ID:

04-0439-9908

Endpoint: Development Rate

CETIS Version:

CETISv1.8.7

Analyzed: 30 Jun-15 9:48

Analysis: Linear Interpolation (ICPIN)

Official Results: Yes

Sample Note: Frozen seawater prepared at Nautilus was used as brine.

Linear	Interpo	lation (Options
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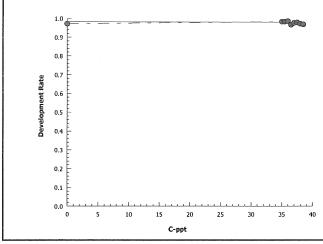
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	2735	1000	Yes	Two-Point Interpolation

Point Estimates

Level	ppt	95% LCL	95% UCL
EC25	>38.5	N/A	N/A
EC50	>38.5	N/A	N/A

Develop	nent Rate Summary	У	Calculated Variate(A/B)								
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.972	0.96	0.98	0.003742	0.008366	0.86%	0.0%	486	500
35		5	0.984	0.96	1	0.007483	0.01673	1.7%	-1.24%	492	500
35.5		5	0.984	0.97	1	0.005099	0.0114	1.16%	-1.24%	492	500
36		5	0.988	0.96	1	0.007349	0.01643	1.66%	-1.65%	494	500
36.5		5	0.968	0.95	0.98	0.005831	0.01304	1.35%	0.41%	484	500
37		5	0.978	0.97	1	0.005831	0.01304	1.33%	-0.62%	489	500
37.5		5	0.98	0.97	1	0.005477	0.01225	1.25%	-0.82%	490	500
38		5	0.974	0.93	1	0.01327	0.02966	3.05%	-0.21%	487	500
38.5		5	0.97	0.94	0.99	0.008367	0.01871	1.93%	0.21%	485	500

Graphics



Report Date:

24 Nov-14 09:36 (p 1 of 2)

Test Code: 1410-5 151 00-7545-0940/1410-S151

Echinoid Er	nbryo-	Larva	l Deve	elopment Te	st	Nautilus Environmental (CA)
Start Date: End Date: Vig 14 Sample Dat	30 03 1 30 0	Oct-14 Nov-14 Oct-14	 1 4	Protoc		aster excentricus Sample Code: Brine 600/R-95/136 (1995) Sample Source: Poseidon I seawater Sample Station:
C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
			1	100	100	
			2	100	100	
			3	100	96	
990 P 99			4	100	98	
			5	100	97	
			6	100	98	
			7	100	99	QC: 25 99/100
			8	100	97	
			9	100	100	
THE RESIDENCE OF THE PARTY OF T			10	100	100	
			11	100	99	
			12	100	98	ac: w 99/100
			13	100	97	
			14	100	97	
			15	100	98	
			16	100	99	
			17	100	98	
			18	100	96	
			19	100	99	
***************************************			20	100	97	
			21	100	97	
			22	100	96	
			23	100	98	
			24	100	100	
			25	100	98	
			26	100	97	
			27	100	96	
			28	100	92	
			29	100	98	
	1		30	100		
			31	100	100 9B	
			32		98	
			33	100	98	
			34	(00)		
			35	100	100	
			36	000		
			37	(00)	98 98	
	+ +		38	100		
			39		97	
	+		40	601	98	
	-		41	100	<i>ab</i>	Q(: 15 92/108 512/5/14 98/100
	-			100	98	Q1: 6 48/108 512/5/14 98/100
	+		42	100	100	
	+		43	100	97	
	-		44	(00)	100	
	++		45	100	94	
			46	100	98	
			47	100	97	

Report Date: Test Code:

24 Nov-14 09:36 (p 2 of 2) 00-7545-0940/1410-S151

C-ppt	Code	Rep	Pos	# Counted	# Normal			Notes	
			48	100	93	QC: 8 48/1	00 8 12/5/19	94/100	
			49	100	95		. 16		
			50	100	96	Q.C. 1 15 89111	D 812/5/1	93/100	

Report Date: Test Code:

28 Oct-14 16:41 (p 1 of 2) 00-7545-0940/1410-S151

Nautilus Environmental (CA)

Start Date: 30 Oct-14 End Date: 30 Nov-14

Echinoid Embryo-Larval Development Test

Species: Dendraster excentricus Protocol: EPA/600/R-95/136 (1995) Sample Code: Brine

Sample Source: Poseidon

nd Date:					al: Brined	20/R-95/136 (1995) Sample Source: Poseidon seawater Sample Station:
C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
0	ВС	1	41			
0	ВС	2	6			
0	ВС	3	32			
0	ВС	4	28			
0	ВС	5	36			
0	LC	1	35			
0	LC	2	40			
0	LC	3	13			
0	LC	4	46			
0	LC	5	27			
35		1	4			
35		2	24		****	
35		3	34			
35		4	31			
35		5	3			
35.5		1	15			
35.5		2	42			
35.5		3	8			
35.5		4	29			
35.5		5	16			
36		1	18			
36		2	44			
36		3	11			
36		4	30			
36		5	19			
36.5		1	50			
36.5		2	5	1000		,
36.5		3	33			
36.5		4	49			
36.5		5	17			
37		1	12			
37		2	21			
37		3	43			
37		4	47			
37		5	1			
37.5		1	10			
37.5	-	2	25			,
37.5		3	37			
37.5		4	38			
37.5		5	20			
38		1	48			
38		2	9			
38		3	2			
38	-	4	39			
38		5	22			
38.5		1	7			

QC=AC

Report Date: Test Code: 28 Oct-14 16:41 (p 2 of 2) 00-7545-0940/1410-S151

C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
38.5		3	26			
38.5		4	23			
38.5		5	45			

QCEAL

QA:KAP12/4/14

Marine Chronic Bioassay

Water Quality Measurements

Client/Project: Poseidon/Salinity Tolerance Study

Sample ID: Brine (frozen seawater)

Test No.: 1410-S151

Test Species: D. excentricus

Start Date/Time: 10/30/2014

End Date/Time: 11/8/2014 17: 20

1112/14

Concentration (ppt)			inity pt)			Temp	erature C) as allet	D	issolve (m	d Oxyg g/L) 👑	en //-			oH units)	
	0	24	48	72	0	24	48 72	0	24	48	72	0	24	48	72
Lab Control	33.3	33.3	33.4	33.6	156	15.7	Tr. 355	8.7	7.9	995	7-8	8.04	7.98	8.07	793
Brine Control	33.7	33.7	33.8	34-3	15.4	15.7	157/5/1811	8.8	7.8	7.8	7.8	8.06	8.00	8.11	794
35	349	34.9	35.0	35.0	14.8	15.8	J5.65.0 18.05	8.8	7.7	7.7	7.9	8.07	799	8.11	7-96
35.5	35,5	35.5	35.5	35.5	15.0	15.7	98950 149	8.8	8.0	7.7	7.8	8.07	8.01	8.09	7.98
36	36.0	36.0	36.1	36.1	14.7	15.7	\$ 7148 15.0	8.7	8.1	78	7.9	8.13	815	8.11	7.98
36.5	36.5	365	36.5	36.5	14.7	15.8	88 H.7 15.1	8.8	7.8	7.8	7.7	8.10	8.05	8.10	7.98
37	37.0	37,0	31.1	37.1	14.8	15.9	189 H.815.0	8,8	7.7	7.9	7.9	8.09	7.99	8.11	7-98
37.5	37.5	375	37.4	37.4	149	15.9	\$9 4.7 15.0	8.8	7.8	7.8	7-9	8.08	8.60	8.19	7.99
38	38.0	37.9	38.0	38.0	154	15.9	159146151	8.7	7.8	7.9	7.9	8,06	8.00	8-17	7.99
38.5	38.5	38.4	38.5	38.5	15.6	16.0	16.014.7151	8.8	7.8	7.9	7.9	8,05	8.00	8.17	8.00

		0	24	48	72
Technician Initials:	WQ Readings:	PA	Aso	KEP	AW
	Dilutions made by:	AC			

Comments:	0 hrs:	
	24 hrs:	
	48 hrs:	
	72 hrs:	
QC Che	ck: AC 12/M14	Final Review: 47 12/4/14

Marine Chronic Bioassay

Echinoderm Laryal Development Worksheet

Client:	Poseidon	Start Date/Time: 10/30/2014 / 1620
Sample ID:	Salinity Tolerance study (Brine)	End Date/Time: 41/3/2014 1 1414 1720
Test No.:	1410-3151	Species: D. excentricus
Tech initials:		Animal Source: Mission Bay
Injection Time:		Date Collected: 1004114
Sperm Absorbance at	400 nm: 0.744 (target range of 0.8 - 1.0 for density of 4x	10 ⁶ sperm/ml)
F 000 4-4.	27 Noon 2 1050	
Eggs Counted:	Mean: X 50 =	eggs/ml
	(target counts of 25 eggs per vertical pass on Sed	
	(target counts of 25 eggs per vertical pass on Sedo	gwick-Rafter
	slide for a final density of 1000 eggs/ml)	· A
	7.5	
	· · · · · · · · · · · · · · · · · · ·	
Initial density:	U050 eggs/ml = 1.05 dilution factor	200 start. 100
Final density:	- Callettott Taotot	egg stock U ml
i mai donary.	(0.05) parts seawater	seawaterml
	D. D. Parts seawater	
Prepare the egg stock a	according to the calculated dilution factor. For example, if the dilution fa	actor is 2.25. use 100 ml of existing stock (1
part) and 125 ml of dilu	tion water (1.25 parts).	?
r.		
Volume of Sperm stock	needed to fertilize eggs:	
Egg Stock (mL) =	00	
Sperm Stock (μL) =	11,0.	
Egg/Sperm Ratio =	Imlilut	
133, 0po radio	[M] ([M]	
Fertilization Time:	1555	
Embryo Stock Fertilizati	ion Checks: No. No.	
	TimeFertUnfert%	
10 minutes (1st fert.)	1605 98 2 98	
20 minutes (2nd fert. If a	needed)	
Test Initiation Time:	Embryo Stock Added: 0.25 ml	
•		
Test Termination:		•
	No. No. %	
	Normal Abnormal Normal	1
72-hour QC check ^a	97 3 97	
End of test QC check		
End of tool QO oncon		
Comments:	^a If the embryo development does not meet the mean test acceptabi	lity critorian of 200/ normally
Comments.	developed, continue the test up to 96-hrs (ASTM 1999).	iny chiefforf of 80 % normally
	developed, continue the test up to so-this (AS TW 1999).	,
		, 1 .
QC Check:	AC12/4/14	Final Review: PTP 12/4/14
		The restriction of the second
Nautilus Environmental. 43	140 Vandever Avenue. San Diego, CA 92120.	

Sand Dollar 72-hour Larval Development

Test Date: July 22, 2015

CETIS Summary Report

Report Date: Test Code: 04 Aug-15 15:01 (p 1 of 1) 1507-S083 | 20-0434-3180

								Test Code:	:	150	7-S083 2	0-0434-318
Echinoid Eml	bryo-Larval Dev	elopmer	nt Test							Nautilus	Environn	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	03-4901-7220 22 Jul-15 15:10 25 Jul-15 15:40 73h	0 i	Test Type: Protocol: Species: Source:	Development EPA/600/R-95/ Dendraster exc Mission Bay	` '			Analyst: Diluent: Brine: Age:		ıral Seawate en Seawate		
Sample ID:	15-0490-2480	(Code:	1507-S083				Client:	Pos	eidon		
Sample Date:			Material:	Brined seawate	r			Project:				
Receive Date			Source:	Poseidon								
Sample Age:	15h	,	Station:									
Comparison	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Meti	nod			
17-8801-0881	Development F	Rate	38.5	>38.5	NA	4.29%		Duni	nett M	ultiple Com	parison Te	st
Point Estimat	te Summary											
Analysis ID	Endpoint		Level	ppt	95% LCL	95% UCL	TU	Meth	emant-ma			
06-2599-1098	Development F	Rate	EC25	>38.5	N/A	N/A		Line	ar Inte	erpolation (IC	CPIN)	
			EC50	>38.5	N/A	N/A			DANIE DE NATIONA			
Test Acceptal	bility											
Analysis ID	Endpoint		Attrib		Test Stat	TAC Limi	ts	Ove	rlap	Decision		14
06-2599-1098	Development F			ol Resp	0.964	0.8 - NL		Yes		Passes Ad		
17-8801-0881	Development F			ol Resp	0.964	0.8 - NL		Yes		Passes Ad		
17-8801-0881	Development F	Rate	PMSD		0.04292	NL - 0.25		No	C. III. A. C.	Passes Ad	cceptability	Criteria
Development	Rate Summary											
C-ppt	Control Type	Count		95% LCL	95% UCL	Min	Max			Std Dev	CV%	%Effect
33.4	Brine Control	5	0.96	0.9228	0.9972	0.91	0.99			0.03	3.13%	0.0%
33.5 34.9	Lab Control	5 5	0.964 0.976	0.9452 0.9552	0.9828 0.9968	0.94 0.95	0.98		6782 7483	0.01517 0.01673	1.57% 1.71%	-0.42% -1.67%
35.4		5	0.962	0.9398	0.9842	0.94	0.99			0.01073	1.86%	-0.21%
35.9		5	0.962	0.9381	0.9859	0.94	0.99		8602	0.01703	2.0%	-0.21%
36.4		5	0.944	0.8939	0.9941	0.9	0.99			0.04037	4.28%	1.67%
36.9		5	0.962	0.9233	1	0.91	0.99			0.03114	3.24%	-0.21%
37.4		5	0.966	0.9549	0.9771	0.96	0.98			0.008944	0.93%	-0.63%
37.9		5	0.93	0.9022	0.9578	0.9	0.96			0.02236	2.4%	3.13%
38.5		5	0.942	0.9111	0.9729	0.92	0.98		114	0.0249	2.64%	1.88%
Development	Rate Detail				M. J							
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
33.4	Brine Control	0.91	0.97	0.97	0.99	0.96						
33.5	Lab Control	0.98	0.97	0.94	0.96	0.97						
34.9		0.99	0.97	0.95	0.98	0.99						
35.4		0.96	0.99	0.94	0.96	0.96						
35.9		0.95	0.94	0.97	0.96	0.99						
36.4		0.99	0.98	0.94	0.9	0.91						
36.9		0.97	0.98	0.96	0.91	0.99						
37.4		0.97	0.96	0.96	0.96	0.98						
37.9		0.92	0.93	0.96	0.94	0.9						
						0.00						

Analyst: AC QA: 15 8/4/19

000-089-170-2 CETIS™ v1.8.7.20

0.92

0.98

0.95

0.92

0.94

38.5

Report Date: Test Code: 04 Aug-15 15:01 (p 1 of 2)

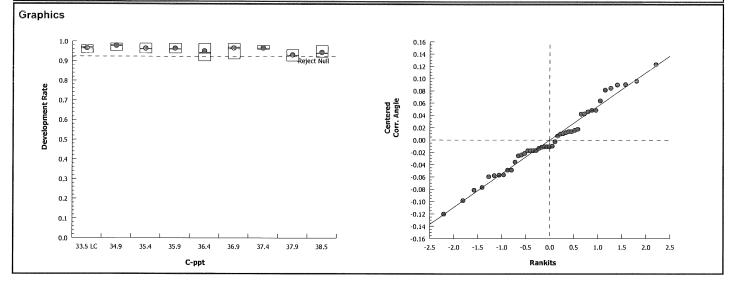
1507-S083 | 20-0434-3180

			EXTENSION OF THE PARTY OF THE P				Test	Code:	150	7-S083 2	0-0434-318
Echinoid En	nbryo-Larval De	velopme	ent Test						Nautilus	Environ	mental (CA)
Analysis ID: Analyzed:	17-8801-0881 04 Aug-15 15		•	velopment F rametric-Cor	Rate ntrol vs Trea	tments		IS Version: cial Results:	CETISv1 Yes	.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cori	rected)	NA	C > T	NA	NA		4.29%	38.5	>38.5	NA	
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-ppt		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(α:5%)		
33.5	34.9		-1.038	2.478	0.094 8	0.9933	CDF	Non-Signif	icant Effect		7-
33.5	35.4		0.06751	2.478	0.094 8	0.8724	CDF	Non-Signif	icant Effect		
33.5	35.9		0.05108	2.478	0.094 8	0.8766	CDF	Non-Signif	icant Effect		
33.5	36.4		0.9325	2.478	0.094 8	0.5351	CDF	Non-Signif	icant Effect		
33.5	36.9		-0.08789	2.478	0.094 8	0.9079	CDF	Non-Signif	icant Effect		
33.5	37.4		-0.09918	2.478	0.094 8	0.9102	CDF	Non-Signif	icant Effect		
33.5	37.9		2.034	2.478	0.094 8	0.1203	CDF	Non-Signif	icant Effect		
33.5	38.5		1.314	2.478	0.094 8	0.3588	CDF	Non-Signif	icant Effect		
ANOVA Tabl	е										
Source	Sum Sq	uares	Mean Squ	Jare	DF	F Stat	P-Value	Decision(a:5%)		
Between	0.048425	526	0.006053	158	8	1.678	0.1376	Non-Signif	icant Effect		
Error	0.129827	7 5	0.0036063	32	36						
Total	0.178252	28			44	_					
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Bartlett	Equality	of Variance	7.868	20.09	0.4464	Equal Var	iances			
Distribution	Shapiro-	-Wilk W	Normality	0.9796	0.9308	0.6045	Normal D	istribution			
Developmen	t Rate Summary	,					***************************************				
C-ppt	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
33.5	Lab Control	5	0.964	0.9452	0.9828	0.97	0.94	0.98	0.006782	1.57%	0.0%
34.9		5	0.976	0.9552	0.9968	0.98	0.95	0.99	0.007483	1.71%	-1.25%
35.4		5	0.962	0.9398	0.9842	0.96	0.94	0.99	0.008	1.86%	0.21%
35.9		5	0.962	0.9381	0.9859	0.96	0.94	0.99	0.008602	2.0%	0.21%
36.4		5	0.944	0.8939	0.9941	0.94	0.9	0.99	0.01806	4.28%	2.08%
36.9		5	0.962	0.9233	1	0.97	0.91	0.99	0.01393	3.24%	0.21%
37.4		5	0.966	0.9549	0.9771	0.96	0.96	0.98	0.004	0.93%	-0.21%
37.9		5	0.93	0.9022	0.9578	0.93	0.9	0.96	0.01	2.4%	3.53%
38.5		5	0.942	0.9111	0.9729	0.94	0.92	0.98	0.01114	2.64%	2.28%
Angular (Cor	rected) Transfo	rmed Su	ımmary								
C-ppt	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
33.5	Lab Control	5	1.383	1.334	1.432	1.397	1.323	1.429	0.01765	2.85%	0.0%
34.9		5	1.422	1.356	1.488	1.429	1.345	1.471	0.02377	3.74%	-2.85%
35.4		5	1.38	1.313	1.448	1.369	1.323	1.471	0.02425	3.93%	0.19%
35.9		5	1.381	1.31	1.452	1.369	1.323	1.471	0.02551	4.13%	0.14%
36.4		5	1.348	1.225	1.47	1.323	1.249	1.471	0.04398	7.3%	2.56%
		_	4 000	4 004	4 400	1.397	1.266	1.471	0.03447	5.56%	-0.24%
		5	1.386	1.291	1.482	1.557	1.200	1.771	0.00 1 17	0.0070	
37.4		5 5	1.387	1.354	1.419	1.369	1.369	1.429	0.01178	1.9%	-0.27%
36.9 37.4 37.9 38.5											

Report Date: Test Code: 04 Aug-15 15:01 (p 2 of 2) 1507-S083 | 20-0434-3180

Echinoid Em	bryo-Larval Dev	elopment	Test					Nautilus Environmental (CA)
Analysis ID: Analyzed:	17-8801-0881 04 Aug-15 15:		•	evelopment arametric-Co		atments	CETIS Version: Official Results:	CETISv1.8.7 Yes
Development	: Rate Detail							
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
33.5	Lab Control	0.98	0.97	0.94	0.96	0.97		
34.9		0.99	0.97	0.95	0.98	0.99		
35.4		0.96	0.99	0.94	0.96	0.96		
35.9		0.95	0.94	0.97	0.96	0.99		
36.4		0.99	0.98	0.94	0.9	0.91		
36.9		0.97	0.98	0.96	0.91	0.99		
37.4		0.97	0.96	0.96	0.96	0.98		
37.9		0.92	0.93	0.96	0.94	0.9		
38.5		0.94	0.92	0.98	0.95	0.92		

Angular (0	Corrected) Transfor	med Detai	il			
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
33.5	Lab Control	1.429	1.397	1.323	1.369	1.397
34.9		1.471	1.397	1.345	1.429	1.471
35.4		1.369	1.471	1.323	1.369	1.369
35.9		1.345	1.323	1.397	1.369	1.471
36.4		1.471	1.429	1.323	1.249	1.266
36.9		1.397	1.429	1.369	1.266	1.471
37.4		1.397	1.369	1.369	1.369	1.429
37.9		1.284	1.303	1.369	1.323	1.249
38.5		1.323	1.284	1.429	1.345	1.284



Report Date: Test Code:

04 Aug-15 15:01 (p 1 of 1)

1507-S083 | 20-0434-3180

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Analysis ID: Analyzed:

06-2599-1098 04 Aug-15 15:01

Analysis: Linear Interpolation (ICPIN)

Endpoint: Development Rate

CETIS Version:

CETISv1.8.7

Official Results:

Yes

Lin	ear li	ntern	olat	ion (Onti	ons

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	1018710	1000	Yes	Two-Point Interpolation

Point Estimates

ppt

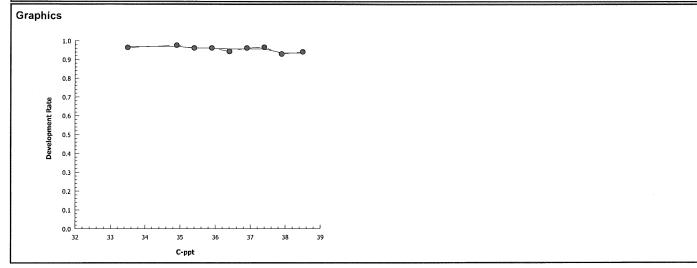
Level

95% LCL 95% UCL

EC25 >38.5 N/A N/A EC50 >38.5 N/A N/A

Developm	nent Rate Summary	/		Calculated Variate(A/B)							
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
33.5	Lab Control	5	0.964	0.94	0.98	0.006782	0.01517	1.57%	0.0%	482	500
34.9		5	0.976	0.95	0.99	0.007483	0.01673	1.71%	-1.25%	488	500
35.4		5	0.962	0.94	0.99	0.008	0.01789	1.86%	0.21%	481	500
35.9		5	0.962	0.94	0.99	0.008602	0.01924	2.0%	0.21%	481	500
36.4		5	0.944	0.9	0.99	0.01806	0.04037	4.28%	2.08%	472	500
36.9		5	0.962	0.91	0.99	0.01393	0.03114	3.24%	0.21%	481	500
37.4		5	0.966	0.96	0.98	0.004	0.008945	0.93%	-0.21%	483	500
37.9		5	0.93	0.9	0.96	0.01	0.02236	2.4%	3.53%	465	500
38.5		5	0.942	0.92	0.98	0.01114	0.0249	2.64%	2.28%	471	500

Developr	nent Rate Detail					
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
33.5	Lab Control	0.98	0.97	0.94	0.96	0.97
34.9		0.99	0.97	0.95	0.98	0.99
35.4		0.96	0.99	0.94	0.96	0.96
35.9		0.95	0.94	0.97	0.96	0.99
36.4		0.99	0.98	0.94	0.9	0.91
36.9		0.97	0.98	0.96	0.91	0.99
37.4		0.97	0.96	0.96	0.96	0.98
37.9		0.92	0.93	0.96	0.94	0.9
38.5		0.94	0.92	0.98	0.95	0.92



Report Date:

21 Jul-15 17:13 (p 1 of 2)

Test Code: 20-0434-3180/7777D98C

Echinoid En	nbryo-	Larva	l Deve	elopment Te	st				Nautilus Environmental (C
Start Date: End Date: Sample Date	25 .	Jul-15 Jul-15 Jul-15		Protoc		aster excer 600/R-95/13 I seawater		Sample Code: Sample Source: Sample Station:	1507-S 083
C-ppt	Code	Rep	Pos	# Counted	# Normal			Notes	
			1	100	92	CH	8/3/15		
	+		2	100	90	- CVI	1 01011		
			3		90		1	**************************************	J-000000000000000000000000000000000000
			4	166	94		-		
*****		!	5	1,00	94		 		
			6	100	97		1		
			7		94				
			8	100	98				
			9	100		-		11 11 11 11 11 11 11 11 11 11 11 11 11	
			10		96	1	/		
			11	100	98	CIL	8/4/15		
			12	100	96	UIT Y	61-0115	Personal and a second	
			13	100	96				
			14		99				
	-		15	100	94				
			16	100			1		
			17		96				
			18	100					
· · · · ·			19	100	99				
			20	100	94				
			21	100					
	-		22	100	96				
			23	100	92				
				100	91				
			24 25	100	91				
			26	100	97			***************************************	
				100	95				
			27	100	91				
			28	100	96			***************************************	
				(00)	98				
			30	100	96				
			31	100	94				
			32	100	99		-	***************************************	
			33	100	97		1		
			34	100	96			***************************************	e verification of the control of the
			35	100	99				
			36	100	93				
			37	100	96				
			38	100	97		<u> </u>		
			39	100	97				
			40	100	95				
			41	100	97				· · · · · · · · · · · · · · · · · · ·
			42	001	99				300000000000000000000000000000000000000
			43	100	96				
			44	100	99				- The state of the
			45	100	97				
			46	100	92				
			47	100	94	1			

Report Date:

21 Jul-15 17:13 (p 2 of 2)

C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
			48	100	98	CH 814116
			49	100	98	
			50	100	96	+

Report Date:

21 Jul-15 17:13 (p 1 of 2)

Test Code: ¡ՏԵՋ-ՏԵՑ-Յ 20-0434-3180/7777D98C

	ibi yo-	Laiva	Deve	elopment Te			Nautilus Environmenta	31 (C
Start Date: End Date: Sample Date	25 .	Jul-15 Jul-15 Jul-15		Specie Protoc Materia	ol: EPA/6	aster excentricus 500/R-95/136 (1995) I seawater	Sample Code: 1507-S Sample Source: Poseidon Sample Station:	
C-ppt	Code	Rep	Pos	# Counted	# Normal		Notes	
0	BC	1	24	100	96	Ac alade		
0	BC	2	33	1.0	. 0	AC 7/28/15		
0	BC	3	25					
0	BC	4	42			45,000,000		
0	BC	5	28					
				(5, 5)	(1)	102/2/2		
0	LC	1	10	100	96	AC7 26/15		
0	LC	2	15					
0	LC	3	20					
0	LC	4	30					
0	LC	5	6					
35		1	32					
35		2	39					
35		3	40					
35		4	8	900				
35		5	19					
35.5		1	12					
35.5		2	44					
35.5		3	5					
35.5		4	21					
35.5		5	11					
36		1	26					
36		2						
36		3	45					
36		4	16					
		5	35					
36								
36.5		1	18				335	
36.5		2	29					
36.5		3	47			AND ADMINISTRATION OF THE PROPERTY AND A SAFETY AND A SAFETY AS A		
36.5		4	2					
36.5		5	27					
37		1	38					
37		2	49					
37		3	34					-
37		4	23					
37		5	13					
37.5		1	41					-
37.5		2	37					
37.5		3	31					
37.5		4	9					
37.5		5	43					
38		1	1					
38		2						
			36					
38		3	50					
38		4	14					
38		5	3					
38.5		1	4					
38.5		2	46					

Report Date: 21 Jul-15 17:13 (p 2 of 2) Test Code: 20-0434-3180/7777D98C

C-ppt	Code	Rep	Pos	# Counted	# Normal	Notes
38.5		3	48			
38.5		4	17			
38.5		5	22			

GC wy

Analyst: A QA: 3 8/4/15

Marine Chronic Bioassay

Water Quality Measurements

Client: Poseidon

Sample ID: Nautilus brine (frozen seawater)

Sample Log No.: 15 N/A

Test No.: 1507

Test Species: D. excentricus

Start Date/Time: -7/21/2015

End Date/Time: 7/24/2015

Concentration (ppt)	Salinity (ppt)					Temperature (°C)				Dissolved Oxygen (mg/L)				pH (pH units)			
	0	24	48	72	0	24	48	72	0	24	48	72	0	24	48	72	
Lab Control	335	33.4	33.5	33.3	16.0	15.5	15.2	154	8.6	7.9	7.9	80	8.09	8.65	793		
Brine Control	334	33.3	33.4	33.3	160	15.2	15.0	15.2	४५	8.0	7.8	8.2	8.10	8.07	294	206	
35.0	34.9	34.8	34.9	34.8	160	15.2	14.9	15.3	8.4	8.6	8.0	8.1	8.09	8.07	1990	8.06	
35.5	35.4	35.4	35.4	35.3	160	15.2	14.8	15.2	8.7	8.1	8.0	£2	8.09	8.09	8.06	8-01	
36.0	35.9	35.9	35.9	35.9	158	15.2	14.8	15.4	8-8	8.1	8.1	8-2	8.09	8.08	8.07	8.0	
36.5	364	36.4	36.4	36.3	15.9	15.1	14.8	15.4	8.8	8-1	8.0	82	8.09	8.09	8.07	8.07	
37.0	36.9	36.9	36.9	36.8	15.9	15.1	14.7	15.4	8.8	8-0	8.0	8-2	8.09	6.69	8.08	8-07	
37.5	37.4	37.4	37.4	37.3	12.8	15.1	14.8	15.4	8.8	8.1	4.0	8.2	8,09	8,09	8.08	8.07	
38.0	37.9	37.9	37.9	37.8	15.6	15-2	14.7	15.4	88	8,0	4.0	8.2	808	809	8.09		
38.5	385	38,5	36.4	383	15.6	15.1	14.7	15.6	8.8	8.1	8.0	8.2	8.05	8.09	8.09		

WQ Readings: Technician Initials: Dilutions made by: Ac

Comments:

Ohrs: Hach Sension 5 Salinity meter

24 hrs:

48 hrs: (A) CH Q18 7/24/15

72 hrs:

QC Check: A(8/4/

Final Review: 8/4/15

Marine Chronic Bioassay

Echinoderm Larval Development Worksheet

Client: Sample ID: Test No.:	POSCIDION Nautius brine (Frozen seawater) 1507-S083	Start Date/Time: 124 151 1510 End Date/Time: 125 151 1540 Species: D. excentricus
Tech initials: Injection Time:	AC 1430	Date Collected: 7 11 15
Sperm Absorbance at	400 nm:/. O(target range of 0.8 - 1.0 for density o	of 4x10 ⁶ sperm/ml)
Eggs Counted:	$\frac{16}{27}$ Mean: $\frac{19.6}{6} \times 50 = \frac{9.80}{20}$ (target counts of 20 eggs per vertical pass on S	eggs/ml
	slide for a final density of 1000 eggs/ml)	cugwick realist
Initial density: Final density:	$\frac{980}{1000} \frac{\text{eggs/ml}}{\text{eggs/ml}} = \frac{6.98}{9} \text{ dilution factor}$	egg stock 100 ml seawater ml
Prepare the egg stock (1 part) and 125 ml of c	according to the calculated dilution factor. For example, if the dilutifilution water (1.25 parts).	ion factor is 2.25, use 100 ml of existing stock
Add 100 μL sperm stoo	k per 100mL of egg stock. For example, if you have 60mL of egg	stock, add 60μL sperm stock.
Embryo Stock Fertilizat	ion Checks (Initiate test only when fertilization is ≥90%):	
10 minutes (1st fert.) 20 minutes (2nd fert. If	$ \begin{array}{cccc} & & \text{No.} & \text{No.} \\ & & \text{Time} & & \text{Fert.} & & \text{Unfert.} & \% \\ \hline 1502 & & 97 & 3 & 97 \end{array} $	<u>7</u>
Fertilization Time:	1452	
Test Initiation Time:	1510 Embryo Stock Added: 0.25	i ml
Test Termination:		
72-hour QC check ^a End of test QC check	No. No. % Normal Abnormal Normal 100	
Comments:	^a If the embryo development does not meet the mean test accep developed, continue the test to 96-hrs (ASTM 1999).	tability criterion of 80% normally
QC Check:	AC 8/4/15	Final Review: 674/15

Purple Urchin Fertilization

Test Date: October 30, 2014

CETIS Summary Report

Report Date: Test Code: 30 Jun-15 09:45 (p 1 of 1)

eport Date.	00 dair 10 00.40 (p 1 01
est Code:	1410-S148 01-7477-75

Echinoid Spe	rm Cell Fertiliza	tion Test 1	5C						Nautilus	Environm	nental (CA)		
Batch ID: Start Date: Ending Date: Duration:	07-7620-3286 30 Oct-14 15:09 30 Oct-14 15:49 40m	5 Pro 5 Sp		Fertilization EPA/600/R-95/ Strongylocentro Pt. Loma	, ,	tus			luent: Natural Seawater rine: Frozen Seawater				
Sample ID: Sample Date: Receive Date: Sample Age:		So	de: terial: urce: tion:	Brine Brined seawate Poseidon Naudilus	BANG			Client: Pos Project:	eidon				
Sample Note:	Frozed seawate	er prepared	at Nauti	lus was used as	brine.								
Comparison S	Summary				·								
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method					
20-1180-9983	Fertilization Ra	te	38.5	>38.5	NA	10.4%		Dunnett M	lultiple Com	parison Te	st		
Point Estimat	e Summary												
Analysis ID	Endpoint		Level ppt		95% LCL	95% UCL	ΤU	Method					
15-0920-7834		te	EC25 >38.5		N/A	N/A		Linear Inte	erpolation (I	CPIN)			
			EC50 >38.5		N/A	N/A							
Test Acceptab	bility												
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Overlap	verlap Decision				
15-0920-7834	<u>.</u>	Fertilization Rate		ol Resp	0.808	0.7 - NL		Yes	Passes Acceptability Crite		Criteria		
20-1180-9983				ol Resp	0.808	0.7 - NL		Yes	Passes A	cceptability	Criteria		
20-1180-9983	-1180-9983 Fertilization Rate PMS			•	0.1039	NL - 0.25		No	Passes A	cceptability	Criteria		
Fertilization R	on Rate Summary												
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect		
0	Brine Control	5	0.836	0.7907	0.8813	0.78	0.87	0.01631	0.03647	4.36%	0.0%		
0	Lab Control	5	0.808	0.7236	0.8924	0.73	0.88	0.0304	0.06797	8.41%	3.35%		
35		5	0.868	0.7893	0.9467	0.77	0.94	0.02835	0.0634	7.31%	-3.83%		
35.5		5	0.828	0.7743	0.8817	0.76	0.87	0.01934	0.04324	5.22%	0.96%		
36		5	0.832	0.8011	0.8629	0.79	0.85	0.01114	0.0249	2.99%	0.48%		
36.5		5	0.848	0.7698	0.9262	0.78	0.95	0.02818	0.06301	7.43%	-1.44%		
37		5	0.842	0.7905	0.8935	0.79	0.89	0.01855	0.04147	4.93%	-0.72%		
37.5		5	0.822	0.772	0.872	0.78	0.86	0.018	0.04025	4.9%	1.68%		
38 38.5		5 5	0.822 0.816	0.7911 0.769	0.8529 0.863	0.79 0.77	0.86 0.86	0.01114 0.01691	0.0249 0.03782	3.03% 4.63%	1.68% 2.39%		
Fertilization R	Pate Detail		2.010	200									
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5							
0	Brine Control	0.87	0.78	0.82	0.86	0.85	d constant						
0	Lab Control	0.75	0.70	0.88	0.73	0.81							
35	235 00/100	0.85	0.89	0.77	0.94	0.89							
35.5		0.86	0.87	0.82	0.76	0.83							
				0.85	0.83	0.84							
36		0.79	0.85	บ.ถอ									
36 36 5		0.79 0.78	0.85 0.82										
36.5		0.78	0.82	0.95	0.84	0.85							
36.5 37		0.78 0.79	0.82 0.89	0.95 0.87	0.84 0.81	0.85 0.85							
36.5		0.78	0.82	0.95	0.84	0.85							

Echinoid Sperm Cell Fertilization Test 15C

Report Date: Test Code: 30 Jun-15 09:44 (p 1 of 2) 1410-S148 | 01-7477-7582

Nautilus Environmental (CA)

Analysis ID: 20-1180-9983 Endpoint: Fertilization Rate CETIS Version: CETISv1.8.7

Analyzed: 30 Jun-15 9:44 Analysis: Parametric-Control vs Treatments Official Results: Yes

Sample Note: Frozed seawater prepared at Nautilus was used as brine.

Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corrected)	NA	C > T	NA	NA	10.4%	38.5	>38.5	NA	

Dunnett Multi	ple C	omparison Test							
Control	vs	C-ppt	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(α:5%)
Lab Control		35	-2.005	2.478	0.104	8	0.9998	CDF	Non-Significant Effect
		35.5	-0.5516	2.478	0.104	8	0.9708	CDF	Non-Significant Effect
		36	-0.6444	2.478	0.104	8	0.9775	CDF	Non-Significant Effect
		36.5	-1.342	2.478	0.104	8	0.9976	CDF	Non-Significant Effect
		37	-1.007	2.478	0.104	8	0.9926	CDF	Non-Significant Effect
		37.5	-0.3573	2.478	0.104	8	0.9512	CDF	Non-Significant Effect
		38	-0.3308	2.478	0.104	8	0.9479	CDF	Non-Significant Effect
		38.5	-0.166	2.478	0.104	8	0.9227	CDF	Non-Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)
Between	0.02892046	0.003615058	8	0.8153	0.5940	Non-Significant Effect
Error	0.1596273	0.004434091	36			
Total	0.1885477		44			

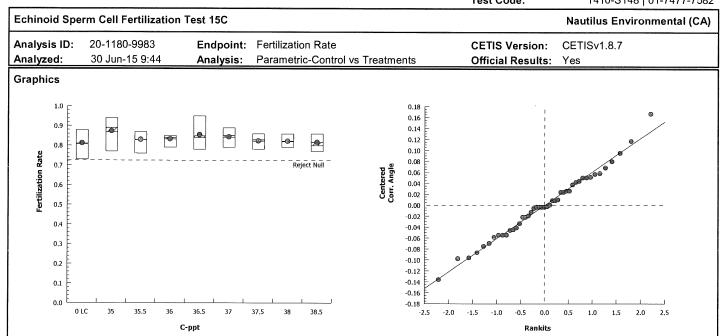
Distributional T	ests				
Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance	9.343	20.09	0.3142	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9902	0.9308	0.9652	Normal Distribution

Fertilization	on Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.808	0.7236	0.8924	0.81	0.73	0.88	0.0304	8.41%	0.0%
35		5	0.868	0.7893	0.9467	0.89	0.77	0.94	0.02835	7.31%	-7.43%
35.5		5	0.828	0.7743	0.8817	0.83	0.76	0.87	0.01934	5.22%	-2.48%
36		5	0.832	0.8011	0.8629	0.84	0.79	0.85	0.01114	2.99%	-2.97%
36.5		5	0.848	0.7698	0.9262	0.84	0.78	0.95	0.02818	7.43%	-4.95%
37		5	0.842	0.7905	0.8935	0.85	0.79	0.89	0.01855	4.93%	-4.21%
37.5		5	0.822	0.772	0.872	0.83	0.78	0.86	0.018	4.9%	-1.73%
38		5	0.822	0.7911	0.8529	0.82	0.79	0.86	0.01114	3.03%	-1.73%
38.5		5	0.816	0.769	0.863	8.0	0.77	0.86	0.01691	4.63%	-0.99%

Angular (0	Corrected) Transfor	med Sumr	nary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.122	1.014	1.231	1.12	1.024	1.217	0.03908	7.79%	0.0%
35		5	1.207	1.091	1.322	1.233	1.071	1.323	0.0416	7.71%	-7.53%
35.5		5	1.145	1.076	1.215	1.146	1.059	1.202	0.02511	4.9%	-2.07%
36		5	1.149	1.109	1.19	1.159	1.095	1.173	0.01452	2.83%	-2.42%
36.5		5	1.179	1.055	1.302	1.159	1.083	1.345	0.04445	8.43%	-5.04%
37		5	1.164	1.094	1.235	1.173	1.095	1.233	0.02549	4.9%	-3.78%
37.5		5	1.137	1.072	1.202	1.146	1.083	1.187	0.02351	4.62%	-1.34%
38		5	1.136	1.095	1.177	1.133	1.095	1.187	0.01477	2.91%	-1.24%
38.5		5	1.129	1.068	1.19	1.107	1.071	1.187	0.02203	4.36%	-0.62%

000-089-170-2 CETIS™ v1.8.7.20

Report Date: Test Code: 30 Jun-15 09:45 (p 2 of 2) 1410-S148 | 01-7477-7582



Report Date: Test Code:

30 Jun-15 09:45 (p 1 of 1)

1410-S148 | 01-7477-7582

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: Analyzed:

15-0920-7834 30 Jun-15 9:44 Endpoint: Fertilization Rate Linear Interpolation (ICPIN) Analysis:

CETIS Version: Official Results:

CETISv1.8.7

Yes

Sample Note: Frozed seawater prepared at Nautilus was used as brine.

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	842477	1000	Yes	Two-Point Interpolation

Point Estimates

Level	ppt	95% LCL	95% UCL
EC25	>38.5	N/A	N/A
EC50	>38.5	N/A	N/A

Fertilizat	ion Rate Summary				Cal	culated Varia	ite(A/B)				
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.808	0.73	0.88	0.0304	0.06797	8.41%	0.0%	404	500
35		5	0.868	0.77	0.94	0.02835	0.0634	7.31%	-7.43%	434	500
35.5		5	0.828	0.76	0.87	0.01934	0.04324	5.22%	-2.48%	414	500
36		5	0.832	0.79	0.85	0.01114	0.0249	2.99%	-2.97%	416	500
36.5		5	0.848	0.78	0.95	0.02818	0.06301	7.43%	-4.95%	424	500
37		5	0.842	0.79	0.89	0.01855	0.04147	4.93%	-4.21%	421	500
37.5		5	0.822	0.78	0.86	0.018	0.04025	4.9%	-1.73%	411	500
38		5	0.822	0.79	0.86	0.01114	0.0249	3.03%	-1.73%	411	500
38.5		5	0.816	0.77	0.86	0.01691	0.03782	4.63%	-0.99%	408	500

Graphics 1.0 ┌ 0.7 0.2 0.1

C-ppt

Report Date:

28 Oct-14 16:43 (p 1 of 2) 01-7477-7582/1410-S148

Test Code: 01-7477-7582/1410-S148

Nautilus Environmental (CA)

Start Date:		Oct-14		Species		centrotus purpuratus Sample Code: Brine	
End Date:	30 (Oct-14	ļ	Protoco		R-95/136 (1995) Sample Source: Poseidon	
Sample Date	: 300	Oct-14	ļ	Materia	I: Brined sea	awater Sample Station:	
C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes	
			1	100	83	1/4/14	
			2	100	83 87	11/ / 1-1	
			3	100	87		
			4	/00	77		
			5	100	84		
			6		95		
			7	100 \$ 26 00	78	•	
·····			8	100	78		
			9	/00	85		
111 - 111 - 111 - 111			10	100	85 87		
			11	00/	82		
			12		76		
			13	/00	70		
			14		94 86 82 82		
			15	100	00		
			16	100	07		
			17	/00	86		
			18	/00/	87 85 77		
			19	/00	<u>85</u>		
			20	/00/	#7		
			21	100	85 75 81		
				100	15		
			22	00 <i>i</i>	81		
			23	/00	79 85		
			24	00	85		
			25	/00	85 82 78		
			26	/00/	88		
			27	100	78		
			28	/00	83		
			29	00/	80		
			30		83 80 79		
			31	/00	86,		
			32	100	86		
			33	/00/	86 86 82 89 82 88 88 86 78 86 86 83		
			34	100	89	11/5/14	
			35	/00	82		
			36	/00	89		
		I	37	/00	88		
			38	/00	85		
			39	/00	95		
			40	.100	78		
			41	100	86		
			42	/00	78		
			43)00	86		
			44	100	26		
			45	100	83		
4			46	100/	25		
			47	000	85 84		

Code Rep Pos

48

49

50

C-ppt

Counted

00/

100

100

Fertilized

79

73

80

Report Date:

28 Oct-14 16:43 (p 2 of 2)

Test Code: 01-7477-7582/1410-S148

Notes

KBOBO Analyst: SG2

Report Date: Test Code:

28 Oct-14 16:43 (p 1 of 2) 01-7477-7582/1410-S148

Nautilus Environmental (CA) **Echinoid Sperm Cell Fertilization Test 15C**

Brine Start Date: 30 Oct-14 Species: Strongylocentrotus purpuratus Sample Code:

Cypit Code Rep Pos # Counted # Fertilized Notes 0 BC 1 10 QOD 3U 1 0 BC 2 40 BC 3 35 0 BC 5 9 10 BC 10 <th>nd Date:</th> <th></th> <th>Oct-14</th> <th></th> <th></th> <th>ol: EPA/600/R-95/136</th> <th></th>	nd Date:		Oct-14			ol: EPA/600/R-95/136	
0	ample Date	e: 30 (Oct-14	ļ	Materia	il: Brined seawater	Sample Station:
0 8C 2 40 0 8C 3 35 5 9 0 0 8C 5 9 9 0 0 1C 1 21 100	C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
0 BC 2 40	0	ВС	1	10	100	86	
0	0	ВС	2	40			
0 BC 5 9 9 100	0	ВС	3	35			
0 LC 1 21 100 76 0 LC 2 3 100 90 0 LC 3 37 0 LC 4 49 0 LC 5 22 35 1 48 100 64 35 2 15 35 3 19 35 5 3 19 35 5 3 3 19 35 5 5 38 35 5 1 3 11 36 6 2 20 36 7 7 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	0	ВС	4	32			
0	0	ВС	5	9			
0	0	LC	1	21	100	76	
0	0	LC	2	3	100	90	
0	0	LC	3	37			
35	0	LC	4	49			
35	0	LC	5	22			
35	35		1	46	loo	84	
35	35		2	15			
35	35	1	3	19			
35.5	35		4	13			
35.5	35	1	5	36			
35.5 2 17 3 3 3 3 3 3 3 3 3		·	1		100	82	
35.5							
36.5			3	38			
36.5 5 1 48 47 48 47 48 <td< td=""><td></td><td></td><td></td><td>12</td><td></td><td></td><td></td></td<>				12			
36 1 48 36 2 20 36 3 25 36 4 28 36.5 1 42 80 36.5 2 16 36.5 3 39 36.5 5 24 37 1 30 37 2 34 37 4 8 .			5				
36 2 20 00 9 36 3 25 36 4 28 36 5 5 36.5 1 42 100 36.5 2 16 36.5 3 39 36.5 4 47 36.5 5 24 37 1 30 100 37 2 34 37 4 8 37.5 1 27 100 76 37.5 2 45 37.5 3 43 3 37.5 4 7 7 38 1 44 100 80 38 1 44 100 80 38 3 3 33 38 4 23 38.5 1 29 100 60						and an artifacture of the second of the seco	
36		+			100	al	
36		+			100		
36.5		-					
36.5							
36.5				L	100	20	
36.5					100	09	
36.5 4 47 36.5 5 24 37 1 30 100 37 2 34 37 3 2 37 4 8 37 5 18 37.5 1 27 37.5 2 45 37.5 3 43 37.5 4 7 37.5 5 41 38 1 44 10 87 38 2 11 38 3 33 38 4 23 38 5 26 38.5 1 29 00							
36.5							
37 1 30 100 3186 37 2 34 34 37 4 8 4 8 37.5 1 27 100 76 37.5 2 45 45 37.5 3 43 343 37.5 4 7 7 38 1 44 100 87 38 2 11 38 38 3 33 33 38 4 23 38 38 5 26 38.5 1 29 100 60		-					
37 2 34 37 4 8 37 5 18 37.5 1 27 00 37.5 2 45 37.5 3 43 37.5 4 7 37.5 5 41 38 1 44 38 2 11 38 3 33 38 4 23 38 5 26 38.5 1 29 00		-			100	Varai	
37 3 2 37 4 8 37 5 18 37.5 1 27 100 76 37.5 2 45 45 37.5 3 43 43 37.5 5 41 41 38 1 44 100 87 38 2 11 41 38 3 33 33 38 4 23 23 38 5 26 26 38.5 1 29 100 60		-			100	1106	
37 4 8 37 5 18 37.5 1 27 100 76 37.5 2 45 37.5 3 43 37.5 4 7 37.5 5 41 38 1 44 100 87 38 2 11 38 3 33 38 4 23 38 5 26 38.5 1 29 100 60		+					
37 5 18 37.5 1 27 100 37.5 2 45 37.5 3 43 37.5 4 7 37.5 5 41 38 1 44 38 2 11 38 3 33 38 4 23 38 5 26 38.5 1 29 100		-					/
37.5 1 27 100 76 37.5 2 45 45 37.5 3 43 43 37.5 5 41 44 100 87 38 1 44 100 87 38 2 11 11 11 38 3 33 33 38 4 23 23 38 5 26 26 38.5 1 29 100 60		-					
37.5 2 45 37.5 3 43 37.5 4 7 38 1 44 100 87 38 2 11 38 3 33 38 4 23 38 5 26 38.5 1 29 100		+			100	76	
37.5 3 43 37.5 4 7 37.5 5 41 38 1 44 100 87 38 2 11 38 3 33 38 4 23 38 5 26 38.5 1 29 100		Market account tomacome			100	70	
37.5 4 7 37.5 5 41 38 1 44 100 38 2 11 38 3 33 38 4 23 38 5 26 38.5 1 29 100		-					
37.5							
38				ļ			
38					100	97	
38 3 33 33 38 4 23 38 5 26 38.5 1 29 100 60					100	OT	
38		-			<u> </u>		
38 5 26 38.5 1 29 100 60							
38.5 1 29 100 60		-					
					:00	(-0	
38.5 2 4		-			100	<u> </u>	

QC SAC

Analyst: A QA: KB11/10/14

Report Date: Test Code: 28 Oct-14 16:43 (p 2 of 2) 01-7477-7582/1410-S148

C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
38.5		3	14			
38.5		4	6			
38.5		5	50			



Marine Chronic Bioassay

Water Quality Measurements

Client/Project: Poseidon/Salinity Tolerance Study Test Species: S. purpuratus

Sample ID: Brine (frozen seawater) Start Date/Time: 10/30/2014 AC 13:05 15:05

Sample Log No.: 44 Not Applicable End Date/Time: 10/30/2014 13.45

Dilutions made by: Test No: 1410-S148

			Analyst:	8
Concentration	DO	Initial R	Readings Salinity	Temperature
(ppt)	(mg/L)	(units)	(ppt)	(°C)
Lab Control	8.7	8.04	33.3	15.6
Brine Control	8.4	8.06	33.7	15.4
35.0	8,4	8.07	34,9	14.8
35.5	8.4	8.07	35.5	15.0
36.0	8.7	8.13	36,0	14.7
36.5	8-8	8.10	36.5	14.7
37.0	6.8	8.09	37.0	14,8
37.5	8.8	8.08	37.5	14.9
38.0	8.7	8.06	38.0	15,4
38.5	8,8	8.05	38,5	15.6

Comments:		
QC Check:	Whiliplia	Final Review: 11/18/14

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

Marine Chronic Bioassay

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	Poseidan Sa Brine (tvoze 1410-5148		nestudy ir)	E	nd Date/Ti Spec	me: 10/30/201 me: 10/30/201 iies: <i>S. purpu</i>	4 / 4 1 5 ! + 5 ratus
Tech initials:	8/AC					rce: <u>Point Lor</u> ted: 10 29	
Injection Time:	1430					- 10 = -	
Sperm Absorbance at 4		(target range of			10 ⁶ sperm/r	nl)	
Eggs Counted:	Me	an: <u>63, 4</u> x :	50 = 31	7 <i>1</i>) eggs	s/ml		
		et counts of 80 eggs p for a final density of 4		on Sedgwick	<-Rafter		
Initial density:	3170 eggs/ml	= 0.63 dilli	tion factor	114	stock	100 ml	1 - d. 7 - 2 - 1 -
Final density:	4000 eggs/ml	- 1.0 par	t egg stock ts seawater	seav		ml	No dilutro required
Prepare the embryo stoc stock (1 part) and 125 m	ck according to the calcu nl of dilution water (1.25 p	lated dilution factor.	For example,	if the dilutio	n factor is 2	2.25, use 100	ml of existing
			Sperm:Egg	Ratio			
Rangefinder Test: ml Sperm Stock	<u>2000:1</u> <u>1600</u>			400:1	200:1	100:1	50:1
ml Seawater	50 40 0.0 10		20 30	10 40	5.0 45	2.5 47.5	1.25 48.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1440 1450	Rangefinder Rati	o: Fert. 73 92	Unfer 27 9	<u>t.</u> 		
NOTE: Choose a sperm this range, choose the ra health, stage of reproduc	atio closest to 90 percent	unless professional	en 80 and 90 ¡ judgment dicta	percent. If i ates consid	more than o eration of o	one concentra ther factors (e	ation is within e.g., organism
<u>Definitive Test</u>		Sperm:Egg Ratio	Used:	01	_		
Sperm Added (100 µl); Eggs Added (0.5 ml):	Time - 1305 1505 - 1345 1545	QC1 QC2 Egg Control 1 Egg Control 2	Fert. 90 74 0	Unfer	<u>t</u> . - - -		
Comments:							

QC Check:	KB 11/10/14				Final Revie	ew: 650 11	18/14
Nautilus Environmental. 4340	T T T T T T T T T T T T T T T T T T T	go, CA 92120.				-	

Purple Urchin Fertilization

Test Date: July 22, 2015

CETIS Summary Report

Report Date:

Test Code:

03 Aug-15 16:36 (p 1 of 1) 1507-S080 | 03-7187-5571

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA)												
	04-1054-0985 22 Jul-15 16:1 1-25 Jul-15 16:5 3734 40 min	7 Pi 7 Si	est Type: rotocol: pecies: purce:	Fertilization EPA/600/R-95/ Strongylocentro Pt. Loma	, ,	itus		Analyst: Diluent: Brine: Age:		ural Seawat zen Seawat	er	
Sample ID: Sample Date Receive Date Sample Age:	: 22 Jul-15	M Se	ode: aterial: ource: ation:	1507-S080 Brined seawate Poseidon Nauhlus P				Client: Project:	Pos	eidon		
Comparison	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Med	thod			
17-4148-2778		ıte	38.5	>38.5	NA	4.94%				lultiple Com	parison Te	st
Doint Fatime	to Current			Part of the second seco								
Point Estimat												
Analysis ID	Endpoint		Level	ppt	95% LCL	95% UCL	TU		hod			· · · · · · · · · · · · · · · · · · ·
02-8620-7509	Fertilization Ra	ite	EC25	>38.5	N/A	N/A		Line	ear Inte	erpolation (I	CPIN)	
			EC50	>38.5	N/A	N/A						
Test Accepta	bility											
Analysis ID	Endpoint		Attribu	ute.	Test Stat	TAC Limi	ts	Ove	erlap	Decision		
02-8620-7509	Fertilization Ra	te	Contro	l Resp	0.936	0.7 - NL		Yes		Passes A	cceptability	Criteria
17-4148-2778			Contro	l Resp	0.936	0.7 - NL		Yes		Passes A	cceptability	Criteria
17-4148-2778	Fertilization Ra	te	PMSD		0.0494	NL - 0.25		No		Passes A	cceptability	Criteria
Fertilization F	Rate Summary											
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std	Err	Std Dev	CV%	%Effect
33.4	Brine Control	5	0.934	0.8961	0.9719	0.91	0.98	0.01	1364	0.0305	3.27%	0.0%
33.5	Lab Control	5	0.936	0.9061	0.9659	0.91	0.97	0.01	1077	0.02408	2.57%	-0.21%
34.9		5	0.946	0.9062	0.9858	0.9	0.98	0.01	1435	0.03209	3.39%	-1.29%
35.4		5	0.934	0.9083	0.9597	0.91	0.96	0.00	9274	0.02074	2.22%	0.0%
35.9		5	0.952	0.9175	0.9865	0.91	0.98	0.01	1241	0.02775	2.92%	-1.93%
36.4		5	0.964	0.9452	0.9828	0.94	0.98	0.00	6782	0.01517	1.57%	-3.21%
36.9		5	0.94	0.9137	0.9663	0.91	0.96	0.00	9487	0.02121	2.26%	-0.64%
37.4		5	0.956	0.9372	0.9748	0.94	0.98		6782	0.01517	1.59%	-2.36%
37.9		5	0.924	0.8928	0.9552	0.9	0.95		122	0.0251	2.72%	1.07%
38.5		5	0.918	0.8688	0.9672	0.88	0.97	0.01	772	0.03962	4.32%	1.71%
Fertilization R	Rate Detail											
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
33.4	Brine Control	0.92	0.95	0.91	0.91	0.98						
33.5	Lab Control	0.93	0.92	0.95	0.91	0.97						
34.9		0.9	0.97	0.98	0.93	0.95						
35.4		0.95	0.96	0.91	0.93	0.92						
35.9		0.94	0.98	0.97	0.96	0.91						
36.4		0.98	0.97	0.94	0.97	0.96						
36.9		0.93	0.96	0.96	0.94	0.91						
37.4		0.98	0.95	0.96	0.94	0.95						
37.9		0.95	0.9	0.92	0.95	0.9						
38.5		0.9	0.97	0.88	0.95	0.89						

@ Q18 2W 8/7/15

Report Date: Test Code: 03 Aug-15 16:34 (p 1 of 2) 1507-S080 | 03-7187-5571

							Test	Code:	150	7-S080 0	3-7187-557
Echinoid Sp	erm Cell Fertiliza	ation Test	15C						Nautilus	Environr	nental (CA
Analysis ID:	17-4148-2778	En	dpoint: Fer	tilization Ra	te		CET	IS Version:	CETISv1.	8.7	
Analyzed:	03 Aug-15 16:	29 A n	alysis : Par	ametric-Cor	trol vs Trea	tments	Offic	ial Results	: Yes		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	rected)	NA	C > T	NA	NA		4.94%	38.5	>38.5	NA	
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-ppt		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(a:5%)		
33.5	34.9		-0.7306	2.478	0.087 8	0.9825	CDF	Non-Signi	ficant Effect		
33.5	35.4		0.1559	2.478	0.087 8	0.8485	CDF	Non-Signi	ficant Effect		
33.5	35.9		-1.082	2.478	0.087 8	0.9942	CDF	Non-Signi	ficant Effect		
33.5	36.4		-1.829	2.478	0.087 8	0.9996	CDF	Non-Signi	ficant Effect		
33.5	36.9		-0.2068	2.478	0.087 8	0.9297	CDF	Non-Signi	ficant Effect		
33.5	37.4		-1.241	2.478	0.087 8	0.9966	CDF	Non-Signi	ficant Effect		
33.5	37.9		0.7002	2.478	0.087 8	0.6433	CDF	Non-Signi	ficant Effect		
33.5	38.5		0.8823	2.478	0.087 8	0.5589	CDF	Non-Signi	ficant Effect		
ANOVA Tabl	e										
Source	Sum Squ	iares	Mean Squ	ıare	DF	F Stat	P-Value	Decision		.,	
Between	0.040866	87	0.0051083	358	8	1.669	0.1400	Non-Signi	ficant Effect		
Error	0.110158	4	0.0030599	955	36	_					
Total	0.151025	3			44						
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(a:1%)			
Variances	Bartlett E	Equality of \	/ariance	4.026	20.09	0.8548	Equal Variances				
Distribution	Shapiro-	Wilk W Nor	mality	0.974	0.9308	0.4007	Normal D	istribution			
Fertilization	Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
33.5	Lab Control	5	0.936	0.9061	0.9659	0.93	0.91	0.97	0.01077	2.57%	0.0%
34.9		5	0.946	0.9062	0.9858	0.95	0.9	0.98	0.01435	3.39%	-1.07%
35.4		5	0.934	0.9083	0.9597	0.93	0.91	0.96	0.009274	2.22%	0.21%
35.9		5	0.952	0.9175	0.9865	0.96	0.91	0.98	0.01241	2.92%	-1.71%
36.4		5	0.964	0.9452	0.9828	0.97	0.94	0.98	0.006782	1.57%	-2.99%
36.9		5	0.94	0.9137	0.9663	0.94	0.91	0.96	0.009487	2.26%	-0.43%
37.4		5	0.956	0.9372	0.9748	0.95	0.94	0.98	0.006782	1.59%	-2.14%
37.9		5	0.924	0.8928	0.9552	0.92	0.9	0.95	0.01122	2.72%	1.28%
38.5		5	0.918	0.8688	0.9672	0.9	0.88	0.97	0.01772	4.32%	1.92%
Angular (Co	rrected) Transfo	rmed Sumi	mary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
33.5	Lab Control	5	1.319	1.254	1.384	1.303	1.266	1.397	0.02346	3.98%	0.0%
34.9		5	1.345	1.255	1.434	1.345	1.249	1.429	0.03216	5.35%	-1.94%
35.4		5	1.314	1.26	1.367	1.303	1.266	1.369	0.01919	3.27%	0.41%
35.9		5	1.357	1.278	1.436	1.369	1.266	1.429	0.02855	4.7%	-2.87%
36.4		5	1.383	1.334	1.432	1.397	1.323	1.429	0.01765	2.85%	-4.85%
36.9		5	1.326	1.271	1.381	1.323	1.266	1.369	0.01987	3.35%	-0.55%
37.4		5	1.362	1.312	1.413	1.345	1.323	1.429	0.01814	2.98%	-3.29%
37.9		5	1.295	1.234	1.355	1.284	1.249	1.345	0.02168	3.75%	1.86%
20.5		5	1 200	1 101	1 386	1 2/10	1 217	1 397	0.03515	6.1%	2 34%

6.1%

2.34%

1.386

1.249

1.217

1.397

0.03515

38.5

5

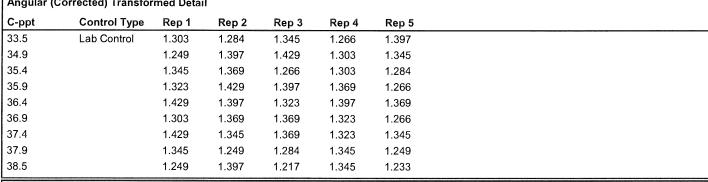
1.288

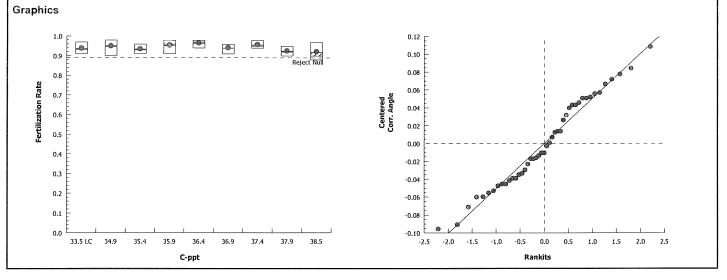
1.191

Report Date: Test Code: 03 Aug-15 16:35 (p 2 of 2)

1507-S080 | 03-7187-5571

Echinoid Sp	erm Cell Fertiliza	Nautilus Environmental (CA)							
		•	Fertilization R Parametric-Co		eatments	CETIS Version: Official Results:	CETISv1.8.7 Yes		
Fertilization	Rate Detail								
C-ppt	Control Type	Rep 1	1 Rep 2	Rep 3	Rep 4	Rep 5			
33.5	Lab Control	0.93	0.92	0.95	0.91	0.97			
34.9		0.9	0.97	0.98	0.93	0.95			
35.4		0.95	0.96	0.91	0.93	0.92			
35.9		0.94	0.98	0.97	0.96	0.91			
36.4		0.98	0.97	0.94	0.97	0.96			
36.9		0.93	0.96	0.96	0.94	0.91			
37.4		0.98	0.95	0.96	0.94	0.95			
37.9		0.95	0.9	0.92	0.95	0.9			
38.5		0.9	0.97	0.88	0.95	0.89			
Angular (Cor	Angular (Corrected) Transformed Detail								
C-ppt	Control Type	Rep 1	l Rep 2	Rep 3	Rep 4	Rep 5			
33.5	Lab Control	1.303	1.284	1.345	1.266	1.397			
34.9		1.249	1.397	1.429	1.303	1 345			





Report Date:

03 Aug-15 16:36 (p 1 of 1)

Test Code:

1507-S080 | 03-7187-5571

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: 02-8620-7509 Analyzed: 03 Aug-15 16:35

Endpoint: Fertilization Rate Linear Interpolation (ICPIN) Analysis:

CETIS Version:

CETISv1.8.7

Official Results: Yes

Linear	Interpo	lation	Options
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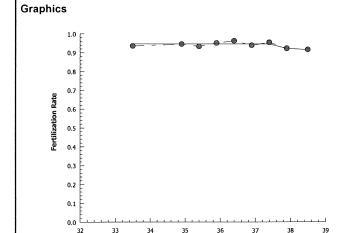
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	45597	1000	Yes	Two-Point Interpolation

Point Estimates

Level	ppt	95% LCL	95% UCI
EC25	>38.5	N/A	N/A
EC50	>38.5	N/A	N/A

Fertilization Rate Summary			Calculated Variate(A/B)								
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
33.5	Lab Control	5	0.936	0.91	0.97	0.01077	0.02408	2.57%	0.0%	468	500
34.9		5	0.946	0.9	0.98	0.01435	0.03209	3.39%	-1.07%	473	500
35.4		5	0.934	0.91	0.96	0.009274	0.02074	2.22%	0.21%	467	500
35.9		5	0.952	0.91	0.98	0.01241	0.02775	2.92%	-1.71%	476	500
36.4		5	0.964	0.94	0.98	0.006782	0.01517	1.57%	-2.99%	482	500
36.9		5	0.94	0.91	0.96	0.009487	0.02121	2.26%	-0.43%	470	500
37.4		5	0.956	0.94	0.98	0.006782	0.01517	1.59%	-2.14%	478	500
37.9		5	0.924	0.9	0.95	0.01122	0.0251	2.72%	1.28%	462	500
38.5		5	0.918	0.88	0.97	0.01772	0.03962	4.32%	1.92%	459	500

Fertilization Rate Detail								
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
33.5	Lab Control	0.93	0.92	0.95	0.91	0.97		
34.9		0.9	0.97	0.98	0.93	0.95		
35.4		0.95	0.96	0.91	0.93	0.92		
35.9		0.94	0.98	0.97	0.96	0.91		
36.4		0.98	0.97	0.94	0.97	0.96		
36.9		0.93	0.96	0.96	0.94	0.91		
37.4		0.98	0.95	0.96	0.94	0.95		
37.9		0.95	0.9	0.92	0.95	0.9		
38.5		0.9	0.97	0.88	0.95	0.89		



C-ppt

Report Date:

21 Jul-15 17:18 (p 1 of 2)

Test Code: ₹₹\$\$7~\$0\\$0 03-7187-5571/162A5EF3

Nautilus Environmental (CA)

Echinoid Sperm Cell Fertilization Test 15C

22 Jul-15

Start Date:

Species: Strongylocentrotus purpuratus

Sample Code: 4507-S N/A

ole Dat	1125 te: 22	Jul-15			al: Brined sea	R-95/136 (1995) awater	Sample S	ource: Poseidon tation:
-ppt	Code	Rep	Pos	# Counted	# Fertilized		N	otes
			1	100	94		7/	29/15
			2	100	91		, /	
			3	100	90			
			4	100	90 95			
			5	100	95			
			6	100	96			
			7	100	95 96 92			
			8	100	95			
			9	100	95			
			10	100	94			
			11	/00	90			
	-		12	100	90 96			
			13	100	98			
			14	100	95			
			15	100	95 96			
			16	100	98			
***			17	100	98 97			
			18	100	97.			
			19	100	9-7			
			20	/00	97 97 95			1.1.100
			21	/00	90			
			22	100	91			
			23	100	91			
			24	100	91 98			
***************************************			25	/00	95			
			26	/00	93			
			27	100	93			
			28	100	93			
			29	100	95			
			30	100	97			
			31	100	9/			
			32	/00/	93 95 97 % 88			
			33	/00	93			
			34	100	<i>95</i>			
			35	100	90			
			36		92			
	+		37	100				
	+ +		38	100	97.			
			39	160	94 92			
			40		96			
			41	/00	91			
	1		42	/00	96			
			43		98			
-			44	/00	94			
	+		45	/00	91			
			46		89			
	1		70	/00	92			

Report Date:

21 Jul-15 17:18 (p 2 of 2)

Test Code: 1507 5080 03-7187-5571/162A5EF3

C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
			48	100	9	7/29/15
			49	100	97	
			50	100	98	4

Report Date: 21 Jul-15 17:18 (p 1 of 2) **Test Code:** 1507-5080 03-7187-5571/162A5EF3

Echinoid Sp	erm C	ell Fe	rtiliza	tion Test 15	3		Nautilus Environmental	(CA)
Start Date: End Date: 7 Sample Date	22 . 2-25 .	Jul-15 Jul-15				centrotus purpuratus R-95/136 (1995)	Sample Code: [©] 1507-S N/小 Sample Source: Poseidon Sample Station:	
			,	# Counted	# Fertilized			
C-ppt 0	Code BC	Rep	Pos 39			1/22/15 AC 9/22/15 AC 9/8	Notes	
0	ВС	2	25	(00	90	AC 71 6018	(7)24)5	
0	BC	3	48					
0	BC	4	41					
0	BC	5	16					
0	LC	1	33	100	96	No alsolve		
0	LC	2	36	100		AC 7/22/15		
0	LC	3	4					
0	LC	4	45					
0	LC	5	30					
35		1	35	100	0.1	10 112-1-		
35	-	2	17	100	92	AC 7/22/15		
35	-	3	43		*****			
35		4	26					
35		5	20					
35.5	-	1	34	1.5	0.4	1-01001-		
		2	/	100	94	AC 7/22/15		
35.5			42					
35.5		3	2		1 - 3\41.4 A - 3.44			
35.5		4	28					
35.5		5	47		0.1	Ne starts		
36		1	10	100	94	Ac 1/20/1		
36		2	50					
36		3	49					
36		4	31					
36		5	22		0 1	1 1 1		
36.5		1	24	100	96	AC 7/27/15		
36.5		2	37					
36.5	-	3	44					
36.5		4	18					
36.5	-	5	12	3 - 0	O 0	<u> </u>		
37		1	27	100	95_	AC7/22/15		
37		2	40					
37	-	3	6					
37		4	38					
37		5	23					
37.5		1	13	100	97	AC7/22/15		
37.5		2	29					
37.5		3	15					
37.5		4	1					
37.5		5	8	h		10		
38		1	9	100	97	AC 7/22/15		
38		2	21					
38		3	7					
38		4	14	,		100		
38		5	3	100	95	AC1/22/15		
38.5		1	11					
38.5		2	19					

 Report Date:
 21 Jul-15 17:18 (p 2 of 2)

 Test Code:
 15ውን - 50%
 03-7187-5571/162A5EF3

C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
38.5		3	32			
38.5		4	5			
38.5		5	46			′

QL WS

Water Quality Measurements

Client:

Poseidon

Test Species: S. purpuratus

Sample ID:

Nautilus brine (frozen seawater)

Start Date/Time: 7/21/2015, 7/22/15 CIB AC 12/15 End Date/Time: 7/21/2015

ALR187122

Dilutions made by: PAIAC

Sample Log No.:

Test No: 1507-5080

			Analyst:	AD
Page consistence and a second			eadings	
Concentration ppt	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	8.6	8.09	33.5	160
Brine Control	8.4	8.10	33.4	16.0
35.0	8.6	8.09	34.9	16.0
35.5	8.7	8-09	35.4	160
36.0	§. §	869	<i>3</i> 5.9	15.8
36.5	8.8	8.09	36.4	15.9
37.0	8.8	8.09	369	15.9
37.5	8.8	8.09	37.4	15.8
38.0	8.8	8.09	37.9	15.6
38.5	8. 8	8.09	38.5	15.6

_					
C_{Ω}	m	m	en	te	•

Hach Sansion # 15 salinity meter

QC Check:

Final Review: ___ 8/7/15

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

Brine Dilution Worksheet

Project:	P	roject:	
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Poseidon

Analyst:

Sample ID:

Nautilus brine (frozen seawater)

Test Date: 7/22/2015

Test No:

Test Type: urchin fertilization and dev.

sand dollar fertilization and dev.

Salinity of Seawater

Test Dilution Volume

33.4

Date of Brine used: 5/21 + 5/29(15; blended 7/21/15

Salinity of Brine

85.2 500

Alkalinity of Brine Control: _____ mg/L as CaCO3

TS = target salinity SE = salinity of effluent

SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
34.0	100.0	250	NA	NA	500
35.0	96.9	484.6	0.03	15.4	500
35.5	95.9	479.7	0.04	20.3	500
36.0	95.0	474.9	0.05	25.1	500
36.5	94.0	470.1	0.06	29.9	500
37.0	93.1	465.3	0.07	34.7	500
37.5	92.1	460.4	0.09	39.6	500
38.0	91.1	455.6	0.10	44.4	500
38.5	90.2	450.8	0.11	49.2	500

DI Volume

	Di Volunio			
Brine Control	76.3	0.64	49.2	500

Total Brine Volume Required (ml):

307.9

QC Check: AL D

Final Review: _____ \$/2/5

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	Poscidon Nantilus brine (Frozen scauciter) 1507-5080 Start Date/Time: 7/2/15/16/7 End Date/Time: 7/2/15/16/57 Species: S. purpuratus
Tech initials: Injection Time:	Animal Source: Point Loma Date Collected: 7 9 15
Sperm Absorbance at 4	00 nm: 0.935 (target range of 0.8 - 1.0 for density of 4x10 ⁶ sperm/ml)
Eggs Counted:	$\frac{G7}{B2}$ Mean: $\frac{G1.6}{S1.6} \times 50 = \frac{4090}{S2}$ eggs/ml
	(target counts of 80 eggs per vertical pass on Sedgwick-Rafter slide for a final density of 4000 eggs/ml)
Initial density: Final density:	
	ck according to the calculated dilution factor. For example, if the dilution factor is 2.25, use 100 ml of existing of dilution water (1.25 parts).
Rangefinder Test: ml Sperm Stock ml Seawater	Sperm:Egg Ratio 2000:1 1600:1 1200:1 800:1 400:1 200:1 100:1 50:1 50 40 30 20 10 5.0 2.5 1.25 0.0 10 20 30 40 45 47.5 48.75
Sperm Added (100 µI): Eggs Added (0.5 mI): Test Ended:	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$
this range, choose the ra	n-to-egg ratio that results in fertilization between 80 and 90 percent. If more than one concentration is within atio closest to 90 percent unless professional judgment dictates consideration of other factors (e.g., organism ctive season, site conditions).
Definitive Test	Sperm:Egg Ratio Used: 350%
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time Fert. Unfert. 1617 QC1 1/1 7 1637 QC2 13 7 1657 Egg Control 1 0 1/0 6 Egg Control 2 0 0 0
Comments:	
QC Check:	AC 8(3)15 Final Review: 1 8/7/15

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

Sand Dollar Fertilization

Test Date: October 30, 2014

CETIS Summary Report

Report Date:

30 Jun-15 09:50 (p 1 of 1)

Test Code:

1410-S150 | 08-5460-9726

Echinoid Spe	rm Cell Fertiliza	ition Test 1	5C					*		Nautilu	s Environr	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	03-5643-7558 30 Oct-14 16:1 30 Oct-14 16:5 40m	0 Pro	tocol: ecies:	Fertilization EPA/600/R-95/ Dendraster exc Mission Bay	, ,			Analyst: Diluent: Brine: Age:	Natu	ural Seawat en Seawate		
Sample ID: Sample Date: Receive Date Sample Age:	: 30 Oct-14	Soi	terial: urce:	Brine Brined seawate Poseidon Www.hus R	-			Client: Project:	Pos	eidon		
Sample Note:	Frozen seawat	er prepared	at Nautil	us was used as	brine.							
Comparison :	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	М	ethod			
17-7819-5516	Fertilization Ra	te	38.5	>38.5	NA	3.75%		D	unnett M	ultiple Com	parison Te	st
Point Estimat	e Summary											
Analysis ID	Endpoint		Level	ppt	95% LCL	95% UCL	ΤU	М	ethod			
20-3895-2473	Fertilization Ra	te	EC25 EC50	>38.5 >38.5	N/A N/A	N/A N/A		Li	near Inte	erpolation (I	CPIN)	
Test Acceptal	oility											
Analysis ID	Endpoint		Attribu	ıte	Test Stat	TAC Limi	ts	0	verlap	Decision		
17-7819-5516	Fertilization Ra	te	Contro	l Resp	0.97	0.7 - NL		Ye	es	Passes A	cceptability	Criteria
20-3895-2473	Fertilization Ra	te	Contro	l Resp	0.97	0.7 - NL		Ye	es	Passes A	cceptability	Criteria
17-7819-5516	Fertilization Ra	te	PMSD		0.03751	NL - 0.25		N)	Passes A	cceptability	Criteria
Fertilization R	late Summary											
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	St	d Err	Std Dev	CV%	%Effect
0	Brine Control	5	0.962	0.9416	0.9824	0.94	0.98	0.	007348	0.01643	1.71%	0.0%
0	Lab Control	5	0.97	0.9485	0.9915	0.94	0.98		007746	0.01732	1.79%	-0.83%
35		5	0.968	0.9458	0.9902	0.95	0.99		800	0.01789	1.85%	-0.62%
35.5		5	0.952	0.9384	0.9656	0.94	0.96		004899	0.01096	1.15%	1.04%
36		5	0.948	0.9184	0.9776	0.91	0.97		01068	0.02387	2.52%	1.46%
36.5 37		5 5	0.96 0.964	0.9368	0.9832	0.94	0.98		008366	0.01871	1.95%	0.21%
37.5		5	0.984	0.9473 0.8943	0.9807 0.9777	0.95 0.88	0.98		006	0.01342 0.03362	1.39% 3.59%	-0.21% 2.7%
38		5	0.938	0.8836	0.9777	0.87	0.97		01503 0196	0.03362	4.67%	2.7%
38.5		5	0.956	0.9181	0.9939	0.92	0.99		01364	0.0305	3.19%	0.62%
Fertilization R	ate Detail											
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Brine Control	0.97	0.95	0.97	0.94	0.98						
0	Lab Control	0.97	0.98	0.94	0.98	0.98						
35		0.97	0.95	0.98	0.95	0.99						
35.5		0.96	0.94	0.94	0.96	0.96						
36		0.97	0.91	0.96	0.96	0.94						
36.5		0.94	0.97	0.94	0.98	0.97						
37		0.95	0.97	0.97	0.95	0.98						
37.5		0.94	0.97	0.88	0.95	0.94						
38		0.96	0.87	0.92	0.98	0.96						
38.5		0.96	0.92	0.93	0.99	0.98						

Echinoid Sperm Cell Fertilization Test 15C

Report Date: Test Code: 30 Jun-15 09:50 (p 1 of 2)

Test Code: 1410-S150 | 08-5460-9726

Nautilus Environmental (CA)

Analysis ID:17-7819-5516Endpoint:Fertilization RateCETIS Version:CETISv1.8.7

Analyzed: 30 Jun-15 9:49 Analysis: Parametric-Control vs Treatments Official Results: Yes

Sample Note: Frozen seawater prepared at Nautilus was used as brine.

Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corrected)	NA	C > T	NA	NA	3.75%	38.5	>38.5	NA	

Dunnett Multi	ple C	omparison Test							
Control	vs	C-ppt	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(a:5%)
Lab Control		35	0.1084	2.478	0.091	8	0.8617	CDF	Non-Significant Effect
		35.5	1.369	2.478	0.091	8	0.3353	CDF	Non-Significant Effect
		36	1.532	2.478	0.091	8	0.2700	CDF	Non-Significant Effect
		36.5	0.7489	2.478	0.091	8	0.6211	CDF	Non-Significant Effect
		37	0.5102	2.478	0.091	8	0.7249	CDF	Non-Significant Effect
		37.5	2.18	2.478	0.091	8	0.0915	CDF	Non-Significant Effect
		38	1.919	2.478	0.091	8	0.1473	CDF	Non-Significant Effect
		38.5	0.8193	2.478	0.091	8	0.5885	CDF	Non-Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)
Between	0.03236474	0.004045592	8	1.196	0.3289	Non-Significant Effect
Error	0.1217972	0.003383255	36			
Total	0.1541619		44			

Distributional Tes	sts				
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)
Variances	Bartlett Equality of Variance	7.847	20.09	0.4485	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.971	0.9308	0.3158	Normal Distribution

Fertilizatio	on Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.97	0.9485	0.9915	0.98	0.94	0.98	0.007746	1.79%	0.0%
35		5	0.968	0.9458	0.9902	0.97	0.95	0.99	0.008	1.85%	0.21%
35.5		5	0.952	0.9384	0.9656	0.96	0.94	0.96	0.004899	1.15%	1.86%
36		5	0.948	0.9184	0.9776	0.96	0.91	0.97	0.01068	2.52%	2.27%
36.5		5	0.96	0.9368	0.9832	0.97	0.94	0.98	0.008366	1.95%	1.03%
37		5	0.964	0.9473	0.9807	0.97	0.95	0.98	0.006	1.39%	0.62%
37.5		5	0.936	0.8943	0.9777	0.94	0.88	0.97	0.01503	3.59%	3.51%
38		5	0.938	0.8836	0.9924	0.96	0.87	0.98	0.0196	4.67%	3.3%
38.5		5	0.956	0.9181	0.9939	0.96	0.92	0.99	0.01364	3.19%	1.44%

Angular (C	orrected) Transfor	med Sumr	nary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.401	1.344	1.458	1.429	1.323	1.429	0.02048	3.27%	0.0%
35		5	1.397	1.33	1.465	1.397	1.345	1.471	0.02428	3.89%	0.28%
35.5		5	1.351	1.32	1.382	1.369	1.323	1.369	0.01129	1.87%	3.59%
36		5	1.345	1.281	1.409	1.369	1.266	1.397	0.02298	3.82%	4.02%
36.5		5	1.374	1.314	1.433	1.397	1.323	1.429	0.02143	3.49%	1.97%
37		5	1.383	1.337	1.428	1.397	1.345	1.429	0.01632	2.64%	1.34%
37.5		5	1.321	1.24	1.402	1.323	1.217	1.397	0.02927	4.95%	5.72%
38		5	1.331	1.221	1.441	1.369	1.202	1.429	0.03962	6.66%	5.04%
38.5		5	1.371	1.272	1.47	1.369	1.284	1.471	0.03568	5.82%	2.15%

000-089-170-2 CETIS™ v1.8.7.20

Report Date: Test Code: 30 Jun-15 09:50 (p 2 of 2) 1410-S150 | 08-5460-9726

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 17-7819-5516 Endpoint: Fertilization Rate **CETIS Version:** CETISv1.8.7 Analyzed: 30 Jun-15 9:49 Parametric-Control vs Treatments Analysis: Official Results: Yes Graphics 0.10 0.08 0.06 8.0 0.04 Fertilization Rate 0.6 -0.02 0.5 -0.04 0,4 -0.06 -0.08 0.3 -0.10 0.2 0.1 -0.14 -0.16 0 LC 35.5 37,5 38,5 -0.5 C-ppt Rankits

Report Date:

30 Jun-15 09:50 (p 1 of 1)

Test Code:

1410-S150 | 08-5460-9726

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: 20-3895-2473 Analyzed: 30 Jun-15 9:49 Endpoint: Fertilization Rate Analysis: Linear Interpolation (ICPIN) **CETIS Version:** Official Results:

CETISv1.8.7

Yes

Sample Note: Frozen seawater prepared at Nautilus was used as brine.

Linear Interpolation Options

X Transform Y Transform Seed Resamples Exp 95% CL Method

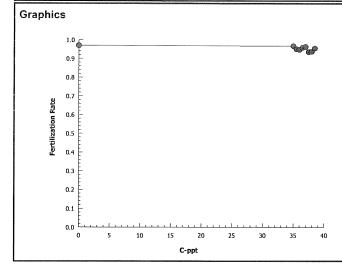
Linear Linear 1475078 1000 Yes Two-Point Interpolation

Point Estimates

Level ppt 95% LCL 95% UCL

EC25 >38.5 N/A N/A EC50 >38.5 N/A N/A

Fertilizati	ion Rate Summary	, , , , , , , , , , , , , , , , , , ,		Calculated Variate(A/B)							
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.97	0.94	0.98	0.007746	0.01732	1.79%	0.0%	485	500
35		5	0.968	0.95	0.99	0.008	0.01789	1.85%	0.21%	484	500
35.5		5	0.952	0.94	0.96	0.004899	0.01096	1.15%	1.86%	476	500
36		5	0.948	0.91	0.97	0.01068	0.02387	2.52%	2.27%	474	500
36.5		5	0.96	0.94	0.98	0.008366	0.01871	1.95%	1.03%	480	500
37		5	0.964	0.95	0.98	0.006	0.01342	1.39%	0.62%	482	500
37.5		5	0.936	0.88	0.97	0.01503	0.03362	3.59%	3.51%	468	500
38		5	0.938	0.87	0.98	0.0196	0.04382	4.67%	3.3%	469	500
38.5		5	0.956	0.92	0.99	0.01364	0.0305	3.19%	1.44%	478	500



CETIS™ v1.8.7.20

Report Date:

28 Oct-14 16:37 (p 1 of 2) 08-5460-9726/1410-S150

Test Code:

Report Date: Test Code: 28 Oct-14 16:37 (p 2 of 2) 08-5460-9726/1410-S150

C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
			48	100	87	11/14/14
			49	000	95	
			50	100	97	

Start Date:

Report Date: Test Code:

28 Oct-14 16:37 (p 1 of 2)

08-5460-9726/1410-S150

Nautilus Environmental (CA)

Echinoid Sperm Cell Fertilization Test 15C

30 Oct-14 30 Oct-14

Species: Dendraster excentricus Protocol: .EPA/600/R-95/136 (1995) Sample Code: Brine

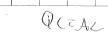
Start Date: End Date:		Oct-14 Oct-14				er excentricus /R-95/136 (1995)	Sample Code: Sample Source:	Brine
Sample Date					al: Brined se		Sample Station:	Poseidon
C-ppt	Code		Pos	# Counted	# Fertilized		Notes	
0	ВС	1	34		***************************************		****	
0	ВС	2	19				VIVIOUS MADE IN CO. L. L. L.	
0	ВС	3	33	100	94			
0	ВС	4	39					
0	ВС	5	24					
0	LC	1	50					
0	LC	2	17	100	96			
0	LC	3	18		, w			
0	LC	4	32				V/9/4	
0	LC	5	6					
35		1	44		THE PART OF THE PA			
35		2	25		V 20001 1		7000000	
35		3	43					
35		4	21					
35		5	12					
35.5		1	16					
35.5		2	10		77WA			
35.5		3	8					
35.5		4	7					
35.5		5	31	-				
36		1	26		T-MATERIAL INC.			
36		2	46					
36		3						
36		4	2 14					
36		5					WAA.	
36.5		1	30		THE WINGS.			
			23		TOWN TOWNS AND ADDRESS OF THE PARTY OF THE P			T-100-000-000-000-000-000-000-000-000-00
36.5		2	3					
36.5		3	42					
36.5		4	13					
36.5		5	27				***	
37		1	49					
37		2	36					
37		3	40					
37		4	38					
37		5	35					
37.5		1	20					
37.5		2	9					
37.5		3	41			The same of the sa		
37.5		4	1					
37.5		5	4					
38		1	22					
38		2	48	100	91			
38		3	11					
38		4	45					
38		5	37		-			
38.5		1	28					
38.5		2	29	100	99		r received	

Report Date:

28 Oct-14 16:37 (p 2 of 2)

Test Code: 08-5460-9726/1410-S150

C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
38.5		3	47			
38.5		4	15			
38.5		5	5			



Water Quality Measurements

Client/Project :

Poseidon/Salinity Tolerance Study

Test Species: D. excentricus

Sample ID:

Brine (frozen seawater)

Start Date/Time: 10/30/2014 \6:10

Sample Log No.:

End Date/Time: 10/30/2014 (6:50)

Dilutions made by: ___AC

Test No: 1410-S150

			Analyst:	5
			eadings	
Concentration (ppt)	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	8.7	8,04	33.3	15.6
Brine Control	8.8	8.06	33,7	15.4
35.0	6.8	8.07	34,9	14.8
35.5	E. B	8,07	35.5	15.0
36.0	8.7	9, 13	36.0	14.7
36.5	8.8	8.10	36.5	14.7
37.0	8-4	8.09	37,0	14.6
37.5	8.8	8.08	37.5	14.9
38.0	8.7	8.06	38.0	15.4
38.5	0.0	8.05	38.5	15.6

Comments:	_	
QC Check:	KBN/14/14	Final Review: 17 11 18 14

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

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	Position/Salinity Tolerance Study Brine (Evozen Securator) 1410-5150 Start Date/Time: 10/30/2014 / 16:50 End Date/Time: 10/30/2014 / 16:50 Species: D. excentricus Animal Source: Mission Bay
Tech initials: Injection Time:	AC Date Collected: 10/24/14
Sperm Absorbance at 4	00 nm: 0,7 44 (target range of 0.8 - 1.0 for density of 4x10 ⁶ sperm/ml)
Eggs Counted:	Mean: $43.8 \times 50 = 2190 \text{ eggs/ml}$
	(target counts of 80 eggs per vertical pass on Sedgwick-Rafter slide for a final density of 4000 eggs/ml)
Initial density: Final density:	2 (90 eggs/ml) = 0.55 dilution factor egg stock ml 4000 eggs/ml - 1.0 part egg stock seawater ml 0.45 parts seawater 45 ml poured of per 100 = 64e
	sk according to the calculated dilution factor. For example, if the dilution factor is 2.25, use 100 ml of existing all of dilution water (1.25 parts).
Rangefinder Test:	<u>Sperm:Egg Ratio</u> 2000:1 1600:1 1200:1 800:1 400:1 200:1 50:1
ml Sperm Stock ml Seawater	50 40 30 20 10 5.0 3.75 2.5 1.25 0.0 10 20 30 40 45 46.25 47.5 48.75
Sperm Added (100 μl): Eggs Added (0.5 ml): Test Ended:	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$
this range, choose the r	n-to-egg ratio that results in fertilization between 80 and 90 percent. If more than one concentration is within atio closest to 90 percent unless professional judgment dictates consideration of other factors (e.g., organism ctive season, site conditions).
Definitive Test	Sperm:Egg Ratio Used: 50:1
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time Fert. Unfert. 16:30 QC1 98 2 16:30 QC2 97 3 16:50 Egg Control 1 0 100 Egg Control 2 0 100
Comments:	
QC Check:	KB 111419 Final Review: KFP 11 18 14

Sand Dollar Fertilization

Test Date: July 22, 2015

CETIS Summary Report

Report Date:

05 Aug-15 09:24 (p 1 of 1)

Test Code:

1507-S082 | 00-0483-7190

r		The system of the superior	A					Test Cod	· .	100	77-0002 00	J-0463-7 190
Echinoid Spe	erm Cell Fertiliza	ation Test	15C							Nautilu	s Environm	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	02-7259-2739 22 Jul-15 15:2 22 Jul-15 16:0 40m	0 P r 0 Տ ք	ecies: D	ertilization PA/600/R-95/ endraster exc lission Bay				Analyst: Diluent: Brine: Age:		ural Seawat zen Seawat		
Sample ID: Sample Date: Receive Date Sample Age:	: 22 Jul-15	Ma So	aterial: B ource: P	9 82FD50 is rined seawate oseidon Jautilus	er	,		Client: Project:	Pos	eidon		
Comparison	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Me	thod			
15-3708-0048	Fertilization Ra	ite	38.5	>38.5	NA	7.59%		Du	nnett M	lultiple Com	parison Tes	st
Point Estimat	te Summary	1										
Analysis ID	Endpoint		Level	ppt	95% LCL	95% UCL	TU	Me	thod			
18-8484-4421	Fertilization Ra	ite	EC25 EC50	>38.5 >38.5	N/A N/A	N/A N/A		Lin	ear Inte	erpolation (l	CPIN)	
Test Acceptal	bility											
Analysis ID	Endpoint		Attribute	9	Test Stat	TAC Limi	ts	Ov	erlap	Decision		
15-3708-0048	Fertilization Ra	ite	Control F	Resp	0.886	0.7 - NL		Ye	s	Passes A	cceptability	Criteria
18-8484-4421	Fertilization Ra		Control F	Resp	0.886	0.7 - NL		Ye	s		cceptability	
15-3708-0048	Fertilization Ra	ite	PMSD		0.07594	NL - 0.25	***************************************	No		Passes A	cceptability	Criteria
Fertilization R	Rate Summary											
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Sto	l Err	Std Dev	CV%	%Effect
33.4	Brine Control	5	0.904	0.8632	0.9448	0.88	0.94		147	0.03286	3.64%	0.0%
33.5	Lab Control	5	0.886	0.8481	0.9239	0.85	0.92		1364	0.0305	3.44%	1.99%
34.9		5	0.906	0.8713	0.9407	0.86	0.93		1249	0.02793	3.08%	-0.22%
35.4 35.9		5	0.91	0.834	0.986	0.85	0.98		2739	0.06124	6.73%	-0.66%
36.4		5 5	0.896	0.8533	0.9387	0.84	0.93		1536	0.03435	3.83%	0.89%
36.9		5 5	0.926 0.92	0.8913	0.9607	0.9	0.96		1249 06324	0.02793	3.02%	-2.43%
37.4		5 5	0.92	0.9024 0.9118	0.9376 0.9602	0.9 0.91	0.93 0.96		06324 08718	0.01414 0.01949	1.54% 2.08%	-1.77% -3.54%
37.9		5	0.886	0.8336	0.9802	0.82	0.90		1887	0.01949	4.76%	-3.54% 1.99%
38.5		5	0.912	0.8704	0.9536	0.82	0.95		1497	0.04219	3.67%	-0.89%
Fertilization R	tate Detail	XXXXIII										
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
33.4	Brine Control	0.88	0.88	0.94	0.88	0.94		***************************************				
33.5	Lab Control	0.92	0.85	0.91	0.86	0.89						
34.9		0.92	0.9	0.93	0.86	0.92						
35.4		0.85	0.86	0.89	0.97	0.98						
35.9		0.84	0.91	0.93	0.91	0.89						
36.4		0.9	0.9	0.92	0.96	0.95						
36.9		0.93	0.93	0.9	0.93	0.91						
37.4		0.91	0.93	0.95	0.96	0.93						
37.9		0.87	0.92	0.82	0.92	0.9						
38.5		0.95	0.87	0.91	0.94	0.89						
		-	•									

@ any sw 8/7/15

Analyst: AC QA: JW 8/7/15

000-089-170-2

Report Date: Test Code: 05 Aug-15 09:23 (p 1 of 2)

•	•	.~9		00.2		(P	•	٠.	_
15	0	7-S	082	2 00)-0)48	3	-71	190

					·		Test	Code:	150	7-S082 C	0-0483-7190
Echinoid Sperm Cell Fertilization Test 15C									Nautilus	Environ	mental (CA)
Analysis ID: Analyzed:	15-3708-0048 05 Aug-15 9:2		•	tilization Ra rametric-Co	te ntrol vs Trea	tments		IS Version cial Results		.8.7	
Data Transfe	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C > T	NA	NA		7.59%	38.5	>38.5	NA	
Dunnett Mul	Itiple Compariso	n Test									
Control	vs C-ppt		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(a:5%)		
33.5	34.9		-0.8331	2.478	0.098 8	0.9871	CDF	Non-Sign	ificant Effect		
33.5	35.4		-1.398	2.478	0.098 8	0.9981	CDF	Non-Sign	ificant Effect		
33.5	35.9		-0.4261	2.478	0.098 8	0.9591	CDF	Non-Sign	ificant Effect		
33.5	36.4		-1.794	2.478	0.098 8	0.9996	CDF	Non-Sign	ificant Effect		
33.5	36.9		-1.426	2.478	0.098 8	0.9982	CDF	Non-Sign	ificant Effect		
33.5	37.4		-2.251	2.478	0.098 8	0.9999	CDF	Non-Sign	ificant Effect		
33.5	37.9		-0.0428	2.478	0.098 8	0.8985	CDF	Non-Sign	ificant Effect		
33.5	38.5		-1.147	2.478	0.098 8	0.9953	CDF	Non-Sign	ificant Effect		
ANOVA Tabl	le										
Source	Sum Squ		Mean Squ	ıare	DF	F Stat	P-Value	Decision	(a:5%)		
Between	0.037433	24	0.0046791	155	8	1.205	0.3240	Non-Sign	ificant Effect		
Error	0.139834	4	0.0038842	29	36	_					
Total	0.177267	7			44						
Distribution	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Bartlett E	Equality	of Variance	10.92	20.09	0.2065	Equal Var	iances			
Distribution	Shapiro-	Wilk W	Normality	0.9805	0.9308	0.6393	Normal D	stribution			
Fertilization	Rate Summary										
C-ppt	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
33.5	Lab Control	5	0.886	0.8481	0.9239	0.89	0.85	0.92	0.01364	3.44%	0.0%
34.9		5	0.906	0.8713	0.9407	0.92	0.86	0.93	0.01249	3.08%	-2.26%
35.4		5	0.91	0.834	0.986	0.89	0.85	0.98	0.02739	6.73%	-2.71%
35.9		5	0.896	0.8533	0.9387	0.91	0.84	0.93	0.01536	3.83%	-1.13%
36.4		5	0.926	0.8913	0.9607	0.92	0.9	0.96	0.01249	3.02%	-4.52%
36.9		5	0.92	0.9024	0.9376	0.93	0.9	0.93	0.006324	1.54%	-3.84%
37.4		5	0.936	0.9118	0.9602	0.93	0.91	0.96	0.008718	2.08%	-5.64%
37.9		5	0.886	0.8336	0.9384	0.9	0.82	0.92	0.01887	4.76%	0.0%
38.5		5	0.912	0.8704	0.9536	0.91	0.87	0.95	0.01497	3.67%	-2.94%
Angular (Co	rected) Transfo	rmed Su	ımmary								
C-ppt	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
33.5	Lab Control	5	1.229	1.169	1.288	1.233	1.173	1.284	0.02154	3.92%	0.0%
34.9		5	1.261	1.205	1.318	1.284	1.187	1.303	0.0205	3.63%	-2.67%
35.4		5	1.284	1.134	1.433	1.233	1.173	1.429	0.05384	9.38%	-4.48%
35.9		5	1.245	1.178	1.313	1.266	1.159	1.303	0.02424	4.35%	-1.37%
		5	1.299	1.23	1.368	1.284	1.249	1.369	0.02482	4.27%	-5.76%
36.4											
		5	1.285	1.253	1.317	1.303	1.249	1.303	0.01146	1.99%	-4.57%
36.9				1.253 1.267		1.303 1.303	1.249 1.266	1.303 1.369	0.01146 0.01807	1.99% 3.07%	-4.57% -7.22%
36.4 36.9 37.4 37.9		5	1.285		1.317						

Analyst: AC QA: _w 8/7/15

Report Date: Test Code: 05 Aug-15 09:24 (p 2 of 2) 1507-S082 | 00-0483-7190

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 15-3708-0048 CETISv1.8.7 Endpoint: Fertilization Rate **CETIS Version:** Analyzed: Parametric-Control vs Treatments 05 Aug-15 9:23 Analysis: Official Results: Yes Graphics 1.0 0.16 0.14 0.12 0.8 0.10 Fertilization Rate 0.08 0.7 0.06 0,6 0.04 0,5 0.02 0.00 -0.02 0.3 -0.04 -0.06 0.2 -0,08 0.1 -0.10 0.0 -0.12 33.5 LC 34.9 35.4 37.4 38.5 -2,5 -2.0 -1.5 1.0 1.5 2.0 C-ppt Rankits

Report Date:

05 Aug-15 09:24 (p 1 of 1)

Test Code:

1507-S082 | 00-0483-7190

Echinoid Sper	m Cell Ferti	ilization Test 15C
----------------------	--------------	--------------------

Nautilus Environmental (CA)

Analysis ID: Analyzed:

18-8484-4421 05 Aug-15 9:23

Endpoint: Fertilization Rate Analysis:

Linear Interpolation (ICPIN)

CETIS Version: Official Results: Yes

CETISv1.8.7

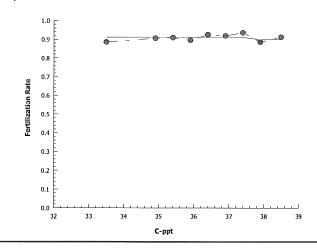
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	1008592	1000	Yes	Two-Point Interpolation

Point Estimates

Level	ppt	95% LCL	95% UC
EC25	>38.5	N/A	N/A
EC50	>29.5	NI/A	NI/A

Fertilizati	on Rate Summary		Calculated Variate(A/B)								
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
33.5	Lab Control	5	0.886	0.85	0.92	0.01364	0.0305	3.44%	0.0%	443	500
34.9		5	0.906	0.86	0.93	0.01249	0.02793	3.08%	-2.26%	453	500
35.4		5	0.91	0.85	0.98	0.02739	0.06124	6.73%	-2.71%	455	500
35.9		5	0.896	0.84	0.93	0.01536	0.03435	3.83%	-1.13%	448	500
36.4		5	0.926	0.9	0.96	0.01249	0.02793	3.02%	-4.52%	463	500
36.9		5	0.92	0.9	0.93	0.006324	0.01414	1.54%	-3.84%	460	500
37.4		5	0.936	0.91	0.96	0.008718	0.01949	2.08%	-5.64%	468	500
37.9		5	0.886	0.82	0.92	0.01887	0.04219	4.76%	0.0%	443	500
38.5		5	0.912	0.87	0.95	0.01497	0.03347	3.67%	-2.94%	456	500

Graphics



Report Date: Test Code: 21 Jul-15 17:15 (p 1 of 2) 00-0483-7190/49CF46

Report Date: 21 Jul-15 17:15 (p 2 of 2) Test Code: (507-506) 00-0483-7190/49CF46

C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
			48	100	91	AC 814/19
			49	100	86	1
			50	100	85	V

Report Date: 21 Jul-15 17:15 (p 1 of 2)
Test Code: 00-0483-7190/49CF46

Echinoid Sp	erm C	ell Fe	rtiliza	tion Test 15	С		Nautilus Environmental (CA)
Start Date: End Date: 7 Sample Date		Jul-15 Jul-15 Jul-15		Protoc	es: Dendrast col: EPA/600/ al: Brined se	R-95/136 (1995)	Sample Code: (A) 1507=3 N/H Sample Source: Poseidon Sample Station:
C-ppt	Code	Rep	Pos	# Counted	# Fertilized		Notes
0	ВС	1	45				
0	ВС	2	11	100	43	37/22/15	
0	ВС	3	40				
0	ВС	4	4				
0	ВС	5	19				
0	LC	1	15				
0	LC	2	44			w no w near the same accommodation of the	
0	LC	3	31	100	90	b 7/22/15	
0	LC	4	42	100	10	0 11 2011	
0	LC	5	22				
35	-	1	20	,			
35		2	3	100	86	57/22/15	
35		3	29	100	96	V 1/66/13	
35		4	49		,,		
35		5	36				
35.5		1	50				
35.5		2	25	100	77	87/22/15	
35.5		3	27	100		0 1700/15	
35.5		4	26				
35.5		5	37				
36		1	39	100	78	AC 7/22	
36		2	24	100	10	76 1166	
36		3	2				
36		4	8				
36		5	6		***************************************		
36.5		1	28	100	86	AC 7/22	
36.5		2	1	100	_00	AC TIFF	
36.5		3	14				
36.5		4	12				
36.5		5	38				
37		1	34				
37		2	23				
37		3	41				
37		4	10	100	92	AC 7/22	
37		5	17	1-0	10	1111	
37.5		1	48	100	an	AC 7/12	
37.5		2	35	100	10	7112	
37.5		3	30				
37.5		4	5				
37.5		5	7				
38	-	1	47				
38		2	33		· · · · · · · · · · · · · · · · · · ·		
38		3	18	100	81	47/V2/19	
38		4	13	1 UU	0 (<i>y y</i> (°)	17.44.494
38		5	21				
38.5		1	9	1122	F= 1	10 7120	
			43	100	gy	AC 7/22	
38.5		2	43				

Report Date: 21 Jul-15 17:15 (p 2 of 2)
Test Code: 1567-566 2.00-0483-7190/49CF46

,						
C-ppt	Code	Rep	Pos	# Counted	# Fertilized	Notes
38.5		3	16			
38.5		4	46			
38.5		5	32			



Water Quality Measurements

Client:

Poseidon

Test Species: D. excentricus

Sample ID:

Nautilus brine (frozen seawater)

Start Date/Time: 7/21/2015 1 22/15 1 520

Sample Log No.: 15- N A

End Date/Time: 7/21/2015 7(22/15 1600

Dilutions made by: __AC______

Test No: 1507-S082-

			Analyst:	Ab
Concentration	62		eadings	
ppt	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	80	8.09	33.5	16.0
Brine Control	8.4	8-10	334	16.0
35.0	8.6	8.09	34.9	16,0
35.5	8.7	9.09	354	16.0
36.0	8.8	8.09	35.9	15.8
36.5	8.8	8.09	36.4	15.9
37.0	8-8	8-09	36.9	15.9
37.5	8.8	9.09	37.4	15-8
38.0	8.8	8.09	37.9	15.6
38.5	8.8	8.09	385	15.6

Comments:	Huch Sension's	Salinity meter	
	,	J	
QC Check:	AC 8/5/15	Final Review: 8/1/15	

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

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	Poseidon Naufilus bri 1507-5087		Rawater)	End Date/Ti Spec	me: 7/22/15 me: 7/22/15 iles: D. excentri	cus
Tech initials: Injection Time:	1430				rce: <u>Mission Bar</u> ted: <u>7/17/2015</u>	У
Sperm Absorbance at 40	00 nm: <u> </u>	(target range of ().8 - 1.0 for dens	ity of 4x10 ⁶ sperm/r	nl)	
Eggs Counted:		n: <u>(03.8</u> x 5	0 = 39	Oeggs/ml		
		t counts of 80 eggs p or a final density of 4		Sedgwick-Rafter		
Initial density: Final density:	4000 eggs/ml eggs/ml	- 1.0 part	ion factor egg stock s seawater	egg stock seawater no dilw	ml ml	
	ck according to the calcula of dilution water (1.25 pa		For example, if t	he dilution factor is	2.25, use 100 m	nl of existing
			Sperm:Egg R	<u>atio</u>		
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1160050400.010	1200:1 30 20	800:1 40 20	0:1 200:1 10 5.0 10 45	100:1 2.5 47.5	50:1 1.25 48.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 11/45 1500	Rangefinder Rati	o: <u>Fert.</u> <u>810</u> <u>914</u>	Unfert.		
this range, choose the ra	n-to-egg ratio that results in atio closest to 90 percent u ctive season, site condition	unless professional				
<u>Definitive Test</u>		Sperm:Egg Ratio	Used: 100)		
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1520 1540 1600	QC1 QC2 Egg Control 1 Egg Control 2	Fert. 917 87	Unfert.		
Comments:				2.40		
QC Check:	AC8/5/15	_		Final Rev	iew:	5/7/15

Giant Kelp 48-hour Germination and Growth

Test Date: May 12, 2015

CETIS Summary Report

Report Date:

28 May-15 11:45 (p 1 of 2)

Test Code:	1505-S092	07-6525-43
	CONTROL SECTION AND ASSESSMENT OF THE PARTY	

								Test Code:	150	5-S092 0	7-6525-431
Macrocystis (Germination an	d Germ	Tube Grow	th Test					Nautilu	s Environr	nental (CA
Batch ID:	00-8657-2759			Growth-Germin				Analyst:			
Start Date:	12 May-15 15:		Protocol:	EPA/600/R-95/	·						
Ending Date:	14 May-15 12:		Species:	Macrocystis py	rifera			Brine: Froz	en Seawate	er	
Duration:	45h		Source:	La Jolla Cove				Age:			
Sample ID:	18-1583-3954		Code:	Nautilus Brine				Client: Pose	eidon		C
Sample Date:			Material:	Natural Seawa	ter			Project: Salir	nty lole	rance	Study
Receive Date:	•		Source:	Poseidon					`		,
Sample Age:	15h		Station:	Nautilus Brine							
Comparison S	Summary										
Analysis ID	Endpoint		NOEL		TOEL	PMSD	ΤU	Method			
01-5183-0033	Germination F	Rate	38.5	>38.5	NA	8.04%			ultiple Com		
12-8384-7212	Mean Length		38.5	>38.5	NA ————	10.2%		Dunnett M	ultiple Com	parison Te	st
Test Acceptal	oility										
Analysis ID	Endpoint		Attrib	ute	Test Stat	THE RESERVE OF THE PERSON OF T	nits	Overlap	Decision		
01-5183-0033	Germination R	Rate		ol Resp	0.894	0.7 - NL		Yes		cceptability	
12-8384-7212	Mean Length			ol Resp	14.55	10 - NL		Yes		cceptability	
01-5183-0033	Germination R	Rate	PMSE		0.08043	NL - 0.2		No		cceptability	
12-8384-7212	Mean Length		PMSD)	0.1025	NL - 0.2		No	Passes A	cceptability	Criteria
Germination I	Rate Summary										
C-ppt	Control Type	Coun	t Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effec
0	Brine Control	5	0.922	0.8804	0.9636	0.87	0.96	0.01497	0.03347	3.63%	0.0%
0	Lab Control	5	0.894		0.9456	0.83	0.94	0.0186	0.04159	4.65%	3.04%
35		5	0.924	0.8893	0.9587	0.89	0.95	0.01249	0.02793	3.02%	-0.22%
35.5		5	0.912	0.8644	0.9596	0.86	0.96	0.01715	0.03834	4.2%	1.09%
36		5	0.91	0.8409	0.9791	0.84	0.96	0.0249	0.05568	6.12%	1.3%
36.5		5	0.942	0.9075	0.9765	0.91	0.97	0.01241	0.02775	2.95%	-2.17%
37 27.5		5	0.904	0.8783	0.9297	0.87	0.92	0.009274	0.02074	2.29%	1.95%
37.5		5	0.918	0.8941	0.9419	0.89	0.94	0.008602	0.01924	2.1%	0.43%
38 38.5		5 5	0.938 0.926	0.8974 0.8588	0.9786 0.9932	0.9	0.98	0.01463	0.03271	3.49%	-1.74%
			0.920	0.0000	0.9932	0.84	0.98	0.02421	0.05413	5.85%	-0.43%
Mean Length C-ppt	•	Count	• BA o = =	050/ 1.01	05% 1101	Min	N#	Ctal Eng	Std Day	CV0/	0/ E&c -
	Brine Control	5	t Mean 14.35	95% LCL 12.95	95% UCL 15.75	Min 13.5	Max 16.25		1.126	CV% 7.85%	%Effec 0.0%
	Lab Control	5	14.55	14.15	14.95	14.25	15.23	0.1458	0.326	7.85% 2.24%	-1.39%
35	Lab Control	5	15.25	13.4	17.1	14.25	17.7		1.49	9.77%	-6.27%
35.5		5	15.25	13.39	17.11	13.75	17.7		1.49	9.84%	-6.27%
36		5	15.25	15.01	16.69	15.75	16.7		0.6755	4.26%	-10.45%
36.5		5	16	15.24	16.76	15.25	16.75		0.6124	3.83%	-11.5%
37		5	16	14.82	17.18	14.75	17.2		0.952	5.95%	-11.5%
37.5		5	16.35	15.71	16.99	15.75	17.2	0.2318	0.5184	3.17%	-13.94%
38		5	15.75	15.09	16.41	15.75	16.25		0.5303	3.37%	-9.76%
30 E		5	15.75	13.03	16.66	10 5	10.20		4.430	7 420/	6.70/0

Analyst: BK QA: ACEDES

7.42%

-6.27%

CETIS™ v1.8.7.20

16.66

13.5

16.25

0.5062

1.132

38.5

5

15.25

13.84

Report Date:

28 May-15 11:45 (p 2 of 2)

Test Code: 1505-S092 | 07-6525-4311

							 1000 0002 07 0020 1011
Macrocys	tis Germination and	d Germ Tu	be Growth	Test			Nautilus Environmental (CA)
Germinati	ion Rate Detail						
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	0.92	0.96	0.87	0.94	0.92	
0	Lab Control	0.94	0.83	0.89	0.92	0.89	
35		0.95	0.93	0.89	0.95	0.9	
35.5		0.93	0.92	0.89	0.96	0.86	
36		0.96	0.95	0.86	0.94	0.84	
36.5		0.94	0.91	0.92	0.97	0.97	
37		0.92	0.9	0.92	0.87	0.91	
37.5		0.92	0.93	0.89	0.94	0.91	
38		0.91	0.98	0.95	0.95	0.9	
38.5		0.84	0.95	0.98	0.95	0.91	
Mean Len	gth Detail						
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	14.25	13.5	13.5	14.25	16.25	
0	Lab Control	14.25	14.75	15	14.5	14.25	
35		14	17.75	14.25	15.25	15	
35.5		15.5	13.75	14	15.5	17.5	
36		16.75	15	15.75	15.5	16.25	
36.5		15.75	15.25	15.75	16.75	16.5	
37		16.5	15.5	16	17.25	14.75	
37.5		16.75	17	15.75	16	16.25	
38		15	16.25	15.5	16.25	15.75	
38.5		15.75	16.25	13.5	14.75	16	

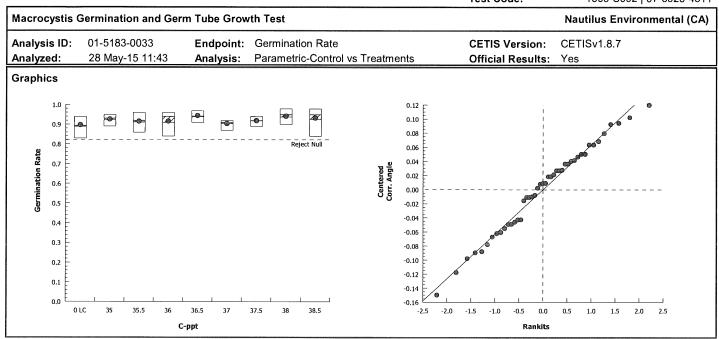
Report Date: Test Code: 28 May-15 11:44 (p 1 of 3) 1505-S092 | 07-6525-4311

							lest	Code:	1505	-5092 07	-0020-4311
Macrocystis	Germination ar	nd Germ	Tube Growth	Test					Nautilus	Environm	nental (CA)
Analysis ID: Analyzed:	01-5183-0033 28 May-15 11			Germination Ra Parametric-Con		ments		S Version: ial Results:	CE T ISv1. Yes	8.7	
Data Transfo		Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	ΤU
Angular (Corr		NA	C > T	NA	NA		8.04%	38.5	>38.5	NA	
Dunnett Mult	tiple Compariso	on Test		440111111111111111111111111111111111111							
Control	vs C-ppt		Test St	at Critical	MSD DF	P-Value	P-Type	Decision(α:5%)		
Lab Control	35		-1.174	2.478	0.108 8	0.9957	CDF	Non-Signif	icant Effect		
	35.5		-0.7222	2.478	0.108 8	0.9820	CDF	Non-Signit	icant Effect		
	36		-0.7592		0.108 8	0.9839	CDF	Non-Signif	ficant Effect		
	36.5		-2.05	2.478	0.108 8	0.9998	CDF	Non-Signif	ficant Effect		
	37		-0.3043		0.108 8	0.9443	CDF	Non-Signif	ficant Effect		
	37.5		-0.8718		0.108 8	0.9886	CDF	-	ficant Effect		
	38		-1.903	2.478	0.108 8	0.9997	CDF	-	ficant Effect		
	38.5		-1.492	2.478	0.108 8	0.9986	CDF	_	ficant Effect		
ANOVA Tabl	е										
Source	Sum Sq	uares	Mean S	Square	DF	F Stat	P-Value	Decision(a:5%)		
Between	0.03649	479	0.00456	61848	8	0.9543	0.4859	Non-Signi	ficant Effect		
Error	0.17208	55	0.00478	30153	36						
Total	0.20858	03			44						
Distribution	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(a:1%)			
Variances	Bartlett	Equality	of Variance	7.736	20.09	0.4597	Equal Va	riances			
Distribution	Shapiro	-Wilk W	Normality	0.983	0.9308	0.7425	Normal D	istribution			
Germination	Rate Summary	1									
C-ppt	Control Type	Coun	nt Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.894	0.8424	0.9456	0.89	0.83	0.94	0.0186	4.65%	0.0%
35		5	0.924	0.8893	0.9587	0.93	0.89	0.95	0.01249	3.02%	-3.36%
35.5		5	0.912	0.8644	0.9596	0.92	0.86	0.96	0.01715	4.2%	-2.01%
36		5	0.91	0.8409	0.9791	0.94	0.84	0.96	0.0249	6.12%	-1.79%
36.5		5	0.942	0.9075	0.9765	0.94	0.91	0.97	0.01241	2.95%	-5.37%
37		5	0.904	0.8783	0.9297	0.91	0.87	0.92	0.009273	2.29%	-1.12%
37.5		5	0.918	0.8941	0.9419	0.92	0.89	0.94	0.008602	2.1%	-2.69%
38		5	0.938	0.8974	0.9786	0.95	0.9	0.98	0.01463	3.49%	-4.92%
38.5		5	0.926	0.8588	0.9932	0.95	0.84	0.98	0.02421	5.85%	-3.58%
Angular (Co	rrected) Transf	ormed S	ummary								
C-ppt	Control Type	Cour	nt Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.244	1.161	1.327	1.233	1.146	1.323	0.02983	5.36%	0.0%
35		5	1.295	1.23	1.361	1.303	1.233	1.345	0.02357	4.07%	-4.13%
		5	1.275	1.189	1.361	1.284	1.187	1.369	0.03104	5.44%	-2.54%
35.5			4 077	1.157	1.397	1.323	1.159	1.369	0.04316	7.56%	-2.67%
35.5 36		5	1.277	1.107							
36		5 5	1.277	1.257	1.41	1.323	1.266	1.397	0.02746	4.61%	-7.21%
						1.323 1.266	1.266 1.202	1.397 1.284	0.02746 0.01523	2.71%	-1.07%
36 36.5 37		5	1.333	1.257	1.41			1.284 1.323	0.01523 0.01555	2.71% 2.71%	-1.07% -3.07%
36 36.5		5 5	1.333 1.257	1.257 1.215	1.41 1.299	1.266	1.202	1.284	0.01523	2.71%	-1.07%

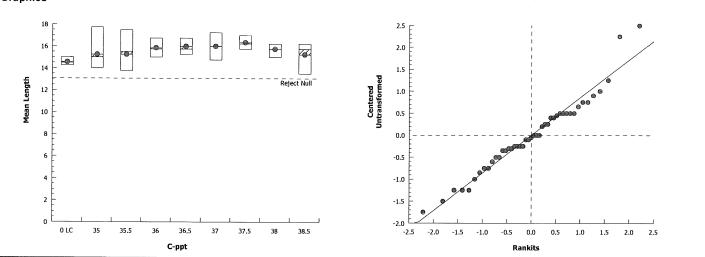
Analyst: 84 QA: AC5 | 28 15

CETIS™ v1.8.7.20

Report Date: Test Code: 28 May-15 11:44 (p 2 of 3) 1505-S092 | 07-6525-4311



CETIS An	alytical Rep		-	ort Date: Code:		28 May-15 11:44 (p 3 of 3) 1505-S092 07-6525-4311						
Macrocystis	Germination ar	d Germ	Tube Growth T	est		VA.						mental (CA)
Analysis ID: Analyzed:	12-8384-7212 28 May-15 11		Endpoint: Me Analysis: Par	an Length ametric-Cor	ntrol vs 1	rea	tments		IS Version		1.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed			PMSD	NOEL	LOEL	TOEL	TU
Untransforme	ed	NA	C > T	NA	NA			10.2%	38.5	>38.5	NA	
Dunnett Mult	tiple Compariso	n Test										
Control	vs C-ppt		Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision	(α:5%)		
Lab Control	35		-1.164	2.478	1.491	8	0.9956	CDF		ificant Effec	t	
	35.5		-1.164	2.478	1.491	8	0.9956	CDF	Non-Sign	ificant Effec	t	
	36		-2.161	2.478	1.491	8	0.9999	CDF	_	ificant Effec		
	36.5		-2.41	2.478	1.491	8	1.0000	CDF	_	ificant Effec		
	37		-2.41	2.478	1.491	8	1.0000	CDF	_	ificant Effec		
	37.5		-2.992	2.478	1.491	8	1.0000	CDF	_	ificant Effec		
	38		-1.995	2.478	1.491	8	0.9998	CDF	_	ificant Effec		
	38.5		-1.164	2.478	1.491	8	0.9956	CDF		ificant Effec		
ANOVA Table	e											
Source	Sum Squ	ıares	Mean Squ	ıare	DF		F Stat	P-Value	Decision	(a:5%)		
Between	12.175 1.521875 8						1.682	0.1367	Non-Sign	ificant Effec	ţ	
Error	32.575		0.9048611		36							
Total	44.75				44							
Distributiona	l Tests											
Attribute	Test			Test Stat	Critica	ı	P-Value	Decision((a:1%)			
Variances	Bartlett B	Equality	of Variance	14.4	20.09		0.0719	Equal Var				
Distribution			Vormality	0.9623	0.9308		0.1491	Normal Di				
Vlean Length	Summary											
C-ppt	Control Type	Coun	t Mean	95% LCL	95% U	CL	Median	Min	Max	Std Err	CV%	%Effect
)	Lab Control	5	14.55	14.15	14.95		14.5	14.25	15	0.1458	2.24%	0.0%
35		5	15.25	13.4	17.1		15	14	17.75	0.6661	9.77%	-4.81%
35.5		5	15.25	13.39	17.11		15.5	13.75	17.5	0.6708	9.84%	-4.81%
36		5	15.85	15.01	16.69		15.75	15	16.75	0.3021	4.26%	-8.94%
86.5		5	16	15.24	16.76		15.75	15.25	16.75	0.2739	3.83%	-9.97%
7		5	16	14.82	17.18		16	14.75	17.25	0.4257	5.95%	-9.97%
7.5		5	16.35	15.71	16.99		16.25	15.75	17	0.2318	3.17%	-12.37%
88		5	15.75	15.09	16.41		15.75	15	16.25	0.2372	3.37%	-8.25%
38.5		5	15.25	13.84	16.66		15.75	13.5	16.25	0.5062	7.42%	-4.81%
Graphics												00000
18 _							2.5 _		F		8	
16							2.0		1		•	



Report Date:

28 May-15 11:45 (p 1 of 2)

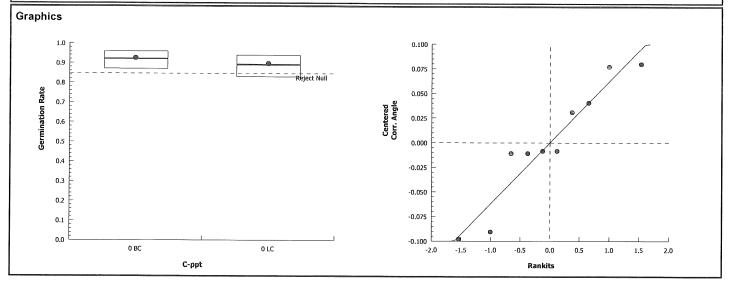
Test Code: 1505-S092 | 07-6525-4311

							rest	Code: 1505-5092 07-6525-431
Macrocystis (Germi	nation and Germ	Tube Growth	Test				Nautilus Environmental (CA
Analysis ID: Analyzed:		5064-8423 May-15 11:43	· ·	Germination F Parametric-Tv				IS Version: CETISv1.8.7
Data Transfor	m	Zeta	Alt Hyp	Trials	Seed		PMSD	Test Result
Angular (Corre	cted)	NA	C > T	NA	NA		5.32%	Passes germination rate
Equal Variand	e t T	wo-Sample Test						
Control	vs	Control	Test St	at Critical	MSD I	DF P-Value	P-Type	Decision(α:5%)
Lab Control		Brine Control	-1.202	1.86	0.076 8	8 0.8681	CDF	Non-Significant Effect
ANOVA Table								
Source		Sum Squares	Mean S	quare	DF	F Stat	P-Value	Decision(α:5%)
Between		0.005960524	0.00596	0524	1	1.445	0.2638	Non-Significant Effect
Error		0.0330071	0.00412	5887	8			-
Total		0.03896762			9			

Distributional	ests				
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)
Variances	Variance Ratio F	1.169	23.15	0.8832	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9076	0.7411	0.2647	Normal Distribution

Germination	Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.894	0.8424	0.9456	0.89	0.83	0.94	0.0186	4.65%	0.0%
0	Brine Control	5	0.922	0.8804	0.9636	0.92	0.87	0.96	0.01497	3.63%	-3.13%

Angular (Cor	rected) Transfor	med Summ	nary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.244	1.161	1.327	1.233	1.146	1.323	0.02983	5.36%	0.0%
0	Brine Control	5	1.293	1.216	1.369	1.284	1.202	1.369	0.02758	4.77%	-3.93%

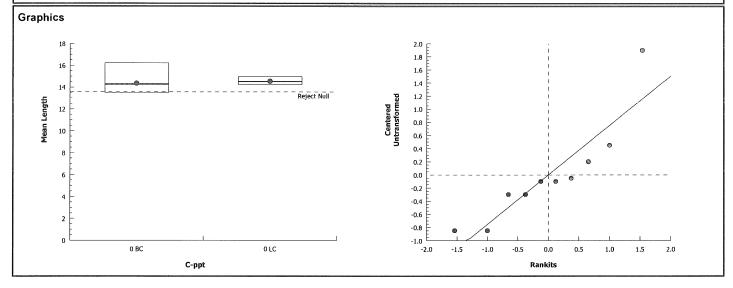


Report Date: Test Code: 28 May-15 11:45 (p 2 of 2) 1505-S092 | 07-6525-4311

Macrocystis (Germi	nation and Germ	Tube Grow	th T	est					Nautilus Environmental (CA
Analysis ID: Analyzed:		9063-4448 May-15 11:44	Endpoint: Analysis:		an Length ametric-Tw	o Sample			IS Version: cial Results:	CETISv1.8.7 Yes
Data Transfor	rm	Zeta	Alt H	lyp	Trials	Seed		PMSD	Test Resul	lt
Untransformed	d	NA	C > T		NA	NA		6.7%	Passes me	ean length
Equal Variand	ce t Tv	wo-Sample Test					Commence of the Commence of th			
Control	vs	Control	Test	Stat	Critical	MSD	DF P-Value	P-Type	Decision(a	r:5%)
Lab Control		Brine Control	0.381	4	1.86	0.975	8 0.3564	CDF	Non-Signifi	cant Effect
ANOVA Table)									
Source		Sum Squares	Mean	Squ	ıare	DF	F Stat	P-Value	Decision(a	r:5%)
Between		0.1	0.1			1	0.1455	0.7128	Non-Signifi	cant Effect
Error		5.5	0.687	5		8				
Total		5.6				9				

Distributional Te	ests				
Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F	11.94	23.15	0.0340	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.8352	0.7411	0.0386	Normal Distribution

Mean Le	ngth Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	14.55	14.15	14.95	14.5	14.25	15	0.1458	2.24%	0.0%
0	Brine Control	5	14.35	12.95	15.75	14.25	13.5	16.25	0.5037	7.85%	1.38%



Analyst: 812 QA: AC5/28/15

000-089-187-4 CETIS™ v1.8.7.20

Report Date:

23 Jun-15 11:24 (p 1 of 1)

Test Code:

1505-S092 | 07-6525-4311

Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental (CA)

Analysis ID:

00-0530-0673 23 Jun-15 11:23

Endpoint: Germination Rate

CETIS Version:

CETISv1.8.7

Analyzed:

Analysis:

Linear Interpolation (ICPIN)

Official Results:

Yes

ı	Linear	Interpolation	Options
1			

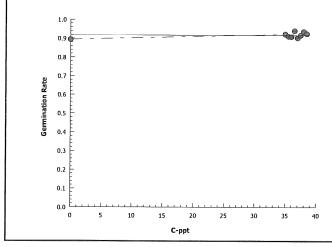
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	778442	1000	Yes	Two-Point Interpolation

Point Estimates

Level	ppt	95% LCL	95% UC
EC25	>38.5	N/A	N/A
EC50	>38.5	N/A	N/A

Germinat	tion Rate Summary										
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.894	0.83	0.94	0.0186	0.04159	4.65%	0.0%	447	500
35		5	0.924	0.89	0.95	0.01249	0.02793	3.02%	-3.36%	462	500
35.5		5	0.912	0.86	0.96	0.01715	0.03834	4.2%	-2.01%	456	500
36		5	0.91	0.84	0.96	0.0249	0.05568	6.12%	-1.79%	455	500
36.5		5	0.942	0.91	0.97	0.01241	0.02775	2.95%	-5.37%	471	500
37		5	0.904	0.87	0.92	0.009273	0.02074	2.29%	-1.12%	452	500
37.5		5	0.918	0.89	0.94	0.008602	0.01923	2.1%	-2.69%	459	500
38		5	0.938	0.9	0.98	0.01463	0.03271	3.49%	-4.92%	469	500
38.5		5	0.926	0.84	0.98	0.02421	0.05413	5.85%	-3.58%	463	500

Graphics



Report Date: Test Code: 23 Jun-15 11:24 (p 1 of 1) 1505-S092 | 07-6525-4311

Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental (CA)

Analysis ID: 04-6246-3634 **Analyzed:** 23 Jun-15 11:23

Endpoint: Mean Length
Analysis: Linear Interpolation (ICPIN)

CETIS Version: Official Results: CETISv1.8.7 Yes

Linear Interpolation Options

X Transform Y Transform Seed Resamples Exp 95% CL Method
Linear Linear 944683 1000 Yes Two-Point Interpolation

Point Estimates

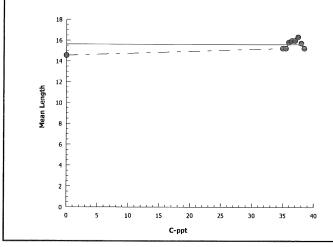
 Level
 ppt
 95% LCL
 95% UCL

 IC25
 >38.5
 N/A
 N/A

 IC50
 >38.5
 N/A
 N/A

Mean Lei	ngth Summary		Calculated Variate							
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	
0	Lab Control	5	14.55	14.25	15	0.1458	0.326	2.24%	0.0%	
35		5	15.25	14	17.75	0.6661	1.49	9.77%	-4.81%	
35.5		5	15.25	13.75	17.5	0.6708	1.5	9.84%	-4.81%	
36		5	15.85	15	16.75	0.3021	0.6755	4.26%	-8.94%	
36.5		5	16	15.25	16.75	0.2739	0.6124	3.83%	-9.97%	
37		5	16	14.75	17.25	0.4257	0.952	5.95%	-9.97%	
37.5		5	16.35	15.75	17	0.2318	0.5184	3.17%	-12.37%	
38		5	15.75	15	16.25	0.2372	0.5303	3.37%	-8.25%	
38.5		5	15.25	13.5	16.25	0.5062	1.132	7.42%	-4.81%	





Analyst: BKAC QA: AC 6000 S

000-089-187-5 CETIS™ v1.8.7.20

Nautilus Environmental - San Diego

Start Date: 12-May-15

Species: Macrocystis pyrifera

Test ID: 1505-S092

End Date: 14-May-15

Protocol: EPA/600/R-95/136 (1995 West Coast Manual)

Sample Source: Poseidon

Sampled: 12-May-15

Sample Station: Nautilus Brine

Random Number	Number Counted	Number Germinated		Tube Length Measurements (micrometer units)										Mean Tube Length (μm)
36	100	98	6	7	8	7	7	7	6	8	4	5	2.5	16.25
37	100	95	9	6	8	6	5	5	6	7	8	5	2.5	16.25
38	100	91	6	6	8	6	5	6	6	7	5	6	2.5	15.25
39	100	83	7	6	7	5	4	5	8	5	6	6	2.5	14.75
40	100	92	8	6	8	5	5	6	5	7	7	7	2.5	16.00
41	100	93	8	5	6	6	5	8	5	6	7	6	2.5	15.50
42	100	94	5	5	8	5	7	5	6	5	4	7	2.5	14.25
43	100	95	7	5	7	5	4	7	5	6	5	5	2.5	14.00
44	100	95	5	7	6	7	5	7	5	7	6	7	2.5	15.50
45	100	96	7	5	8	7	6	7	5	7	9	6	2.5	16.75
46	100	96	6	6	6	6	5	8	7	8	4	6	2.5	15.50
47	100	93	7	8	8	7	8	8	6	5	6	5	2.5	17.00
48	100	90	5	6	6	7	5	6	5	6	9	8	2.5	15.75
49	100	98	4	6	5	5	7	7	6	5	5	4	2.5	13.50
50	100	95	7	6	5	5	7	8	7	6	5	5	2.5	15.25
51	100	91	6	7	5	7	6	5	7	7	5	9	2.5	16.00
52	100	92	6	7	5	5	6	8	6	4	4	6	2.5	14.25
53	100	89	5	6	6	8	7	7	7	5	6	6	2.5	15.75
54	100	89	5	8	4	8	5	4	8	5	5	4	2.5	14.00
55	100	94	7	7	8	6	6	8	4	7	5	6	2.5	16.00
56	100	91	7	7	7	8	8	4	8	7	4	5	2.5	16.25
57	100	89	7	4	6	6	4	5	6	6	7	6	2.5	14.25
58	100	96	5	6	5	5	5	5	7	7	4	5	2.5	13.50
59	100	90	6	5	6	7	5	5	6	7	8	7	2.5	15.50
60	100	84	9	7	7	5	5	7	5	6	6	8	2.5	16.25
61	100	93	6	7	7	8	7	7	6	8	8	7	2.5	17.75
62	100	84	6	7	5	8	5	8	7	5	7	5	2.5	15.75
63	100	92	7	6	6	7	8	6	6	5	8	6	2.5	16.25
64	100	87	7	4	4	6	6	5	4	7	6	5	2.5	13.50
65	100	92	5	7	8	7	6	7	4	8	10	4	2.5	16.50
66	100	92	9	7	7	7	7	4	6	5	7	4	2.5	15.75
67	100	91	6	6	6	6	7	7	6	5	4	6	2.5	14.75
68	100	92	7	7	8	6	7	7	6	6	7	6	2.5	16.75
69	100	91	7	6	6	5	5	4	7	7	7	6	2.5	15.00
70	100	97	7	8	4	7	7	9	7	5	6	7	2.5	16.75

QC:	BK	5/28/15	
wo.		912011.2	

Nautilus Environmental - San Diego

Start Date: 12-May-15

Species: Macrocystis pyrifera

Test ID: 1505-S092

End Date: 14-May-15

Protocol: EPA/600/R-95/136 (1995 West Coast Manual)

Sample Source: Poseidon

Sampled: 12-May-15

Sample Station: Nautilus Brine

Random Number	Number Counted	Number Germinated		Tube Length Measurements (micrometer units)									Calibration	Mean Tube
71	100	86	6	8 7 7 9 7 9 6 6 5									Factor	Length (μm)
72	100	97	7	7	7	7	8	5	9 7	6	6	5	2.5	17.5
73	100	92	6	6	3	7	5	3		6	5	7	2.5	16.5
74	100	95	7	7	5	7	8	5	5	5	8	7	2.5	13.75
75	100	94	6	5	5	7	7	7	6 5		7	6	2.5	16.25
76	100	89	6	5	5	6	7	5		7	7	6	2.5	15.5
77	100	95	4	5	6	8	6	5	6	4	6	7	2.5	14.25
78	100	89	6	7	5	8	5	6	6	4	8	7	2.5	14.75
79	100	94	5	5	8	7	5	5	6	5	7	5	2.5	15
80	100	90	8	8	6	5	5	5	8	7	5	4	2.5	14.25
81	100	86	7	8	6	8	5	6	6	5 5	5	5	2.5	15
82	100	87	5	9	6	8	7	7	6	7	6	6	2.5	15.75
83	100	95	7	6	5	5	6	7	5		7	7	2.5	17.25
84	100	92	5	6	7	6	5	6	7	6	7	6	2.5	15
85	100	94		7	5	7	5	6	7	7	4	6	2.5	14.5
			·	,			<u> </u>	0		/	5	7	2.5	15.75
								 						
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QC:	BK	5	28	115

Nautilus Environmental - San Diego

Start Date: 12-May-15

Species: Macrocystis pyrifera

Test ID: 1505-5092

End Date: 14-May-15

Protocol: EPA/600/R-95/136 (1995 West Coast Manual)

Sample Source: Poseidon

Sampled: 12-May-15

Sample Station: Nauhlus Brine

				_								11		
Random	Number	Number		Tube Length Measurements (micrometer units)									Calibration	Mean Tube
Number	Counted	Germinated		· · · · · · · · · · · · · · · · · · ·									Factor	Length (μm)
36	100	98	0	7	4	7		7	0	8	4	5	A	#DIV/0!
37	100	95	9	9	,8	6	5	C-3	6	7	8	5		#DIV/0!
38	100	91	6	6	8	6	15	6	0	7	6	6		#DIV/0!
39	100	43		0	17	5	4	5	8	5	φ	6		#DIV/0!
40	100	92	4	6	8	65	5	6	5	7		7		#DIV/0!
41	100	93	8	5	6	U	5	8	5	6	7	6		#DIV/0!
42	100	94	5	5	8	5	-1	5	6	15.	4	7		#DIV/0!
43	100	95	-	5	7	5	4	17	5	0	5	5		#DIV/0!
44	100	95	5	7	(0.	17	5,	7	5	7	6	7		#DIV/0!
45	100	910		5	8	1	0	7.	5	7.	9	6		#DIV/0!
46	100	90	9	Q,	6	4	9	8,	7	8	4	6		#DIV/0!
47	100	93		8	8	1	8	8	0	5	6	5,		#DIV/0!
48	100	90,	5	0	6		5	6	5	0	9	8		#DIV/0!
49	100	98	4	6	5	5	7	7	6	5	5	14		#DIV/0!
50	100	45	7	6	5	5	1-7	8	7	6	5	5		#DIV/0!
51	100	91	(0	7	5	_7	6	5	7	7	5	19		#DIV/0!
52	100	92	6	7	5	5.	Q	8	6	14	4	6		#DIV/0!
53	100	89	5	6,	6	8	17	7	7	5	6	6		#DIV/0!
54	100	89	5	8	4	8	5	14	8	5	5	4		#DIV/0!
55	100	94	7	7	8	6	6.	8	1	17	5	6		#DIV/0!
56	100	91		7	7	18	9	4	8	1-7	4	5		#DIV/0!
57	100	89	20	4	le	10	4	5	6	6		6		#DIV/0!
58	100	96	5	0	5	5	600	5	7	17	4	5		#DIV/0!
59	100	40	Q_	5	0	17	5	5	6	7	8	7		#DIV/0!
60	100	84	9	17	1	5	5	17	5	6	6	8		#DIV/0!
61	100	93	0	1	7	4	17	17	6	8	8	7		#DIV/0!
62	100	84	0	1	5	8	5	8	. 7	5	17	5		#DIV/0!
63	100	92	7	6	6		4	10	6	5	8	6		#DIV/0!
64	100	87	1900	4	14,	0	6	15	4	17	4	5		#DIV/0!
65	100	92	5	1	8	17	6	1.7	4-	8	10	14		#DIV/0!
66	100	92	9		1	1	1	4	Q	5	7	14		#DIV/0!
67	100	91	0	6	6	6	7	7	0		4	6		#DIV/0!
68	100	92	7	17	8	6	7	1	6	6	•7	6		#DIV/0!
69	100	91	7	6	0	5	5	i-	1	7	7	6		#DIV/0!
70	100	97		8	1			9	7	5	6	7	2,5	#DIV/0!

00	Die	5/28/15	
QC:	BL	5128/15	

Nautilus Environmental - San Diego

Start Date: 12-May-15

lay-15 Species: Macrocystis pyrifera

Test ID: 1505 - 5092

End Date: 14-May-15

Protocol: EPA/600/R-95/136 (1995 West Coast Manual)

Sample Source: Poseidon

Sampled: 12-May-15

Sample Station: Nautilus Brive

Random Number	Number Counted	Number Germinated		Tube Length Measurements (micrometer units)								Calibration Factor	Mean Tube Length (μm)	
71 72 73 74 75	100 100 100 100 100	97 97 92 95 94	Si Si	8 1	71355	1	5	535	5	957	5,	5176		#DIV/0! #DIV/0! #DIV/0! #DIV/0!
76 77 78 79 80	100 100 100 100 100	49 95 49 94	1950 1950	55	5 6 5,	84	7 9 5	5 6 5	6	4 5 7	1 5	677		#DIV/0! #DIV/0! #DIV/0! #DIV/0!
81 82 83 84 85	100 100 100 100	\$1 81 95 92 94	757	89 49 49 7	9995	5 8 8 5 V	5 5 5	797	800	551091	5077774	59799	2.5	#DIV/0!

QC. 01- 7108 117	QC:	BK	5/28	15	
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Final Review: <u>AC 5/28/15</u>

Analyst FP

Marine Chronic Bioassay

Water Quality Measurements

1500

Client:

Poseidon

Test Species: Macrocystis pyrifera

Sample ID:

Nautilus brine (frozen seawater)

Start Date/Time: 5/6 2019

Sample Log No.: 45 Nautilus Brie

End Date/Time: 5/8/2015 5/14/15 1215

Test No.:

1505-5092

Dilutions made by: By

			Analyst:]		Analyst:	
	-1			8K	Final Re		,a.y o (.	ンい
Salinity	Initial R	eadings bH	Salinity	Temperature	leeggaageee			
(ppt)	(mg/L)	pn (units)	Salifility (ppt)	remperature (°C)	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	8.5	8,05	33,0	15.5	7.7	7.97	33.4	15.6
Brine Control	8.0	7.95	34.0	15.7	7.7	7.97	34.3	15:6
35.0	8.2	8,00	35,0	15,3	7.6	7.99	355	15.6
35.5	8,4	8.01	35,5	15.2	7.6	801	36.0	15.6
36.0	8.4	8,63	36.0	15.1	7.7	8.02	36.6	15.6
36.5	8,5	8,03	366	15,2	7.6	8.03	37.2	15.6
37.0	8,5	8,02	37.0	15.2	7.7	8.03	37,5	156
37.5	8.4	8,62	37.5	15.2	76	8.02	38.1	15.6
38.0	8.5	8.61	38.1	15,4	すぶ	8.03	38.6	15,6
38.5	8.5	8,00	38,5	15.5	7,6	8.04	39.1	1516

Comments: Hach Sension 5 salinity meter used.

BBK GIS 5/28/15

QC Check: BK \$128/18

Final Review: <u>AC 5 | 28 | 15 | </u>

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

CETIS Test Data Worksheet

Macrocystis Germination and Germ Tube Growth Test

Report Date: Test Code:

11 May-15 16:03 (p 1 of 2) Q -07-6525-4311/1505-5092

Nautilus Environmental (CA)

Sample Code: 4504 Noutilus Brive Sample Source: Poseidon Species: Macrocystis pyrifera 12 May-15 Start Date:

Protocol: EPA/600/R-95/136 (1995) End Date: 14 May-15

ample Date					al: Natural Se		Sample Station: Nautilus Brine
C-ppt	Code		Pos	# Counted	# Germinated	CalFactor	Notes
0	ВС	1	52	100		1	
0	ВС	2	58	100		1	11.44
0	ВС	3	64	100		1	
0	ВС	4	42	100		1	
0	ВС	5	63	100		1	
0	LC	1	79	100		 1	
0	LC	2	39	100		1	
0	LC	3	78	100		1	
0	LC	4	84	100		1	
0	LC	5	57	100		1	
35		1	43	100		1	
35		2	61	100		1	- Alexander - Alex
35	-	3	76	100		1	
35		4	50	100		 1	
35	-	5	80	100		1	
35.5		1	41	100		1	
		2	73	100		1	
35.5		3	54	100		1	
35.5			46	100		1	
35.5		4	1	100		1	
35.5		5	71			1	
36	-	1	45	100		1	
36		2	83	100		1	
36		3	81	100		 1	
36		4	75	100			
36		5	60	100		1 1	
36.5		1	85	100		1	
36.5		2	38	100		1	
36.5		3	66	100		1	
36.5		4	70	100		1	
36.5		5	72	100		1	
37		1	65	100		1	
37		2	59	100		1	
37		3	40	100		1	
37		4	82	100		1	
37		5	67	100		1	
37.5		1	68	100		1	
37.5		2	47	100		1	
37.5		3	53	100		1	
37.5		4	55	100		1	
37.5		5	56	100		1	
38		1	69	100		1	
38		2	36	100		1	
38		3	44	100		1	
38		4	37	100		1	
38		5	48	100		1	
38.5		1	62	100		1	
38.5		2	74	100		1	

Analyst: KB QA: AC5/28/15

CETIS Test Data Worksheet

Report Date:

11 May-15 16:03 (p 2 of 2)

Test Code:

@ 07-6525-4311/1505-509Z

C-ppt	Code	Rep	Pos	# Counted	# Germinated	Mean Length	CalFactor	Notes
38.5		3	49	100			1	
38.5		4	77	100			1	
38.5		5	51	100			1	

QC:BK

@ QN BK 5/28/15

Analyst: KB QA: AC 5/28/15

Marine Chronic Bioassay

Brine Dilution Worksheet

_	PA:	~	
_	roi	œι	aL.

Poseidon

Analyst: BK

Sample ID:

frozen seawater

Test Date: 5/12/2015

Test No:

1505 - 5092

Test Type: M. pyrifera

Salinity of Seawater

33.1

Salinity of Brine

89.7

Date of Brine used: 5/8/2015

Test Dilution Volume

250

Alkalinity of Brine Control: \(\langle \text{\gamma}\) mg/L as CaCO3

TS = target salinity

SE = salinity of effluent

SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
34.0	100.0	250	NA	NA	250
35.0	96.6	241.6	0.03	8.4	250
35.5	95.8	239.4	0.04	10.6	250
36.0	94.9	237.2	0.05	12.8	250
36.5	94.0	235.0	0.06	15.0	250
37.0	93.1	232.8	0.07	17.2	250
37.5	92.2	230.6	0.08	19.4	250
38.0	91.3	228.4	0.09	21.6	250
38.5	90.5	226.1	0.11	23.9	250

DI Volume

	Di volunie			
Brine Control	40.8	0.58	23.9	250

Total Brine Volume Required (ml):

152.8

QC Check: 8K 5/28/15

Final Review: <u>AC 5/28/15</u>

Marine Chronic Bioass	ay	K	Celp Spore Germina	tion & Growth Worksheet
Client: Poseidon			Start Date/Time: 5	/12/2015 / <i>\Sc</i> o
Test No.: 1505 - 3				14/2015 / 12/5
Tech. Initials: BK	,		Test Species:	· · · · · · · · · · · · · · · · · · ·
rech. miliais.			rest opedes.	Macrocystis pyrilera
Date Collected:	5/12/15			
	N-			
Kelp Collectors:	La Jolla Cove			
Collection Location:	La sella corre			
The cold Wel Discher and D	5/	1415 0930	•	
Time of Initial Rinsing and De	~1	1415 0450		
Time of Rinsing and Transfe		_/·····		
Conditions of Zoospore Dens	sity and Motility:	gor density and	poor motility	
Time of Blade Removal Fron	1 Release Beaker:	415 1456	·	
Density Counts (target = 90):	58 64 67	<u>56</u> <u>70</u>	Mean: <u>63</u>	
Maan 63 * 10,000 -	530,000 spores/ml (density	v of spore rolesso)		
Weari 10,000 = _	spores/fill (defisit)	y of spore release)		
If spore release > 900,000 s To calculate the dilution factor	spores/ml: Calculate a dilution factor or: Nひ Piluhibへ Necesse	·	tock of 900,000 spores/n	nl and inoculate with 0.25 ml.
Density of spore release		· spor	res = - (x)	dil.factor
, ,	* <u>0.25 ml</u> = 1 container	225,000 spor		- 1.0 part spore stock
				part(s) seawater
				pan(s) seawater
	·	it may sometimes be nec		
Time of inoculation:1500	Amount ino	culated: <u>05m\</u>		
Location in Environmental do not split up tests among s				
Shelf number	Measured Light Intensity Range (must be between 160 and 240 ft-		24-hour ge	ermination check
1	175 - 231	86-115	QC dish #	
2	184-226	116-150	3	92
3	180 - 237	36-85		
(A) \(\frac{1}{2} \)	178 - 228	151-185		
5				
6				
Timers Checked? '\	Should be on 16:8 light:dark cycle	e initials: BK		

Comments:

QC Check: 1 5/25/15

Pacific Topsmelt 7-day Survival and Growth

Test Date: May 5, 2015

CETIS Summary Report

Report Date: Test Code: 26 Jun-15 09:20 (p 1 of 2) 1505-S091 | 11-2970-9154

Pacific Toner	THE SHOW OF THE STREET STREET,					******************************		Test Code:			
Tacine Topsi	melt 7-d Surviva	l and G	rowth Test						Nauti	lus Environn	nental (CA
Batch ID: Start Date: Ending Date: Duration:	05-8756-2032 05 May-15 13:5 12 May-15 11:0 6d 22h		Test Type: Protocol: Species: Source:	Growth-Surviva EPA/600/R-95/ Atherinops affir Aquatic Biosyst	136 (1995) nis			Brine:	Natural Seaw Frozen Seaw 15 d		
Sample ID: Sample Date: Receive Date Sample Age:	: 05 May-15		Code: Material: Source: Station:	Nautilus Brine Natural Seawat Poseidon Nautilus Brine	er			Client: Project:	Poseidon		
Comparison :											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Meth	od		
06-8392-5997	7d Survival Rat	te	38.5	>38.5	NA	18.6%		Steel	Many-One Ra	ank Sum Test	
08-3759-0266	Mean Dry Biom	nass-mo	38.5	>38.5	NA	19.6%		Dunne	ett Multiple Co	omparison Te	st
Point Estimat	te Summary										
Analysis ID	Endpoint	VINDY III VII III III	Level	ppt	95% LCL	95% UCL	TU	Metho	od		
09-2555-2009	7d Survival Rat	te	EC25 EC50	>38.5 >38.5	N/A N/A	N/A N/A		Linea	r Interpolation	(ICPIN)	
10-9737-7367	Mean Dry Biom	nass-mo	······	>38.5	N/A	N/A		Linea	r Interpolation	(ICPIN)	
	•		IC50	>38.5	N/A	N/A			•	· ,	
Test Acceptal	bility	***************************************									
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Overl	ap Decisio	on	
06-8392-5997		te		ol Resp	0.88	0.8 - NL		Yes		Acceptability	
09-2555-2009				ol Resp	0.88	0.8 - NL		Yes		Acceptability	
08-3759-0266	•	_		ol Resp	1.161	0.85 - NL		Yes		Acceptability	
10-9737-7367 06-8392-5997	•	-		ol Resp	1.161 0.1858	0.85 - NL		Yes		Acceptability Acceptability	
08-3759-0266			PMSC PMSC		0.1856	NL - 0.25 NL - 0.5		No No		Acceptability Acceptability	
			,								
7d Survival R	-		, , , , , , ,				James Park				
7d Survival R C-ppt	-	Coun		95% LCL	95% UCL	Min	Max	Std E	rr Std De	v CV%	%Effect
	ate Summary				95% UCL	Min 0.8	Max 1	Std E	rr Std De		%Effec 0.0%
C-ppt 0 0	ate Summary Control Type	Coun	t Mean	95% LCL						9.32% 20.33%	0.0% 8.33%
C-ppt 0 0 35	ate Summary Control Type Brine Control	Coun 5 5 5	t Mean 0.96	95% LCL 0.8489	1	0.8	1	0.04 0.08 0	0.0894	9.32% 20.33% 0.0%	0.0% 8.33% -4.17%
C-ppt 0 0 35 35.5	ate Summary Control Type Brine Control	Coun 5 5 5 5	t Mean 0.96 0.88 1	95% LCL 0.8489 0.6579 1	1 1 1 1	0.8 0.6 1	1 1 1	0.04 0.08 0	0.0894 0.1789 0	9.32% 20.33% 0.0% 0.0%	0.0% 8.33% -4.17% -4.17%
C-ppt 0 0 35 35.5 36	ate Summary Control Type Brine Control	Coun 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96	95% LCL 0.8489 0.6579 1 1 0.8489	1 1 1 1	0.8 0.6 1 1 0.8	1 1 1 1	0.04 0.08 0 0 0.04	0.0894 0.1789 0 0 0 0.0894	9.32% 20.33% 0.0% 0.0% 4 9.32%	0.0% 8.33% -4.17% -4.17% 0.0%
C-ppt 0 0 35 35.5 36 36.5	ate Summary Control Type Brine Control	Coun 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1	95% LCL 0.8489 0.6579 1 1 0.8489	1 1 1 1 1	0.8 0.6 1 1 0.8	1 1 1 1 1	0.04 0.08 0 0 0.04	0.0894- 0.1789 0 0 0.0894-	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0%	0.0% 8.33% -4.17% -4.17% 0.0% -4.17%
C-ppt 0 0 35 35.5 36 36.5 37	ate Summary Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5	0.96 0.88 1 1 0.96 1 0.92	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979	1 1 1 1 1 1	0.8 0.6 1 1 0.8 1	1 1 1 1 1 1	0.04 0.08 0 0 0.04 0	0.0894- 0.1789 0 0 0.0894- 0 0.1789	4 9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44%	0.0% 8.33% -4.17% -4.17% 0.0% -4.17%
C-ppt 0 0 35 35.5 36 36.5 37 37.5	ate Summary Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784	1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8	1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095	4 9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91%	0.0% 8.33% -4.17% -4.17% 0.0% -4.17% 4.17%
C-ppt	ate Summary Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5	0.96 0.88 1 1 0.96 1 0.92	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979	1 1 1 1 1 1	0.8 0.6 1 1 0.8 1	1 1 1 1 1 1	0.04 0.08 0 0 0.04 0	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894-	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91%	0.0% 8.33% -4.17% -4.17% 0.0% -4.17%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5	ate Summary Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.96	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489	1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8	1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894-	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32%	0.0% 8.33% -4.17% -4.17% 0.0% -4.17% 4.17% 0.0%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio	ate Summary Control Type Brine Control Lab Control	Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.96 0.92	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489	1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8	1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.04	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91%	0.0% 8.33% -4.17% -4.17% 0.0% -4.17% 4.17% 0.0%
C-ppt 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio	ate Summary Control Type Brine Control Lab Control mass-mg Summ Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 7 7 7 8 8 8 8 8 8 8 8 8	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.92 0.92 t Mean 1.078	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.04	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV%	0.0% 8.33% -4.17% -4.17% 0.0% -4.17% 4.17% 0.0% 4.17% %Effec 0.0%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio C-ppt 0 0	ate Summary Control Type Brine Control Lab Control	Coun 5 5 5 5 5 5 5 5 Coun Coun 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.92 0.92 t Mean 1.078 1.161	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784 95% LCL 0.9036 1.031	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8 0.8 0.8	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.048 Std E 2 0.062 4	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De 66 0.1401 69 0.1044	4 9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV% 13.0% 9.0%	0.0% 8.33% -4.17% 0.0% -4.17% 4.17% 0.0% 4.17% 0.0% 4.17%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio C-ppt 0 0 35	ate Summary Control Type Brine Control Lab Control mass-mg Summ Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 Coun 6 Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.92 0.92 t Mean 1.078 1.161 1.041	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784 95% LCL 0.9036 1.031 0.9807	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8 0.8 0.8 0.922 1.066 0.962	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.04	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De 66 0.1401 69 0.1044 65 0.0484	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV% 13.0% 9.0% 1 4.65%	0.0% 8.33% -4.17% 0.0% -4.17% 4.17% 4.17% 0.0% 4.17% %Effec 0.0% -7.72% 3.42%
C-ppt 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio C-ppt 0 0 35 35.5	ate Summary Control Type Brine Control Lab Control mass-mg Summ Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.92 0.96 1.078 1.161 1.041 1.106	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784 95% LCL 0.9036 1.031 0.9807 1.049	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8 0.8 0.8 0.922 1.066 0.962 1.064	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.048 Std E 2 0.062 4 0.021 2 0.020	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De 66 0.1401 69 0.1044 65 0.0484- 43 0.04566	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV% 13.0% 9.0% 1 4.65% 8 4.13%	0.0% 8.33% -4.17% 0.0% -4.17% 4.17% 4.17% 0.0% 4.17% %Effect 0.0% -7.72% 3.42% -2.6%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio C-ppt 0 0 35 35.5	ate Summary Control Type Brine Control Lab Control mass-mg Summ Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.92 0.96 1.078 1.161 1.041 1.106 1.139	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784 95% LCL 0.9036 1.031 0.9807 1.049 0.8754	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8 0.8 0.8 0.922 1.066 0.962 1.064 0.97	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.048 Std E 2 0.062 4 0.021 2 0.020 4 0.095	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De 66 0.1401 69 0.1044 65 0.0484- 43 0.04566 02 0.2125	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV% 13.0% 9.0% 1 4.65% 8 4.13% 18.65%	0.0% 8.33% -4.17% 0.0% -4.17% 4.17% 4.17% 0.0% 4.17% %Effec 0.0% -7.72% 3.42% -2.6% -5.72%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio C-ppt 0 0 35 35.5 36 36.5 36 36.5	ate Summary Control Type Brine Control Lab Control mass-mg Summ Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.96 0.92 t Mean 1.078 1.161 1.041 1.106 1.139 1.199	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784 95% LCL 0.9036 1.031 0.9807 1.049 0.8754 0.9086	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8 0.8 0.8 0.9 1.066 0.962 1.064 0.97 0.978	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.048 Std E 2 0.062 4 0.021 2 0.020 4 0.095 2 0.104	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De 66 0.1401 69 0.1044 65 0.0484 43 0.04566 02 0.2125 0.2338	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV% 13.0% 9.0% 1 4.65% 18.65% 19.5%	0.0% 8.33% -4.17% 0.0% -4.17% 4.17% 4.17% 0.0% 4.17% %Effec 0.0% -7.72% 3.42% -2.6% -11.25%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5 Mean Dry Bio C-ppt 0 0 35 35.5 36 36.5 37	ate Summary Control Type Brine Control Lab Control mass-mg Summ Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.96 0.92 t Mean 1.078 1.161 1.041 1.106 1.139 0.898	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784 95% LCL 0.9036 1.031 0.9807 1.049 0.8754 0.9086 0.7274	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8 0.8 0.9 1.066 0.962 1.064 0.97 0.978 0.756	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.048 Std E 2 0.062 4 0.021 2 0.020 4 0.095 2 0.104 6	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De 66 0.1401 69 0.1044 65 0.0484- 43 0.04566 02 0.2125 5 0.2338 45 0.1374	4 9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV% 13.0% 9.0% 1 4.65% 18.65% 19.5% 15.3%	0.0% 8.33% -4.17% 0.0% -4.17% 4.17% 4.17% 0.0% 4.17% %Effec 0.0% -7.72% 3.42% -2.6% -5.72% -11.25% 16.67%
C-ppt 0 0 35 35.5 36 36.5 37 37.5 38 38.5	ate Summary Control Type Brine Control Lab Control mass-mg Summ Control Type Brine Control	Coun 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	t Mean 0.96 0.88 1 1 0.96 1 0.92 0.92 0.96 0.92 t Mean 1.078 1.161 1.041 1.106 1.139 1.199	95% LCL 0.8489 0.6579 1 1 0.8489 1 0.6979 0.784 0.8489 0.784 95% LCL 0.9036 1.031 0.9807 1.049 0.8754 0.9086	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.8 0.6 1 1 0.8 1 0.6 0.8 0.8 0.8 0.8 0.8 0.9 1.066 0.962 1.064 0.97 0.978	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	0.04 0.08 0 0 0.04 0 0.08 0.048 0.04 0.048 2 0.062 4 0.021 2 0.020 4 0.095 2 0.104 0.061 4 0.055	0.0894- 0.1789 0 0 0.0894- 0 0.1789 99 0.1095 0.0894- 99 0.1095 rr Std De 66 0.1401 69 0.1044 65 0.0484- 43 0.04566 02 0.2125 5 0.2338 45 0.1374 57 0.1243	9.32% 20.33% 0.0% 0.0% 4 9.32% 0.0% 19.44% 11.91% 4 9.32% 11.91% v CV% 13.0% 9.0% 1 4.65% 18.65% 19.5%	0.0% 8.33% -4.17% 0.0% -4.17% 4.17% 4.17% 0.0% 4.17% %Effec 0.0% -7.72% 3.42%

Report Date: Test Code: 26 Jun-15 09:20 (p 2 of 2) 1505-S091 | 11-2970-9154

Pacific To	psmelt 7-d Surviva	l and Grov	vth Test				Nautilus Environmental (CA)
7d Surviv	al Rate Detail				Was Addressed		
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	1	0.8	1	1	1	
0	Lab Control	8.0	1	1	1	0.6	
35		1	1	1	1	1	
35.5		1	1	1	1	1	
36		8.0	1	1	1	1	
36.5		1	1	1	1	1	
37		1	1	0.6	1	1	
37.5		1	8.0	8.0	1	1	
38		1	1	8.0	1	1	
38.5		1	1	1	8.0	0.8	
Mean Dry	Biomass-mg Detail						
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Brine Control	1.17	0.922	1.262	1.064	0.97	
0	Lab Control	1.066	1.334	1.112	1.116	1.176	
35		1.068	0.962	1.026	1.074	1.074	
35.5		1.172	1.064	1.09	1.07	1.132	
36		1.012	1.504	1.118	0.97	1.092	
36.5		0.978	1.404	1.02	1.492	1.1	
37		1.066	0.774	0.756	1.006	0.888	
37.5		1.16	0.992	0.916	1.224	1.088	
38		1.238	0.926	1.036	1.274	1.02	
38.5		1.09	1.084	1.212	1.11	0.856	

Report Date: Test Code: 26 Jun-15 09:19 (p 1 of 4) 1505-S091 | 11-2970-9154

								Test	Code:	150	5-S091 11	1-2970-915
Pacific Topsi	melt 7-d Surviva	l and Gro	wth Test							Nautilu	s Environn	nental (CA
Analysis ID: Analyzed:	06-8392-5997 23 Jun-15 11:1		ndpoint: 7d :	Survival Rat		vs T	reatments		IS Version		.8.7	
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed			PMSD	NOEL	LOEL	TOEL	TU
Angular (Corre	ected)	NA	C > T	NA	NA			18.6%	38.5	>38.5	NA	
Steel Many-O	ne Rank Sum T	est										
Control	vs C-ppt		Test Stat	Critical	Ties	DF	P-Value	P-Type	Decision	ı(α:5%)		
Lab Control	35		32.5	16	1	8	0.9941	Asymp	Non-Sign	ificant Effect	•	
	35.5		32.5	16	1	8	0.9941	Asymp		ificant Effect		
	36		30.5	16	2	8	0.9771	Asymp	Non-Sign	ificant Effect		
	36.5		32.5	16	1	8	0.9941	Asymp	Non-Sign	ificant Effect		
	37		29.5	16	2	8	0.9588	Asymp	-	ificant Effect		
	37.5		28.5	16	2	8	0.9304	Asymp	-	ificant Effect		
	38		30.5	16	2	8	0.9771	Asymp	-	ificant Effect		
	38.5		28.5	16	2	8	0.9304	Asymp	_	ificant Effect		
ANOVA Table	e			2 3 30-40-40 34-50 32 4-50 32 4-50 32 5-50 5-50 5-50 5-50 5-50 5-50 5-50 5-5								
Source	Sum Squ	ares	Mean Squ	are	DF		F Stat	P-Value	Decision	(α:5%)		
Between	0.104312	2	0.0130390	2	8		0.8296	0.5825	Non-Sign	ificant Effect		
Error	0.565848	8	0.0157180	2	36							
Total	0.670161				44		MAILE .					
Distributiona	l Tests											
Attribute	Test			Test Stat	Critica	al	P-Value	Decision	(α:1%)			
Variances	Mod Lev	ene Equal	ity of Variance	0.8755	3.256		0.5489	Equal Var	iances			
Variances	Levene E	quality of	Variance	5.86	3.052		<0.0001	Unequal \	/ariances			
Distribution	Shapiro-	Wilk W No	ormality	0.8352	0.9308	3	<0.0001	Non-norm	al Distributi	on		
7d Survival R	Rate Summary											
C-ppt	Control Type	Count	Mean	95% LCL	95% L	JCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.88	0.6579	1		1	0.6	1	0.08	20.33%	0.0%
35		5	1	1	1		1	1	1	0	0.0%	-13.64%
35.5		5	1	1	1		1	1	1	0	0.0%	-13.64%
36		5	0.96	0.8489	1		1	0.8	1	0.04	9.32%	-9.09%
36.5		5	1	1	1		1	1	1	0	0.0%	-13.64%
37		5	0.92	0.6979	1		1	0.6	1	80.0	19.44%	-4.55%
37.5		5	0.92	0.784	1		1	0.8	1	0.04899	11.91%	-4.55%
38		5	0.96	0.8489	1		1	0.8	1	0.04	9.32%	-9.09%
38.5		5	0.92	0.784	1		1	0.8	1	0.04899	11.91%	-4.55%
Angular (Cor	rected) Transfor	med Sum	mary									
C-ppt	Control Type	Count	Mean	95% LCL	95% L	JCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.206	0.9496	1.462		1.345	0.8861	1.345	0.09228	17.11%	0.0%
35		5	1.345	1.345	1.346		1.345	1.345	1.345	0	0.0%	-11.57%
35.5		5	1.345	1.345	1.346		1.345	1.345	1.345	0	0.0%	-11.57%
										0.04700	0.240/	-7.62%
		5	1.298	1.165	1.43		1.345	1.107	1.345	0.04763	8.21%	-7.02/0
36		5 5						1.107		0.04763	0.0%	
36 36.5		5	1.345	1.345	1.346		1.345	1.345	1.345			-11.57%
36 36.5 37		5 5	1.345 1.253	1.345 0.9984	1.346 1.508		1.345 1.345	1.345 0.8861	1.345 1.345	0	0.0%	-11.57% -3.95%
36 36.5 37 37.5 38		5	1.345	1.345	1.346		1.345	1.345	1.345	0 0.09184	0.0% 16.38%	-11.57%

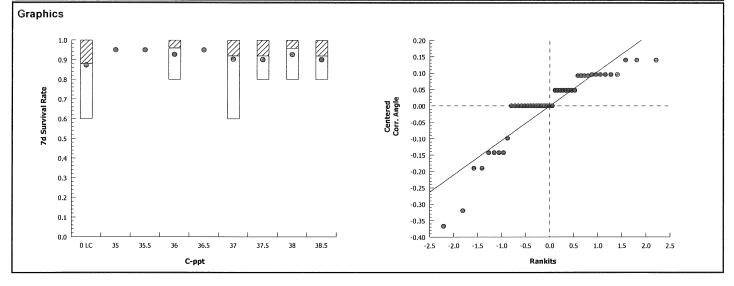
Report Date:

26 Jun-15 09:20 (p 2 of 4)

1505-S091 | 11-2970-9154 **Test Code:**

Pacific Topsi	melt 7-d Surviva	l and G	rowth Test					Nautilus Environmental (CA)
Analysis ID: Analyzed:	06-8392-5997 23 Jun-15 11:1		•		l Survival Rate onparametric-Control vs Treatments		CETIS Version: Official Results:	CETISv1.8.7 Yes
7d Survival R	Rate Detail							
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
0	Lab Control	0.8	1	1	1	0.6		
35		1	1	1	1	1		
35.5		1	1	1	1	1		
36		8.0	1	1	1	1		
36.5		1	1	1	1	1		
37		1	1	0.6	1	1		
37.5		1	8.0	8.0	1	1		
38		1	1	0.8	1	1		
38.5		1	1	1	0.8	0.8		

Angular (Co	Angular (Corrected) Transformed Detail										
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
0	Lab Control	1.107	1.345	1.345	1.345	0.8861					
35		1.345	1.345	1.345	1.345	1.345					
35.5		1.345	1.345	1.345	1.345	1.345					
36		1.107	1.345	1.345	1.345	1.345					
36.5		1.345	1.345	1.345	1.345	1.345					
37		1.345	1.345	0.8861	1.345	1.345					
37.5		1.345	1.107	1.107	1.345	1.345					
38		1.345	1.345	1.107	1.345	1.345					
38.5		1.345	1.345	1.345	1.107	1.107					



Report Date: Test Code:

26 Jun-15 09:20 (p 3 of 4) 1505-S091 | 11-2970-9154

							Test	Code:	150)5-S091 1	1-2970-9154
Pacific Tops	melt 7-d Surviva	al and G	rowth Test						Nautilu	s Environn	nental (CA)
Analysis ID: Analyzed:	08-3759-0266 23 Jun-15 11:			an Dry Biom ametric-Cor	_	tments		IS Version		.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Untransforme		NA	C > T	NA	NA		19.6%	38.5	>38.5	NA	
Dunnett Mul	tiple Compariso	n Test				Water Control of the					
Control	vs C-ppt		Test Stat	Critical	MSD DE	P-Value	P-Type	Decision	ı(a:5%)		
Lab Control	35		1.309	2.478	0.227 8	0.3608	CDF	Non-Sigr	ificant Effec	t	
	35.5		0.6023	2.478	0.227 8	0.6864	CDF	Non-Sigr	nificant Effec	t	
	36		0.2357	2.478	0.227 8	0.8244	CDF	Non-Sigr	ificant Effec	t	
	36.5		-0.4146	2.478	0.227 8	0.9579	CDF	Non-Sigr	ificant Effec	t	
	37*		2.867	2.478	0.227 8	0.0208	CDF	Significa	nt Effect		
	37.5		0.9253	2.478	0.227 8	0.5386	CDF	Non-Sigr	ificant Effec	t	
	38		0.6765	2.478	0.227 8	0.6539	CDF	Non-Sigr	ificant Effec	t	
	38.5		0.9864	2.478	0.227 8	0.5096	CDF	Non-Sigr	ificant Effec		
ANOVA Table	е										
Source	Sum Sqւ	ıares	Mean Squ	ıare	DF	F Stat	P-Value	Decision	ı(<i>a</i> :5%)		
Between	0.297022	3	0.0371277	'9	8	1.768	0.1162	Non-Sigr	ificant Effec		
Error	0.755974	2	0.0209992	28	36						
Total	1.052997	470,40	With the State of		44				***************************************		
Distributiona	al Tests			**************************************							
Attribute	Test			Test Stat	Critical	P-Value	Decision	(a:1%)			WWW.
Variances	Bartlett E	Equality of	of Variance	14.6	20.09	0.0673	Equal Var	iances			
Distribution	Shapiro-	Wilk W I	Normality ————————————————————————————————————	0.9685	0.9308	0.2559	Normal D	istribution			
Mean Dry Bio	omass-mg Sumr	пагу									
C-ppt	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.161	1.031	1.29	1.116	1.066	1.334	0.04669	9.0%	0.0%
35		5	1.041	0.9807	1.101	1.068	0.962	1.074	0.02165	4.65%	10.34%
35.5		5	1.106	1.049	1.162	1.09	1.064	1.172	0.02043	4.13%	4.76%
36		5	1.139	0.8754	1.403	1.092	0.97	1.504	0.09502	18.65%	1.86%
36.5		5	1.199	0.9086	1.489	1.1	0.978	1.492	0.1045	19.5%	-3.27%
37		5	0.898	0.7274	1.069	0.888	0.756	1.066	0.06145	15.3%	22.64%
37.5		5	1.076	0.9217	1.23	1.088	0.916	1.224	0.05557	11.55%	7.31%
38		5	1.099	0.9125	1.285	1.036	0.926	1.274	0.06711	13.66%	5.34%
38.5		5	1.07	0.9083	1.232	1.09	0.856	1.212	0.05838	12.19%	7.79%
Mean Dry Bio	omass-mg Detai	I									
C-ppt	Control Type	Rep 1		Rep 3	Rep 4	Rep 5					000000000000000000000000000000000000000
0	Lab Control	1.066		1.112	1.116	1.176					
35		1.068		1.026	1.074	1.074					
35.5		1.172	1.064	1.09	1.07	1.132					
36		1.012	1.504	1.118	0.97	1.092					
36.5		0.978	1.404	1.02	1.492	1.1					
37		1.066	0.774	0.756	1.006	0.888					
37.5		1.16	0.992	0.916	1.224	1.088					
38		1.238		1.036	1.274	1.02					
38.5		1.09	1.084	1.212	1.11	0.856					
JU.J		1.08	1.004	1.414	1.11	0.000					

Report Date: Test Code: 26 Jun-15 09:20 (p 4 of 4) 1505-S091 | 11-2970-9154

Pacific Topsmelt 7-d Survival and Growth Test Nautilus Environmental (CA) Analysis ID: 08-3759-0266 Endpoint: Mean Dry Biomass-mg **CETIS Version:** CETISv1.8.7 Analyzed: 23 Jun-15 11:20 Parametric-Control vs Treatments Analysis: Official Results: Yes Graphics 0.40 0.35 0.30 Mean Dry Biomass-mg 0.25 0.20 0.10 8.0 0.05 0,6 0.00 -0.05 -0.10 -0.15 0.2 -0.20 -0.25 0 LC 35.5 36.5 37.5 38.5 -2.5 -2.0 -1.5 -1.0 -0.5 0.0 1.0 1.5 2.0 C-ppt Rankits

Report Date:

26 Jun-15 09:20 (p 1 of 2)

Test Code:

1505-S091 | 11-2970-9154

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental (CA)

Analysis ID: 09-2555-2009 Analyzed: 23 Jun-15 11:19

Endpoint: 7d Survival Rate Analysis:

CETIS Version:

CETISv1.8.7

Linear Interpolation (ICPIN)

Official Results:

Yes

Linear Interpol	ation Options
X Transform	Y Transform

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	196443	1000	Yes	Two-Point Interpolation

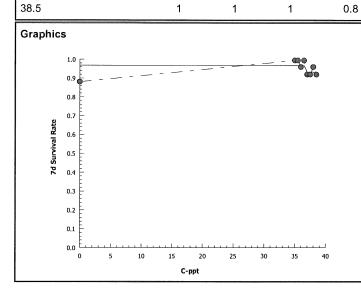
Point Estimates

Level	ppt	95% LCL	95% UC
EC25	>38.5	N/A	N/A
EC50	>38.5	N/A	N/A

7d Survival Rate Summary			Calculated Variate(A/B)								
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.88	0.6	1	0.08	0.1789	20.33%	0.0%	22	25
35		5	1	1	1	0	0	0.0%	-13.64%	25	25
35.5		5	1	1	1	0	0	0.0%	-13.64%	25	25
36		5	0.96	8.0	1	0.04	0.08944	9.32%	-9.09%	24	25
36.5		5	1	1	1	0	0	0.0%	-13.64%	25	25
37		5	0.92	0.6	1	0.08	0.1789	19.44%	-4.55%	23	25
37.5		5	0.92	8.0	1	0.04899	0.1095	11.91%	-4.55%	23	25
38		5	0.96	8.0	1	0.04	0.08944	9.32%	-9.09%	24	25
38.5		5	0.92	8.0	1	0.04899	0.1095	11.91%	-4.55%	23	25

7d Surviva	7d Survival Rate Detail								
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5			
0	Lab Control	0.8	1	1	1	0.6			
35		1	1	1	1	1			
35.5		1	1	1	1	1			
36		8.0	1	1	1	1			
36.5		1	1	1	1	1			
37		1	1	0.6	1	1			
37.5		1	8.0	8.0	1	1			
38		1	1	8.0	1	1			

8.0



Report Date:

26 Jun-15 09:20 (p 2 of 2)

Test Code:

1505-S091 | 11-2970-9154

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental (CA)

Analysis ID: 10-9737-7367 Analyzed: 23 Jun-15 11:20

Endpoint: Mean Dry Biomass-mg Analysis: Linear Interpolation (ICPIN)

CETIS Version: CETISv1.8.7 Official Results: Yes

Linear	Interpo	lation	Options
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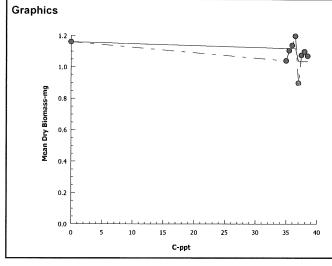
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	1863856	1000	Yes	Two-Point Interpolation

Point Estimates

Levei	ppt	95% LCL	95% 00
IC25	>38.5	N/A	N/A
IC50	>38.5	N/A	N/A

Mean Dry Biomass-mg Summary				Calculated Variate							
C-ppt	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect		
0	Lab Control	5	1.161	1.066	1.334	0.04669	0.1044	9.0%	0.0%		
35		5	1.041	0.962	1.074	0.02165	0.04841	4.65%	10.34%		
35.5		5	1.106	1.064	1.172	0.02043	0.04568	4.13%	4.76%		
36		5	1.139	0.97	1.504	0.09502	0.2125	18.65%	1.86%		
36.5		5	1.199	0.978	1.492	0.1045	0.2338	19.5%	-3.27%		
37		5	0.898	0.756	1.066	0.06145	0.1374	15.3%	22.64%		
37.5		5	1.076	0.916	1.224	0.05557	0.1243	11.55%	7.31%		
38		5	1.099	0.926	1.274	0.06711	0.1501	13.66%	5.34%		
38.5		5	1.07	0.856	1.212	0.05838	0.1305	12.19%	7.79%		

Mean Dry	/ Biomass-mg Deta	il				
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Lab Control	1.066	1.334	1.112	1.116	1.176
35		1.068	0.962	1.026	1.074	1.074
35.5		1.172	1.064	1.09	1.07	1.132
36		1.012	1.504	1.118	0.97	1.092
36.5		0.978	1.404	1.02	1.492	1.1
37		1.066	0.774	0.756	1.006	0.888
37.5		1.16	0.992	0.916	1.224	1.088
38		1.238	0.926	1.036	1.274	1.02
38.5		1.09	1.084	1.212	1.11	0.856



Client: Poseidon	Test Species: A. affinis
Sample ID: Nautilus brine (frozen seawater)	Start Date/Time: 5/5/2015
Test No.: \505-5091	End Date/Time: 5/12/2015

Conc.	D.,,	Rand	Test Day / No. Organisms Alive							Percent	
(<u>ppt</u>)	Rep.	#	0	1	2	3	4	5	6	7	Survival
Lab Control	а	55	5	4	1-1	4	4	4	4	4	80
	b	32	5	5	5	5	5	5	5	5	100
	С	58	5	5	5	5	5	5	5	5	(00)
	d	34	5	5	5	5	5	5	5	5	100
	е	35	5	3	3	B	3	3	3	3	60
Brine Control	а	72	5	5	5	5	S	5	5	5	100
	b	48	5	4	4	4	4	4	4	4	60
	С	38	5	5	5	5	5	5	5	5	100
	d	63	5	5	5	2	5	5	5	5	(00)
	е	46	5	5	5	5	5	5	5	5	100
35.0	а	39	5	5	5	<i>-</i> 5'	5	5	5	5	0
	b	78	5	5	5	5	5	5	5	5	100
	С	74	5	5	S	5	Ī	5	5	5	100
	d	51	5	5	5	5	5	5	5	5	160
	е	64	5	5	5	5	S	5	5	5	100
35.5	а	71	5	5	5	5	5	5	5	5	00
	b	66	5	5	5	5	5	5	3	5	100
	С	47	5	5	5	5	5	5	5	5	(00)
	d	40	5	5	\$	5	5	5	5	5	100
	е	56	5	5	5	5	5	5	5	5	100
36.0	а	73	5	4	Ц	U	ч	4	4	Ť	80
	b	43	5	5	5	5	5	5	5	5	C0]
	С	68	5	5	2	5	5	5	5	5	[00]
	d	60	5	5	S	S	\$	5	5	5	100
	е	61	5	5	5	_5	5	5	5	5	100
36.5	а	36	5	5	,5	5	5	5	5	5	100
	b	59	5	5	5	5	5	5	5	5	100
	С	54	5	5	5	5	5	5	5	5	100
	d	49	5	5	2	S	5	5	5	5	100
	е	41	5	5	5	?	5	5	5	5	100
Rand # QC:	Tech	Initials	EG	CH	NH	NH	NIT	3/L	CH	CH	

Time Fed (day):	0	1	2	3	4	5	01.26018
morning:	- Labor	0815	0820	0930	0825	0830	+0830
evening:	1540	1530	1655	1530	1730	1545	+1700
						6	Shallbala

Γare wt. Initials/Date:	362	5/1	46
Date/Time in:	5/12/	15	1305
Date/Time out:	5h311	5	<u>155</u> 0
Temp (°C):	600	2	
		. 1	

Drying Oven Info

QC Check: ACOLOIS

Comments:

Client: Poseidon	Test Species: A. affinis
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Start Date/Time: 5/5/2015 \\ \frac{3.50}{}

Sample ID: Nautilus brine (frozen seawater)

End Date/Time: 5/12/2015 1130

Test No.: 1605-5091

Conc.	Rep.	Rand			Test	Day / No	. Organ	isms Al	ive		Percent
(ppt)	Kep.	#	0	1	2	3	4	5	6	7	Survival
37.0	а	76	5	645	5	5	5	5	5	5	100
	b	67	5	65 684	S	5	5	5	5	5	100
	С	69	5	884	3	3	3	3	3	3	60
	d	33	5	5	5	5	5	5	5	5	100
	е	80	5	885	5	5	_\$	5	5	5	100
37.5	а	77	5	5	5	5	5	5	5	5	00
	b	65	5	4	4	S. Commercial Commerci	·g	4	4	4	80
	С	52	5	5	5	5	5	5	5	L	80
	d	31	5	5	5	5	5	5	5	5	100
	е	57	5	5	2	S	5	5	5	5	(00)
38.0	а	45	5	5	5	5	5	5	5	5	100
	b	75	5	5	5	5	5	5	5	5	(60
	. с	50	5	4	4	3	1	4	4	4	80
	d	37	5	5	5	5	5	5	5	5	100
	е	62	5	5	5	S	5	5	5	5	100
38.5	a	44	5	5	5	5	5	5	5	5	100
	b	53	5	5	2	2	5	5	5	3	100
	С	70	5	5	5	5	5	5	5	5	100
	d	42	5	4	4	4	7	4	4	Ч	80
	е	79	5	4	4	d	7	4	4	Ч	90
	а		5								
A. A	b		5								
	С		5								
	d		5								
	е		5								
***************************************	а		5								
	b		5			:					
	С		5								
	d		5 💥	ò	20-11-1						
	е		5 %								
Rand # QC: CH	Tech	Initials	EG.	CH	NA	NA	иH	BK	CH	CH	
Initial Count QC: 1950		Time	1350	1145	1402	1579	1600	1226	1315	1205	

Time Fed (day): 0 1 2 3 4 5 6

morning: - 0416 0820 0430 0825 0830 0830

evening: 1540 1630 1665 1530 1730 1645 + 1700

Comments: ACH SIGUS GAS

Drying Oven Info

Tare wt. Initials/Date: 562 5/12/15

Date/Time in: 5/12/15 13:05

Date/Time out: 5/13/15/15/50

Temp (°C): 66.0

QC Check: VB521116 Final Review: AL 422015 Client: Poseidon Test Species: A. affinis

Sample ID: Nautilus Brine (frozen seawater) Start Date/Time: 5/5/2015 1350

Test No.: 1505-S091 **End Date/Time**: 5/12/2015 1130

Conc.	Rep.	pan weight	pan + fish weight	total organism
(<u>ppt</u>)		(mg)	(mg)	weight (mg)
	a	21.68	27.01	5.33
Lab Control	b	22.59	29.26	6.67
	С	22.72	28.28	5.56
	d	22.6	28.18	5.58
	е	22.36	28.24	5.88
	а	24.24	30.09	5.85
Brine Control	b	23.31	27.92	4.61
	С	22.81	29.12	6.31
	d	22.46	27.78	5.32
	е	23.63	28.48	4.85
	а	23.73	29.07	5.34
35	b	23.51	28.32	4.81
	С	22.74	27.87	5.13
	d	22.95	28.32	5.37
	е	23.36	28.73	5.37
	а	22.75	28.61	5.86
35.5	b	23.62	28.94	5.32
	С	23.86	29.31	5.45
	d	22.63	27.98	5.35
	е	22.49	28.15	5.66
	а	22.72	27.78	5.06
36	b	22.12	29.64	7.52
	С	23.36	28.95	5.59
	d	22.81	27.66	4.85
	е	21.42	26.88	5.46
	a	22.74	27.63	4.89
36.5	b	20.67	27.69	7.02
	С	21.05	26.15	5.1
	d	21.37	28.83	7.46
	е	22.98	28.48	5.5
	а	24.04	29.37	5.33
37	b	24.47	28.34	3.87
	С	24.94	28.72	3.78
	d	24.67	29.7	5.03
	e	24.93	29.37	4.44
	1			

 Tech Initials:
 SG
 VCR

 Date/Time:
 5/12/15 0945
 5/13/2015 1550

QC Check: VB 5/21/5
Final Review: ACC126/15

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

Client: Poseidon Test Species: A. affinis

Sample ID: Nautilus Brine (frozen seawater) Start Date/Time: 5/5/2015 1350

Test No.: 1505-S091 **End Date/Time:** 5/12/2015 1130

	1			
Conc.	Rep.	pan weight	pan + fish weight	total organism
(ppt)	Kep.	(mg)	(mg)	weight (mg)
	а	22.06	27.86	5.8
37.5	b	21.35	26.31	4.96
	С	21.37	25.95	4.58
	d	22.73	28.85	6.12
	е	23.93	29.37	5.44
	а	22.03	28.22	6.19
38	b	22.42	27.05	4.63
	С	21.14	26.32	5.18
	d	20.03	26.4	6.37
	е	23.5	28.6	5.1
	а	22.99	28.44	5.45
38.5	b	22.89	28.31	5.42
	С	21.26	27.32	6.06
	d	22.28	27.83	5.55
	е	22.11	26.39	4.28
	а			0
	b			0
	С			0
	d			0
	е			0
	а			0
	b			0
	С			0
	d			0
	е			0
	а			0
	b			0
	С			0
	d			0
	е		000000000000000000000000000000000000000	0
	а			0
	b			0
	С			0
	d			0
	е			0
				Y

 Tech Initials:
 SG
 VCR

 Date/Time:
 5/12/15 0945
 5/13/2015 1550

QC Check: 108 5 2115

Final Review: A((2 | 24) 5

Client: Poseidon

Sample ID: Nautilus brine (frozen seawater)

Test No: 1505-9091

Concentration		Lab Control						
Day	0	1	2	3	4	5	6	7
			- 64 km	Ini	tial			
рН	8.06	7.85	740	7.99	8.00	7.83	794	
DO (mg/L)	7.7	7.2	6.8	7.6	6.9	7.4	8.0	
Salinity (ppt)	33,3	33,4	234	334	33-1	334	33.4	
Temp (°C)	19.0	19.9	20.0	19.6	20.2	20.1	20.2	
				Fi	nal			
рН		7-,94	7.81	7.70	7.79	7.88	7.80	7.7
DO (mg/L)		Deptu.	6.0	6.0	6.5	6.6	7.3	64
Salinity (ppt)		33.W	33.4	33.5	33.6	33.3	33.3	33.4
Temp (°C)	7	19.8	20.0	19.8	19,8	19.8	19.7	20.1

Concentration		Brine Control						
Day	0	1	2	3	4	5	6	7
				lni	tial			
pH	8.03	7.88	7.95	8.00	4.00	7.91	7.95	
DO (mg/L)	7.5	7.2	6.8	7.6	6.9	7. Car	G. (
Salinity (ppt)	33.3	33.4	33.4	334	33.4	33.4	33.4	
Temp (°C)	20.3	20.2	20.4	19.6	20.3	20.0	20.6	
				Fi	nal			
pН		4.91	7.78	7.71	7.73	7.84	7.50	7:79
DO (mg/L)		4.8	6.0	5.0	5,9	6.0	7.2	6.2
Salinity (ppt)		33.8	33.4	33,6	33.6	33.4	34.1	33.6
Temp (°C)		19.3	20.0	196	19.9	19.7	19.8	19.7

Concentration		35.0 ppt						
Day	0	1	2	3	4	5	6	7
				lmi	tial			
рН	8.02	7.86	7.87	8.00	9.00	7.94	7.94	
DO (mg/L)	7.5	7.2	6.4	7.6	7.	7.5	8.1	
Salinity (ppt)	35.0	35.0	36.0	350	36.0	35.0	35.0	
Temp (°C)	19.4	20.0	20,1	19.5	20.2	119.8	20.5	
				Fi	nal			
рН		7.95	7.81	7.75	7.78	7.89	7.81	7.81
DO (mg/L)		le. le	5.9	5.9	6.1	6.1	7.1	6.2
Salinity (ppt)		39.2	35.0	35.2	35.0	34.9	351	35.1
Temp (°C)		7719.8	19.9	19.1	20,0	11.3	19.7	199

Animal Source/Date Received:

Animal Age at Initiation:

ABS / 5/1/15

Sample Log-in Numbers: A: NA

A: N/A C: N/A

Test Species: A. affinis

End Date/Time: 5/12/2015 1/30

Concentration				35.5	ppt			
Day	0	1	2	3	4	5	6	7
				Ini				
pН	8.02	7.86	7.92	2001	799	7.75	7.94	
DO (mg/L)	7.4	701	6.7	7.6	7.0	7.4	0.1	
Salinity (ppt)	35.5	35.5	35.5	35.5	355	35.5	35.5	
Temp (°C)	19.3	20.0	20.0	19.4	7.05	19.7	7-0.5	
				Fi	nal			
рН		7.93	7.63	7.73	7.77	7.89	7.82	7.84
DO (mg/L)		ie-7-	5.6	5.6	5,9	6.1	7.0	6.4
Salinity (ppt)		74.0	35.6	35.7	35,5	35.4	35.7	35.7
Temp (°C)	1	19.4	20.2	19.9	20.1	19.9	19.6	19.8

Concentration			100.440011	36.0) ppt			
Day	0	1	2	3	4	5	6	7
				lni	tial			
pН	8.02	7.88	7.92	4.51	40	7.96	793	
DO (mg/L)	7.5	7.0	6.8	76	7.0	7.5	G. 1	
Salinity (ppt)	36.0	36,0	36.0	36.0	36.0	36,0	36.0	
Temp (°C)	19.4	20.0	20.0	19.4	201	19.7	20.3	
				Fi	nal			
pН		7-91	7.83	776	7.81	7.90	7.83	7.84
DO (mg/L)		U.5	6.4	6.0	6.3	6.3	7.4	6.7
Salinity (ppt)	1	34.3	36.1	36.2	36.1	36.0	36.4	36.2
Temp (°C)		14.2	19.9	19.7	19.7	19.7	195	19.6

Concentration				36.5	5 ppt			
Day	0	1	2	3	4	5	6	7
				ln:	itial			
На	8.00	7.88	7.93	15,0	799	7.97	7.93	
DO (mg/L)	7.4	7.1	68	7.6	69	7.4	8.1 0	SHIP
Salinity (ppt)	36.5	36.5	36,5	36.5	36.5	36.5	36.7	5
Temp (°C)	19,4	20.0	19.9	1904	20.1	19.6	Zo. 3	
				FI	nal			
pН		7.15	7.84	7.74	7.79	7.88	7.62	7.79
DO (mg/L)		4.4	5.8	5.6	5,8	6.0	7.0	6.1
Salinity (ppt)		34.U	345	36.6	36.5	36.5	ع) عائز	367
Temp (°C)		19.8	20.1	14.8	20,2	19.8	19.9	19.8

Analysts: Initial: CH CH CH NH NH VH CH CB AG

Dilutions made by: CH CH CH NH NH NH NH CH

Sample Used (A, B, C): Brize Brize Brize Brize Brize Brize Brize

Comments:

@ Hach sension 5 used for all water quality readings (salinity).

QC Check:

KB Blailis

Final Review: AC Ce (2015

Client: Poseidon Test Species: A. affinis Sample ID: Nautilus brine (frozen seawater) 1350 Start Date/Time: 5/5/2015 Test No: 1505-5091 End Date/Time: 5/12/2015 11200 Concentration 37.0 ppt Concentration 38.5 ppt Day 0 6 Day 6 Initial. Initial рΗ .00 7.88 10 7.98 100 194 На DO (mg/L) 7.1 7.3 6.9 6. 9 DO (mg/L) 37,0 37,0 37.6 37.0 37.0 37.0 Salinity (ppt) 38.5 Salinity (ppt) 38.5 38.5 70.2 19.9 Temp (°C) 6 19.6 Temp (°C) 19.8 19.9 19.1 9,5 147 70.3 Final Final рΗ 7.79 7.907.82 7.81 7-14-1 7.86 7.79 7.83 рΗ DO (mg/L) 5. 5. 62 5.6 DO (mg/L) 5.6 6.4 0 Salinity (ppt) 37.2 31.2 37.0 37.1 38.7 36.5 37 , C 37.2 Salinity (ppt) 38.0 38.7 38,5 10.4 19.4 19.9 Temp (°C) 19.9 19.8 (9.0) 19.7 Temp (°C) 19.4 19.9 Concentration 37.5 ppt Concentration Day 0 Day 0 3 4 5 6 Initial 4.00 рΗ DO (mg/L) 13 7.5 DO (mg/L) Ь Salinity (ppt) 37.5 37 Salinity (ppt) Temp (°C) 20.0 146 202 Temp (°C) Final рΗ 7.79 7.80 pН 5.8 59 DO (mg/L) 2 DO (mg/L) 6-0 37.5 37.5 Salinity (ppt) **38.0** 37.7 Salinity (ppt) 19.7 Temp (°C) 19.2 20.0 19.8 Temp (°C) Concentration 38.0 ppt Concentration Day 5 6 Day 3 5 6 Initial 7.90 7.93 8.01 рΗ рΗ DO (mg/L) 6.9 DO (mg/L) 38.0 38.0 Salinity (ppt) 36.0 Salinity (ppt) 19.5 19.9 19.9 70.2 19,6 20.3 19.5 Temp (°C) Temp (°C) Final рΗ 4.13 7.05 7,80 рΗ DO (mg/L) 5.8 6.0 (g. DO (mg/L) 200.0 38,0 293 Salinity (ppt) Salinity (ppt) Temp (°C) 19. 4 20.1 19.9 20.0 19,8 19 Temp (°C) 3 7 NIK NIE KB Animal Source/Date Received: Analysts: Initial: ALB KB NH KB AG Animal Age at Initiation: Final CH NH NA Dilutions made by NH MA Sample Log-in Numbers: ~1 Brine Bline MA brine Anne Brine Sample Used (A, B, C): Dire for all water quality readings (salinity) Comments: Final Review: 406/26/15 QC Check:

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

Marine Chronic Bioassay

Brine Dilution Worksheet

Project:

Poseidon

Analyst: CH

Sample ID:

frozen seawater

Test Date: 5/5/2015

Test No:

Test Type: Chronic Topsmelt

Salinity of Seawater

33.3

Salinity of Brine

86.7

Date of Brine used: Composited on S14119 using brine from: 4/2/15 - 4/24/15

Test Dilution Volume

1250

Alkalinity of Brine Control: 128

mg/L as CaCO3

TS = target salinity

SE = salinity of effluent

SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
34.0	100.0	250	NA	NA	1250
35.0	96.8	1210.2	0.03	39.8	1250
35.5	95.9	1198.5	0.04	51.5	1250
36.0	94.9	1186.8	0.05	63.2	1250
36.5	94.0	1175.1	0.06	74.9	1250
37.0	93.1	1163.4	0.07	86.6	1250
37.5	92.1	1151.7	0.09	98.3	1250
38.0	91.2	1140.0	0.10	110.0	1250
38.5	90.3	1128.3	0.11	121.7	1250

0.099485019

0.128745318

0.158005618

0.187265918

0.216526217

0.245786517

0.275046816

0.304307116

Q18 KB 921/15

DI	Volume

Brine Control 195.2 0.62 121.7 1250
--

Total Brine Volume Required (ml):

646.1

QC Check: KB5 21

Final Review: Acle Holl

5/6/15 renewal through 5/11/15 Brine Dilution Worksheet

Marine Chronic Bioassay

Project:	Poseidon	

Analyst: CH

Sample ID:

frozen seawater

Test Date: 5/5/2015

Test No:

1505-5091

Test Type: Chronic Topsmelt

Salinity of Seawater

33.4

Salinity of Brine

86.7

Date of Brine used: composited on 5/4/15 using lonive from:

Test Dilution Volume

1000

Alkalinity of Brine Control: 122

mg/L as CaCO3

TS = target salinity

SE = salinity of effluent

SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)	
34.0	100.0	CH 5/6/15 ^{Q/8} 250 (000	NA	NA	1000	
35.0	97.0	970.0	0.03	30.0	1000	0\075046904
35.5	96.1	960.6	0.04	39.4	1000	0.098499062
36.0	95.1	951.2	0.05	48.8	1000	0.12195122
36.5	94.2	941.8	0.06	58.2	1000	0.145403377
37.0	93.2	932.5	0.07	67.5	1000	0.1688\$5535
37.5	92.3	923.1	0.08	76.9	1000	0.19230 692
38.0	91.4	913.7	0.09	86.3	1000	0.21575985
38.5	90.4	904.3	0.11	95.7	1000	0.239212008

DI Volume

Brine Control 152.7 0.63 95.7 1000

Total Brine Volume Required (ml):

502.8

QC Check: 185 3 15

Final Review: AC 6/26/15

APPENDIX C

Glossary of Lab 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 ≤ 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. H owever, 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.

Updated: 6/30/15

APPENDIX D

Reference Toxicant
Test Data and Statistical Summaries



CETIS Summary Report

Report Date: Test Code: 16 Dec-14 18:07 (p 1 of 1) 141210hrrt | 18-3651-9027

								Test Code:		141	1210hrrt 1	8-3651-902
Red Abalone	Larval Develop	ment Test								Nautilu	s Environr	mental (CA)
Batch ID: Start Date: Ending Date: Duration:	00-7981-3930 10 Dec-14 15:0 12 Dec-14 17:0 50h	30 Pro 00 Spe	t Type: tocol: cies: rce:	Development EPA/600/R-95 Haliotis rufeso American Aba	ens					ral Seawat Applicable	er	
Sample ID: Sample Date: Receive Date: Sample Age:		Sou	le: erial; rce: ion:	141210hrrt Zinc sulfate Reference To Zinc sulfate	xicant			Client: Project:	Inter	nal		
Comparison S	Summary									in the second se		
Analysis ID	Endpoint		NOEL		TOEL	PMSD	TU	Metho	od			· ·
20-3619-3450	Development F	Rate	18	32	24	4.79%		Dunne	ett Mı	ultiple Com	parison Te	st
Point Estimate	e Summary											
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Metho	od			
05-5260-7606	Development F	Rate	EC50	40.8	40.06	41.55		Spear	rman-	Kärber		
Test Acceptab	oility											
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	its	Overl	ар	Decision		
05-5260-7606	Development F	Rate	Contro	ol Resp	0.946	0.8 - NL	Yes	Yes Passes Acceptability Criter			Criteria	
20-3619-3450	Development R	late	Contro	ol Resp	0.946	0.946 0.8 - NL			Yes Passes Acceptability Criter			
20-3619-3450	Development R	late	NOEL		18	NL - 56		No			cceptability	
20-3619-3450	Development F	tate	PMSD)	0.04793	NL - 0.2		No		Passes A	cceptability	Criteria
Development	Rate Summary	0.000										
C-μg/L	Control Type	Count	Mean	95% LCL	. 95% UCL	Min	Max	Std E	rr	Std Dev	CV%	%Effect
0	Lab Control	5	0.946	0.9366	0.9554	0.92	0.97	0.0112	22	0.0251	2.65%	0.0%
10		5	0.964	0.9583	0.9697	0.95	0.99	0.006	782	0.01517	1.57%	-1.9%
18		5	0.952	0.9459	0.9581	0.94	0.97	0.0073	348	0.01643	1.73%	-0.63%
32		5	0.86	0.8423	0.8777	0.79	0.92	0.0212	21	0.04743	5.52%	9.09%
56		5	0.036	0.02492	0.04708	0	0.08	0.0132	27	0.02966	82.4%	96.19%
100		5	0	0	0	0	0	0		0		100.0%
Development I	Rate Detail											
C-μg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	0.97	0.92	0.92	0.95	0.97						,
10		0.96	0.96	0.95	0.99	0.96						
18		0.97	0.94	0.97	0.94	0.94						
32		0.85	0.86	0.92	0.88	0.79						
56		0	0.02	0.04	0.08	0.04						
100		0	0.02	0.04	0.00	0.04						
100		U	U	U	U	U						

Report Date: Test Code: 16 Dec-14 18:06 (p 1 of 2) 141210hrrt | 18-3651-9027

									Test	Code:	14	+1210nmt	18-3051-9027
Red Abalone	Larva	l Developr	nent T	est		, ,				. ,	Nautil	us Enviro	nmental (CA)
Analysis ID: Analyzed:	, ,						tments	CET Offic		/1.8.4			
Data Transfo	rm		Zeta	Alt Hyp	Trials	Seed			NOEL	LOEL	TOEL	TU	PMSD
Angular (Corr	ected)		NA	C > T	NA	NA			18	32	24		4.79%
Dunnett Mult	iple C	omparison	Test										
Control	vs	C-µg/L		Test Stat	Critical	MSD	DF	P-Value	P-Type	Decisio	n(α:5%)		
Lab Control		10		-1.099	2.305	0.091	8	0.9814	CDF	Non-Sig	nificant Effe	ct	
		18		-0.2861	2,305	0.091	8	0.8795	CDF	Non-Sig	nificant Effe	ct	
		. •											
		32*		3.792	2.305	0.091	8	0.0020	CDF	Significa	int Effect		

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)
Between	5.321269	1.330317	4	339.7	<0.0001	Significant Effect
Error	0.07833021	0.00391651	20			
Total	5.399599		24			

Distributional To	Distributional Tests											
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)							
Variances	Bartlett Equality of Variance	2.625	13.28	0.6224	Equal Variances							
Distribution	Shapiro-Wilk W Normality	0.9785	0.8877	0.8533	Normal Distribution							

Developme	Development Rate Summary													
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect			
0	Lab Control	5	0.946	0.9148	0.9772	0.95	0.92	0.97	0.01122	2.65%	0.0%			
10		5	0.964	0.9452	0.9828	0.96	0.95	0.99	0.006783	1.57%	-1.9%			
18		5	0.952	0.9316	0.9724	0.94	0.94	0.97	0.007348	1.73%	-0.63%			
32		5	0.86	0.8011	0.9189	0.86	0.79	0.92	0.02121	5.52%	9.09%			
56		5	0.036	0	0.07283	0.04	0	80.0	0.01327	82.4%	96.19%			
100		5	0	0	0	0	0	0	0		100.0%			

Angular (C	Angular (Corrected) Transformed Summary													
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect			
0	Lab Control	5	1.341	1.271	1.411	1.345	1.284	1.397	0.02521	4.2%	0.0%			
10		5	1.385	1.324	1.446	1.369	1.345	1.471	0.02195	3.54%	-3.24%			
18		5	1.353	1.303	1.403	1.323	1.323	1.397	0.01798	2.97%	-0.84%			
32		5	1.191	1.106	1.277	1.187	1.095	1.284	0.03077	5.78%	11.19%			
56		5	0.1763	0.0677	0.2849	0.2014	0.05002	0.2868	0.03911	49.61%	86.86%			
100		5	0.05002	0.05001	0.05003	0.05002	0.05002	0.05002	0	0.0%	96.27%			

Report Date: Test Code: 16 Dec-14 18:06 (p 2 of 2) 141210hrrt | 18-3651-9027

Red Abalone Larval Development Test Nautilus Environmental (CA) Analysis ID: 20-3619-3450 Endpoint: **Development Rate CETIS Version:** CETISv1.8.4 Analyzed: 16 Dec-14 18:06 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 1.0 0,12 **-0**-**_**@__ 0.10 Reject Null 0.08 8.0 0.06 0.7 0.04 0.02 0.6 0.00 0.5 -0.02 -0.04 0,4 -0.06 0.3 -0,08 0.2 -0.12 0.1 -0.14 0.0 -2,0 -2.5 -1,5 -1.0 -0.5 0,0 0,5 1.0 1.5 2.0 C-µg/L Rankits

Report Date: Test Code:

16 Dec-14 18:07 (p 1 of 1)

141210hrrt | 18-3651-9027

Red Abalone Larval Development Test

Nautilus Environmental (CA)

Analysis ID: Analyzed:

05-5260-7606 16 Dec-14 18:06

Endpoint: Development Rate Analysis: Untrimmed Spearman-Kärber

CETIS Version: Official Results:

CETISv1.8.4

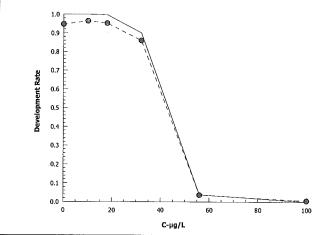
Yes

Spearman-Kärber	Estimate	95
-----------------	----------	----

Threshold Option	Inresnoid	Irim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0.054	0.00%	1.611	0.003965	40.8	40.06	41.55

Developm	nent Rate Summar	/	Calculated Variate(A/B)								
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.946	0.92	0.97	0.01122	0.0251	2.65%	0.0%	473	500
10		5	0.964	0.95	0.99	0.006783	0.01517	1.57%	-1.9%	482	500
18		5	0.952	0.94	0.97	0.007348	0.01643	1.73%	-0.63%	476	500
32		5	0.86	0.79	0.92	0.02121	0.04743	5.52%	9.09%	430	500
56		5	0.036	0	0.08	0.01327	0.02966	82.4%	96.19%	18	500
100		5	0	0	0	0	0		100.0%	0	500

Graphics



Protocol:

Report Date:

16 Dec-14 18:11 (1 of 1)

Red Abaione Larval Development Test

Sigma:

11.69

CV:

20.60%

Nautilus Environmental (CA)

Test Type: Development

Organism: Haliotis rufescens (Red Abalone) EPA/600/R-95/136 (1995)

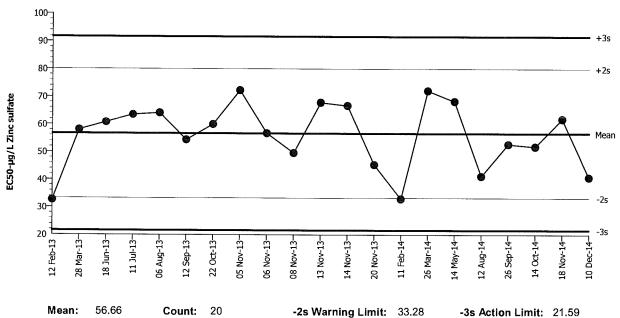
Endpoint: Development Rate

Material: Source:

Zinc sulfate Reference Toxicant-REF

+3s Action Limit: 91.73

Red Abalone Larval Development Test



+2s Warning Limit:

80.04

Quality Control Data										
Point	Year	Month	Day	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2013	Feb	12	32.63	-24.03	-2.056	(-)		12-6287-3770	13-5482-5100
2		Mar	28	58.01	1.351	0.1156			02-1898-0832	19-4308-5407
3		Jun	18	60.76	4.101	0.3509			15-0077-4900	00-2560-4372
4		Jul	11	63.48	6.821	0.5835			20-1873-1666	04-6373-4064
5		Aug	6	64.09	7.426	0.6352			07-0122-1486	08-7879-6283
6		Sep	12	54.37	-2.289	-0.1958			01-8875-4392	00-0984-0680
7		Oct	22	59.96	3.297	0.282			00-1950-7526	03-4814-7235
8		Nov	5	72.27	15.61	1.336			13-0598-2106	06-6008-6070
9			6	56.71	0.04992	0.00427			17-8546-9636	17-3071-8592
10			8	49.64	-7.018	-0.6004			20-4825-5447	15-3343-8191
11			13	67.88	11.22	0.9595			01-9285-3290	05-2114-7000
12			14	66.73	10.07	0.8613			12-4955-9047	05-7865-8140
13			20	45.42	-11.24	-0.9611			15-8538-2252	14-5629-7331
14	2014		11	33.01	-23.65	-2.023	(-)		00-8191-4476	07-3868-3337
15		Mar	26	72.1	15.44	1.321			11-0783-9458	08-4579-6000
16		May	14	68.33	11.67	0.9985			14-0092-0578	07-8756-4120
17		Aug	12	41.25	-15.41	-1.318			09-7316-9900	19-6875-2864
18		Sep	26	52.78	-3.877	-0.3316			12-0077-1970	07-6392-1596
19		Oct	14	51.91	-4.753	-0.4066			01-1692-6353	05-8596-4968
20		Nov		61.92	5.259	0.4499			12-7477-5365	16-0305-1770
21		Dec	10	40.8	-15.86	-1.357			18-3651-9027	05-5260-7606

CETIS Test Data Worksheet

Report Date: Test Code:

09 Dec-14 16:07 (p 1 of 1)

18-3651-9027/141210hrrt

No.	
Red Abalone Larval Development Test	Nautilus Environmental (CA)

10 Dec-14 (530 Sample Code: 141210hrrt Start Date: Species: Haliotis rufescens

12 Dec-14 17/9/9 Protocol: EPA/600/R-95/136 (1995) Sample Source: Reference Toxicant End Date:

d Date:	12 (Jec-14	4 170		iol: EPA/600/R-95/136 (1995)	Sample Source: Reference Toxicant
mple Dat					al: Zinc sulfate	Sample Station: Zinc sulfate
C-µg/L	Code	Rep	Pos		# Normal	Notes
			1	100	0	
			2	}	0	
			3	1	0	
			4		94	
			5		95	
			6		96	
			7		99	
			8		92	
			9		0	
			10		92	
			11		92	
			12		0	
			13		8	
			14		94	
			15		4	
			16		86	
			17		8 4 914 6/14	
			18		₹ ₹₹	
			19		44	
			20		97	
			21		96	
			22		97	
			23		85	
			24		92	
			25		97	
			26		76	
			27		79	
		-	28		95	
			29	\\.	97	1
			30	V	99	

Report Date:

09 Dec-14 16:07 (p 1 of 1) 18-3651-9027/141210hrrt

Test Code:

Red Abalone	Larva	al Dev	elopn	nent Test				Nautilus Environmental (C
Start Date: 10 Dec-14 1530 Species: Haliotis rufescens End Date: 12 Dec-14 1700 Protocol: EPA/600/R-95/136 (1995) Sample Date: 10 Dec-14 Material: Zinc sulfate				Sample Code: Sample Source: Sample Station:	141210hrrt Reference Toxicant Zinc sulfate			
C-µg/L	Code	Rep	Pos	# Counted	# Normal		Notes	
0	LC	1	25	100	94	AC 12/12/14	**************************************	
0	LC	2	24					
0	LC	3	8				***************************************	CONTRACTOR OF CONTRACTOR
0	LC	4	28					
0	LC	5	29					
10		1	26	100	95	AC 12/12/14		
10		2	21	1 5				
10		3	5				# # A A A A A A A A A A A A A A A A A A	
10		4	30		~			
10		5	6					
18		1	22	100	99	AC 14/2/14		
18		2	7	,,,,		11-119		- 100 mg - 110 mg - 1
18		3	20				- WARE BILL	
18		4	14					
18		5	4				7447-7411-74111	
32		1	23	94	19	Ac 12/12/14		
32		2	16			11010119		
32		3	11					
32		4	19					
32		5	27				THE STATE OF THE S	
56		1	1	100	0	AC12/12/14		
56		2	13	100	**	- 14/11		
56		3	15	Tarris		- AND I		
56		4	18			w		
56		5	17					The state of the s
100		1	10	100	0	XC12/12/14	7 to W 10 to	
100		2	3	100		10111	The state of the s	
100		3	2	1		784 - 10 - 10 - 10 - 10 - 10 - 10 - 10 - 1		
100		4	12					

QC-VCR

100

Marine Chronic Bioassay

Water Quality Measurements

Client:	Internal	Test Species:	Haliotis rufeso	ens
Sample ID:	ZnSO₄	Start Date/Time:	12/10/2014	1530
Test ID:	141210hrrt	End Date/Time:	12/12/2014	1700

Concentration (μg/L)	· · · · · · · · · · · · · · · · · · ·			٦	Temperature (°C)			Dissolved Oxygen (mg/L)			pH (pH units)		
	0	24	48	(A) 0	24	48	0	24	48	0	24	48	
Lab Control	329	330	32.9	15.5	15.3	14,9	8.1	ಶ.೦	8.0	8. <i>0</i> 5	7.98	7.97	
10	32.8	33.0	330	15.5	15.1	14.4	හ.0	7.9	7,9	8.04	7.98	7,98	
18	32.8	32.0	33.0	15.5	14.9	14,9	8.0	4.0	7.9	8.04	7.99	7,28	
32	32.8	32.9	33.0	15.5	15,0	14.4	8.0	8.0	7.9	8.04	7.99	7.98	
56	32.6	32.7	32.9	15.5	14,9	14,6	8.0	4.0	7.9	8.04	7.19	7.28	
100	32.5	32.7	32.7	15.5	14.9	14.9	8.0	8,0	7.9	8.04	7.99	7.97	

Technician Initials:	WQ Readings:	VCR/AG NH	νH	Conc.	10	18	32	56	100
	Dilutions made by:[VCRYAY -		Vol. Zn	0.53	0.95	1.7	3,0	5,3
				stock (mL):	0.57				
				Zn Stock Concentration	n (μg/L):		9,	480	
Comments:	0 hrs: A Temp	xerature taken	from a	i sumaate vial	on test	-trial.			
	24 hrs:			J					
	48 hrs:								
QC Check:	is 12/16/14				Fin	al Review:	KB1114	6	

48

24

0

Dilution calcs. (final volume 500 mL):

nt

Warine Chroni	c Bioassay	Abalone Embryo-	Larval Dev	elopme
Client:	Internal	Test Species	: Haliotis rufes	scens
Sample ID:	ZNSOy Reference Towent	Start Date/Time	12/10/2014	15:3
Test No.:	141210 hart	End Date/Time	: 12/12/2014	17:0
Animal Source/D	ate Received: American Abalone/12/09/14			
Number of abalo	ne and condition upon receipt/holding:			
Males:	416 cood condition			
Females:	41 bood condition			
				_
		Males:	Females:	
	Tris & peroxide addition time	1100	1030	
	Spawn time	1330	1400	
	Number of spawners	4		
	Condition of spawn (light, moderate, heavy)	Heury	un oderate	
	Fertilization time	143	5	
	Embryo counts (per 0.5	ml)		

Embryo counts (per 0.5 ml)						
1	121					
2	131					
3	145					
Mean	132.3					

Time of test initiation:	1530
	~

48 hr. QC 97%

Technician Initials:

Comments:

QC Check:

W 12/16/14

Final Review: VD 1/14/15

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

CETIS Summary Report

Report Date:

29 May-15 09:59 (p 1 of 1)

Test Code:	150520hrdv 08-9621-28

								rest oode.	1000	22011101 00	
Red Abalone	Larval Developr	nent Test							Nautilus	s Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	08-7453-2163 20 May-15 14: 22 May-15 15:5 50h	15 Pro 55 Sp	st Type: otocol: ecles: urce:	Development EPA/600/R-95 Haliotis rufesc American Abal	ens				tural Seawate t Applicable	er	
Sample ID: Sample Date: Receive Date: Sample Age:	: 20 May-15	Ma So	de: terial: urce: ition:	150520hrdv Zinc sulfate Reference Tox Zinc sulfate	dicant			Client: Int Project:	ernal		
Comparison S	Summary										
Analysis ID 10-6271-9277	Endpoint Development R		NOEL 32	L OE L 56	TOEL 42.33	PMSD 2.58%	TU	Method Dunnett	Multiple Com	parison Tes	st
Point Estimate	e Summary		70		2.00000000 - 1000000000 - 100000000000000						
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Method			
00-2924-1270	Development R	tate	EC50		54.11	57.01		Trimmed	Spearman-	Kärber	
Test Acceptab	oility				1,500,000						
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Overlap	Decision		
00-2924-1270	Development R	ate	Contro	ol Resp	0.988	0.8 - NL		Yes	Passes A	cceptability	Criteria
10-6271-9277	Development R	ate	Contro	ol Resp	0.988	0.8 - NL		Yes	Passes A	cceptability	Criteria
10-6271-9277	Development R	ate	NOEL		32	NL - 56		No	Passes A	cceptability	Criteria
10-6271-9277	Development R	ate	PMSD)	0.02582	NL - 0.2		No	Passes A	cceptability	Criteria
Development	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	0.988	0.9744	1	0.97	1	0.004899	0.01095	1.11%	0.0%
10		5	0.986	0.9672	1	0.96	1	0.006782	0.01517	1.54%	0.2%
18		5	0.982	0.9716	0.9924	0.97	0.99	0.003742	0.008367	0.85%	0.61%
32		5	0.984	0.9673	1	0.97	1	0.006	0.01342	1.36%	0.4%
56		5	0.478	0.354	0.602	0.36	0.57	0.04465	0.09985	20.89%	51.62%
100		5	0	0	0	0	0	0	0		100.0%
		J			**						
Development	Rate Detail	J					***************************************				
•	Rate Detail Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
C-µg/L				Rep 3	Rep 4 0.97	Rep 5 0.99					
C-µg/L	Control Type	Rep 1	Rep 2								
С-µg/L 0	Control Type	Rep 1	Rep 2 0.99	0.99	0.97	0.99					
C-μg/L 0 10	Control Type	Rep 1 1 0.99	Rep 2 0.99 0.99	0.99 1	0.97 0.99	0.99 0.96					
C-μg/L 0 10 18	Control Type	Rep 1 1 0.99 0.97	Rep 2 0.99 0.99 0.99	0.99 1 0.98	0.97 0.99 0.98	0.99 0.96 0.99					

Analyst: AC QA: MTP 6/11/15

000-089-180-4 CETIS™ v1.8.7.20

Report Date:

29 May-15 09:58 (p 1 of 1) 150520hrdy | 08-9621-2840

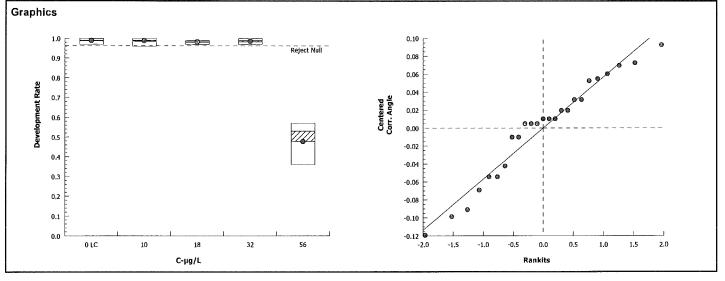
Test Code: 150520hrdv | 08

									lest	Code:	150	1520nrdv 0	8-9621-2840
Red Abalone	Larva	l Development	Test								Nautil	us Environr	nental (CA)
Analysis ID; Analyzed:		6271-9277 May-15 9:58	Endpoint: Analysis:		elopment R ametric-Cor		Trea	tments		IS Versior		1.8.7	
Data Transfor	m	Zet	a Alt l	Нур	Trials	Seed			PMSD	NOEL	LOEL	TOEL	TU
Angular (Corre	ected)	NA	C > .	Т	NA	NA			2.58%	32	56	42.33	
Dunnett Multi	ple C	omparison Tes											
Control	vs	C-µg/L	Test	Stat	Critical	MSD	DF	P-Value	P-Type	Decisio	n(a:5%)		
Lab Control		10	0.13	97	2.305	0.09	8	0.7516	CDF	Non-Sig	nificant Effe	ct	
		18	0.68	43	2.305	0.09	8	0.5207	CDF	Non-Sig	nificant Effe	ct	
		32	0.37	86	2.305	0.09	8	0.6564	CDF	Non-Sig	nificant Effe	ct	
		56*	18		2.305	0.09	8	<0.0001	CDF	Significa	int Effect		
ANOVA Table						X					-W	ACCUSION AND AND AND AND AND AND AND AND AND AN	
Source		Sum Squares	Mea	n Squ	are	DF		F Stat	P-Value	Decisio	n(a:5%)		
Between		1.913564	0.478	8391		4		125.5	<0.0001	Significa	int Effect		
Error		0.07624391	0.00	38121	95	20				-			
Total		1.989808				24		_					
Distributional	Tests	S			The state of the s								
Attelbuto		Tool			T4 C4-4	.		5.4.1	Danislass	40/1			

Diomibutional i	00,0					
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)	
Variances	Bartlett Equality of Variance	5.534	13.28	0.2367	Equal Variances	
Distribution	Shapiro-Wilk W Normality	0.954	0.8877	0.3081	Normal Distribution	
Development R	ate Summary	Emplement of the Control of the Cont	WALL TO SHARE THE SHARE TH			

Developm	ent Rate Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.988	0.9744	1	0.99	0.97	1	0.004899	1.11%	0.0%
10		5	0.986	0.9672	1	0.99	0.96	1	0.006783	1.54%	0.2%
18		5	0.982	0.9716	0.9924	0.98	0.97	0.99	0.003742	0.85%	0.61%
32		5	0.984	0.9673	1	0.99	0.97	1	0.006	1.36%	0.4%
56		5	0.478	0.354	0.602	0.53	0.36	0.57	0.04465	20.89%	51.62%

Angular (C	Corrected) Transfor	med Sumr	nary								
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.466	1.411	1.521	1.471	1.397	1.521	0.01983	3.03%	0.0%
10		5	1.46	1.392	1.529	1.471	1.369	1.521	0.02473	3.79%	0.37%
18		5	1.439	1.4	1.478	1.429	1.397	1.471	0.01413	2.2%	1.82%
32		5	1.451	1.384	1.518	1.471	1.397	1.521	0.02401	3.7%	1.01%
56		5	0.7628	0.6377	0.888	0.8154	0.6435	0.8556	0.04507	13.21%	47.96%



Analysis ID:

Report Date: Test Code: 29 May-15 09:59 (p 1 of 1)

est Code: 150520hrdv | 08-9621-2840

Nautilus Environmental (CA)

Red Abalone Larval Development Test

00-2924-1270

Endpoint: Development Rate CETIS Version: CETISv1.8.7

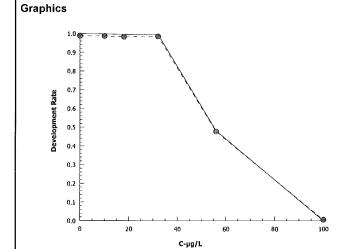
Analyzed: 29 May-15 9:58 Analysis: Trimmed Spearman-Kärber Official Results: Yes

Trimmed Spearman-Kärber Estimates

 Threshold Option
 Threshold
 Trim
 Mu
 Sigma
 EC50
 95% LCL
 95% UCL

 Control Threshold
 0.012
 0.20%
 1.745
 0.005671
 55.54
 54.11
 57.01

Developn	nent Rate Summary										
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.988	0.97	1	0.004899	0.01095	1.11%	0.0%	494	500
10		5	0.986	0.96	1	0.006783	0.01517	1.54%	0.2%	493	500
18		5	0.982	0.97	0.99	0.003742	0.008367	0.85%	0.61%	491	500
32		5	0.984	0.97	1	0.006	0.01342	1.36%	0.4%	492	500
56		5	0.478	0.36	0.57	0.04465	0.09985	20.89%	51.62%	238	500
100		5	0	0	0	0	0		100.0%	0	500



Analyst: AC QA: MP6/11/15

CETIS™ v1.8.7.20

CETIS QC PlotReport Date: 29 May-15 09:59 (1 of 1)

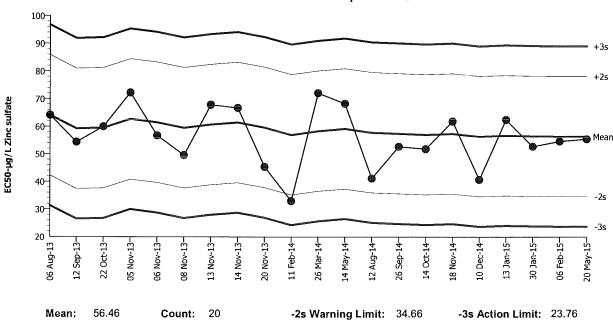
Red Abalone Larval Development Test

Nautilus Environmental (CA)

Test Type: Development Organism: Haliotis rufescens (Red Abalone) Material: Zinc sulfate

Protocol: EPA/600/R-95/136 (1995) Endpoint: Development Rate Source: Reference Toxicant-REF

Red Abalone Larval Development Test



Mean:	56.46	Count:	20	-2s Warning Limit:	34.66	-3s Action Limit:	23.76
Sigma:	10.9	CV:	19.30%	+2s Warning Limit:	78.26	+3s Action Limit:	89.16

Quality Con	trol Data	а			
Point Year	Month	Day Time	QC Data	Delta	Sig

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2013	Aug	6	15:35	64.09	7.626	0.6996			07-0122-1486	08-7879-6283
2		Sep	12	14:40	54.37	-2.089	-0.1917			01-8875-4392	00-0984-0680
3		Oct	22	14:50	59.96	3.497	0.3208			00-1950-7526	03-4814-7235
4		Nov	5	14:25	72.27	15.81	1.451			13-0598-2106	06-6008-6070
5			6	15:30	56.71	0.2499	0.02293			17-8546-9636	17-3071-8592
6			8	0:00	49.64	-6.818	-0.6255			20-4825-5447	15-3343-8191
7			13	16:00	67.88	11.42	1.047			01-9285-3290	05-2114-7000
3			14	14:50	66.73	10.27	0.9421			12-4955-9047	05-7865-8140
9			20	15:15	45.42	-11.04	-1.012			15-8538-2252	14-5629-7331
10	2014	Feb	11	15:00	33.01	-23.45	-2.151	(-)		00-8191-4476	07-3868-3337
11		Mar	26	15:25	72.1	15.64	1.435			11-0783-9458	08-4579-6000
12		May	14	15:35	68.33	11.87	1.089			14-0092-0578	07-8756-4120
13		Aug	12	15:35	41.25	-15.21	-1.396			09-7316-9900	19-6875-2864
14		Sep	26	16:10	52.78	-3.677	-0.3373			12-0077-1970	07-6392-1596
15		Oct	14	15:30	51.91	-4.553	-0.4177			01-1692-6353	05-8596-4968
16		Nov	18	15:05	61.92	5.459	0.5008			12-7477-5365	16-0305-1770
17		Dec	10	15:30	40.8	-15.66	-1.436			18-3651-9027	05-5260-7606
18	2015	Jan	13	14:48	62.52	6.058	0.5558			11-7205-2664	10-2598-5960
19			30	15:22	52.83	-3.632	-0.3332			06-2409-9903	03-0729-5027
20		Feb	6	13:33	54.69	-1.771	-0.1625			19-4508-4987	07-8543-5535
21		Mav	20	14:15	55.54	-0.923	-0.08468			08-9621-2840	00-2924-1270

CETIS Test Data Worksheet

Report Date:

19 May-15 15:21 (p 1 of 1) 08-9621-2840/150520hrdv

Test Code:

Nautilus Environmental (CA)

Start Date: 20 May-15

Red Abalone Larval Development Test

Species: Haliotis rufescens

Sample Code: 150520hrdv

Reference Toyin

ample Date: 22 May-15				ol: EPA/6 al: Zinc si	00/R-95/136 (1995) ulfate	Sample Source: Reference Toxicant Sample Station: Zinc sulfate	
C-μg/L	Code	Rep	Pos	# Counted	# Normal		Notes
			1	100	96		
			2	(00)	53	6100=AC	
~			3	100	0		
			4	100	99		
			5	100	99		
			6	100	0	O QCEAC	
			7	loo	97		
			8	100	99		
			9	100	97		
			10	100	99		
			11	100	99		
			12	160	99		
			13	loo	38	34 QC=4C	
			14	(60	57		
			15	100	99	ł.	
			16	100	99		
			17	100	36		
			18	100	97		
			19	100	99		
			20	100	6		
			21	100	100		
			22	100	100		
			23	(00	97		
			24	100	98		
			25	100			
			. 26	100	100	99 OCAC	
			27	100	0		
			28	100	0		
			29	100	55		
			30	100	aa		

CETIS Test Data Worksheet

Report Date: Test Code: 19 May-15 15:20 (p 1 of 1) 08-9621-2840/150520hrdv

Red Abalone Larval Development Test Nautilus Environmental (CA) Start Date: 20 May-15 150520hrdv Species: Haliotis rufescens Sample Code: End Date: 22 May-15 Protocol: EPA/600/R-95/136 (1995) Sample Source: Reference Toxicant Sample Date: 20 May-15 Material: Zinc sulfate Sample Station: Zinc sulfate # Normal C-µg/L Code Rep Pos # Counted Notes LC LC LC LC LC COJ

QCAL

SIMIS

Analyst:___

QA: ACE/29/15

Marine Chronic Bioassay

Water Quality Measurements

Client: Internal	Test Species: Haliotis rufescens
Sample ID: ZnSO ₄	Start Date/Time: 5/20/2015 1415
Test ID: 150520hrdv	End Date/Time: 5/22/2015 1555

Concentration (μg/L)	Salinity (ppt)			Temperature (°C)			Dissolved Oxygen (mg/L)			pH (pH units)		
	0	24	48	0	24	48	0	24	48	0	24	48
Lab Control	33.3	33.3	33.6	157	15.4	15.5	8.4	8.3	83	8,06	8.06	806
10	33.3	35.3	33.8	15.7	15.1	15.0	8.3	8.3	8.3	8.05	8.07	807
18	33.3	33,4	33.8	157	5.0	15.1	8.3	8.3	8.3	8.06	8.08	808
32	33.3	33.3	33.8	157	15.0	15.0	8.3	8.3	84	8.06	8.08	808
56	33.2	33.3	33.7	15:7	isi	15.1	8.3	8.3	કે. પ	8.06	807	808
100	33-1	33.2	33.6	15.7	15.2	150	8.2	83	8.4	8.07	જ.08	8.08
AW.			1									
-												

		0	24	48	Dilution calc	s. (final vo	<u>lume 500 r</u>	nL):		
Technician Initials:	WQ Readings:	AC	A6	AD	Conc.	10	18	32	56	100
	Dilutions made by:				Vol. Zn stock (mL):	0-50	0,90	1.6	2.8	5.0
					Zn Stock Concentration	n (μg/L):		10,00	0	
Comments:	0 hrs:									·
	24 hrs:									
	48 hrs:									
QC Check:	AC ST	4/15				Fir	nal Review	: MP 6	1115	
									•	

Client:	Internal	Test Species: Haliotis rufe	escens
Sample ID:	Zn 804	Start Date/Time: 5/20/2015	1415
Test No.:	150520 hrdV	End Date/Time: <u>5/22/2015</u>	1555
Animal Source/Dat	e Received: American Abalone/ ちっしょう		·
Number of abalone	e and condition upon receipt/holding:		
Males:	4, spawned 5/19/15 in holding	·	
Females:	4		

	Males:	Females:
Tris & peroxide addition time	1035	1030
Spawn time	1312	1314
Number of spawners	3	2-4
Condition of spawn (light, moderate, heavy)	moderate	heavy
Fertilization time	132	5

Embryo counts (per 0	5 ml)
1	124.
2	160
3	292
Mean	192

Time of test Initiat	ion: 1415	48 hr. QC <u>97 /.</u>
Technician Initia	als: _Ac	
Comments:		
QC Check:	AC 5/29/15	Final Review: KFP 6 11 5

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



CETIS Summary Report

Report Date:

17 Nov-14 14:05 (p 1 of 1)

Test Code: 141030spdv | 16-2563-7748

								rest Code:			0-2563-774
Echinoid Emb	oryo-Larval Dev	elopment	Test						Nautilu	s Environn	nental (CA
Batch ID: Start Date: Ending Date: Duration:	14-1104-4410 30 Oct-14 15:0 02 Nov-14 17:0 74h	0 Pr 00 Sp	st Type: otocol: pecies: purce:	Development EPA/600/R-95/ Strongylocentro Pt. Loma		itus			atural Seawal	ter	
Sample ID:	20-7882-8985	Co	de:	141030spdv				Client: In	ternal		
Sample Date:	30 Oct-14	Ma	aterial:	Copper chloride	e			Project:			
Receive Date:	30 Oct-14	Sc	urce:	Reference Toxi	icant			•			
Sample Age:	15h	St	ation:	Copper Chlorid	е						
Comparison S	Summary										
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method			
17-9042-1561	Development F	Rate	<2.5	2.5	NA	3.24%		Dunnet	Multiple Com	parison Tes	st
Point Estimate	e Summary					***************************************					
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Method			
00-5600-3113	Development R	late	EC50	6.16	5.917	6.413		Trimme	d Spearman-l	Kärber	, 110, 100
Test Acceptab	oility										
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Overlag	Decision		
00-5600-3113	Development R	ate	Contro	ol Resp	0.964	0.8 - NL	-	Yes	Passes A	cceptability	Criteria
17-9042-1561	Development R			ol Resp	0.964	0.8 - NL		Yes	Passes A	cceptability	Criteria
17-9042-1561	Development R	ate	PMSD)	0.03243	NL - 0.25		No	Passes A	cceptability	Criteria
Development	Rate Summary										
	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0			0 0 0 4	0.055	0.973	0.00					0.00/
	Lab Control	5	0.964	0.955	0.010	0.93	0.99	0.01077	0.02408	2.5%	0.0%
2.5	Lab Control	5	0.856	0.955	0.8683	0.93	0.99 0.88	0.01077 0.0147	0.02408 0.03286	2.5% 3.84%	0.0% 11.2%
2.5 5	Lab Control	5 5	0.856 0.704	0.8437 0.6794					0.03286		
2.5 5 10	Lab Control	5 5 5	0.856 0.704 0.114	0.8437 0.6794 0.1054	0.8683	0.82	0.88	0.0147	0.03286	3.84%	11.2%
2.5 5 10 20	Lab Control	5 5 5 5	0.856 0.704 0.114 0	0.8437 0.6794	0.8683 0.7286	0.82 0.61	0.88 0.78	0.0147 0.02943	0.03286 0.0658	3.84% 9.35%	11.2% 26.97%
2.5 5 10 20 40	Lab Control	5 5 5 5 5	0.856 0.704 0.114 0	0.8437 0.6794 0.1054	0.8683 0.7286 0.1226	0.82 0.61 0.09	0.88 0.78 0.14	0.0147 0.02943 0.0103	0.03286 0.0658 0.02302	3.84% 9.35%	11.2% 26.97% 88.17%
2.5 5 10 20 40 80	Lab Control	5 5 5 5	0.856 0.704 0.114 0	0.8437 0.6794 0.1054 0	0.8683 0.7286 0.1226 0	0.82 0.61 0.09 0	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0%
2.5 5 10 20 40 80		5 5 5 5 5	0.856 0.704 0.114 0	0.8437 0.6794 0.1054 0	0.8683 0.7286 0.1226 0	0.82 0.61 0.09 0	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%
2.5 5 10 20 40 80 Development I	Rate Detail Control Type	5 5 5 5 5 5 7 8	0.856 0.704 0.114 0 0 0	0.8437 0.6794 0.1054 0 0 0	0.8683 0.7286 0.1226 0 0 0	0.82 0.61 0.09 0 0 0	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%
2.5 5 10 20 40 80 Development I C-µg/L	Rate Detail	5 5 5 5 5 5	0.856 0.704 0.114 0 0	0.8437 0.6794 0.1054 0 0	0.8683 0.7286 0.1226 0 0	0.82 0.61 0.09 0 0	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%
2.5 5 10 20 40 80 Development I C-µg/L	Rate Detail Control Type	5 5 5 5 5 5 7 8	0.856 0.704 0.114 0 0 0	0.8437 0.6794 0.1054 0 0 0	0.8683 0.7286 0.1226 0 0 0	0.82 0.61 0.09 0 0 0	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%
2.5 5 10 20 40 80 Development I C-µg/L 0 2.5	Rate Detail Control Type	5 5 5 5 5 5 5 7 8 8 8 9 1 0.93	0.856 0.704 0.114 0 0 0	0.8437 0.6794 0.1054 0 0 0 0 Rep 3 0.99	0.8683 0.7286 0.1226 0 0 0 Rep 4	0.82 0.61 0.09 0 0 0 Rep 5	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%
2.5 5 10 20 40 80 Development I C-µg/L 0 2.5	Rate Detail Control Type	5 5 5 5 5 5 5 7 8 8 8 93 0.88	0.856 0.704 0.114 0 0 0 Rep 2 0.98 0.88	0.8437 0.6794 0.1054 0 0 0 0 Rep 3 0.99 0.82	0.8683 0.7286 0.1226 0 0 0 Rep 4 0.97 0.82	0.82 0.61 0.09 0 0 0 Rep 5 0.95 0.88	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%
2.5 5 10 20 40 80 Development I	Rate Detail Control Type	5 5 5 5 5 5 5 7 8 8 0.93 0.88 0.78	0.856 0.704 0.114 0 0 0 Rep 2 0.98 0.88 0.61	0.8437 0.6794 0.1054 0 0 0 0 Rep 3 0.99 0.82 0.72	0.8683 0.7286 0.1226 0 0 0 Rep 4 0.97 0.82 0.67	0.82 0.61 0.09 0 0 0 Rep 5 0.95 0.88 0.74	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%
2.5 5 10 20 40 80 Development I C-µg/L 0 2.5 5	Rate Detail Control Type	5 5 5 5 5 5 5 7 8 8 0.93 0.88 0.78 0.09	0.856 0.704 0.114 0 0 0 Rep 2 0.98 0.88 0.61 0.12	0.8437 0.6794 0.1054 0 0 0 0 Rep 3 0.99 0.82 0.72 0.13	0.8683 0.7286 0.1226 0 0 0 Rep 4 0.97 0.82 0.67 0.09	0.82 0.61 0.09 0 0 0 Rep 5 0.95 0.88 0.74 0.14	0.88 0.78 0.14 0	0.0147 0.02943 0.0103 0	0.03286 0.0658 0.02302 0	3.84% 9.35%	11.2% 26.97% 88.17% 100.0% 100.0%

Analyst: <u>VB</u> QA: <u>ACII/18</u>/14

Report Date:

17 Nov-14 11:26 (p 1 of 2)

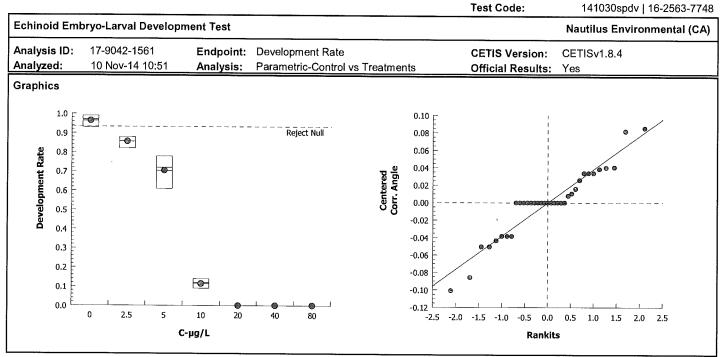
•	\·
est Code:	141030spdv 16-2563-774

							Test	Code:	1410	030spdv 1	6-2563-7748
Echinoid En	nbryo-Larval De	velopment	Test						Nautilu	s Environr	nental (CA)
Analysis ID:	17-9042-1561	Er	idpoint: De	velopment F	Rate		CET	IS Version	: CETISv	1.8.4	····
Analyzed:	10 Nov-14 10	:51 A r	alysis: Pa	rametric-Co	ntrol vs Trea	tments	Offic	ial Results	s: Yes		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		NOEL	LOEL	TOEL	TU	PMSD
Angular (Cor	rected)	NA	C > T	NA	NA		<2.5	2.5	NA		3.24%
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*		5.692	2.227	0.080 8	<0.0001	CDF	Significar	<u> </u>		
	5*		10.84	2.227	0.080 8	<0.0001	CDF	Significar			
	10*		28.95	2.227	0.080 8	<0.0001	CDF	Significar			
ANOVA Tabl	е	V-100									
Source	Sum Sq	uares	Mean Squ	uare	DF	F Stat	P-Value	Decision	(α:5%)		
Between	3.07234		1.024113		3	313.9	<0.0001	Significar	nt Effect		****
Error	0.052206	649	0.0032629	905	16			-			
Total	3.124547	,			19						
Distributiona	l Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(a:1%)			
Variances	Bartlett I	Equality of \	/ariance	2.003	11.34	0.5718	Equal Var	iances			
Distribution	Shapiro-	Wilk W Nor	mality	0.9359	0.866	0.2004	Normal Di	stribution			
Developmen	t Rate Summary	,					T-1				
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.964	0.9341	0.9939	0.97	0.93	0.99	0.01077	2.5%	0.0%
2.5		5	0.856	0.8152	0.8968	0.88	0.82	0.88	0.0147	3.84%	11.2%
5		5	0.704	0.6223	0.7857	0.72	0.61	0.78	0.02943	9.35%	26.97%
10		5	0.114	0.08541	0.1426	0.12	0.09	0.14	0.0103	20.19%	88.17%
20		5	0	0	0	0	0	0	0		100.0%
40		5	0	0	0	0	0	0	0		100.0%
80		5	0	0	0	0	0	0	0		100.0%
Angular (Cor	rected) Transfo	med Sumr	mary				-			-	
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.389	1.307	1.471	1.397	1.303	1.471	0.02968	4.78%	0.0%
2.5		5	1.183	1.126	1.241	1.217	1.133	1.217	0.02068	3.91%	14.8%
		5	0.9973	0.9081	1.087	1.013	0.8963	1.083	0.03216	7.21%	28.19%
5		3	0.00.0								
		5	0.3431	0.2976	0.3885	0.3537	0.3047	0.3835	0.01637	10.67%	75.3%
5 10 20				0.2976 0.05001	0.3885 0.05003	0.3537 0.05002	0.3047 0.05002	0.3835 0.05002	0.01637 0		
10		5	0.3431							10.67% 0.0% 0.0%	75.3% 96.4% 96.4%

Report Date:

17 Nov-14 11:26 (p 2 of 2)

141030spdv | 16-2563-7748



Report Date:

17 Nov-14 11:26 (p 1 of 1)

Test Code: 141030spdv | 16-2563-7748

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Analysis ID: Analyzed:

00-5600-3113 10 Nov-14 10:52

Endpoint: Development Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version:

CETISv1.8.4

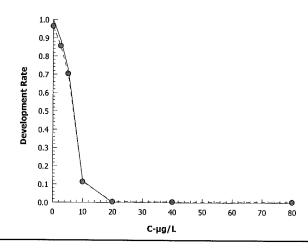
Official Results: Yes

Trimmed	Spearman-Kärber	Estimates
---------	-----------------	-----------

Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0.036	11.20%	0.7896	0.008736	6.16	5.917	6.413

Developm	nent Rate Summar	y			Cal	culated Varia	ite(A/B)				
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	A	В
0	Lab Control	5	0.964	0.93	0.99	0.01077	0.02408	2.5%	0.0%	482	500
2.5		5	0.856	0.82	0.88	0.0147	0.03286	3.84%	11.2%	428	500
5		5	0.704	0.61	0.78	0.02943	0.0658	9.35%	26.97%	352	500
10		5	0.114	0.09	0.14	0.0103	0.02302	20.19%	88.17%	57	500
20		5	0	0	0	0	0		100.0%	0	500
40		5	0	0	0	0	0		100.0%	0	500
80		5	0	0	0	0	0		100.0%	0	500

Graphics



Protocol:

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Test Type: Development

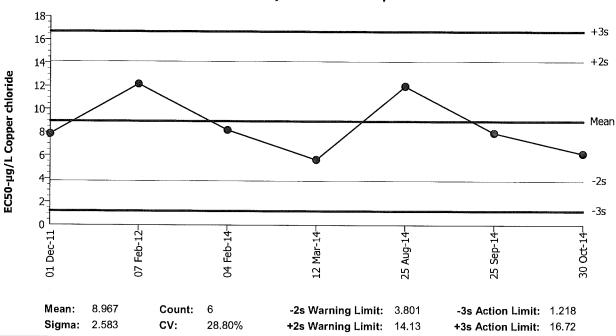
Organism: Strongylocentrotus purpuratus (Purpl

Endpoint: Development Rate

Material: Copper chloride

Source: Reference Toxicant-REF

Echinoid Embryo-Larval Development Test



Quali	ty Con	trol Data	a								
Point	Year	Month	Day	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID	
1	2011	Dec	1	7.852	-1.115	-0.4316			07-8281-3338	17-7265-1210	
2	2012	Feb	7	12.17	3.203	1.24			12-7990-2055	07-7286-1647	
3	2014		4	8.195	-0.7721	-0.2989			14-5835-0600	07-6515-7453	
4		Mar	12	5.64	-3.327	-1.288			16-4219-4884	02-2584-6206	
5		Aug	25	11.99	3.028	1.172			11-2159-2788	17-1870-3217	
6		Sep	25	7.95	-1.017	-0.3938			09-4928-6784	08-7961-1534	
7		Oct	30	6.16	-2.807	-1.087			16-2563-7748	00-5600-3113	

CETIS Test Data Worksheet

Report Date: Test Code: 28 Oct-14 16:04 (p 1 of 1) 16-2563-7748/141030spdv

Echinoid En	nbryo-l	arval	Deve	lopment Te	st			Nautilus Environmental (CA
Start Date: End Date: Sample Date	30 (25 0 3 (30 (e: 30 (Oct-14	10114	Specie Protoc Materi	-	ylocentrotus p 00/R-95/136 (r chloride	Sample Code: Sample Source: Sample Station:	141030spdv Reference Toxicant Copper Chloride
C-µg/L	Code		Pos	# Counted	# Normal		Notes	
			1	(00	Ø			
			2	100	Ø CA	1115114	 	
			3	loo	15 Q18	39		
			4	100	97			
			5	100	93			
			6	(00)	Ø		 	
			7	(oo	\mathscr{D}		 	
			8	100	Ø		 	
			9	(00	98			
			10	(06	88			
			11	100	72		1000//5000	
			12	(00	Ø			
			13	loo	74			
			14	100	Ø		101010	
			15	100	82			
	-		16	(00	67			
			17	loo	61			
	-		18 19	lou	Ø		 	
	-		20	100			 	
	-		21	100	Ø			
	-		22	100	99		 	
			23	100	14		 	· · · · · · · · · · · · · · · · · · ·
			24	100	1 1		 	
	-		25		78 ø		 	100 per
			26	(00	Ø			
			27	loo ioo	82	-	 	
	-		28		02		 	
				(00 (00	95			
			30	100	(2			
**			31	100	0		 	
			32	100	9		 	1 / 2 / 2 · · · · · · · · · · · · · · · ·
	+		33	(00	88		 	
			34	(00)	13		 	
	+	-	35	toc	වරු		 	

CETIS Test Data Worksheet

Report Date: Test Code:

28 Oct-14 16:04 (p 1 of 1) 16-2563-7748/141030spdv

Echinoid Em	bryo-l	_arval	Deve	lopment Te	st		Nautilus Environmental (CA)
Start Date: End Date: Sample Date	30 (-93-N 21-8 30 (Oct-14 Voy-14 Oct-14	1 1 1 10 1 10	Specie Protoc Materi		00/R-95/136 (1995) Sample Source:	141030spdv Reference Toxicant Copper Chloride
C-µg/L	Code		Pos	# Counted	# Normal	Notes	
0	LC	1	5				
0	LC	2	9				
0	LC	3	22	10 ²	99		
0	LC	4	4		•		
0	LC	5	29				
2.5		1	10				
2.5		2	35				
2.5		3	15				
2.5		4	27				
2.5		5	33				
5		1	24				
5		2	17	~			
5		3	11	100	78		
5		4	16				
5		5	13				
10		1	3	100	25		
10		2	30				
10		3	34				
10		4	32				
10		5	23				
20		1	6				
20		2	26				
20		3	2	100	0		
20		4	18				
20		5	28				
40		1	7				
40		2	8				:
40		3	31				
40		4	14				
40		5	19				
80		1	21				
80		2	12	,			
80	1	3	25				
80		4	20	100	0		
80		5	1				

Analyst VCR QA: KBI 10/14

OCEAC

Marine Chronic Bioassay

Water Quality Measurements

Client: Internal	Test Species:	S. purpuratus
Sample ID: CuCl ₂	Start Date/Time:	10/30/2014 15:00
Test No.: 141030spdv	End Date/Time:	11/3/2014 7:00

Concentration			Salinity (ppt)		W	p Heldre	† Te	mperati (°C)	ıre			Diss	olved Ox (mg/L)	kygen			(pH pH units	s)	· · · · · · · · · · · · · · · · · · ·
	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
Lab Control	33,5	33.3	33.2	32.9	1	14,5	157	15.8	16.2		8-8	7.7	7.9	7.7		8,60	801	8.06	7.85	
2.5	33.6	33. b	33.4	33.4		14.5	15.5	15.4	16.1		8.7	7.1	7.9	7.9		3.02	8.01	8.05	7.91	
5	33.6	33.L	33.5	33-4		14,6	13.2	15.2	15.2		8.7	8.1	7.8	7.5		4.01	802	8.06	7.94	
10	33,5	33.7	33.1	33.8		14,6	15,2	15.0	15.3		8.6	7.8	7.8	7.5		8.60	8.02	807	7.94	
20	33.6	33 b	33.7	33.8		14.6	15.3	15.0	16.0		8.7	8.0	7.9	7.7	**	8.01	801	8.07	7.94	
40	33.5	33.b	33.60	33.6		14,6	15.3	14.8	15-0		8.7	4.0	8.0	7.7	-	8.03	8.63	8.07	7.95	
80	33,4			33.6		14.7	15.3	14.8	15.0	/	8.7	8.)	8.1	7.6		8.04	8.0 Z	8.08	7.95	/

Technician Initials:	W
•	Dilution

	0	24	48	72	96	. 1
WQ Readings:	ど	AL	FP	.AW		Emin
utions made by:	AC					

High conc. made (μg/L): Vol. Cu stock added (mL): Final Volume (mL): Cu stock concentration (μg/L):

	_
80	
4,4	
500	
1,050	

Comments:	0 hrs:			
	24 hrs:			
	48 hrs:	-		
	72 hrs:			
QC Ch	eck: V.B 111/2/14	Final Review:	AC 11/18/14	

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

Marine Chronic Bioassay

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

Echinoderm Larval Development Worksheet

Client: Sample ID: Test No.:	Internal Cucia 1410305pdv	Start Date/Time: 10/30/2014 / 15 . CO End Date/Time: 11/3/2014 17 00 Species: S. purpuratus
Tech initials: Injection Time:	PA 	Animal Source: Point Loma Date Collected: 10(6)
Sperm Absorbance at 4	00 nm: 0,823 (target range of 0.8 - 1.0 for density of 4)	x10 ⁶ sperm/ml)
Eggs Counted:	Mean: 77.7 x 50 = 110 Wean: 77.7 x 50 = 110 (target counts of 26 eggs per vertical pass on Sed slide for a final density of 1000 eggs/ml)	eggs/ml gwick-Rafter
Initial density: Final density:	1000 eggs/ml = 1.0 dilution factor 1.0 part egg stock 0, 11 parts seawater	egg stock 160 ml seawater ml
Prepare the egg stock a part) and 125 ml of dilut	according to the calculated dilution factor. For example, if the dilution fion water (1.25 parts).	actor is 2.25, use 100 ml of existing stock (1
Volume of Sperm stock	needed to fertilize eggs:	
Egg Stock (mL) = Sperm Stock (μL) = Egg/Sperm Ratio =	= 725 	
Fertilization Time:	1995	
Embryo Stock Fertilizati 10 minutes (1st fert.) 20 minutes (2nd fert. If I	$\begin{array}{c cccc} \hline \text{Time} & \hline \text{Fert.} & \hline \text{Unfert.} & \% \\ \hline \hline 14.55 & \underline{100} & \bigcirc & \underline{i00} \\ \hline \end{array}$	- - -
Test Initiation Time:	Embryo Stock Added: 0.25 m	n <u>l</u>
Test Termination:	No. No. %	
72-hour QC check ^a End of test QC check	Normal Abnormal Normal 97 3 97 3 97	
Comments:	^a If the embryo development does not meet the mean test acceptal developed, continue the test up to 96-hrs (ASTM 1999).	bility criterion of 80% normally
QC Check:	KB 1116114	Final Review: A.C. 111814

CETIS Summary Report

Report Date: Test Code: 05 Aug-15 17:32 (p 1 of 1) 150722spdv | 11-3114-3362

Brine: Not Age: Client: Inte Project: Method Dunnett M	Nautilus tural Seawater t Applicable ernal Multiple Compa	r parison Tes	st
Diluent: Nati Brine: Not Age: Client: Inte Project: Method Dunnett M Method Trimmed	t Applicable ernal Multiple Compa	parison Tes	st
Method Dunnett M Method Trimmed	Multiple Comp Spearman-Kä		st
Method Dunnett M Method Trimmed	Spearman-Kä		st
Dunnett M Method Trimmed	Spearman-Kä		st
Dunnett M Method Trimmed	Spearman-Kä		st
Dunnett M Method Trimmed	Spearman-Kä		st
Method Trimmed	Spearman-Kä		st
Trimmed		ärber	
Trimmed		ärber	
		ärber	
Overlap	Decision		
Overlap	Decision		
Yes	Passes Acc	ceptability	Criteria
Yes	Passes Acc	ceptability	Criteria
No	Passes Acc	ceptability	Criteria
Std Err	Std Dev	CV%	%Effect
0.003742		0.85%	0.0%
0.003162		0.73%	1.82%
0.003162	0.007071	0.76%	5.87%
0.01503	0.03362	5.66%	39.88%
0.003742		104.6%	99.19%
0	0		100.0%
0	0		100.0%
	0	0 0	0 0

Report Date: 05 Aug-15 17:31 (p 1 of 2)
Test Code: 150722spdv | 11-3114-3362

							Test	Code:	1507	22spdv 11	1-3114-3362
Echinoid Em	bryo-Larval Dev	elopme	nt Test						Nautilus	Environn	nental (CA)
Analysis ID: Analyzed:	08-4881-4252 05 Aug-15 17:	31	•	velopment R ametric-Cor		tments		IS 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		1.1%	<2.5	2.5	NA	
Dunnett Mult	iple Compariso	n Test									
Control	vs C-μg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(a:5%)		
Lab Control	2.5*		3.39	2.305	0.045 8	0.0051	CDF	Significant			
	5*		8.218	2.305	0.045 8	<0.0001	CDF	Significant	Effect		
	10*		29.86	2.305	0.045 8	<0.0001	CDF	Significant	Effect		
	20*		70.35	2.305	0.045 8	<0.0001	CDF	Significant	Effect		
ANOVA Table											***************************************
Source	Sum Squ	ıares	Mean Squ	ıare	DF	F Stat	P-Value	Decision(z:5%)		
Between	6.535633		1.633908		4	1710	<0.0001	Significant	Effect		
Error	0.019112	2	0.0009556	3102	20	_					
Total	6.554745				24						
Distributiona	l Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision((a:1%)			
Variances	Bartlett B	Equality	of Variance	4.658	13.28	0.3242	Equal Var	iances			
Distribution	Shapiro-	Wilk W I	Normality	0.9459	0.8877	0.2026	Normal Di	stribution			
Development	Rate Summary										
C-µg/L	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Мах	Std Err	CV%	%Effect
0	Lab Control	5	0.988	0.9776	0.9984	0.99	0.98	1	0.003742	0.85%	0.0%
2.5		5	0.97	0.9612	0.9788	0.97	0.96	0.98	0.003162	0.73%	1.82%
5		5	0.93	0.9212	0.9388	0.93	0.92	0.94	0.003162	0.76%	5.87%
10		5	0.594	0.5523	0.6357	0.59	0.55	0.64	0.01503	5.66%	39.88%
20		5	0.008	0	0.01839	0.01	0	0.02	0.003742	104.6%	99.19%
Angular (Cori	ected) Transfo	med Su	ımmary								
C-μg/L	Control Type	Coun		95% LCL	95% UCL	Median	Min	Мах	Std Err	CV%	%Effect
0	Lab Control	5	1.464	1.417	1.511	1.471	1.429	1.521	0.01699	2.6%	0.0%
2.5		5	1.398	1.372	1.424	1.397	1.369	1.429	0.009421	1.51%	4.53%
5		5	1.303	1.286	1.321	1.303	1.284	1.323	0.006214	1.07%	10.98%
10		5	0.8801	0.8376	0.9227	0.8759	0.8355	0.9273	0.01534	3.9%	39.88%
20		5	0.08845	0.04003	0.1369	0.1002	0.05002	0.1419	0.01744	44.09%	93.96%
Development											
C-μg/L	Control Type	Rep 1	· · · · · · · · · · · · · · · · · · ·	Rep 3	Rep 4	Rep 5					
0	Lab Control	0.99	0.98	0.98	0.99	1					
2.5		0.97	0.98	0.97	0.97	0.96					
5		0.93	0.94	0.93	0.92	0.93					
10		0.55	0.59	0.58	0.61	0.64					
20		0.01	0	0.02	0	0.01					
Angular (Corr	ected) Transfor	med De	etail								
C-μg/L	Control Type	Rep 1		Rep 3	Rep 4	Rep 5	4000		LANGUE,		
0	Lab Control	1.471		1.429	1.471	1.521					
2.5		1.397		1.397	1.397	1.369					
5		1.303		1.303	1.284	1.303					
10		0.835		0.8657	0.8963	0.9273					
20		0.100	2 0.05002	0.1419	0.05002	0.1002					

000-089-170-2 CETIS™ v1.8.7.20

0 LC

2.5

C-µg/L

10

20

Report Date: Test Code:

05 Aug-15 17:32 (p 2 of 2) 150722spdv | 11-3114-3362

Echinoid Embryo-Larval Development Test Nautilus Environmental (CA) Analysis ID: 08-4881-4252 Endpoint: Development Rate **CETIS Version:** CETISv1.8.7 Analyzed: 05 Aug-15 17:31 Parametric-Control vs Treatments Analysis: Official Results: Yes Graphics 1,0 0.06 Reject Null 0.9 0.05 0,04 8.0 0,03 0.6 0,5 0.00 0.4 -0,01 0,3 -0.02 -0.03 0.1 -0.04 -0.05

-2,0

-1.0

Rankits

Report Date: **Test Code:**

05 Aug-15 17:32 (p 1 of 1)

150722spdv | 11-3114-3362

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Analysis ID: Analyzed:

14-3278-2605 05 Aug-15 17:31 Endpoint: Development Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version: Official Results: Yes

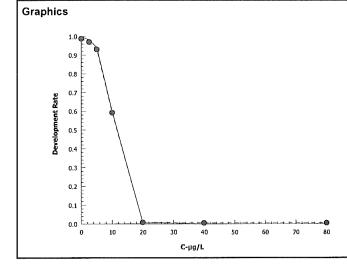
CETISv1.8.7

Trimmed	Spearman-Kärber	Estimates
---------	-----------------	-----------

Threshold Option Threshold Sigma EC50 95% LCL 95% UCL Trim Mu 0.012 Control Threshold 1.82% 1.017 0.007669 10.41 10.05 10.78

Developm	nent Rate Summary	Calculated Variate(A/B)									
C-µg/L	Control Type	Count	Mean	Min	Max Std Err		Std Dev CV%		%Effect	Α	В
0	Lab Control	5	0.988	0.98	1	0.003742	0.008367	0.85%	0.0%	494	500
2.5		5	0.97	0.96	0.98	0.003162	0.00707	0.73%	1.82%	485	500
5		5	0.93	0.92	0.94	0.003162	0.00707	0.76%	5.87%	465	500
10		5	0.594	0.55	0.64	0.01503	0.03362	5.66%	39.88%	296	500
20		5	0.008	0	0.02	0.003742	0.008367	104.6%	99.19%	4	500
40		5	0	0	0	0	0		100.0%	0	500
80		5	0	0	0	0	0		100.0%	0	500

Developm	nent Rate Detail					
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	Lab Control	0.99	0.98	0.98	0.99	1
2.5		0.97	0.98	0.97	0.97	0.96
5		0.93	0.94	0.93	0.92	0.93
10		0.55	0.59	0.58	0.61	0.64
20		0.01	0	0.02	0	0.01
40		0	0	0	0	0
80		0	0	0	0	0



Analyst: AC QA: VBB 6 5

Report Date:

05 Aug-15 17:32 (1 of 1)

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Test Type: Development

2.465

Sigma:

Protocol: EPA/600/R-95/136 (1995)

Organism: Strongylocentrotus purpuratus (Purpl

Endpoint: Development Rate

25.00%

CV:

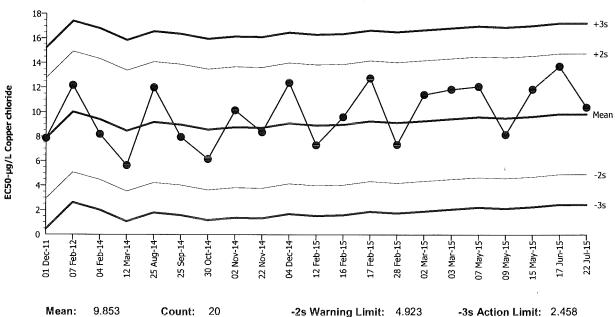
Material: Copper chloride

Source:

Reference Toxicant-REF

+3s Action Limit: 17.25

Echinoid Embryo-Larval Development Test



+2s Warning Limit: 14.78

Quali	ty Con	trol Data	a								
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2011	Dec	1	17:40	7.852	-2.001	-0.8117			07-8281-3338	17-7265-1210
2	2012	Feb	7	15:45	12.17	2.317	0.94			12-7990-2055	07-7286-1647
3	2014		4	14:15	8.195	-1.658	-0.6727			14-5835-0600	07-6515-7453
4		Mar	12	18:20	5.64	-4.213	-1.709			16-4219-4884	02-2584-6206
5		Aug	25	19:10	11.99	2.142	0.8689			11-2159-2788	17-1870-3217
6		Sep	25	17:30	7.95	-1.903	-0.7721			09-4928-6784	08-7961-1534
7		Oct	30	15:00	6.16	-3.693	-1.498			16-2563-7748	00-5600-3113
8		Nov	2	17:00	10.14	0.2904	0.1178			05-9121-9644	01-7691-0405
9			22	16:45	8.358	-1.495	-0.6066			06-8410-0954	18-0830-4230
10		Dec	4	14:10	12.4	2.547	1.033			15-8916-5237	11-1209-5739
11	2015	Feb	12	14:25	7.308	-2.545	-1.033			21-1011-3319	03-9708-7225
12			16	18:25	9.603	-0.2503	-0.1015			12-8378-8021	11-4706-6613
13			17	15:12	12.77	2.912	1.181			12-8622-3584	03-8986-1540
14			28	15:00	7.345	-2.508	-1.018			04-6319-9803	10-6603-8112
15		Mar	2	18:50	11.43	1.572	0.6378			14-8443-1027	00-7020-5756
16			3	19:43	11.86	2.008	0.8145			06-2006-0576	21-3581-4807
17		May	7	15:55	12.1	2.243	0.9098			18-5128-4001	14-3953-8938
18			9	14:30	8.171	-1.682	-0.6822			08-8892-0966	06-5858-7490
19			15	16:37	11.87	2.017	0.8183			06-5352-8837	20-0018-4829
20		Jun	17	16:40	13.75	3.895	1.58			10-5901-9632	03-4943-7748
21		Jul	22	16:10	10.41	0.5562	0.2257			11-3114-3362	14-3278-2605

Report Date:

21 Jul-15 10:57 (p 1 of 1)

Test Code: 11-3114-3362/150722spdv **Echinoid Embryo-Larval Development Test** Nautilus Environmental (CA) Start Date: 22 Jul-15 Species: Strongylocentrotus purpuratus Sample Code: 150722spdv 25 Jul-15 End Date: Protocol: EPA/600/R-95/136 (1995) Sample Source: Reference Toxicant Sample Date: 22 Jul-15 Material: Copper chloride Sample Station: Copper Chloride C-µg/L Code Rep Pos # Counted # Normal Notes A6 8/5/15 (00 \bigcirc O Ò O \circ

Analyst: AC QA: AC \$15

 \Diamond

CETIS Test Data Worksheet

Report Date: Test Code:

21 Jul-15 10:57 (p 1 of 1) 11-3114-3362/150722spdv

Echinoid Em	bryo-	Larva	i Deve	elopment Te	st			Nautilus Environmental (C.
Start Date: End Date: Sample Date	25 .	Jul-15 Jul-15 Jul-15				gylocentrotus purpuratus 600/R-95/136 (1995) er chloride	Sample Code: Sample Source: Sample Station:	150722spdv Reference Toxicant Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Normal		Notes	
0	LC	1	16	100	100	AC7 28/15		
0	LC	2	35			7,5		
0	LC	3	2					
0	LC	4	28					
0	LC	5	30				×	
2.5		1	20	100	97	AC 7/28/15	Hart Sarahada	
2.5		2	31			1,201.3		
2.5		3	4			777444	110110 504444	
2,5		4	1					
2.5		5	21	· · · · · · · · · · · · · · · · · · ·				
5		1	10	100	95	AC 7/28/15		
5		2	26		1 3	2001/1		1100 = 4
5		3	23					
5		4	13		-			Value of the second of the sec
5		5	25					
10		1	5	100	H9	AC7/28/15		
10		2	12	100		, , , , of 10		
10		3	24					
10		4	33					
10		5	19			***************************************		
20		1	17	100	0	AC 7/28/15		
20		2	22					
20		3	7					
20		4	15					
20		5	9			The second secon		
40		1	18	100	0			
40		2	11	1 40	**eaget#		·	
40		3	8					
40		4	3					Washes and
40		5	14			10		
80		1	32	100	0			
80		2	34		Nagara -			

618

3

4

5 6

29

27

80

80

80

Marine Chronic Bioassay

Water Quality Measurements

Client: Internal

Sample ID: CuCl₂

Test No.: 150722spdv

Test Species: S. purpuratus

Start Date/Time: 7/22/2015 | 6 ()

End Date/Time: <u>7/25/2015</u> \\(\(\sigma\)\(\)

Concentration			linity		Temperature				Dissolved Oxygen				рН			
(μg/L)	(ppt)				(°C)			(mg/L)				(pH units)				
	0	24	48	72	0	24	48	72	0	24	48	72	0	24	48	72
Lab Control	33.3	33,4	33.3	33,2	14.9	14,8	14.7	15.5	8.9	8.0	7.9	8.2	8.09	6.67	8,05	8.03
2.5	33.4	33.5	33.5	33.4	146	14.5	14.5	15.0	8-8	છ.1	8.0	8.4	8.08	8.07	4.06	8.05
5	33.5	33.6	33.6	33.5	14.8	14.4	14,4	14.8	8-8	ų.2	8.0	8.4	8-08	6.06	8.06	8.05
10	33.5	33.5	33.6	33.5	145	14.3	14.3	14.9	88	8.2	3.1	8.4	8.08	€.07	8,06	8.05
20	33.5	33.6	33,6	33.5	14.9	14.6	14.5	15.1	8.7	8.2	8.0	8.3	808		8.07	8.06
40	33.4	33.4	33.5	33.4	14.8	14.5	14.4	15-1	8.7	8.3	8.1	8.4	8.09	8.08	8.07	8.ae
80	333	33.3	33,4	33.3	14.7	14,4	14.3	15.1	8.7	٤.3	8,1	8.4	8.09	40.8	8,07	8:06

Technician Initials:

	0	24	48	72
WQ Readings:	AD	ÉG	4	14D
Dilutions made by:	- AC			

High conc. made (μg/L):

Vol. Cu stock added (mL):

Final Volume (mL):

Cu stock concentration (μg/L):

80

4.5

Final Volume (mL):

80

4.5

Comments:

Ohrs: Hach Sension 5 Salinity muter

24 hrs:

48 hrs:

72 hrs:

QC Check: AC 8(3) 15

Final Review: KB 8 6 15

Marine Chronic Bioassay

Echinoderm Larval Development Worksheet

Client: Sample ID: Test No.: Tech initials: Injection Time: Sperm Absorbance at	PA/A/ 1507235001 PA/A/ 1525 ACRIBATE (target range of 0.8 - 1.0 for density)	Start Date/Time: 12915 160 End Date/Time: 12515 160 Species: S. purpuratus Date Collected: 1915
Eggs Counted:	Mean: 3 , 2 x 50 = 50 H Order (target counts of 20 eggs per vertical pass on slide for a final density of 1000 eggs/ml)	-99
Initial density: Final density:	$\frac{1560 \text{ eggs/ml}}{1000 \text{ eggs/ml}} = \frac{1.6 \text{ dilution factor}}{1.0 \text{ Stock}}$ 0.6 Sw	egg stock 50 ml seawater 30 60 ml
Prepare the egg stock a (1 part) and 125 ml of d	according to the calculated dilution factor. For example, if the dilution water (1.25 parts).	ution factor is 2.25, use 100 ml of existing stock
Add 100 μL sperm stoc	k per 100mL of egg stock. For example, if you have 60mL of eg	g stock, add 60μL sperm stock.
Embryo Stock Fertilizati	ion Checks (Initiate test only when fertilization is ≥90%):	
10 minutes (1st fert.) 20 minutes (2nd fert. If I	No. No. Time Fert. Unfert. 1665 98 2 9	%
Fertilization Time:	1555	
Test Initiation Time:	Lelo Embryo Stock Added: 0.2	25 ml
Test Termination: 72-hour QC check ^a End of test QC check	No. No. % Normal Abnormal Normal O(0)	
Comments:	^a If the embryo development does not meet the mean test accedeveloped, continue the test to 96-hrs (ASTM 1999).	eptability criterion of 80% normally
QC Check:	ACB 3 15	Final Review: KBB615



CETIS Summary Report

Report Date:

07 Aug-15 10:27 (p 1 of 1)

Test Code:

141030dedv | 14-4718-7348

Echinoid Embryo-Larval Development Test Nautilus Environment Batch ID: 00-8568-0963 Test Type: Development Analyst: Start Date: 30 Oct-14 16:20 Protocol: EPA/600/R-95/136 (1995) Diluent: Natural Seawater Ending Date: 02 Nov-14 17:20 Species: Dendraster excentricus Brine: Not Applicable Duration: 73h Source: Mission Bay Age: Sample ID: 02-2472-0325 Code: 141030dedv Client: Internal Sample Date: 30 Oct-14 Material: Copper chloride Project: Receive Date: 30 Oct-14 Source: Reference Toxicant Sample Age: 16h Station: Copper Chloride Comparison Summary Analysis ID Endpoint NOEL LOEL TOEL PMSD TU Method Point Estimate Summary Analysis ID Endpoint Level μg/L 95% LCL 95% UCL TU	ntal (CA)
Start Date: 30 Oct-14 16:20 Protocol: EPA/600/R-95/136 (1995) Ending Date: 02 Nov-14 17:20 Species: Dendraster excentricus Duration: 73h Source: Mission Bay Age: Sample ID: 02-2472-0325 Code: 141030dedv Sample Date: 30 Oct-14 Material: Copper chloride Receive Date: 30 Oct-14 Source: Reference Toxicant Sample Age: 16h Station: Copper Chloride Comparison Summary Analysis ID Endpoint NOEL LOEL TOEL PMSD TU Method Project: PMSD TU Method Dunnett Multiple Comparison Test Point Estimate Summary Analysis ID Endpoint Level µg/L 95% LCL 95% UCL TU Method	
Sample Date: 30 Oct-14 Receive Date: 30 Oct-14 Sample Age: 16h Station: Copper Chloride Comparison Summary Analysis ID Endpoint NOEL LOEL TOEL PMSD TU Method 07-8842-6412 Development Rate 10 >10 NA 5.16% Dunnett Multiple Comparison Test Project: Projec	
Analysis IDEndpointNOELLOELTOELPMSDTUMethod07-8842-6412Development Rate10>10NA5.16%Dunnett Multiple Comparison TestPoint Estimate SummaryAnalysis IDEndpointLevelμg/L95% LCL95% UCLTUMethod	
07-8842-6412 Development Rate 10 >10 NA 5.16% Dunnett Multiple Comparison Test Point Estimate Summary Analysis ID Endpoint Level μg/L 95% LCL 95% UCL TU Method	
Analysis ID Endpoint Level μg/L 95% LCL 95% UCL TU Method	
06-3337-3291 Development Rate EC50 13.95 13.81 14.09 Trimmed Spearman-Kärber	
Test Acceptability	Accessor and the second
Analysis ID Endpoint Attribute Test Stat TAC Limits Overlap Decision	
06-3337-3291 Development Rate Control Resp 0.93 0.8 - NL Yes Passes Acceptability Cri	iteria
07-8842-6412 Development Rate Control Resp 0.93 0.8 - NL Yes Passes Acceptability Cri	
07-8842-6412 Development Rate PMSD 0.05164 NL - 0.25 No Passes Acceptability Cri	iteria
Development Rate Summary	
С-µg/L Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV% %	%Effect
0 Lab Control 5 0.93 0.8983 0.9617 0.9 0.96 0.0114 0.0255 2.74% 0	0.0%
2.5 5 0.918 0.8884 0.9476 0.89 0.95 0.01068 0.02387 2.6% 1	1.29%
5 0.928 0.8788 0.9772 0.88 0.98 0.01772 0.03962 4.27% C	0.22%
10 5 0.908 0.8664 0.9496 0.87 0.95 0.01497 0.03347 3.69% 2	2.37%
20 5 0 0 0 0 0 0 0 1	100.0%
40 5 0 0 0 0 0 0 0 1	100.0%
80 5 0 0 0 0 0 0 0 1	100.0%
Development Rate Detail	
C-µg/L Control Type Rep 1 Rep 2 Rep 3 Rep 4 Rep 5	
0 Lab Control 0.91 0.93 0.96 0.9 0.95	
2.5 0.89 0.95 0.93 0.9 0.92	
5 0.9 0.95 0.88 0.98 0.93	
10 0.87 0.93 0.95 0.88 0.91	
20 0 0 0 0	
40 0 0 0 0 0	
80 0 0 0 0	

Report Date:

07 Aug-15 10:27 (p 1 of 2)

511 115 In the second s								Code:	141030dedv 14-4718-734			
Echinoid Em	nbryo-Larval Dev	velopment	Test				Nautilus	Environ	mental (C			
Analysis ID: Analyzed:	07-8842-6412 07 Aug-15 10:			velopment R ametric-Cor		ıtments		IS Version:	CETISv1 Yes	.8.7		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU	
Angular (Corr	rected)	NA	C > T	NA	NA		5.16%	10	>10	NA		
Dunnett Mul	tiple Compariso	n Test					MWW.	A CONTRACTOR OF THE SECOND				
Control	vs C-μg/L		Test Stat	Critical	MSD DE	P-Value	P-Type	Decision(a:5%)			
Lab Control	2.5		0.6118	2.227	0.086 8	0.4953	CDF	Non-Signif	icant Effect			
	5		-0.05363	2.227	0.086 8	0.7688	CDF	Non-Significant Effect				
	10		1.028	2.227	0.086 8	0.3211	CDF	Non-Signif	icant Effect			
ANOVA Tabl	е											
Source	Sum Squ	Jares	Mean Squ	ıare	DF	F Stat	P-Value	Decision(a:5%)		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	
Between	0.006063	994	0.0020213	331	3	0.5363	0.6641	Non-Signif	icant Effect			
Error	0.060302	:32	0.0037688	395	16							
Total	0.066366	31			19	*************************						
Distributiona	al Tests											
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)				
Variances	Bartlett I	Equality of \	Variance	1.669	11.34	0.6439	Equal Variances					
Distribution	Shapiro-	Wilk W No	rmality	0.9646	0.866	0.6394	Normal D	stribution				
Developmen	t Rate Summary	1		1 1 11 11 11 11 11 11 11 11 11 11 11 11								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effec	
0	Lab Control	5	0.93	0.8983	0.9617	0.93	0.9	0.96	0.0114	2.74%	0.0%	
2.5		5	0.918	0.8884	0.9476	0.92	0.89	0.95	0.01068	2.6%	1.29%	
5		5	0.928	0.8788	0.9772	0.93	0.88	0.98	0.01772	4.27%	0.22%	
10		5	0.908	0.8664	0.9496	0.91	0.87	0.95	0.01497	3.69%	2.37%	
Angular (Cor	rected) Transfo	rmed Sumi	mary									
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effec	
0	Lab Control	5	1.307	1.243	1.37	1.303	1.249	1.369	0.02281	3.9%	0.0%	
2.5		5	1.283	1.227	1.338	1.284	1.233	1.345	0.01995	3.48%	1.82%	
5		5	1.309	1.205	1.412	1.303	1.217	1.429	0.03727	6.37%	-0.16%	
10		5	1.267	1.193	1.341	1.266	1.202	1.345	0.0266	4.7%	3.05%	
Developmen	t Rate Detail											
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	0.91	0.93	0.96	0.9	0.95						
2.5		0.89	0.95	0.93	0.9	0.92						
5		0.9	0.95	0.88	0.98	0.93						
10		0.87	0.93	0.95	0.88	0.91						
Angular (Cor	rected) Transfo	rmed Detai	1									
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	1.266	1.303	1.369	1.249	1.345						
2.5		1.233	1.345	1.303	1.249	1.284						
5		1.249	1.345	1.217	1.429	1.303						
4.0												

Analyst: 4C QA: 45 8 7 6

000-089-170-2 CETIS™ v1.8.7.20

1.303

1.345

1.217

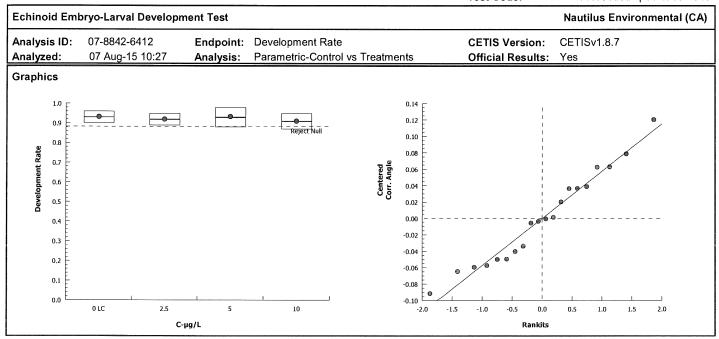
1.266

1.202

10

Report Date: Test Code: 07 Aug-15 10:27 (p 2 of 2)

141030dedv | 14-4718-7348



Report Date: Test Code:

07 Aug-15 10:27 (p 1 of 1)

141030dedv | 14-4718-7348

Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Analysis ID: Analyzed:

06-3337-3291 07 Aug-15 10:27

Endpoint: Development Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version: Official Results:

CETISv1.8.7

Yes

Trimmed	Spearman-Kärber	Estimates
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Threshold Option Threshold Trim Mu Sigma EC50 95% LCL 95% UCL Control Threshold 0.07 0.75% 1.144 0.00216 13.95 13.81 14.09

Developm	nent Rate Summary	y									
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.93	0.9	0.96	0.0114	0.02549	2.74%	0.0%	465	500
2.5		5	0.918	0.89	0.95	0.01068	0.02387	2.6%	1.29%	459	500
5		5	0.928	0.88	0.98	0.01772	0.03962	4.27%	0.22%	464	500
10		5	0.908	0.87	0.95	0.01497	0.03347	3.69%	2.37%	454	500
20		5	0	0	0	0	0		100.0%	0	500
40		5	0	0	0	0	0		100.0%	0	500
80		5	0	0	0	0	0		100.0%	0	500

Developm	nent Rate Detail						
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	
0	Lab Control	0.91	0.93	0.96	0.9	0.95	
2.5		0.89	0.95	0.93	0.9	0.92	
5		0.9	0.95	0.88	0.98	0.93	
10		0.87	0.93	0.95	0.88	0.91	
20		0	0	0	0	0	
40		0	0	0	0	0	
80		0	0	0	0	0	

Graphics 0.6 0.3 0.1 C-µg/L

CETIS QC Plot Report Date: 07 Aug-15 10:29 (1 of 1)

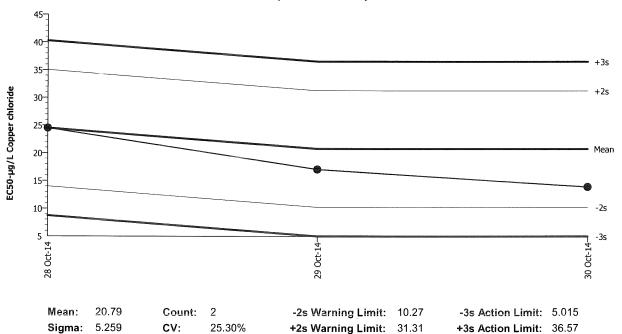
Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Test Type: Development Organism: Dendraster excentricus (Sand Dollar) Material: Copper chloride

Protocol: EPA/600/R-95/136 (1995) Endpoint: Development Rate Source: Reference Toxicant-REF

Echinoid Embryo-Larval Development Test



Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Oct	28	12:35	24.51	3.721	0.7075			19-0774-1141	02-3565-1966
2			29	15:45	17.07	-3.717	-0.7068			05-2123-6408	20-1447-5780
3			30	16:20	13.95	-6.844	-1.301			14-4718-7348	06-3337-3291

Report Date: Test Code: 28 Oct-14 15:51 (p 1 of 1) 14-4718-7348/141030dedv

Nautilus Environmental (CA)

Echinoid Embryo-Larval Development Test Nautilus E

Start Date: 30 Oct-14
End Date: 02 93 Nov-14

Species: Dendraster excentricus
Protocol: EPA/600/R-95/136 (1995

Sample Code: 141030dedv
Sample Source: Reference Toxicant
Sample Station: Copper Chloride

d Date: «	te: 30	Oct-14	+ 	Materi	al: Coppe	00/R-95/136 (1995) r chloride		Reference Toxicant Copper Chloride
C-µg/L		Rep			# Normal		Notes	
			1	100	9+5A	114 O18 0 119/14 Q18 0		
			2	100	0	11) Q10		
			3	100	80 5A	19/14 00 P		
			4		0	P-411 Q10 -		
			5		0			
			6		90			
			7		98			
			8		98			
			9		93			
			10		E)			
			11		98			
			12		Ø	44.00		
			13		96			
			14		95			
			15		90		,	
			16	100000	89			
			17		91			
1.119/2			18		95	A QIB	i	
			19		90	111114 Ø	. \	
			20		90			
			21		Ø			
			22		93			
			23		93			
			24		Ø		,	
			25		Ø			
		-	26		95		alara de la companya	
			27		9			
			28		Ø			
			29		87			
			30		93			
			31		95			
			32		91			
		-	33		92			
			34		93			
			35		88			

Report Date: Test Code: 28 Oct-14 15:51 (p 1 of 1) 14-4718-7348/141030dedv

				elopment Tes			
Start Date:		Oct-14			: Dendraster excentricus	Sample Code:	141030dedv
End Date:		Nov-14			I: EPA/600/R-95/136 (1995)		Reference Toxicant
Sample Date	e: 30 C	Oct-14		Materia	: Copper chloride	Sample Station:	Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Normal	Notes	
0	LC	1	17				
0	LC	2	30				
0	LC	3	13				
0	LC	4	20				
0	LC	5	26				
2.5		1	16				
2.5		2	18				
2.5		3	23				
2.5		4	15				
2.5		5	33				
5		1	6				
5		2	14				
5		3	11				
5		4	8				
5		5	9				
10		1	29				
10		2	34			11 10 10 10 10 10 10 10 10 10 10 10 10 1	
10		3	31				
10		4	35				
10		5	32				
20		1	1				
20		2	28				
20		3	19				
20		4	3				
20		5	7				
40		1	25				
40		2	10				
40		3	4			Alla M. Alla A. Alla A	
40		4	27			1	
40		5	22				
80		1	12				
80		2	5				
80		3	21				
80		4	2				
80		5	24				



Water Quality Measurements

Client: Internal	Test Species:	D. excentricus
Sample ID: CuCl ₂	Start Date/Time:	10/30/2014 6 20
Test No.: 141030dedv	End Date/Time:	11/3/2014 1720
		OTA,
		11/2/14

Concentration		S	Salinity				Te	mperati	ure		Δ.	Disso	olved Ox	ygen		0		рН		
(µg/L)			(ppt)		V	(P		(°C)		ď	K		(mg/L)		·Ø	Υ	(pH units	s)	XX.
	0	24	48	72	96	0	24	48	72	96 ,	0	24	48	72	96	, 0	24	48	72	96
Lab Control	335 8	3.5 2	3.5	33.4		14.5	15.6	15.4	16.0		\$.8°	8.1	7.8	7.7	/	8,00	7,99	8.4	7.94	
2.5	33,63	3.63	33.W	33 F		14.5	15.6	150	16.0		8,7	8.1	7.9	79		8,02	7.94	8.05	7-97	
5	33.6	33.6 3	35.1	23.7		14.6	15.6	14.9	16.0		8.7	8.1	8.0	79		8:01	8.01	8.07	7.97	
10	33.5 \$	33.6 3	33.7	33.7		14.6	15.8	14.8	15.1		8,6	8.1	8.0	79		8.00	8.00	8.64	7-97	
20	33.6 ³	3.88	33.6	33.7		14.60	15.8	15.0	14.9		8.7	8.2	7.9	7.9		8,01	800	812	7.96	
40	33.5 3	35.5	33.5	33.5		14.6	15.9	14.9	14.9		87	8.2	8.0	7.9		8.03	8.00	8.11	797	
80	33.4 3	33,3	33.3	33.4.	/	14.7	15.7	14.9	15.0		8.7	8.1	8.0	7.9		8.04	8,00	8.11	7-97	/

Technician Initials: WQ Readings: PA A W NA DIlutions made by: A V Cu stock concentration (μg/L): 80

Vol. Cu stock added (mL): 4. 4

Final Volume (mL): 500

Cu stock concentration (μg/L): 9,050

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

Echinoderm Larval Development Worksheet

Client: Sample ID: Test No.:	Internal Cycir 191030dodu	Start Date/Time: 10/30/2014 / 1620 End Date/Time: 41/3/2014 / 1414 1720 Species: D. excentricus
Tech initials: Injection Time:	AC 15:15	Animal Source: Mission Bay Date Collected: 1000414
Sperm Absorbance at	400 nm: 0,744 (target range of 0.8 - 1.0 f	or density of 4x10 ⁶ sperm/ml)
Eggs Counted:	Mean: Z X 50 = Y	eggs/ml cal pass on Sedgwick-Rafter gs/ml)
Initial density: Final density:	1000 eggs/ml = 1.05 dilution factors and the second segments are searched as a second segment of the second segments.	ock seawater ml
Prepare the egg stock part) and 125 ml of dilu	according to the calculated dilution factor. For example, ion water (1.25 parts).	if the dilution factor is 2.25, use 100 ml of existing stock (1
Volume of Sperm stock	needed to fertilize eggs:	
Egg Stock (mL) = Sperm Stock (µL) = Egg/Sperm Ratio = Fertilization Time:	100 100 1m1:1u1 1555	
Embryo Stock Fertilizati 10 minutes (1st fert.) 20 minutes (2nd fert. If I	$\begin{array}{ccc} \text{Time} & \text{Fert.} & \text{Unfer} \\ 1605 & 98 & 2 \end{array}$	<u>t. </u>
Test Initiation Time:	16:20 Embryo Stock Add	ed: <u>0.25 ml</u>
Test Termination:	No. No. % Normal Abnormal Normal	
72-hour QC check ^a End of test QC check	97 3 97	
Comments:	^a If the embryo development does not meet the mean developed, continue the test up to 96-hrs (ASTM 1999)	
QC Check:	Ac 8/7/15	Final Review: 45 8715

CETIS Summary Report

Report Date:

05 Aug-15 17:33 (p 1 of 1)

Test Code: 150722dedv | 13-7165-0540

								rest Code:			-7 165-054
Echinoid Emb	oryo-Larval Dev	elopment ⁻	est						Nautilus	s Environm	ental (CA
Batch ID:	07-5278-8592	Tes	t Type:	Development				Analyst:			
Start Date:	22 Jul-15 15:10		tocol:	EPA/600/R-95/	136 (1995)			Diluent: Natu	ıral Seawat	er	
•	25 Jul-15 15:40) Sp	ecies:	Dendraster exc	entricus			Brine: Not	Applicable		
Duration:	73h	So	urce:	Mission Bay				Age:			
Sample ID:	11-6268-5099	Co	de:	150722dedv	\			Client: Inter			
Sample Date:	22 Jul-15	Ma	terial:	Copper chloride	9			Project:			
Receive Date:	: 25 Jul-15	So	ırce:	Reference Toxi	cant						
Sample Age:	15h	Sta	tion:	Copper Chlorid	e						
Comparison S	Summary										
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Method			
04-0931-8852	Development F	Rate	10	20	14.14	10.2%		Dunnett M	ultiple Com	parison Tes	t
Point Estimat	e Summary										
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Method			
17-1529-7817	Development F	Rate	EC50	14.59	14.4	14.78		Spearman	-Kärber		
Test Acceptab	oility										
Analysis ID	-			ute	Test Stat	TAC Limi	ts	Overlap	Decision		
04-0931-8852	Development R	Rate	Control Resp		0.92	0.8 - NL		Yes	Passes A	cceptability	Criteria
17-1529-7817	Development R		Contro	ol Resp	0.92	0.8 - NL		Yes		cceptability	
04-0931-8852	Development R	Rate	PMS)	0.1019	NL - 0.25		No	Passes A	cceptability	Criteria
Development	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	0.92	0.846	0.994	0.83	0.99	0.02665	0.05958	6.48%	0.0%
2.5		5	0.93	0.853	1	0.84	0.98		0.06205	6.67%	-1.09%
5		5	0.908	0.7881	1				0.09654	10.63%	1.3%
10		5			•	0.74	0.98				
00		5	0.97	0.9524	0.9876	0.95	0.98	0.006325	0.01414	1.46%	-5.44%
		5 5	0.97 0.042	0.9524 0.001384	0.9876 0.08262	0.95 0	0.98 0.09	0.006325 0.01463	0.01414 0.03271		95.43%
20 40 80		5 5 5	0.97 0.042 0	0.9524 0.001384 0	0.9876 0.08262 0	0.95 0 0	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80		5 5	0.97 0.042	0.9524 0.001384	0.9876 0.08262	0.95 0	0.98 0.09	0.006325 0.01463	0.01414 0.03271	1.46%	95.43%
40 80 Development		5 5 5 5	0.97 0.042 0 0	0.9524 0.001384 0 0	0.9876 0.08262 0	0.95 0 0	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80 Development C-μg/L	Control Type	5 5 5 5 Rep 1	0.97 0.042 0 0	0.9524 0.001384 0 0 Rep 3	0.9876 0.08262 0 0	0.95 0 0 0 Rep 5	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80 Development C-μg/L		5 5 5 5 7 8 Rep 1	0.97 0.042 0 0 Rep 2	0.9524 0.001384 0 0 Rep 3	0.9876 0.08262 0 0 Rep 4 0.83	0.95 0 0 0 0 Rep 5 0.99	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80 Development C-μg/L 0 2.5	Control Type	5 5 5 5 Rep 1 0.94 0.97	0.97 0.042 0 0 Rep 2 0.94 0.84	0.9524 0.001384 0 0 	0.9876 0.08262 0 0 Rep 4 0.83 0.89	0.95 0 0 0 0 Rep 5 0.99 0.97	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80 Development C-μg/L 0 2.5	Control Type	5 5 5 5 Rep 1 0.94 0.97 0.98	0.97 0.042 0 0 Rep 2 0.94 0.84 0.92	0.9524 0.001384 0 0 Rep 3	0.9876 0.08262 0 0 Rep 4 0.83	0.95 0 0 0 0 Rep 5 0.99 0.97 0.94	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80 Development C-μg/L 0 2.5	Control Type	5 5 5 5 5 Rep 1 0.94 0.97 0.98 0.98	0.97 0.042 0 0 Rep 2 0.94 0.84	0.9524 0.001384 0 0 	0.9876 0.08262 0 0 Rep 4 0.83 0.89	0.95 0 0 0 0 Rep 5 0.99 0.97	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80 Development C-μg/L 0 2.5 5	Control Type	5 5 5 5 Rep 1 0.94 0.97 0.98	0.97 0.042 0 0 Rep 2 0.94 0.84 0.92	0.9524 0.001384 0 0 8 Rep 3 0.9 0.98 0.74	0.9876 0.08262 0 0 Rep 4 0.83 0.89 0.96	0.95 0 0 0 0 Rep 5 0.99 0.97 0.94	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%
40 80 Development	Control Type	5 5 5 5 5 Rep 1 0.94 0.97 0.98 0.98	0.97 0.042 0 0 Rep 2 0.94 0.84 0.92 0.95	0.9524 0.001384 0 0 8 Rep 3 0.9 0.98 0.74 0.96	0.9876 0.08262 0 0 Rep 4 0.83 0.89 0.96 0.98	0.95 0 0 0 0 Rep 5 0.99 0.97 0.94 0.98	0.98 0.09 0	0.006325 0.01463 0	0.01414 0.03271 0	1.46%	95.43% 100.0%

Analyst: AC QA: 497/16

Report Date:

05 Aug-15 17:33 (p 1 of 2)

CETIS An	alytical Rep	ort		•	ort Date: Code:			33 (p 1 of <i>2</i> 3-7165-054			
Echinoid Em	nbryo-Larval Dev	/elopment	Test							•	nental (CA
Analysis ID: Analyzed:	04-0931-8852 05 Aug-15 17:		-	evelopment R arametric-Cor		tments		IS Version:		.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	ected)	NA	C > T	NA	NA		10.2%	10	20	14.14	
Dunnett Mult	tiple Compariso	n Test									
Control	vs C-μg/L		Test Sta	t Critical	MSD DF	P-Value	P-Type	Decision	(a:5%)		
Lab Control	2.5		-0.2915	2.305	0.162 8	0.8808	CDF	Non-Sign	ificant Effect		
	5		0.2017	2.305	0.162 8	0.7283	CDF	Non-Sign	ificant Effect		
	10		-1.396	2.305	0.162 8	0.9918	CDF	Non-Sign	ificant Effect		
	20*	Managaran ()	15.85	2.305	0.162 8	<0.0001	CDF	Significar	t Effect		****
ANOVA Table	e										
Source	Sum Squ		Mean Sc	luare	DF	F Stat	P-Value	Decision	(a:5%)		
Between	5.211512		1.302878		4	106	<0.0001	Significar	t Effect		
Error	0.245789		0.012289	945	20	_					
Total	5.457301				24						
Distributiona	l Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Bartlett Equality of Variance 5.39 Shaniro-Wilk W Normality 0.95				13.28	0.2496	Equal Var	iances			
Distribution	Shapiro-Wilk W Normality 0.9527				0.8877	0.2881	Normal Di	stribution			
Developmen	t Rate Summary	•									
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.92	0.846	0.994	0.94	0.83	0.99	0.02665	6.48%	0.0%
2.5		5	0.93	0.853	1	0.97	0.84	0.98	0.02775	6.67%	-1.09%
5		5	0.908	0.7881	1	0.94	0.74	0.98	0.04317	10.63%	1.3%
10		5	0.97	0.9524	0.9876	0.98	0.95	0.98	0.006325	1.46%	-5.44%
20		5	0.042	0.001384	0.08262	0.04	0	0.09	0.01463	77.88% 	95.43%
Angular (Cor	rected) Transfor	rmed Sumr	mary								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Мах	Std Err	CV%	%Effect
0	Lab Control	5	1.302	1.155	1.45	1.323	1.146	1.471	0.05317	9.13%	0.0%
2.5		5	1.323	1.175	1.471	1.397	1.159	1.429	0.0534	9.03%	-1.57%
5		5	1.288	1.101	1.476	1.323	1.036	1.429	0.0676	11.73%	1.09%
10 20		5 5	1.4 0.1911	1.35 0.07601	1.45 0.3063	1.429 0.2014	1.345 0.05002	1.429 0.3047	0.01794 0.04146	2.86% 48.51%	-7.51% 85.32%
		-	0.1311	0.07001	0.3003	0.2014	0.03002	0.3047	0.04140	40.5170	
Development		D 4	D 0		5 4	D =					
C-µg/L	Control Type Lab Control	Rep 1 0.94	Rep 2 0.94	Rep 3 0.9	Rep 4 0.83	Rep 5 0.99		ng mujua nuun na aun sasalake (iik kohiste et e			
2.5	Lan Colling	0.94	0.94	0.9	0.89	0.99					
z.5 5		0.98	0.84	0.98	0.89	0.97					
10		0.98	0.92	0.74	0.98	0.94					
20		0.98	0.95	0.90	0.98	0.98					
	41\ T ?				3.00	3.00					
• ,	rected) Transfor			D 0	D 4	D 5					
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5					
0	Lab Control	1.323	1.323	1.249	1.146	1.471					
2.5 -		1.397	1.159	1.429	1.233	1.397					
5		1.429	1.284	1.036	1.369	1.323					
10		1.429	1.345	1.369	1.429	1.429					

0.2014

20

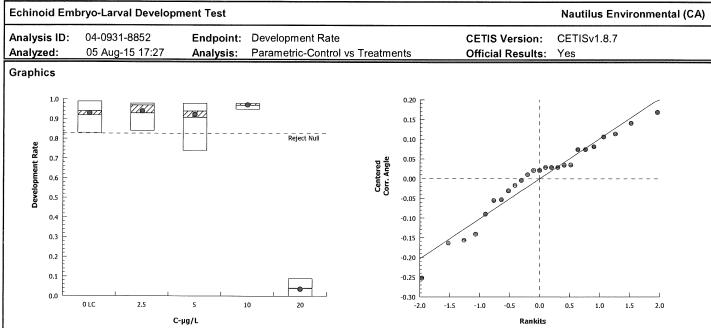
0.1741

0.2255 0.05002 0.3047

Report Date:

05 Aug-15 17:33 (p 2 of 2)

Test Code: 150722dedv | 13-7165-0540



Echinoid Embryo-Larval Development Test

Report Date: Test Code: 05 Aug-15 17:33 (p 1 of 1)

150722dedv | 13-7165-0540

Nautilus Environmental (CA)

17-1529-7817 Endpoint: Development Rate CETIS Version: CETISv1.8.7

Analyzed: 05 Aug-15 17:32 Analysis: Untrimmed Spearman-Kärber Official Results: Yes

Spearman-Kärber Estimates

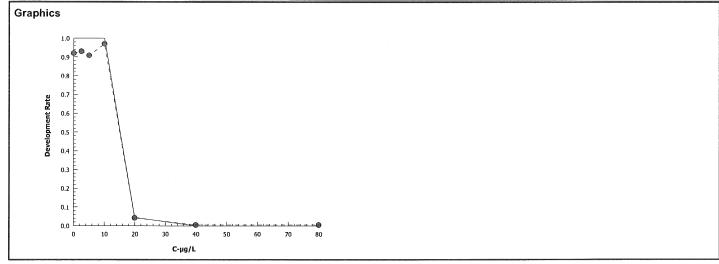
Analysis ID:

 Threshold Option
 Threshold
 Trim
 Mu
 Sigma
 EC50
 95% LCL
 95% UCL

 Control Threshold
 0.08
 0.00%
 1.164
 0.002793
 14.59
 14.4
 14.78

Developn	nent Rate Summary	Calculated Variate(A/B)									
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.92	0.83	0.99	0.02665	0.05958	6.48%	0.0%	460	500
2.5		5	0.93	0.84	0.98	0.02775	0.06205	6.67%	-1.09%	465	500
5		5	0.908	0.74	0.98	0.04317	0.09654	10.63%	1.3%	454	500
10		5	0.97	0.95	0.98	0.006325	0.01414	1.46%	-5.44%	485	500
20		5	0.042	0	0.09	0.01463	0.03271	77.88%	95.43%	21	500
40		5	0	0	0	0	0		100.0%	0	500
80		5	0	0	0	0	0		100.0%	0	500

Development Rate Detail										
C-μg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5				
0	Lab Control	0.94	0.94	0.9	0.83	0.99				
2.5		0.97	0.84	0.98	0.89	0.97				
5		0.98	0.92	0.74	0.96	0.94				
10		0.98	0.95	0.96	0.98	0.98				
20		0.04	0.05	0	0.09	0.03				
40		0	0	0	0	0				
80		0	0	0	0	0				



Report Date:

07 Aug-15 16:32 (1 of 1)

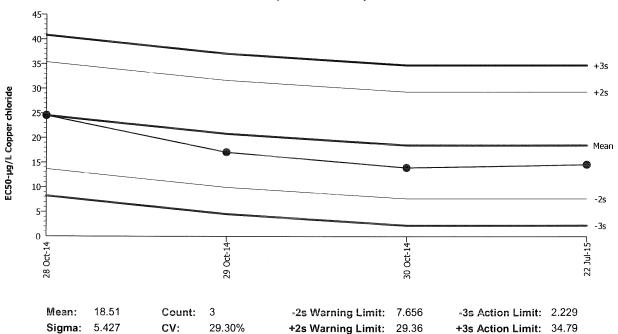
Echinoid Embryo-Larval Development Test

Nautilus Environmental (CA)

Test Type: Development Organism: Dendraster excentricus (Sand Dollar) Material: Copper chloride

Protocol: EPA/600/R-95/136 (1995) Endpoint: Development Rate Source: Reference Toxicant-REF

Echinoid Embryo-Larval Development Test



Quality	Control	Data
Quanty	Control	vata

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Oct	28	12:35	24.51	6.001	1.106			19-0774-1141	02-3565-1966
2			29	15:45	17.07	-1.437	-0.2648			05-2123-6408	20-1447-5780
3			30	16:20	13.95	-4.564	-0.841			14-4718-7348	06-3337-3291
4	2015	Jul	22	15:10	14.59	-3.919	-0.7222			13-7165-0540	17-1529-7817

Report Date: Test Code:

21 Jul-15 10:55 (p 1 of 1) 13-7165-0540/150722dedv

Echinoid En	nbryo-	Larva	l Deve	elopment Te	est			Nautilus Environmental (CA
Start Date: End Date: Sample Date	25 .	Jul-15 Jul-15 Jul-15		Specie Protoc Materi	ol: EPA/6	aster excentricus 00/R-95/136 (1995) or chloride	Sample Code: Sample Source: Sample Station:	150722dedv Reference Toxicant Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Normal		Notes	
0	LC	1	17	100	98	AC 7/28/15	OPPOSED TO THE PARTY OF THE PAR	
0	LC	2	30			1/-5/13		
0	LC	3	13				**************************************	
0	LC	4	20					
0	LC	5	26					
2.5		1	16	(00)	95	AC 7/28/15		
2.5		2	18					
2.5		3	23					
2.5		4	15					
2.5		5	33			,		
5		1	6	100	96	AC 7/28/15		
5		2	14			***		
5		3	11					
5		4	8					
5		5	9			100000		
10		1	29	(00)	94	AC 7/28/15		
10		2	34					
10		3	31					
10		4	35					
10		5	32					
.20		1	1	100	23	AC 7/28/15	94294200	
20		2	28			1		
20		3	19					
20		4	3					
20		5	7					
40		1	25	100	0	AC 7/28/18		
40		2	10					
40		3	4					
40		4	27					
40		5	22					
80		1	12	100	0	AC 7/28/15		
80		2	5					
80		3	21			-		
80		4	2					
80		5	24					

QC.2

Report Date: Test Code: 21 Jul-15 10:55 (p 1 of 1)

13-7165-0540/150722dedv

Echinoid En	nbryo-l	Larva	l Deve	elopment Te	st			Nautilus Environmental (CA	
Start Date: End Date: Sample Date	25 .	Jul-15 Jul-15 Jul-15		Protoc		aster excent 600/R-95/136 er chloride			150722dedv Reference Toxicant Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Normal			Notes	
			1	100	4	CH -	1/30/15		
			2	100	0				
			3	100	9		11100		
			4	100	0				
			5	100	0				
			6 7	100	99	2187/30/15			
			8	100	965				
			9	(00	96				
			10	100	79			10740-009000	
			11	100	74				
			12	100	0				
,			13	100	90				
			14	100	az				
			15	100	30				
			16	100	97				
			17	100	94				
-10-02-44 k			18	100	84				
			19	100	0				
		-	20	100	83	V			
			21	100	0	CH 8	5/6/16		
			22	100	ව				77.00
			23	100	98				
			25	100	Ó				
			26	100	0				
			27	100	99		7,00	***************************************	
			28	100	5	1	416/162		A
			29	100	985	CHGIS	で(りハソ		**************************************
			30	100	10				
			31	100	94				
			32	100	98				
			33	100	97				
			34	100	95	1		***************************************	
			35	100	98	V			,

Analyst: CH

Water Quality Measurements

Client: Internal Sample ID: CuCl₂

Test No.: 150722dedv

Test Species: D. excentricus

Start Date/Time: 7/22/2015 1510

End Date/Time: 7/25/2015

Concentration (µg/L)	Salinity (ppt)						erature C)		Dissolved Oxygen pH (mg/L) (pH units)							
(/	0	24	48	72	0	24	48	72	0	24	9/L) 48	72	Ö	(p⊓ 24	48	72
Lab Control	333	33.4	33.3	33.2	14.9	14-8	14.7	155	89	4.U	7.9	8.2	8-09	8.07	8.05	8-03
2.5	33.4	33.5	33.6	33.4	146	14.5	14.5	15.0	8.8	8.1	8.0	8.4	8.08	8.67	8.06	8.05
5	335	33.6	33.6	33.5	14.8	14.4	14,4	148	8-8	8.2	8.0	8.4	80.8	8,06	8.06	8.05
10	33.5	33.5	33.6	<i>33.</i> 5	145	14.3	14.3	14.9	8-8	٤.2	8.1.	8.4	80.6	8.07	8.06	8.05
20	33.5	33-6	33.6	33.5	14.9	14.6	14.5	15.1	8.7	8.2	8.0	8.3	8,06	807	8.07	8.06
40	33.4	33.4	33.5	33.4	14.8	14.5	14.4	15, 1	8.7	8.3	8.1	8.4	8.09	80,8	8.07	8.06
80	33.3	33.3	33.4	33.3	1	14.4	14.3	15.1	8.7	8.3	8.1	8.4	F.09	8,07	4.07	2.00

Technician Initials:

24 48 72 EG WQ Readings: at AD Dilutions made by:

High conc. made (µg/L): 80 4.5 Vol. Cu stock added (mL): Final Volume (mL): 500 Cu stock concentration (µg/L): 8,850

Comments:

0 hrs:_____

24 hrs:_____

48 hrs:

72 hrs:

QC Check:

Final Review: 6 971/15

Echinoderm Larval Development Worksheet

Client: Sample ID: Test No.: Tech initials:	Memal Cull2 150722 ded NC 018 813/15	Start Date/Time: 122 157 151D End Date/Time: 123 151 1540 Species: D. excentricus Date Collected: 7 11 15
Injection Time:	1436	
Sperm Absorbance at	400 nm: / · 🔾 (target range of 0.8 - 1.0 for density of 4	4x10 ⁶ sperm/ml)
Eggs Counted:	$\frac{16}{27}$ Mean: $\frac{19.6}{50} \times 50 = \frac{980}{27}$	_eggs/ml
	(target counts of 20 eggs per vertical pass on Sed slide for a final density of 1000 eggs/ml)	lgwick-Rafter
Initial density: Final density:	$\frac{940}{1000} \frac{\text{eggs/ml}}{\text{eggs/ml}} = \frac{6.98}{0.98} \text{ dilution factor}$	egg stock (00 ml seawater ml
Prepare the egg stock a (1 part) and 125 ml of c	according to the calculated dilution factor. For example, if the dilution lilution water (1.25 parts).	n factor is 2.25, use 100 ml of existing stock
Add 100 μL sperm stoo	k per 100mL of egg stock. For example, if you have 60mL of egg st	tock, add 60μL sperm stock.
Embryo Stock Fertilizati	ion Checks (Initiate test only when fertilization is ≥90%):	
	No. No. Time Fert. Unfert. %	
10 minutes (1st fert.) 20 minutes (2nd fert. If	1502 47 3 97	- - -
Fertilization Time:	1452	
Test Initiation Time:	Embryo Stock Added: 0.25 r	nl
Test Termination:	No. No. %	
72-hour QC check ^a End of test QC check	Normal Abnormal Normal 100 0 100	
Comments:	^a If the embryo development does not meet the mean test acceptadeveloped, continue the test to 96-hrs (ASTM 1999).	ability criterion of 80% normally
QC Check:	AC815115	Final Review: 8/7/15



CETIS Summary Report

Report Date:

10 Nov-14 10:32 (p 1 of 1)

Test Code:

141030sprt | 00-9558-0596

Echinoid Spe	erm Cell Fertiliz	ation Te	est 15C						Nautil	us Environi	nental (CA
Batch ID: Start Date: Ending Date: Duration:	13-0452-0527 30 Oct-14 15: 30 Oct-14 15: 40m	05	Test Type: Protocol: Species: Source:	Fertilization EPA/600/R-95 Strongylocentr Pt. Loma		atus		Analyst: Diluent: Brine: Age:	Natural Seawa Not Applicable	nter	
Sample ID: Sample Date: Receive Date: Sample Age:			Code: Material: Source: Station:	141030sprt Copper chlorid Reference Tox Copper Chloric	dcant			Client: Project:	Internal		
Comparison S	Summary	-									
Analysis ID 01-4265-2489	Endpoint Fertilization Ra	ate	NOEL <10	LOEL 10	TOEL NA	PMSD 10.0%	TU	Metho	od ett Multiple Cor	nparison Te	st
Point Estimat Analysis ID 15-2024-8469	e Summary Endpoint Fertilization Ra	ate	Level EC50	μ g/L 19.02	95% LCL 17.82	95% UCL 20.3	TU	Metho	od		
Test Acceptab				19.02	17.02	20.3		Irimm	ed Spearman-	Kärber	
Analysis ID 01-4265-2489 15-2024-8469	Endpoint Fertilization Ra	Endpoint Attrib Fertilization Rate Contro Fertilization Rate Contro			Resp 0.858 0.7 - NL			Overla Yes Yes No	Passes A	Acceptability Acceptability Acceptability	Criteria
Fertilization R	ate Summary								. 455557	ioooptaDiiity	Ontona
	Control Type Lab Control	5 5 5 5 5 5	0.858 0.636 0.446 0.06 0	95% LCL 0.8387 0.6055 0.4238 0.04706 0	95% UCL 0.8773 0.6665 0.4682 0.07294 0	0.8 0.57 0.35 0	Max 0.92 0.73 0.5 0.09 0	0.0231	1 0.05167 5 0.08173 7 0.05941	6.02% 12.85% 13.32% 57.74%	%Effect 0.0% 25.87% 48.02% 93.01% 100.0%
Fertilization Ra	ate Detail										
	Control Type Lab Control	Rep 1 0.81 0.73 0.35 0.09 0	Rep 2 0.92 0.59 0.5 0.07 0	Rep 3 0.87 0.72 0.48 0 0	Rep 4 0.89 0.57 0.47 0.07 0	Rep 5 0.8 0.57 0.43 0.07 0					

Analysis ID: 01-4265-2489

Echinoid Sperm Cell Fertilization Test 15C

Report Date: **Test Code:**

10 Nov-14 10:32 (p 1 of 1) 141030sprt | 00-9558-0596

1001 00001	1410003prt	100-9006-0090
	Nautilus Enviro	nmental (CA)

Analysis ID: Analyzed:	01-4265-2489 10 Nov-14 10:32	Endpoint: Analysis:		tilization R ametric-Co	ate ontrol vs Treatments	CE1 Offi		1.8.4		
Data Transfor		a Alt I	Т ур	Trials	Seed	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corre	cted) NA	C > 1	Γ	NA	NA	<10	10	NA		10.0%

Dunnett Multiple Comparison Test

	pic companson res	-						
Control	vs C-μg/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(a:5%)
Lab Control	10*	5.058	2.227	0.116	8	0.0002	CDF	Significant Effect
	20*	8.774	2.227	0.1 1 6	8	<0.0001	CDF	Significant Effect
	40*	18.33	2,227	0.116	8	<0.0001	CDF	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)
Between	2.455379	0.8184596	3	120	<0.0001	Significant Effect
Error	0.1091171	0.006819817	16	0	0.0001	Oigninoant Enect
Total	2.564496		19			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)
Variances	Bartlett Equality of Variance	1.059	11.34	0.7869	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9249	0.866	0.1230	Normal Distribution

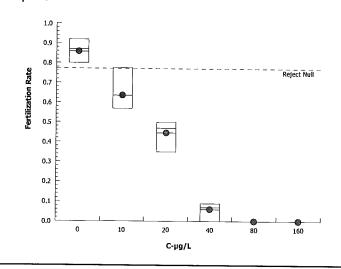
Fertilization Rate Summary

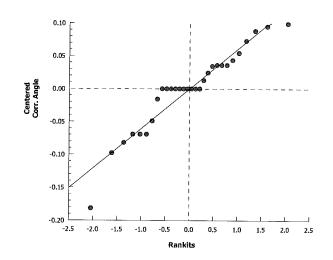
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.858	0.7938	0.9222	0.87	0.8	0.92	0.02311	6.02%	0.0%
10		5	0.636	0.5345	0.7375	0.59	0.57	0.73	0.03655	12.85%	25.87%
20		5	0.446	0.3722	0.5198	0.47	0.35	0.5	0.02657	13.32%	48.02%
40		5	0.06	0.01699	0.103	0.07	0	0.09	0.01549	57.74%	93.01%
80		5	0	0	0	0	0	0	0	0.11.170	100.0%
160		5	0	0	0	0	0	0	0		100.0%

Angular (Corrected) Transformed Summary

,	,	oui.ii	· ··u· y								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.189	1.096	1.282	1.202	1.107	1.284	0.03362	6.32%	0.0%
10		5	0.9249	0.818	1.032	0.8759	0.8556	1.024	0.03853	9.32%	22.22%
20		5	0.7309	0.6559	0.8058	0.7554	0.6331	0.7854	0.027	8.26%	38.54%
40		5	0.2316	0.104	0.3592	0.2678	0.05002	0.3047	0.04595	44.37%	80.52%
80		5	0.05002	0.05001	0.05003	0.05002	0.05002	0.05002	0	0.0%	95.79%
160		5	0.05002	0.05001	0.05003	0.05002	0.05002	0.05002	0	0.0%	95.79%

Graphics





Report Date:

10 Nov-14 10:32 (p 1 of 1)

Test Code:

141030sprt | 00-9558-0596

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: Analyzed:

15-2024-8469 10 Nov-14 10:32

Endpoint: Fertilization Rate

Analysis: Trimmed Spearman-Kärber

CETIS Version:

CETISv1.8.4

Official Results: Yes

	ırımmea	Spearman-Karber	Estima	tes
--	---------	-----------------	--------	-----

Threshold Option	Inreshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL	
Control Threshold	0.142	25.87%	1.279	0.01415	19.02	17.82	20.3	•

Fertilizati	Fertilization Rate Summary			Calculated Variate(A/B)							
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.858	0.8	0.92	0.02311	0.05167	6.02%	0.0%	429	500
10		5	0.636	0.57	0.73	0.03655	0.08173	12.85%	25.87%	317	500
20		5	0.446	0.35	0.5	0.02657	0.05941	13.32%	48.02%	223	500
40		5	0.06	0	0.09	0.01549	0.03464	57.74%	93.01%	30	500
80	,	5	0	0	0	0	0		100.0%	0	500
160		5	0	0	0	0	0		100.0%	0	500

Graphics 8.0 0,6 0.5 0.3 0.2 40 120 C-µg/L

Report Date:

10 Nov-14 10:33 (1 of 1)

Echinoid Sperm Cell Fertilization Test 15C

Test Type: Fertilization

Protocol: EPA/600/R-95/136 (1995)

Organism: Strongylocentrotus purpuratus (Purpl

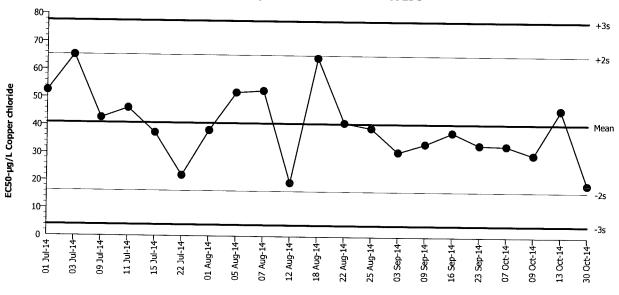
Endpoint: Fertilization Rate

Nautilus Environmental (CA)

Copper chloride Material:

Source: Reference Toxicant-REF

Echinoid Sperm Cell Fertilization Test 15C



Mean: Sigma:

40.82 12.25

Count: CV:

20 30.00%

-2s Warning Limit: +2s Warning Limit:

16.32 65.32

-3s Action Limit: 4.07 +3s Action Limit: 77.57

Quality Control Data

Point	Year	Month	Day	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Jul	1	52.45	11.63	0.9492		,	03-1633-9040	04-6714-9615
2			3	65.26	24.44	1.995			16-7036-5031	17-9179-4003
3			9	42.65	1.832	0.1496			16-1504-3840	07-5767-6661
4			11	46.17	5.351	0.4368			10-5811-3549	06-0498-7042
5			15	37.29	-3.529	-0.2881			13-2908-2037	08-6489-1251
6			22	21.9	-18.92	-1.545			12-6705-4933	01-8116-2108
7		Aug	1	38.18	-2.641	-0.2156			14-6400-5139	20-5348-7458
8			5	51.86	11.04	0.9013			13-0687-0962	02-1578-6969
9			7	52.56	11.74	0.958			00-5563-1682	00-5377-5202
10			12	19.39	-21.43	-1.749			14-9442-2864	07-1676-7162
11			18	64.39	23.57	1.924			16-6489-0031	15-5233-2184
12			22	41.11	0.2913	0.02378			02-4639-6440	00-1755-8985
13			25	39.27	-1.554	-0.1269			17-8206-5737	15-9942-8515
14		Sep	3	30.64	-10.18	-0.831			15-0487-5875	08-0190-3799
15			9	33.59	-7.226	-0.5899			20-3803-1763	15-4983-2355
16			16	37.75	-3.069	-0.2505			01-5049-2617	15-5471-0022
17			23	33.24	-7.576	-0.6184			02-0667-4002	20-3091-9441
18		Oct	7	32.97	-7.845	-0.6404			14-5637-2531	13-7332-7338
19			9	29.68	-11.14	-0.9091			13-5792-5239	01-7979-4948
20			13	45.96	5.136	0.4193			11-7429-5626	04-4094-9558
21			30	19.02	-21.8	-1.78			00-9558-0596	15-2024-8469

Report Date:

28 Oct-14 15:59 (p 1 of 1)

Test Code: 00-9558-0596/141030sprt **Echinoid Sperm Cell Fertilization Test 15C** Nautilus Environmental (CA) Start Date: 30 Oct-14 Species: Strongylocentrotus purpuratus Sample Code: 141030sprt **End Date:** 30 Oct-14 Protocol: EPA/600/R-95/136 (1995) Sample Source: Reference Toxicant Sample Date: 30 Oct-14 Copper chloride Sample Station: Copper Chloride # Counted C-µg/L Code Rep Pos # Fertilized Notes 1 Ø 60/10/31/14 2 /00 48 3 100 Ø 4 Ø 100 7 5 100 6 Ø 100

Report Date: Test Code:

28 Oct-14 15:59 (p 1 of 1) 00-9558-0596/141030sprt

Start Date: End Date: Sample Date	30 C	Oct-14 Oct-14 Oct-14		Protoc		entrotus purpuratus -95/ 1 36 (1995) oride	Sample Code: Sample Source: Sample Station:	141030sprt Reference Toxicant Copper Chloride
C-µg/L		Rep		# Counted	# Fertilized		Notes	
0	LC	1	7					
0	LC	2	8	100	81		1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	
0	LC	3	20	10 -	<i>V</i> 1			
0	LC	4	9					
0	LC	5	28					
10		1	19			and the second s		
10		2	26					
10		3	22					
10		4	27					
10		5	10	100	78			
20		1	15					
20		2	30					
20		3	2	100	44			
20		4	12			AA-0		
20		5	17					
40		1	29					
40		2	25					
40		3	24	100	4			
40		4	5		i i			
40		5	18					
80		1	1					
80		2	4					1200-
80		3	11	100	0			
80		4	21					
80		5	14					
160		1	16				a/	V
160		2	6					~
160		3	3	100	0		and 1 (10) 1 (10) 1 (10)	
160		4	13					
160		5	23	4				

Analyst: 17 | QA: 18 11 | 7 | 14

Water Quality Measurements

-			
7.	10	nf	

Internal

Test Species: S. purpuratus

Sample ID:

CuCl₂

Start Date/Time: 10/30/2014 AC 3:05 15:05

Test No:

141030sprt

End Date/Time: 10/30/2014 + 43:45 15:45

Dilutions made by:

AC

Final Volume (mL):

High conc. made (μg/L):
Vol. Cu stock added (mL):

160 9.8

Cu stock concentration (μg/L):

9050

Analyst:

	Initial Readings										
Concentration (μg/L)	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)							
Lab Control	8.4	8.00	33.5	14,5							
10	8.6	8.00	33,5	14.6							
20	8.7	8.01	33.6	14.6							
40	8.7	8.03	33.5	14,6							
80	8.7	8.04	33.4	14.6							
160	8.7	8.05	33.2	14.7							

Comme	ents:
-------	-------

QC Check:

KB WIOLH

Final Review: 11/18/14

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

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	Internal Cuci2			End Date/Ti	me: 10/30/2014 me: 10/30/2014	1/4ct 3:45 15	
163(110.,	1410 20 Dec				ies: <u>S. purpura</u>		
Tech initials:	8/AC				rce: Point Lom	1a	
Injection Time:	1430			Date Collec	ted: 10/29/	19	
injection rime.	<u> </u>						
Sperm Absorbance at 4	00 nm: <u>0.873</u>	(target range of 0.8		•	nl)		
Eggs Counted:		an: $\frac{1}{6}$ $\frac{3}{3}$ $\frac{4}{1}$ x 50	= 3110	eggs/ml			
	107						
		et counts of 80 eggs per for a final density of 4000		Sedgwick-Rafter			
	71	0.79					
	2170		VB 117111	8	14.0		
Initial density:	3[70 eggs/ml	= 0.67 dilution	n factor	egg stock 🕺	. [<i>UD</i>] ml	No delition	
Final density:	4000 eggs/ml		gg stock	seawater	ml	No delution required,	,
		0.37 parts s				very orives,	
Prepare the embryo stock stock (1 part) and 125 m	ck according to the calcul nl of dilution water (1.25 p	$Q \ \mathcal{Z}O \ \mathcal{N} \mathcal{P}$ ated dilution factor. For arts).	or example, if the	ne dilution factor is :	2.25, use 100 r	ml of existing	
Dangafindar Taati	0000.4 4000		Sperm:Egg Ra				
Rangefinder Test: ml Sperm Stock	2000:1 1600 50 40		<u>00:1</u> <u>400</u>		100:1	<u>50:1</u> 1.25	
ml Seawater	0.0 10		20 10 30 40		2.5 47.5	48.75	
				0 10	17.0	40.70	
	Time	Rangefinder Ratio:	Fert.	<u>Unfert.</u>			
Sperm Added (100 µl):	1940	(100)()	73	27			
Eggs Added (0.5 ml):	<u> (456</u>	150:1	92	<u>4</u>			
Test Ended:	<u> 1500 </u>	150'.1	91	9			
			P*****				
this range, choose the ra	n-to-egg ratio that results i atio closest to 90 percent ctive season, site conditio	unless professional jud	80 and 90 perdigment dictates	cent. If more than of c	one concentrat other factors (e.	tion is within .g., organism	
Definitive Test		Sperm:Egg Ratio U	sed: (50°				
		-p					
	Time		Fert.	Unfert.			
Sperm Added (100 µI):p	U13091505	QC1	90	to			
Eggs Added (0.5 ml):	1325, 1525	QC2	84	1/2			
Test Ended:	1345 1545	Egg Control 1	0	100			
		Egg Control 2	<u>0</u>	100			
Comments:							
Commonto.						77.001.00	
		Partition of the second of the		A			
QC Check:	KB (1/10/14			Final Revi	A.C.	ulalid	
ao Olieon.	10.10	<u> </u>		rınai Kevi	5W / V 🗸 _	HOLL C	

CETIS Summary Report

Report Date:

05 Aug-15 10:59 (p 1 of 1)

Test Code:

150722sprt | 00-0842-3841

					William III			rest Code:		130	zzspit j ud	J-004Z-304
Echinoid Spe	rm Cell Fertiliza	tion Test	15C							Nautilus	Environm	iental (CA)
Batch ID: Start Date: Ending Date: Duration:	07-0709-1109 22 Jul-15 16:1 22 Jul-15 16:5 40m	7 P 7 S	Protocol:EPA/600/R-95/136 (1995)ESpecies:Strongylocentrotus purpuratusE			Analyst: Diluent: Brine: Age:		ıral Seawate Applicable	er			
Sample ID: Sample Date: Receive Date: Sample Age:	: 22 Jul-15	M S	ode: aterial: ource: tation:	150722sprt Copper chlorid Reference Tox Copper Chlorid	icant			Client: Project:	Inter	nal		
Comparison S	Summary				(2)							
Analysis ID 21-1097-1868	Endpoint Fertilization Ra	te	NOEL <10	. LOEL	TOEL NA	PMSD 4.04%	TU	Meth Dunn		ultiple Com	parison Tes	st
Point Estimat Analysis ID	e Summary Endpoint		Level	μg/L	95% LCL	95% UCL	ΤU	Meth	od			
01-2722-3396	Fertilization Ra	te	EC50	35.94	33.88	38.13		Trim	med S	Spearman-K	ärber	
Test Acceptal	oility	4				- Washington						
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	its	Over	lap	Decision		
01-2722-3396	Fertilization Ra			ol Resp	0.962	0.7 - NL		Yes		Passes Ad	ceptability	Criteria
21-1097-1868	Fertilization Ra			ol Resp	0.962	0.7 - NL		Yes			ceptability	
21-1097-1868	Fertilization Ra	te	PMSD)	0.04035	NL - 0.25		No	to the total section of	Passes Ac	ceptability	Criteria
Fertilization R	tate Summary											
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std E	Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.962	0.9436	0.9804	0.94	0.98			0.01483	1.54%	0.0%
10		5	0.814	0.7462	0.8818	0.74	0.87			0.05459	6.71%	15.38%
20 40		5 5	0.75	0.6905	0.8095	0.7	0.8	0.021		0.04796	6.39%	22.04%
80		5 5	0.492 0.008	0.3812 0,002447	0.6028 0.01355	0.39	0.61			0.08927	18.15% 55.9%	48.86% 99.17%
160		5	0.008	0.002447	0.01333	0 0	0.01 0	0.002 0	•	0.004472 0	55.5%	100.0%
Fertilization R	ate Detail			***************************************	CHANGE TO THE PARTY OF THE PART							
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	0.94	0.97	0.96	0.98	0.96						
10		0.74	0.82	0.86	0.87	0.78						
20		0.8	0.79	0.76	0.7	0.7						
40		0.39	0.61	0.54	0.42	0.5						
40												
80		0.01	0.01	0.01	0	0.01						

Report Date:

05 Aug-15 10:59 (p 1 of 2)

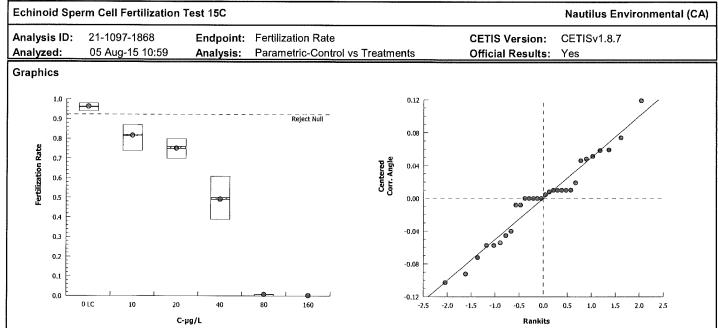
himaid Course Call Partitionation Tank 480			-		
	Test Code:	15	0722	sprt	00-0842-384
ino Analytical Nepolt					(–

Echinoid Sp	erm Ce	ell Fertiliza	ition Te	st 15C						Nautilus	Environn	nental (CA
Analysis ID: Analyzed:		097-1868 .ug-15 10:		•	ertilization Ra arametric-Cor		tments	_	IS Version: cial Results	CETISv1 : Yes	.8.7	
Data Transfo	rm		Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	ected)		NA	C > T	NA	NA		4.04%	<10	10	NA	
Dunnett Mul	tiple Co	omparisor	n Test									
Control	vs	C-μg/L		Test Sta	t Critical	MSD DF	P-Value	P-Type	Decision	(a:5%)		
Lab Control		10*		6.563	2.305	0.088 8	<0.0001	CDF	Significan	t Effect		
		20*		8.653	2.305	0.088 8	<0.0001	CDF	Significan	t Effect		
		40*		15.79	2.305	0.088 8	<0.0001	CDF	Significan	t Effect		
		80*		33.86	2.305	0.088 8	<0.0001	CDF	Significan	t Effect		
ANOVA Table	e										****	
Source		Sum Squ	ares	Mean S	quare	DF	F Stat	P-Value	Decision	(a:5%)		
Between		4.859248		1.21481	2	4	336.2	<0.0001	Significan	t Effect		
Error		0.0722656	64°	0.00361	3282	20						
Total		4.931514		117		24	_					
Distributiona	l Tests		MMm00000		***************************************					***		
Attribute		Test			Test Stat	Critical	P-Value	Decision	(a:1%)			
Variances		Bartlett E	quality o	f Variance	6.887	13.28	0.1420	Equal Var	iances			
Distribution		Shapiro-\	Nilk W Ν	lormality	0.9687	0.8877	0.6133	Normal D	istribution			
Fertilization l	Rate S	ummary				Orași - adelarea comunica - acosmi						
C-μg/L	Conti	ol Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab C	Control	5	0.962	0.9436	0.9804	0.96	0.94	0.98	0.006633	1.54%	0.0%
10			5	0.814	0.7462	0.8818	0.82	0.74	0.87	0.02441	6.71%	15.38%
20			5	0.75	0.6905	0.8095	0.76	0.7	8.0	0.02145	6.39%	22.04%
40			5	0.492	0.3812	0.6028	0.5	0.39	0.61	0.03992	18.15%	48.86%
80			5	0.008	0.002447	0.01355	0.01	0	0.01	0.002	55.9%	99.17%
160			5	0	0	0	0	0	0	0		100.0%

Angular (C	orrected) Transfor	med Sumr	nary								
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.378	1.329	1.426	1.369	1.323	1.429	0.01743	2.83%	0.0%
10		5	1.128	1.041	1.215	1.133	1.036	1.202	0.03129	6.2%	18.11%
20		5	1.049	0.9799	1.117	1.059	0.9912	1.107	0.02476	5.28%	23.88%
40		5	0.7773	0.6657	0.889	0.7854	0.6745	0.8963	0.04021	11.57%	43.57%
80		5	0.09014	0.06229	0.118	0.1002	0.05002	0.1002	0.01003	24.88%	93.46%
160		5	0.05002	0.05001	0.05003	0.05002	0.05002	0.05002	0	0.0%	96.37%

Report Date: Test Code: 05 Aug-15 10:59 (p 2 of 2)

150722sprt | 00-0842-3841



Report Date: Test Code:

05 Aug-15 10:59 (p 1 of 1)

150722sprt | 00-0842-3841

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: Analyzed:

01-2722-3396 05 Aug-15 10:59

Endpoint: Fertilization Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version: Official Results:

CETISv1.8.7

Yes

Trimmed	Spearman-Kärber	Estimates
---------	-----------------	-----------

Threshold Option Threshold Trim Мu Sigma EC50 95% LCL 95% UCL Control Threshold 0.038 15.38% 1.556 0.01281 35.94 33.88 38,13

Fertilizati	on Rate Summary				Cal	culated Varia	te(A/B)				
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.962	0.94	0.98	0.006633	0.01483	1.54%	0.0%	481	500
10		5	0.814	0.74	0.87	0.02441	0.05459	6.71%	15.38%	407	500
20		5	0.75	0.7	0.8	0.02145	0.04796	6.39%	22.04%	375	500
40		5	0.492	0.39	0.61	0.03992	0.08927	18.15%	48.86%	246	500
80		5	0.008	0	0.01	0.002	0.004472	55.9%	99.17%	4	500
160		5	0	0	0	0	0		100.0%	0	500

Graphics 0.6 0.5 0.3 0.2 0.0 20 40 60 100 120 140

C-µg/L

CETIS QC Plot Report Date: 05 Aug-15 10:59 (1 of 1)

Echinoid Sperm Cell Fertilization Test 15C

Sigma:

8.445

CV:

36.80%

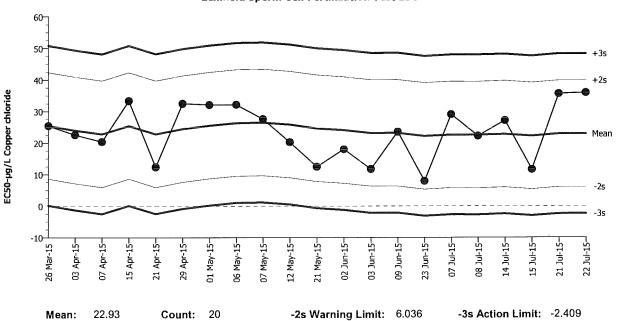
Nautilus Environmental (CA)

+3s Action Limit: 48.26

Test Type: Fertilization Organism: Strongylocentrotus purpuratus (Purpl Material: Copper chloride

Protocol: EPA/600/R-95/136 (1995) Endpoint: Fertilization Rate Source: Reference Toxicant-REF

Echinoid Sperm Cell Fertilization Test 15C



+2s Warning Limit:

39.82

Quali	ty Con	trol Data	a								
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2015	Mar	26	14:14	25.45	2.524	0.2989			18-4459-4447	20-9665-3202
2		Apr	3	14:01	22.56	-0.3686	-0.04365			07-8763-8572	13-9279-9843
3			7	13:45	20.36	-2.572	-0.3046			14-7381-4605	00-4113-7336
4			15	12:21	33.41	10.48	1.242			18-4629-1325	01-3982-8535
5			21	14:45	12.37	-10.56	-1.25			04-6687-0700	00-9564-1182
6			29	13:12	32.5	9.574	1.134			00-7279-3461	06- 1 313-6314
7		May	1	14:45	32.13	9.201	1.09			08-3887-8784	07-0987-3806
8			6	16:10	32.21	9.284	1.099			07-9821-3669	18-2350-8328
9			7	15:45	27.64	4.712	0.558			05-8209-5022	06-2905-6100
10			12	14:42	20.26	-2.668	-0.3159			20-1583-4383	17-4186-2489
11			21	15:38	12.5	-10.43	-1.236			03-1600-9362	13-2344-9592
12		Jun	2	14:10	18.03	-4.896	-0.5797			03-8226-7406	18-7623-453 1
13			3	15:00	11.77	-11.16	-1.322			01-6843-4438	09-0678-9638
14			9	15:50	23.6	0.6746	0.07988			01-7239-4208	17-5901-6261
15			23	16:10	7.931	-15	-1.776			10-4967-2005	14-2593-5380
16		Jul	7	14:33	29.07	6.143	0.7274			18-8004-9621	11-4682-8456
17			8	14:38	22.27	-0.6565	-0.07774			00-9935-8934	02-7693-7150
18			14	15:00	27.18	4.252	0.5035			17-8977-4567	05-9270-4497
19			15	14:45	11.62	-11.31	-1.34			03-6006-9976	15-4075-1359
20			21	10:41	35.64	12.71	1.505			15-5568-4316	12-3562-8162
21			22	16:17	35.94	13.01	1.541			00-0842-3841	01-2722-3396

Report Date: Test Code: 21 Jul-15 10:43 (p 1 of 1)

00-0842-3841/150722sprt

Echinoid Sp	erm C	ell Fe	rtiliza	tion Test 15	C			Nautilus Environmental (CA
Start Date: 22 Jul-15 End Date: 22 Jul-15 Sample Date: 22 Jul-15 С-µg/L Code Rep Pos					ol: EPA/600/i	150722sprt Reference Toxicant Copper Chloride		
C-µg/L	Code	Rep	Pos	# Counted	# Fertilized		Notes	The company of the co
			1	100	98		7/3//5	Ď
			2	/∞	1			
			3	100	74			
			4	/00	D			
The second secon			5	/00	B			
			6	/06	78			
			7	/60	79 96	110000		
			8	/00	96	10.3413130000000000000000000000000000000		
			9	/00	50			
			10	/∞	80			
			11	/00	70			-
			12	/00	42			
			13	100	97			
			14	/00	70			
			15	/00	<i>8</i>			
	-		16	/00	76 87			
			17	/00	3f			
			18 19	100	82			
			20	\00	54			
			21	/60				
			22	/00	86			
			23	/00	<u> </u>			
			24	/00	D'			
	-		25		4			
			26	100	\ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \	(10°44 = 10°44		
			27		94			
			28	/00	77			
			29	/00	39			and a relative
			30	/00	96			

Test Code: 00-0842-3841/150722sp	Report Date:	21 Jul-15 10:43 (p 1 of 1)
	Test Code:	00-0842-3841/150722sprt

Echinoid Sp	erm C	ell Fe	rtiliza	tion Test 15	Ü				Nautilus Environmental (CA
Start Date: End Date: Sample Date	22 、	Jul-15 Jul-15 Jul-15		Protoc	es: Strongylo ol: EPA/600/ al: Copper ch	R-95/13		Sample Code: Sample Source: Sample Station:	150722sprt Reference Toxicant Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Fertilized			Notes	
0	LC	1	27						49.
0	LC	2	13	100	96	13	7/21/15		
0	LC	3	30						
0	LC	4	1						
0	LC	5	8						
10		1	3				1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1		
10		2	18						
10		3	21	100	95				
10		4	17						
10		5	6						
20		1	10					1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	
20		2	7	(60	84			——————————————————————————————————————	
20		3	16						
20		4	14		-				
20		5	11		170 110 110				
40		1	29						
40		2	22						
40		3	19	100	3 &		***************************************		
40		4	12	, _	- 4				
40		5	9						
80		1	20 .		- MAY Viscous a				
80		2	23					dan	
80		3	2	100	0			. ((1)	
80		4	26	1					
80		5	28		******				1
160		1	5	-					
160		2	24						
160		3	25						
160		4	4	100	0				

Water Quality Measurements

Client :	Internal	

Test Species: S. purpuratus

Sample ID:

CuCl₂

Start Date/Time: 7/22/2015 1617

Test No:

150722dert លិស្សមក្សមក្រុទ

End Date/Time: 7/22/2015

1657

Dilutions made by: AC

High conc. made (μg/L): 160
Vol. Cu stock added (mL): 9.0

Final Volume (mL): 500

Cu stock concentration (µg/L): 8.80

Analyst:

AD Initial Readings Concentration DO Salinity Temperature рΗ (μg/L) (mg/L) (units) (ppt) (°C) **Lab Control** 8.9 809 33.3 149 10 8.8 8.08 8-7 20 8.08 149 8.7 8.09 40 33.4 14.8 8.7 33.3 80 8.09 147 8.8 160 14.8. 810 331

Comments:		
QC Check:	KB814115	Final Review: VCR 8/6/15

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

Marine Chronic Bio	passay			Ech	inoderm S	Sperm-Cell	Fertilizatio	n Worksheet
Client: Sample ID: Test No.: Tech initials:	Internal CuC12 150722sp	<u>(</u>		 			ne: 7 }} ies: <i>S. purpu</i> rce: Point Lo	S 1657 ratus
Injection Time:	1525					Date Collect	eu. <u>/ (</u>	113
Sperm Absorbance at 4	100 nm: <u>0,93</u>) (ta	arget range o	f 0.8 - 1.0 foi	density of	1x10 ⁶ sperm/n	nl)	
Eggs Counted:	47	Mean:	81.8_x	50 = <u>U</u>)	gs/ml		
	80 79 81		nts of 80 eggs inal density of		~	vick-Rafter		
Initial density: Final density:	4090 egg	gs/ml gs/ml		ution factor Irt egg stock Irts seawater	se	gg stock	m	
Prepare the embryo sto stock (1 part) and 125 r				. For examp	le, if the dilu	ution factor is 2	2.25, use 100	0 ml of existing
				Sperm:E	gg Ratio			
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1 50 0.0	1600:1 40 10	1200:1 30 20	800:1 20 30	400:1 10 40	200:1 5.0 45	100:1 2.5 47.5	50:1 1.25 48.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1538 1553 1603		ingefinder Ra . UU \ ZUU \ LUU \	tio: Fel	rt. <u>Un</u> 3 1 7 5 5 5	fert. 7 6		
NOTE: Choose a sperr this range, choose the r health, stage of reprodu	atio closest to 90 pe	ercent unles						
Definitive Test		Sp	erm:Egg Rat	io Used:	3504			

<u>Definitive Test</u>	Sperm:Egg Ratio Use		Sperm:Egg Ratio Used:		
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1617 1637 1657	QC1 QC2 Egg Control 1 Egg Control 2	Fert. (1) (1) (1) (1) (1)	Unfert. 9 7 100	
Comments:					

QC Check:

KB8/4/15

Final Review: VCR 816119



CETIS Summary Report

Report Date:

10 Nov-14 10:46 (p 1 of 1)

Test Code:

141030dert | 15-5280-7209

								rest Code:	141	Jouann 1	7 0200 720
Echinoid Spe	erm Cell Fertiliza	tion Test	15C						Nautilus	Environm	ental (CA)
Batch ID: Start Date:	08-7037-7425 30 Oct-14 16:1		st Type: otocol:	Fertilization EPA/600/R-95/	136 (1995)			Analyst: Diluent: Na	atural Seawate	er	
Ending Date:	30 Oct-14 16:5	0 S p	ecies:	Dendraster exc					ot Applicable		
Duration:	40m	Sc	urce:	Mission Bay				Age:			
Sample ID:	04-7273-8124	Co	de:	141030dert				Client: In	ternal	<u></u>	
Sample Date:	30 Oct-14	Ma	iterial:	Copper chloride	e			Project:			
Receive Date	: 30 Oct-14	So	urce:	Reference Toxi	cant						
Sample Age:	16h	Sta	ation:	Copper Chlorid	е						
Comparison S	Summary			A Security of the control of the con		10 000000		Caracana Santa Santa (Car	n in netter producer and a second financial a	is adver	
Analysis ID	Endpoint		NOEL	. LOEL	TOEL	PMSD	TU	Method			
19-1439-3601	Fertilization Ra	te	10	20	14.14	3.85%		Dunnett	Multiple Com	parison Tes	st
Point Estimat	e Summary		- 1111								
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Method			
15-6995-0711	Fertilization Ra	te	EC50	26.22	25.25	27.23		Trimme	d Spearman-K	ärber	
Test Acceptal	bility										
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Overlap	Decision		
15-6995-0711	Fertilization Ra	te	Contro	Control Resp 0.962 0.7 - NL				Yes	Passes Ad	ceptability	Criteria
19-1439-3601	Fertilization Ra	te		ol Resp	0.962	0.7 - NL		Yes	Passes A	ceptability	Criteria
19-1439-3601	Fertilization Ra	te	PMSE)	0.03849	NL - 0.25		No	Passes A	ceptability	Criteria
Fertilization F	Rate Summary										
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max		Std Dev	CV%	%Effect
0	Lab Control	5	0.962	0.9523	0.9717	0.92	0.99			2.69%	0.0%
2.5		5	0.984	0.9807	0.9873	0.97	0.99		0.008945	0.91%	-2.29%
5		5	0.986	0.984	0.988	0.98	0.99			0.56%	-2.5%
10		5	0.964	0.9506	0.9774	0.91	1	0.016	0.03578	3.71%	-0.21%
20								0.02421	0.05413	7.69%	26.82%
40		5	0.704	0.6838	0.7242	0.67	0.8				
40		5	0.172	0.1572	0.1868	0.12	0.21	0.01772	0.03962	23.04%	82.12%
80								0.01772	0.03962	23.04% 104.6%	82.12% 99.17%
Fertilization R		5 5	0.172 0.008	0.1572 0.004876	0.1868 0.01112	0.12	0.21	0.01772	0.03962		
80 Fertilization R C-μg/L	Control Type	5 5 Rep 1	0.172 0.008 Rep 2	0.1572 0.004876 Rep 3	0.1868 0.01112 Rep 4	0.12 0 Rep 5	0.21	0.01772	0.03962		
Fertilization R C-µg/L 0		5 5 Rep 1 0.97	0.172 0.008 Rep 2 0.96	0.1572 0.004876 Rep 3 0.92	0.1868 0.01112 Rep 4 0.97	0.12 0 Rep 5 0.99	0.21	0.01772	0.03962		
Fertilization R C-µg/L 0 2.5	Control Type	5 5 Rep 1 0.97 0.98	0.172 0.008 Rep 2 0.96 0.99	0.1572 0.004876 Rep 3 0.92 0.97	0.1868 0.01112 Rep 4 0.97 0.99	0.12 0 Rep 5 0.99 0.99	0.21	0.01772	0.03962		
Fertilization R C-µg/L 0 2.5 5	Control Type	5 5 Rep 1 0.97	0.172 0.008 Rep 2 0.96 0.99 0.99	0.1572 0.004876 Rep 3 0.92 0.97 0.98	0.1868 0.01112 Rep 4 0.97 0.99 0.99	0.12 0 Rep 5 0.99 0.99 0.99	0.21	0.01772	0.03962		
Fertilization R C-µg/L 0 2.5	Control Type	5 5 Rep 1 0.97 0.98	0.172 0.008 Rep 2 0.96 0.99	0.1572 0.004876 Rep 3 0.92 0.97	0.1868 0.01112 Rep 4 0.97 0.99	0.12 0 Rep 5 0.99 0.99	0.21	0.01772	0.03962		
Fertilization R C-µg/L 0 2.5 5	Control Type	5 5 Rep 1 0.97 0.98 0.98	0.172 0.008 Rep 2 0.96 0.99 0.99	0.1572 0.004876 Rep 3 0.92 0.97 0.98	0.1868 0.01112 Rep 4 0.97 0.99 0.99	0.12 0 Rep 5 0.99 0.99 0.99	0.21	0.01772	0.03962		
Fertilization R C-μg/L 0 2.5 5	Control Type	5 5 Rep 1 0.97 0.98 0.98	0.172 0.008 Rep 2 0.96 0.99 0.99	0.1572 0.004876 Rep 3 0.92 0.97 0.98 0.95	0.1868 0.01112 Rep 4 0.97 0.99 0.99 0.97	0.12 0 Rep 5 0.99 0.99 0.99 0.99	0.21	0.01772	0.03962		

Report Date:

10 Nov-14 10:46 (p 1 of 2)

							Test	Code:	1410	030dert 18	5-5280-720
Echinoid Sp	perm Cell Fertiliz	ation Test	15C						Nautilus	Environn	nental (CA
Analysis ID: Analyzed:	19-1439-3601 10 Nov-14 10:		•	tilization Ra ametric-Cor		tments		IS Version: cial Results:	CETISv1 Yes	.8.4	
Data Transfe	orm	Zeta	Alt Hyp	Trials	Seed		NOEL	LOEL	TOEL	TU	PMSD
Angular (Cor	rected)	NA	C > T	NA	NA		10	20	14.14		3.85%
Dunnett Mu	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		-1.709	2.407	0.090 8	0.9988	CDF		icant Effect		
	5		-1.88	2.407	0.090 8	0.9993	CDF	_	icant Effect		
	10		-0.4377	2.407	0.090 8	0.9442	CDF	_	icant Effect		
	20*		10.32	2.407	0.090 8	<0.0001	CDF	Significant			
	40*		25.58	2.407	0.090 8	<0.0001	CDF	Significant			
	80*		34.58	2.407	0.090 8	<0.0001	CDF	Significant			
ANOVA Tabl	le										
Source	Sum Sqւ	ıares	Mean Squ	ıare	DF	F Stat	P-Value	Decision(a:5%)		
Between	9.343492		1.557249		6	444.1	<0.0001	Significant	Effect		
Error	0.098192	94	0.0035068	39	28			J			
Total	9.441685	-	· · · · · · · · · · · · · · · · · · ·		34	-					
Distribution	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision((a:1%)			
Variances	Bartlett E	quality of '	√ariance	9.709	16.81	0.1374					
Distribution							⊏quai var	lances			
Distribution	Snapiro-	Wilk W No	mality	0.9771	0.9146	0.6641	Equal Var Normal Di				
	Rate Summary	Wilk W No	mality		0.9146		-				
		Wilk W No	mality Mean		0.9146 95% UCL		-		Std Err	CV%	%Effect
Fertilization	Rate Summary			0.9771		0.6641	Normal Di	stribution	Std Err 0.01158		
Fertilization C-µg/L	Rate Summary Control Type	Count	Mean	0.9771 95% LCL	95% UCL	0.6641 Median	Normal Di	stribution Max		2.69%	0.0%
Fertilization C-µg/L 0 2.5	Rate Summary Control Type	Count 5	Mean 0.962	0.9771 95% LCL 0.9299	95% UCL 0.9941	0.6641 Median 0.97	Min 0.92	Max 0.99	0.01158	2.69% 0.91%	0.0% -2.29%
Fertilization C-µg/L 0 2.5	Rate Summary Control Type	Count 5 5	Mean 0.962 0.984	0.9771 95% LCL 0.9299 0.9729	95% UCL 0.9941 0.9951	0.6641 Median 0.97 0.99 0.99	Min 0.92 0.97 0.98	Max 0.99 0.99	0.01158 0.004	2.69% 0.91% 0.56%	0.0% -2.29% -2.5%
Fertilization C-µg/L 0 2.5 5	Rate Summary Control Type	Count 5 5 5	Mean 0.962 0.984 0.986	0.9771 95% LCL 0.9299 0.9729 0.9792	95% UCL 0.9941 0.9951 0.9928	0.6641 Median 0.97 0.99 0.99 0.97	Min 0.92 0.97 0.98 0.91	Max 0.99 0.99 0.99	0.01158 0.004 0.00245	2.69% 0.91%	0.0% -2.29% -2.5% -0.21%
Fertilization C-µg/L 0 2.5 5 10	Rate Summary Control Type	Count 5 5 5 5 5	Mean 0.962 0.984 0.986 0.964	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196	95% UCL 0.9941 0.9951 0.9928 1	0.6641 Median 0.97 0.99 0.99 0.97 0.68	Min 0.92 0.97 0.98	Max 0.99 0.99 0.99 1	0.01158 0.004 0.00245 0.016	2.69% 0.91% 0.56% 3.71%	0.0% -2.29% -2.5% -0.21% 26.82%
Fertilization C-µg/L 0 2.5 5 10 20 40	Rate Summary Control Type	Count 5 5 5 5 5 5 5	Mean 0.962 0.984 0.986 0.964 0.704	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368	95% UCL 0.9941 0.9951 0.9928 1 0.7712	0.6641 Median 0.97 0.99 0.99 0.97	Min 0.92 0.97 0.98 0.91 0.67	Max 0.99 0.99 0.99	0.01158 0.004 0.00245 0.016 0.02421	2.69% 0.91% 0.56% 3.71% 7.69%	0.0% -2.29% -2.5% -0.21%
Fertilization C-μg/L 0 2.5 5 10 20 40 80	Rate Summary Control Type	Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19	Min 0.92 0.97 0.98 0.91 0.67	Max 0.99 0.99 0.99 1 0.8 0.21	0.01158 0.004 0.00245 0.016 0.02421 0.01772	2.69% 0.91% 0.56% 3.71% 7.69% 23.04%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12%
Fertilization C-µg/L 0 2.5 5 10 20 40 80 Angular (Cor	Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19	Min 0.92 0.97 0.98 0.91 0.67	Max 0.99 0.99 0.99 1 0.8 0.21	0.01158 0.004 0.00245 0.016 0.02421 0.01772	2.69% 0.91% 0.56% 3.71% 7.69% 23.04%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12% 99.17%
Fertilization C-µg/L 0 2.5 5 10 20 40 80 Angular (Cor	Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 5 med Sum	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228 0	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212 0.01839	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19 0.01	Min 0.92 0.97 0.98 0.91 0.67 0.12	Max 0.99 0.99 0.99 1 0.8 0.21 0.02	0.01158 0.004 0.00245 0.016 0.02421 0.01772 0.003742	2.69% 0.91% 0.56% 3.71% 7.69% 23.04% 104.6%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12%
Fertilization C-µg/L 0 2.5 5 10 20 40 80 Angular (Cor	Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 5 5 Count Count	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008 mary Mean	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228 0	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212 0.01839	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19 0.01 Median	Min 0.92 0.97 0.98 0.91 0.67 0.12 0	Max 0.99 0.99 0.99 1 0.8 0.21 0.02	0.01158 0.004 0.00245 0.016 0.02421 0.01772 0.003742 Std Err	2.69% 0.91% 0.56% 3.71% 7.69% 23.04% 104.6%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12% 99.17% %Effect 0.0%
Fertilization C-µg/L 0 2.5 5 10 20 40 80 Angular (Cor C-µg/L 0 2.5	Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 5 7 med Sum Count 5	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008 mary Mean 1.384	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228 0 95% LCL 1.3	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212 0.01839 95% UCL 1.467	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19 0.01 Median 1.397	Min 0.92 0.97 0.98 0.91 0.67 0.12 0 Min 1.284 1.397	Max 0.99 0.99 0.99 1 0.8 0.21 0.02 Max 1.471 1.471	0.01158 0.004 0.00245 0.016 0.02421 0.01772 0.003742 Std Err 0.03003	2.69% 0.91% 0.56% 3.71% 7.69% 23.04% 104.6% CV% 4.85% 2.33%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12% 99.17% %Effect 0.0% -4.63%
Fertilization C-µg/L 0 2.5 5 10 20 40 80 Angular (Cor C-µg/L 0 2.5	Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 5 Count Count 5 5 5	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008 mary Mean 1.384 1.447	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228 0 95% LCL 1.3 1.406	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212 0.01839 95% UCL 1.467 1.489	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19 0.01 Median 1.397 1.471 1.471	Min 0.92 0.97 0.98 0.91 0.67 0.12 0 Min 1.284 1.397 1.429	Max 0.99 0.99 1 0.8 0.21 0.02 Max 1.471 1.471 1.471	0.01158 0.004 0.00245 0.016 0.02421 0.01772 0.003742 Std Err 0.03003 0.01505 0.01022	2.69% 0.91% 0.56% 3.71% 7.69% 23.04% 104.6% CV% 4.85% 2.33% 1.57%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12% 99.17% %Effect 0.0% -4.63% -5.09%
Fertilization C-μg/L 0 2.5 5 10 20 40 80	Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 5 Count Count 5 5 5 5	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008 mary Mean 1.384 1.447 1.454	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228 0 95% LCL 1.3 1.406 1.426 1.275	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212 0.01839 95% UCL 1.467 1.489 1.482 1.525	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19 0.01 Median 1.397 1.471	Min 0.92 0.97 0.98 0.91 0.67 0.12 0 Min 1.284 1.397 1.429 1.266	Max 0.99 0.99 1 0.8 0.21 0.02 Max 1.471 1.471 1.521	0.01158 0.004 0.00245 0.016 0.02421 0.01772 0.003742 Std Err 0.03003 0.01505 0.01022 0.045	2.69% 0.91% 0.56% 3.71% 7.69% 23.04% 104.6% CV% 4.85% 2.33% 1.57% 7.19%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12% 99.17% %Effect 0.0% -4.63% -5.09% -1.19%
Fertilization C-μg/L 0 2.5 5 10 20 40 80 Angular (Cor C-μg/L 0 2.5 5	Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 5 Count Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.962 0.984 0.986 0.964 0.704 0.172 0.008 mary Mean 1.384 1.447 1.454 1.4	0.9771 95% LCL 0.9299 0.9729 0.9792 0.9196 0.6368 0.1228 0 95% LCL 1.3 1.406 1.426	95% UCL 0.9941 0.9951 0.9928 1 0.7712 0.2212 0.01839 95% UCL 1.467 1.489 1.482	0.6641 Median 0.97 0.99 0.99 0.97 0.68 0.19 0.01 Median 1.397 1.471 1.471 1.397	Min 0.92 0.97 0.98 0.91 0.67 0.12 0 Min 1.284 1.397 1.429	Max 0.99 0.99 1 0.8 0.21 0.02 Max 1.471 1.471 1.471	0.01158 0.004 0.00245 0.016 0.02421 0.01772 0.003742 Std Err 0.03003 0.01505 0.01022	2.69% 0.91% 0.56% 3.71% 7.69% 23.04% 104.6% CV% 4.85% 2.33% 1.57%	0.0% -2.29% -2.5% -0.21% 26.82% 82.12% 99.17% %Effect 0.0% -4.63% -5.09%

0.3

0,1

0.0

2.5

10

C-µg/L

Report Date: Test Code: 10 Nov-14 10:46 (p 2 of 2) 141030dert | 15-5280-7209

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 19-1439-3601 Endpoint: Fertilization Rate **CETIS Version:** CETISv1.8.4 Parametric-Control vs Treatments Analyzed: 10 Nov-14 10:46 Analysis: Official Results: Yes Graphics 0.16 F 0.14 Reject Null 0.9 0.12 0.10 8,0 Fertilization Rate 0.08 -@-0.06 0.04 0,6 0.02 0.5 0.00 -0.02

•

-0.04 -0.06

-0,08

-0.10 -0.12

-0.14

-0.16

-1.5

-0.5 0.0

Rankits

Report Date:

10 Nov-14 10:46 (p 1 of 1)

Test Code:

141030dert | 15-5280-7209

Echinoid Spe	erm Cell Fertilization	n Test 15C

Nautilus Environmental (CA)

Analysis ID: Analyzed:

15-6995-0711 10 Nov-14 10:46

Endpoint: Fertilization Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version: Official Results:

CETISv1.8.4

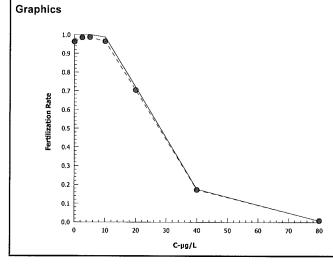
•	•	,	9		• •	•	•	C	•	
	***	Ξ	-	. 1.				į,	j.	

Trimmed Spearman-Kä	rber Estimates					
Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCI

CL 95% UCL

THE CONTRACT OF MAIN	71110011010	* * * * * * * * * * * * * * * * * * * *	mu	Olgina	L000	30 /0 LOL	33 /0 00
Control Threshold	0.038	0.82%	1.419	0.008184	26.22	25.25	27.23

Fertilization	on Rate Summary	Calculated Variate(A/B)									
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.962	0.92	0.99	0.01158	0.02588	2.69%	0.0%	481	500
2.5		5	0.984	0.97	0.99	0.004	0.008945	0.91%	-2.29%	492	500
5		5	0.986	0.98	0.99	0.00245	0.005479	0.56%	-2.5%	493	500
10		5	0.964	0.91	1	0.016	0.03578	3.71%	-0.21%	482	500
20		5	0.704	0.67	0.8	0.02421	0.05413	7.69%	26.82%	352	500
40		5	0.172	0.12	0.21	0.01772	0.03962	23.04%	82.12%	86	500
80		5	0.008	0	0.02	0.003742	0.008367	104.6%	99.17%	4	500



+3s Action Limit: 74.91

Sigma:

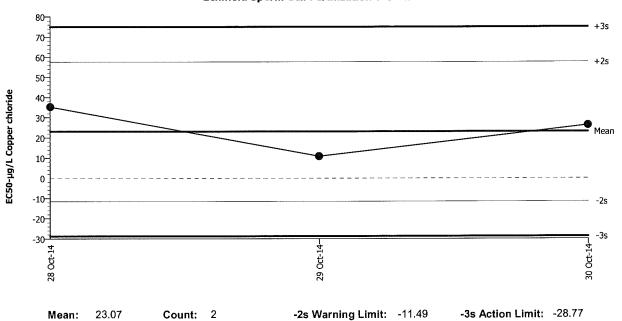
17.28

CV:

74.90%

Nautilus Environmental (CA) **Echinoid Sperm Cell Fertilization Test 15C** Organism: Dendraster excentricus (Sand Dollar) Copper chloride Material: Test Type: Fertilization Reference Toxicant-REF Endpoint: Fertilization Rate Source: Protocol: EPA/600/R-95/136 (1995)

Echinoid Sperm Cell Fertilization Test 15C



Quali	ty Con	trol Data	а						
Point	Year	Month	Day	QC Data	Delta	Sigma	Warning Action	Test ID	Analysis ID
1	2014	Oct	28	35.29	12.22	0.7071		13-6517-2086	18-6351-1063
2			29	10.85	-12.22	-0.7069		08-2984-3114	08-2412-4232
3			30	26.22	3.151	0.1823		15-5280-7209	15-6995-0711

+2s Warning Limit: 57.63

CETIS Test Data Worksheet

Report Date: Test Code: 28 Oct-14 15:56 (p 1 of 1)

15-5280-7209/141030dert

Echinoid Sp	erm C	ell Fei	rtilizat	tion Test 150				Nautilus Environmental (CA
Start Date: End Date: Sample Date	30 (Oct-14 Oct-14 Oct-14				er excentricus R-95/136 (1995) Noride		141030dert Reference Toxicant Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Fertilized		Notes	
			1	/00	1		11/7/10	4
			2	100	100			
			3	100	20			
			4	100	18	(2C Ohok: 19.1	CBH10/14	
			5	100	6 <i>9</i>	QC check: 19,10 QC Check: 12,61	3 1110 14	
			6	100	91	QCO SP 151, D.		
-			7	\60	91 2			
A CONTRACTOR OF THE PROPERTY O			8	100	1			
			9	700	97			
			10	100	80			
			11	,100	99			
			12	100	99			
******			13	,00	99 \2			
			14	/00	(2			
			15	/00	99			
			16	700	98			
			17	/00/	67			
			18	00/	67.			
			19	/00	99 \9 68			
			20	/00/	\9			
			21	00/	68			
			22	/00	95			
			23	/00	99			
			24	/00	98			
			25	/\pi	99			
			26	00/	97			
			27	100	97			
· · · · · · · · · · · · · · · · · · ·			28	100	68			
			29		99			
			30	/00	14			
	1		31	100	99			
			32	/00 /00	97			
			33	000	96			
			34	700	92			
	1		35	100	98		•	

CETIS Test Data Worksheet

Report Date: Test Code:

28 Oct-14 15:56 (p 1 of 1) 15-5280-7209/141030dert

Echinoid Sp	erm C	ell Fe	rtiliza	tion Test 150				Nautilus Environmental (CA)
Start Date: End Date: Sample Date	30 (Oct-14 Oct-14 Oct-14	ļ			r excentricus R-95/136 (1995) Ioride		141030dert Reference Toxicant Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Fertilized		Notes	
0	LC	1	9	100	97			
0	LC	2	33		1,			
0	LC	3	34				4.00	
0	LC	4	32					
0	LC	5	23				MA AMERICAN DE CONTRACTOR DE C	
2.5		1	35	100	97			
2.5		2	15					
2,5		3	27					
2,5		4	13					
2,5		5	25					
5		1	16	100	(0)			
5		2	29					
5		3	24					
5		4	19					
5		5	12					
10		1	2	100	92			
10		2	6					
10		3	22					
10		4	26					
10		5	31					
20		1	10	100	72			
20		2	28					
20		3	21					
20		4	5					
20		5	18					
40		1	14	100	10			
40		2	4					
40		3	20					
40		4	30					
40		5	3					
80		1	1					
80		2	7					
80		3	11	100]			
80		4	17					
80		5	8					

QCSAC

Marine Chronic Bioassay

Water Quality Measurements

Client :	Internal	Test Species: D. excentricus
		1001 openion B. oxeorxirode

Sample ID: CuCl₂ Start Date/Time: 10/30/2014 \ \(\chi^{-1} \) \(\chi^{-1} \)

Test No: 141030dert End Date/Time: 10/30/2014 16:50

Dilutions made by:

High conc. made (μg/L): 80

Vol. Cu stock added (mL): 4,4

Final Volume (mL): 500

Cu stock concentration (μg/L): 9 6 5 0

		Initial R	eadings	
Concentration (μg/L)	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	8.5	8.02	33.4	14.8
2.5	8.7	8.03	33.4	14.5
5	8.8	8.04	33.4	14.4
10	G.4	G.06	33.5	14.4
20	8.8	8.06	33.5	14.4
40	G .4	8.07	33.4	14,3
80	4.9	8.08	33.3	14,4

Comments:			10000
			M 1.1
QC Check:	KB 11/10/14	Final Review:	AC 11/18/14

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

Marine Chronic Bioassay

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client:	Internal		Sta	rt Date/Time: <u>10/30/201</u>	4/ (4:10
Sample ID:	141030 dert		Er	nd Date/Time: 10/30/201	
Test No.:	9 <u>CUC12</u>			Species: D. excen	
—	۸.			nimal Source: Mission B	Bay
Tech initials:	100		Da	ate Collected: <u>「ぴん</u> ぴ	1114
Injection Time:	15:15				
Sperm Absorbance at 4		_(target range of 0.8 - 1.0			
Eggs Counted:	37 Mean	: 43.8 x 50 =	2190 eggs/	ml	
	41				
		counts of 80 eggs per vertic		Rafter	
	slide fo	r a final density of 4000 egg	s/ml)		
	64				
	<u>w</u>	,			
Initial density:	$\mathcal{I}(\mathcal{O})$ eggs/ml	= 0.55 dilution fact	tor egg s	tock 🗂 ml	
Final density:	4000 eggs/ml	 1.0 part egg ste 	ock seawa	ater ml	
		0,45 parts seaw	ater 45 mil pa	oured of per	100 able
D		Se	things	overed of per	
	ck according to the calculat nl of dilution water (1.25 pai	ed dilution lactor, For exa	ample, if the dilutior	i factor is 2.25, use 100	ml of existing
Stock (1 part) and 120 fi	ii oi diidiiori water (1.25 pai	13).			
		Sper	m:Egg Ratio		
Rangefinder Test:	2000:1 1600:			200:1 50/100:1	50:1
ml Sperm Stock	50 40	30 20	10	5.0 3.75 2.5	1.25
ml Seawater	0.0 10	20 30	40	45 46.25 47.5	48.75
	Time	Rangefinder Ratio:	Fert. Unfert		
Sperm Added (100 µl):	1535	57	707 33	:	
Eggs Added (0.5 ml):	15:50	100		•	
Test Ended:	10:00	150	7/2 H	•	
Tool Endod.			142	•	
				•	
NOTE: Choose a spern	n-to-egg ratio that results in	fertilization between 80 a	nd 90 percent. If n	nore than one concentra	ation is within
this range, choose the ra	atio closest to 90 percent u	nless professional judgme			
health, stage of reprodu-	ctive season, site condition	s).			
Definition Test		Sperm:Egg Ratio Used:	150:1		
Definitive Test		Sperm:Egg Ratio Used:	150.1		
	Time		Fert, Unfert		
Sperm Added (100 µl):	1/0:10	QC1	98 2	!	
Eggs Added (0.5 ml):	112:30	QC2	95 a	•	
Test Ended:	16:50	Egg Control 1	100		
rest Ended.		Egg Control 2	$\frac{1}{2}$)	
		Egg Control 2		,	
Comments:					
					_
	VB WINE.			E. 10 . ~1 ~ .	1. 1.16
QC Check:	10 110 14	-		Final Review: AC /	111977

CETIS Summary Report

Report Date: Test Code: 05 Aug-15 09:29 (p 1 of 1)

t Code: 150722dert | 10-2804-3517

								Test Code:		150	722dert 10	J-2804-351
Echinoid Spe	rm Cell Fertiliza	tion Test '	5C							Nautilus	s Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	06-2792-8877 22 Jul-15 15:20 22 Jul-15 16:00 40m	Pro Spe	st Type: otocol: ecies: urce:	Fertilization EPA/600/R-95 Dendraster ex Mission Bay	` ,			Analyst: Diluent: Brine: Age:		ural Seawate Applicable	er	
Sample ID: Sample Date: Receive Date: Sample Age:	: 22 Jul-15	So	de: terial: urce: tion:	150722dert Copper chloride Reference Toxicant Copper Chloride				Client: Project:	Inte	rnal		
Comparison S	Summary											
Analysis ID	Endpoint		NOEL	. LOEL	TOEL	PMSD	TU	Metl	hod			
14-8653-0565	Fertilization Rat	e	<10	10	NA	8.25%		Duni	nett M	lultiple Com	parison Tes	st
Point Estimat	e Summary							XXX T AND X				
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Metl	hod			
10-4495-5773	Fertilization Rat	e	EC50		16.14	17.45		Trim	med	Spearman-k	(ärber	
Test Acceptab	oility											
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Ove	rlap	Decision		
10-4495-5773	Fertilization Rat	e	Contro	ol Resp	0.874	0.7 - NL		Yes		Passes A	cceptability	Criteria
14-8653-0565	Fertilization Rat	e	Contro	ol Resp	0.874	0.7 - NL		Yes		Passes A	cceptability	Criteria
14-8653-0565	Fertilization Rat	е	PMSE)	0.08252	NL - 0.25		No		Passes A	cceptability	Criteria
Fertilization R	late Summary											
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Мах	Std	Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.874	0.8332	0.9148	0.83	0.92	0.01	47	0.03286	3.76%	0.0%
10		5	0.768	0.6745	0.8615	0.65	0.84	0.03	367	0.0753	9.81%	12.13%
20		5	0.304	0.2092	0.3988	0.21	0.41	0.03	415	0.07635	25.12%	65.22%
40		5	0	0	0	0	0	0		0		100.0%
80		5	0	0	0	0	0	0		0		100.0%
160		5	0	0	0	0	0	0		0		100.0%
Fertilization R	ate Detail											
C-μg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		**************************************				
0	Lab Control	0.92	0.88	0.88	0.83	0.86						
10		0.74	8.0	0.84	0.81	0.65						
20		0.3	0.34	0.26	0.21	0.41						
40		0	0	0	0	0						
80		0	0	0	0	0						
160		0	0	0	0	0						

Analyst: 40 QA: 1587/15

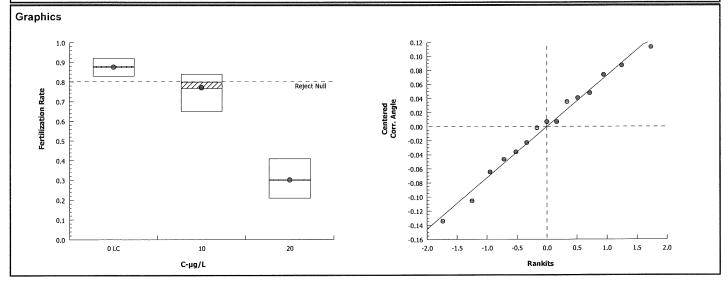
CETIS™ v1.8.7.20

Report Date: Test Code:

05 Aug-15 09:29 (p 1 of 1) 150722dert | 10-2804-3517

Echinoid Sp	erm C	ell Fertiliza	tion Te	est 150	3							Nautilus	Environm	nental (CA)
Analysis ID: Analyzed:		3653-0565 Aug-15 9:28	3	Endp Analy		tilization Rat ametric-Con		reat	tments		S Version: ial Results:	CETISv1 Yes	.8.7	
Data Transfo	orm		Zeta		Alt Hyp	Trials	Seed			PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	ected)		NA		C > T	NA	NA			8.25%	<10	10	NA	
Dunnett Mul	tiple C	omparisor	n Test											
Control	vs	C-µg/L			Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(a:5%)		
Lab Control		10*			2.894	2.108	0.101	8	0.0123	CDF	Significant	Effect		
		20*			13.15	2.108	0.101	8	<0.0001	CDF	Significant	Effect		
ANOVA Tabl	е										<u> </u>		d. in the second second	
Source		Sum Squ	ares		Mean Squ	are	DF		F Stat	P-Value	Decision(a:5%)		
Between		1.091132			0.5455661		2		95.55	<0.0001	Significant	Effect		
Error		0.0685191	11		0.0057099	26	12							
Total		1.159651					14					***************************************		
Distributiona	al Test	s												
Attribute		Test				Test Stat	Critica	1	P-Value	Decision((a:1%)			
Variances		Bartlett E	quality	of Var	iance	1.147	9.21		0.5636	Equal Var	iances			
Distribution		Shapiro-\	Nilk W	Norma	ality	0.982	0.8328		0.9812	Normal Di	stribution			
Fertilization	Rate S	ummary												
C-µg/L	Cont	rol Type	Cour	nt	Mean	95% LCL	95% U	CL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab (Control	5		0.874	0.8332	0.9148		0.88	0.83	0.92	0.0147	3.76%	0.0%
10			5		0.768	0.6745	0.8615		8.0	0.65	0.84	0.03367	9.81%	12.13%
20			5		0.304	0.2092	0.3988		0.3	0.21	0.41	0.03415	25.12%	65.22%

Angular (Corrected) Transformed Summary											
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.21	1.147	1.273	1.217	1.146	1.284	0.0226	4.18%	0.0%
10		5	1.072	0.9636	1.18	1.107	0.9377	1.159	0.03904	8.14%	11.43%
20		5	0.5816	0.4781	0.6852	0.5796	0.476	0.6949	0.0373	14.34%	51.94%



Report Date:

05 Aug-15 09:29 (p 1 of 1)

Test Code:

150722dert | 10-2804-3517

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: Analyzed:

10-4495-5773 05 Aug-15 9:28 Endpoint: Fertilization Rate Trimmed Spearman-Kärber Analysis:

CETIS Version: Official Results:

Yes

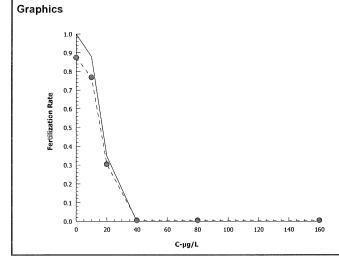
CETISv1.8.7

Trimmed Spearman-Kärber Estimates

EC50 95% UCL **Threshold Option** Threshold Trim Mu Sigma 95% LCL

Control Threshold 0.126 12.13% 1.225 0.008463 16.78 16.14 17.45

ntrol 5	Mean 0.874	Min 0.83	Max	Std Err	Std Dev	CV%	%Effect	Α	В
	0.874	0.83							
_		0.03	0.92	0.0147	0.03286	3.76%	0.0%	437	500
5	0.768	0.65	0.84	0.03367	0.0753	9.81%	12.13%	384	500
5	0.304	0.21	0.41	0.03415	0.07635	25.12%	65.22%	152	500
5	0	0	0	0	0		100.0%	0	500
5	0	0	0	0	0		100.0%	0	500
5	0	0	0	0	0		100.0%	0	500
	5 5 5	5 0.304 5 0 5 0	5 0.304 0.21 5 0 0 5 0 0	5 0.304 0.21 0.41 5 0 0 0 5 0 0 0	5 0.304 0.21 0.41 0.03415 5 0 0 0 0 5 0 0 0 0	5 0.304 0.21 0.41 0.03415 0.07635 5 0 0 0 0 0 5 0 0 0 0 0	5 0.304 0.21 0.41 0.03415 0.07635 25.12% 5 0 0 0 0 0 5 0 0 0 0 0	5 0.304 0.21 0.41 0.03415 0.07635 25.12% 65.22% 5 0 0 0 0 100.0% 5 0 0 0 0 100.0%	5 0.304 0.21 0.41 0.03415 0.07635 25.12% 65.22% 152 5 0 0 0 0 100.0% 0 5 0 0 0 0 100.0% 0



CETIS™ v1.8.7.20

CETIS QC Plot Report Date: 07 Aug-15 10:06 (1 of 1)

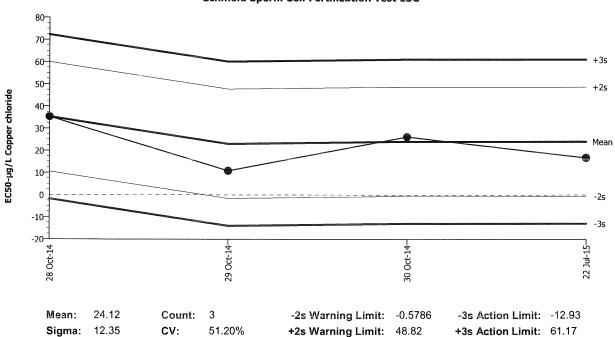
Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Test Type: Fertilization Organism: Dendraster excentricus (Sand Dollar) Material: Copper chloride

 Protocol:
 EPA/600/R-95/136 (1995)
 Endpoint:
 Fertilization Rate
 Source:
 Reference Toxicant-REF

Echinoid Sperm Cell Fertilization Test 15C



Quality Control Data	a	a	D	Ы	c	tr	n	o	C	tγ	li	ıa	Q	
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Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning Action	Test ID	Analysis ID
1	2014	Oct	28	13:00	35.29	11.17	0.9043		13-6517-2086	18-6351-1063
2			29	15:45	10.85	-13.27	-1.074		08-2984-3114	08-2412-4232
3			30	16:10	26.22	2.101	0.1701		15-5280-7209	15-6995-0711
4	2015	Jul	22	15:20	16.78	-7.338	-0.5941		10-2804-3517	10-4495-5773

000-089-170-2 CETIS™ v1.8.7.20

CETIS Test Data Worksheet

Report Date:

21 Jul-15 10:52 (p 1 of 1)

Test Code: 10-2804-3517/150722dert

Echinoid Sp	erm C	ell Fe	rtiliza	tion Test 15	С				Nautilus Environmental (CA)
Start Date: End Date: Sample Date	22 .	Jul-15 Jul-15 Jul-15		Protoc	es: Dendraste col: EPA/600/f al: Copper ch	R-95/136 (1995)			150722dert Reference Toxicant Copper Chloride
C-µg/L	Code	Rep	Pos	# Counted	# Fertilized			Notes	
			1	100	83				
			2	100	26				
			3	100	0				
			4	100	74				
			5	100	21				
10000000000000000000000000000000000000			6	100	<u> </u>				
			7	100	Q				
			8	100	65	Q 18	BJ	5/4/5	
				100		AC 8/4/15			
			10	100	O			***************************************	
			11	100	83				
			12	100	<u>0</u>				
-			13	100	0				
			14 15	100	0				
			16						
			17	100	84				
			18		0				
			19	100	30			40.000	
			20	100)	80				
			21	100	00				
			22	100	86			_	
			23	100	0				
			24	100	92				
			25	100	81	is a manufacture of the state o			
			26	100	0				
			27	100	41				~~~
			28	(00)	0	i i			
			29	100	0				
			30	100	34	y			

CETIS Test Data Worksheet

Report Date: Test Code: 21 Jul-15 10:52 (p 1 of 1) 10-2804-3517/150722dert

Echinoid Sperm Cell Fertilization Test 15CNautilus Environmental (CA)Start Date:22 Jul-15Species:Dendraster excentricusSample Code:150722dertEnd Date:22 Jul-15Protocol:EPA/600/R-95/136 (1995)Sample Source:Reference ToxicantSample Date:22 Jul-15Material:Copper chlorideSample Station:Copper Chloride

C-μg/L	Code	Rep	Pos	# Counted	# Fertilized	Notes
0	LC	1	24	100	89	AC 7122/15
0	LC	2	9			
0	LC	3	1			
0	LC	4	11			
0	LC	5	22			
10		1	4	100	89	Ac 1/22/11
10		2	20			
10		3	16			
10		4	25			
10		5	8			
20		1	18	(00)	27	AC 7/22/15
20		2	30)	
20		3	2			
20		4	5			
20		5	27			
40		1	17	iw	0	AC 7/22/15
40		2	29			
40		3	6			
40		4	12			
40		5	10			
80		1	19	100	0	AC 7/22 15
80		2	14			
80		3	7			
80		4	23			
80		5	3			
160		1	26	100	Ũ	dc 7/22/15
160		2	28			
160		3	13			
160		4	15			
160		5	21			

QC1, y

Analyst: AC QA: 4587/15

000-089-170-2

CETIS™ v1.8.7.20

Marine Chronic Bioassay

Water Quality Measurements

Analyst:

Client :	Internal	Test Species: D. excentricus
Sample ID:	CuCl ₂	Start Date/Time: 7/22/2015 520
Test No:	150722dert And Q18 7/21/15	End Date/Time: <u>7/22/2015</u> しん

Dilutions made by: ____A

High conc. made (μg/L): 160

Vol. Cu stock added (mL): 9.0

Final Volume (mL): 500

Cu stock concentration (μg/L): 3,850

	9			AD
			eadings	
Concentration (μg/L)	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	8.9	8.09	33-3	14.9
10	8.8	8.08	33.5	14.5
20	8.7	8.08	335	14.9
40	8.7	8.09	33.4	14.8
80	8.7	8.09	33.3	H.7
160	8.8	8-10	33.1	14.8

Comments:		
QC Check:	AC 8/5/15	Final Review: Y5 8715

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

Marine Chronic Bioassay

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	Internal CuC12 150722 ded	0 AC 6188	- <u>]</u> 3/15	E	nd Date/Tir Speci	me: 7/22/19 me: 7/22/ des: <i>D. excer</i> rce: Mission I	ntricus
Tech initials: Injection Time:	AC 1430					ed: <u>7/17/201</u>	
Sperm Absorbance at 40	00 nm: <u>(6</u>	_(target range of 0).8 - 1.0 for de	ensity of 4x1	0 ⁶ sperm/n	nl)	
Eggs Counted:	<u>(67</u> 54 (target	n: <u>(03</u> & x 5 counts of 80 eggs pe or a final density of 40	er vertical pass				
Initial density: Final density:	3100 eggs/ml 4000 eggs/ml	- <u>1.0</u> part	ion factor egg stock s seawater	egg s seaw		ml ml	
Prepare the embryo stock stock (1 part) and 125 m	ck according to the calcula all of dilution water (1.25 pa	ted dilution factor. rts).	For example,	if the dilutio	n factor is 2	2.25, use 100) ml of existing
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1 1600: 50 40 0.0 10	1 1200:1 30 20	Sperm:Egg 800:1 20 30	Ratio 400:1 10 40	200:1 5.0 45	100:1 2.5 47.5	50:1 1.25 48.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1445 1500	Rangefinder Ratio	5: <u>Fert.</u> 8 (4	Unfer	<u>t.</u> - - -		
this range, choose the ra	n-to-egg ratio that results in atio closest to 90 percent u ctive season, site condition	nless professional j					
Definitive Test		Sperm:Egg Ratio	Used: 10	0	_		
Sperm Added (100 µI): Eggs Added (0.5 mI): Test Ended:	Time 1520 1540 1600	QC1 QC2 Egg Control 1 Egg Control 2	Fert. 97 87 0	Unfer 3 13	<u>t.</u> - - -		
Comments:							100000000000000000000000000000000000000
QC Check:	AC815/15	_			Final Revie	ew:	1 15



CETIS Summary Report

Report Date:

02 Jun-15 17:34 (p 1 of 2) 150513mprt | 08-7733-4797

Test Code:	150513mprt 08-

Macrocystis (Germination and	l Germ	Tube Grow	th Test						Nautilus	s Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	18-3814-7553 13 May-15 12:2 15 May-15 11:1 47h	20 5	Test Type: Protocol: Species: Source:	Growth-Germin EPA/600/R-95/ Macrocystis py La Jolla Cove	136 (1995)			Analyst: Diluent: Brine: Age:		oratory Seav Applicable	water	
Sample ID: Sample Date: Receive Date: Sample Age:	: 13 May-15		Code: Material: Source: Station:	150513mprt Copper chloride Reference Toxi Copper Chlorid	cant			Client: Project:	Inter	nal		
Comparison S	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Me	thod			
12-4945-1639 02-9211-9979	Germination Ra	ite	10 10	32 32	17.89 17.89	7.2% 8.57%				Adj t Test Adj t Test		
Point Estimat	e Summary		· · · · · · · · · · · · · · · · · · ·									
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Me	thod			
07-4758-4339	Germination Ra	ıte	EC50	118.1	111.6	124.9				-Kärber		
14-5916-3987	Mean Length		IC25 IC50	82.9 280.1	61.05 249	114.7 300.2		<u>i</u> _		erpolation (I	CPIN)	
Test Acceptat	oility		NHIA.								14.66	COMMON TO STATE OF THE STATE OF
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	ts	Ov	erlap	Decision		
07-4758-4339	Germination Ra	ite	Contro	ol Resp	0.816	0.7 - NL		Ye	3	Passes A	cceptability	Criteria
12-4945-1639	Germination Ra	ite	Contro	l Resp	0.816	0.7 - NL		Ye	3	Passes A	cceptability	Criteria
02-9211-9979	Mean Length		Contro	l Resp	12.1	10 - NL		Ye	3	Passes A	cceptability	Criteria
14-5916-3987	Mean Length		Contro	l Resp	12.1	10 - NL		Ye	6	Passes A	cceptability	Criteria
02-9211-9979	Mean Length		NOEL		10	NL - 35		No		Passes A	cceptability	Criteria
12-4945-1639	Germination Ra	ite	PMSD	•	0.07205	NL - 0.2		No		Passes A	cceptability	Criteria
02-9211-9979	Mean Length		PMSD		0.08566	NL - 0.2		No		Passes A	cceptability	Criteria
Germination F	Rate Summary											
C-μg/L	Control Type	Coun	t Mean	95% LCL	95% UCL	Min	Max	Sto	l Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.816	0.7802	0.8518	0.78	0.84		1288	0.02881	3.53%	0.0%
10		5	0.818	0.7911	0.8449	8.0	0.85		09695	0.02168	2.65%	-0.25%
32		5	0.75	0.7222	0.7778	0.72	0.78			0.02236	2.98%	8.09%
100		4	0.562		0.6643	0.49	0.63		3198	0.06397	11.37%	31.07%
180		5	0.286	0.2561	0.3159	0.26	0.32		1077	0.02408	8.42%	64.95%
320		5	0.046	0.005195	0.08681	0.02	0.1		147	0.03286	71.44%	94.36%
560		5	0	0	0	0	0	0		0		100.0%
Mean Length	-											
C-µg/L	Control Type	Coun		95% LCL	95% UCL	Min	Max		Err	Std Dev	CV%	%Effect
0	Lab Control	5	12.1	11.49	12.71	11.5	12.7		179	0.4873	4.03%	0.0%
10		5	12.05	11.19	12.91	11.25	13		102	0.6937	5.76%	0.41%
32		5	10.6	9.885	11.31	10	11.5		574	0.5755	5.43%	12.4%
100		4	8.563	7.467	9.658	7.75	9.25		442	0.6884	8.04%	29.24%
180		5	8.15	6.751	9.549	7.25	9.5		037	1.126	13.82%	32.64%
320		5	5.213	4.803	5.622	5	5.75		474	0.3295	6.32%	56.92%
560		5	5	5	5	5	5	0		0	0.0%	58.68%

Analyst: BK QA: Sc 6/4/15

CETIS Summary Report

Report Date: Test Code: 02 Jun-15 17:34 (p 2 of 2) 150513mprt | 08-7733-4797

							rest Code:	1505 15mprt 06-7753-4797
Macrocys	tis Germination an	d Germ Tu	be Growth	Test				Nautilus Environmental (CA)
Germinati	on Rate Detail							
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
0	Lab Control	0.79	0.83	0.84	0.78	0.84		
10		0.83	8.0	0.81	0.85	0.8		
32		0.72	0.75	0.74	0.78	0.76		
100		0.63	0.53		0.49	0.6		
180		0.28	0.26	0.32	0.27	0.3		
320		0.05	0.02	0.04	0.1	0.02		
560		0	0	0	0	0		
Mean Leng	gth Detail							
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
0	Lab Control	12.25	11.75	12.75	11.5	12.25		
10		12.5	11.75	13	11.75	11.25		
32		10.25	10.75	10.5	11.5	10		
100		9	8.25		7.75	9.25		
180		7.25	9.5	7.25	9.25	7.5		
320		5	5	5.313	5.75	5		
560		5	5	5	5	5		

Analyst: BY QA:

Report Date:

02 Jun-15 17:34 (p 1 of 3)

Test Code:

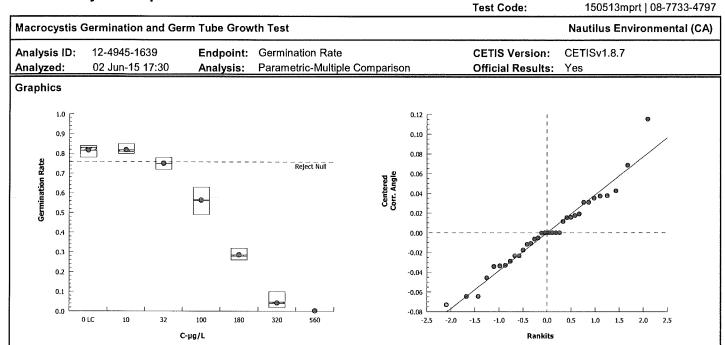
150513mprt | 08-7733-4797

Macrocystis	Germination an	d Germ T	ube Growth	Test		-			Nautilus	Environn	nental (CA)
Analysis ID: Analyzed:	12-4945-1639 02 Jun-15 17:		•	ermination Ra arametric-Mul		arison		IS Version: cial Results	CETISv1 : Yes	.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	Š	PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C > T	NA	NA		7.2%	10	32	17.89	
Bonferroni A	∖dj t Test				·						
Control	vs C-μg/L		Test Sta	t Critical	MSD DF	P-Value	P-Type	Decision((a:5%)		
Lab Control	10		-0.07729	2.5	0.073 8	1.0000	CDF	Non-Signi	ficant Effect		
	32*		2.777	2.5	0.073 8	0.0268	CDF	Significan			
	100*		9.064	2.5	0.077 7	<0.0001	CDF	Significan			
	180*		19.38	2.5	0.073 8	<0.0001	CDF	Significan	t Effect		
	320*		31.66	2.5	0.073 8	<0.0001	CDF	Significan	t Effect		
ANOVA Tabl	е									* *************************************	
Source	Sum Squ	ares	Mean Sc	quare	DF	F Stat	P-Value	Decision(a:5%)		
Between	3.429466		0.685893	33	5	323.7	<0.0001	Significan	t Effect		
Error	0.048738	44	0.002119	9063	23						
Total	3.478205				28						
Distributiona	al Tests										
Distributiona Attribute	al Tests Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
	Test	Equality of	Variance	Test Stat	Critical 15,09	P-Value 0.1592	Decision Equal Var	<u>`</u>			
Attribute	Test Bartlett E	Equality of Wilk W N						iances			
Attribute Variances Distribution	Test Bartlett E			7.947	15.09	0.1592	Equal Var	iances			
Attribute Variances Distribution	Test Bartlett E Shapiro-			7.947	15.09	0.1592	Equal Var	iances	Std Err	CV%	%Effect
Attribute Variances Distribution Germination	Test Bartlett E Shapiro- Rate Summary	Wilk W N	ormality	7.947 0.9691	15.09 0.9004	0.1592 0.5364	Equal Var Normal D	iances istribution	Std Err 0.01288	CV% 3.53%	%Effect 0.0%
Attribute Variances Distribution Germination C-µg/L	Test Bartlett E Shapiro- Rate Summary Control Type	Wilk W N	ormality Mean	7.947 0.9691 95% LCL	15.09 0.9004 95% UCL	0.1592 0.5364 Median	Equal Var Normal D	iances istribution			
Attribute Variances Distribution Germination C-µg/L 0	Test Bartlett E Shapiro- Rate Summary Control Type	Count	Mean 0.816	7.947 0.9691 95% LCL 0.7802	15.09 0.9004 95% UCL 0.8518	0.1592 0.5364 Median 0.83	Equal Var Normal D Min 0.78	iances istribution Max 0.84	0.01288	3.53%	0.0%
Attribute Variances Distribution Germination C-µg/L 0 10	Test Bartlett E Shapiro- Rate Summary Control Type	Count 5 5	Mean 0.816 0.818	7.947 0.9691 95% LCL 0.7802 0.7911	15.09 0.9004 95% UCL 0.8518 0.8449	0.1592 0.5364 Median 0.83 0.81	Equal Var Normal D Min 0.78 0.8	Max 0.84 0.85	0.01288 0.009695	3.53% 2.65%	0.0% -0.25%
Attribute Variances Distribution Germination C-µg/L 0 10 32	Test Bartlett E Shapiro- Rate Summary Control Type	Count 5 5 5	Mean 0.816 0.818 0.75	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222	15.09 0.9004 95% UCL 0.8518 0.8449 0.7778	0.1592 0.5364 Median 0.83 0.81 0.75	Min 0.78 0.8 0.72	Max 0.84 0.85 0.78	0.01288 0.009695 0.01	3.53% 2.65% 2.98%	0.0% -0.25% 8.09%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100	Test Bartlett E Shapiro- Rate Summary Control Type	Count 5 5 4	Mean 0.816 0.818 0.75 0.5625	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607	15.09 0.9004 95% UCL 0.8518 0.8449 0.7778 0.6643	0.1592 0.5364 Median 0.83 0.81 0.75 0.565	Equal Var Normal D Min 0.78 0.8 0.72 0.49	Max 0.84 0.85 0.78 0.63	0.01288 0.009695 0.01 0.03198	3.53% 2.65% 2.98% 11.37%	0.0% -0.25% 8.09% 31.07%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180	Test Bartlett E Shapiro- Rate Summary Control Type	Count 5 5 5 4 5 5	Mean 0.816 0.818 0.75 0.5625 0.286	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561	15.09 0.9004 95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28	Equal Var Normal D Min 0.78 0.8 0.72 0.49 0.26	Max 0.84 0.85 0.78 0.63 0.32	0.01288 0.009695 0.01 0.03198 0.01077	3.53% 2.65% 2.98% 11.37% 8.42%	0.0% -0.25% 8.09% 31.07% 64.95%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180 320 560	Test Bartlett E Shapiro- Rate Summary Control Type	Count 5 5 4 5 5 5	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195	15.09 0.9004 95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04	Equal Var Normal D Min 0.78 0.8 0.72 0.49 0.26 0.02	Max 0.84 0.85 0.78 0.63 0.32 0.1	0.01288 0.009695 0.01 0.03198 0.01077 0.0147	3.53% 2.65% 2.98% 11.37% 8.42%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180 320 560	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control	Count 5 5 4 5 5 5	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195 0	15.09 0.9004 95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04 0	Equal Var Normal D Min 0.78 0.8 0.72 0.49 0.26 0.02	Max 0.84 0.85 0.78 0.63 0.32 0.1	0.01288 0.009695 0.01 0.03198 0.01077 0.0147	3.53% 2.65% 2.98% 11.37% 8.42%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36%
Attribute Variances Distribution Germination C-μg/L 0 10 32 100 180 320 560 Angular (Cor	Test Bartlett E Shapiro-¹ Rate Summary Control Type Lab Control	Count 5 5 4 5 5 5 4 5 5 med Sun	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195 0	15.09 0.9004 95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681 0	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04 0	Min 0.78 0.8 0.72 0.49 0.26 0.02 0	Max 0.84 0.85 0.78 0.63 0.32 0.1 0	0.01288 0.009695 0.01 0.03198 0.01077 0.0147 0	3.53% 2.65% 2.98% 11.37% 8.42% 71.44%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36% 100.0%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180 320 560 Angular (Cor	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 4 5 5 5 med Sum	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0 mmary Mean	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195 0	15.09 0.9004 95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681 0	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04 0	Min 0.78 0.8 0.72 0.49 0.26 0.02	Max 0.84 0.85 0.78 0.63 0.32 0.1 0	0.01288 0.009695 0.01 0.03198 0.01077 0.0147 0	3.53% 2.65% 2.98% 11.37% 8.42% 71.44%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36% 100.0%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180 320 560 Angular (Cor C-µg/L 0 10	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 4 5 5 5 med Sun Count 5	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0 nmary Mean 1.128	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195 0	95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681 0	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04 0	Min 0.78 0.8 0.72 0.49 0.26 0.02 0	Max 0.84 0.85 0.78 0.63 0.32 0.1 0 Max	0.01288 0.009695 0.01 0.03198 0.01077 0.0147 0	3.53% 2.65% 2.98% 11.37% 8.42% 71.44% CV% 3.27%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36% 100.0% %Effect 0.0%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180 320 560 Angular (Cor C-µg/L 0 10 32	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 4 5 5 5 med Sun Count 5 5	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0 nmary Mean 1.128 1.131	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195 0 95% LCL 1.083 1.095	95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681 0 95% UCL 1.174 1.166	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04 0 Median 1.146 1.12	Min 0.78 0.8 0.72 0.49 0.26 0.02 0 Min 1.083 1.107	Max 0.84 0.85 0.78 0.63 0.32 0.1 0 Max 1.159 1.173	0.01288 0.009695 0.01 0.03198 0.01077 0.0147 0 Std Err 0.01649 0.01276	3.53% 2.65% 2.98% 11.37% 8.42% 71.44% CV% 3.27% 2.52%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36% 100.0% -0.0% -0.2%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180 320 560 Angular (Cor C-µg/L 0 10 32 100 10 32 100	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 4 5 5 med Sun Count 5 5 5	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0 nmary Mean 1.128 1.131 1.048	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195 0 95% LCL 1.083 1.095 1.015	95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681 0 95% UCL 1.174 1.166 1.08	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04 0 Median 1.146 1.12 1.047	Min 0.78 0.8 0.72 0.49 0.26 0.02 0 Min 1.083 1.107 1.013	Max 0.84 0.85 0.78 0.63 0.32 0.1 0 Max 1.159 1.173 1.083	0.01288 0.009695 0.01 0.03198 0.01077 0.0147 0 Std Err 0.01649 0.01276 0.01156	3.53% 2.65% 2.98% 11.37% 8.42% 71.44% CV% 3.27% 2.52% 2.47%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36% 100.0% *Effect 0.0% -0.2% 7.16%
Attribute Variances Distribution Germination C-µg/L 0 10 32 100 180 320 560 Angular (Cor C-µg/L 0	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 4 5 5 med Sun Count 5 5 4 4 5 5 4 5 5 4 6 6 7 7 8 7 8 8 8 8 8 8 8 8 8 8 8 8 8 8	Mean 0.816 0.818 0.75 0.5625 0.286 0.046 0 mary Mean 1.128 1.131 1.048 0.8485	7.947 0.9691 95% LCL 0.7802 0.7911 0.7222 0.4607 0.2561 0.005195 0 95% LCL 1.083 1.095 1.015 0.7456	95% UCL 0.8518 0.8449 0.7778 0.6643 0.3159 0.08681 0 95% UCL 1.174 1.166 1.08 0.9513	0.1592 0.5364 Median 0.83 0.81 0.75 0.565 0.28 0.04 0 Median 1.146 1.12 1.047 0.8507	Min 0.78 0.8 0.72 0.49 0.26 0.02 0 Min 1.083 1.107 1.013 0.7754	Max 0.84 0.85 0.78 0.63 0.32 0.1 0 Max 1.159 1.173 1.083 0.9169	0.01288 0.009695 0.01 0.03198 0.01077 0.0147 0 Std Err 0.01649 0.01276 0.01156 0.03231	3.53% 2.65% 2.98% 11.37% 8.42% 71.44% CV% 3.27% 2.52% 2.47% 7.62%	0.0% -0.25% 8.09% 31.07% 64.95% 94.36% 100.0% **Effect 0.0% -0.2% 7.16% 24.81%

Analyst: B/C QA: SC

Report Date:

02 Jun-15 17:34 (p 2 of 3)



000-089-187-4 CETIS™ v1.8.7.20

Report Date: Test Code:

02 Jun-15 17:34 (p 1 of 1)

150513mprt | 08-7733-4797

Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental (CA)

Analysis ID: 07-4758-4339 Analyzed:

02 Jun-15 17:30

Endpoint: Germination Rate

Analysis: Untrimmed Spearman-Kärber

CETIS Version: Official Results:

CETISv1.8.7

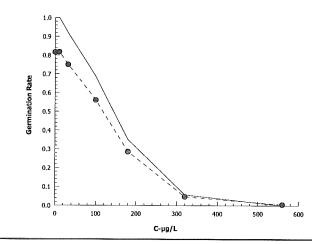
Yes

Spearman-Kärber	Est	imat	les
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Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0.184	0.00%	2.072	0.01219	118.1	111.6	124 9

Germination Rate Summary			Calculated Variate(A/B)								
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.816	0.78	0.84	0.01288	0.02881	3.53%	0.0%	408	500
10		5	0.818	8.0	0.85	0.009695	0.02168	2.65%	-0.25%	409	500
32		5	0.75	0.72	0.78	0.01	0.02236	2.98%	8.09%	375	500
100		4	0.5625	0.49	0.63	0.03198	0.06397	11.37%	31.07%	224	400
180		5	0.286	0.26	0.32	0.01077	0.02408	8.42%	64.95%	143	500
320		5	0.046	0.02	0.1	0.0147	0.03286	71.44%	94.36%	23	500
560		5	0	0	0	0	0		100.0%	0	500

Graphics



Analyst: BIC QA:

Report Date:

02 Jun-15 17:34 (p 3 of 3) 150513mprt | 08-7733-4797

Test Code:

Macrocystis	Germination and G	erm Tube Growth Test		Nautilus Environmental (CA)
Analysis ID:	02-9211-9979	Endocint: Mean Length	CETIS Varaion	CETICU1 9.7

Analyzed:	02 Jun-15 17:31	Analysis:	Parametric-Multiple Comparison	Official Results: Yes

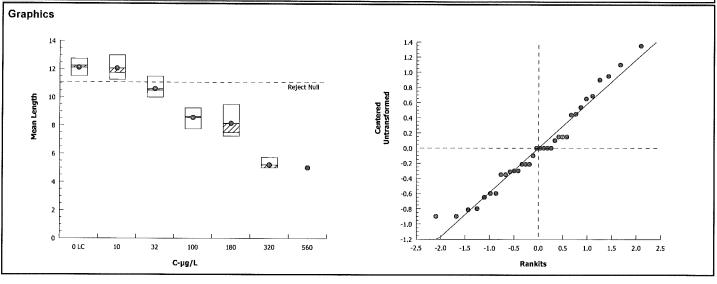
Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU
Untransformed	NA	C > T	NA	NA	8.57%	10	32	17.89	

Bonferroni Adj t Test Control C-µg/L Test Stat Critical MSD DF P-Value P-Type Decision(a:5%) Lab Control 10 0.1231 2.552 1.037 8 1.0000 CDF Non-Significant Effect 32* 3.694 2.552 1.037 8 0.0030 CDF Significant Effect 100* 8.213 2.552 1.099 7 < 0.0001 CDF Significant Effect 180* 9.727 2.552 1.037 8 < 0.0001 CDF Significant Effect 320* 16.96 2.552 1.037 8 < 0.0001 CDF Significant Effect 560* 17.48 2,552 1.037 8 < 0.0001 CDF Significant Effect

ANOVA Table							
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)	
Between	262.3404	43.7234	6	106.1	<0.0001	Significant Effect	
Error	11.13125	0.4122685	27				
Total	273.4716		33	111 has			

Distributional Te	sts				
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)
Variances	Mod Levene Equality of Variance	1.917	3.812	0.1251	Equal Variances
Variances	Levene Equality of Variance	7.891	3.558	<0.0001	Unequal Variances
Distribution	Shapiro-Wilk W Normality	0.9618	0.9125	0.2741	Normal Distribution

C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	12.1	11.49	12.71	12.25	11.5	12.75	0.2179	4.03%	0.0%
10		5	12.05	11.19	12.91	11.75	11.25	13	0.3102	5.76%	0.41%
32		5	10.6	9.885	11.31	10.5	10	11.5	0.2574	5.43%	12.4%
100		4	8.563	7.467	9.658	8.625	7.75	9.25	0.3442	8.04%	29.24%
180		5	8.15	6.751	9.549	7.5	7.25	9.5	0.5037	13.82%	32.64%
320		5	5.213	4.803	5.622	5	5	5.75	0.1474	6.32%	56.92%
560		5	5	5	5	5	5	5	0	0.0%	58.68%



Report Date:

02 Jun-15 17:34 (p 1 of 1)

Test Code:

150513mprt | 08-7733-4797

Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental (CA)

Analysis ID: Analyzed:

14-5916-3987 02 Jun-15 17:31 Endpoint: Mean Length

Analysis: Linear Interpolation (ICPIN)

CETIS Version:

CETISv1.8.7

Official Results: Yes

Linear	Interpolation	n Options
	_	

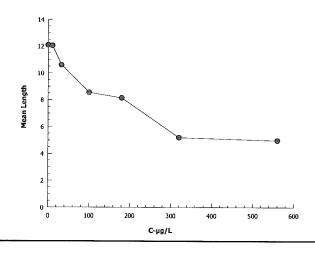
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	985217	1000	Yes	Two-Point Interpolation

Point Estimates

Level	μg/L	95% LCL	95% UC
IC25	82.9	61.05	114.7
IC50	280.1	249	300.2

Mean Len	ngth Summary		Calculated Variate							
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	
0	Lab Control	5	12.1	11.5	12.75	0.2179	0.4873	4.03%	0.0%	
10		5	12.05	11.25	13	0.3102	0.6937	5.76%	0.41%	
32		5	10.6	10	11.5	0.2574	0.5755	5.43%	12.4%	
100		4	8.563	7.75	9.25	0.3442	0.6884	8.04%	29.24%	
180		5	8.15	7.25	9.5	0.5037	1.126	13.82%	32.64%	
320		5	5.213	5	5.75	0.1474	0.3295	6.32%	56.92%	
560		5	5	5	5	0	0	0.0%	58.68%	

Graphics



Analyst: BL QA:

Protocol:

Report Date:

02 Jun-15 17:35 (1 of 1)

Macrocystis Germination and Germ Tube Growth Test

EPA/600/R-95/136 (1995)

Nautilus Environmental (CA)

Test Type: Growth-Germination

Organism: Macrocystis pyrifera (Giant Kelp)

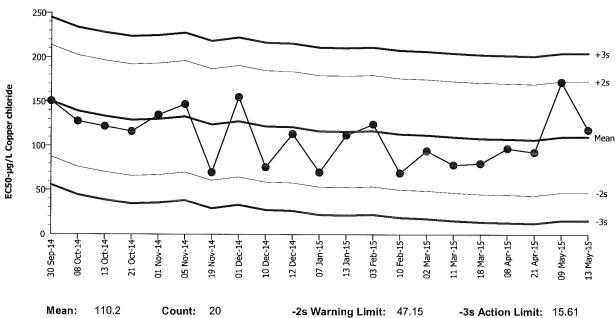
Endpoint: Germination Rate

Material: Copper chloride

Source: Reference Toxicant-REF

+3s Action Limit: 204.9

Macrocystis Germination and Germ Tube Growth Test



+2s Warning Limit:

173.3

Quality	Control	Data
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Sigma:

31.54

CV:

28.60%

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Sep	30	12:15	150.5	40.33	1.279			13-9787-7056	02-8487-1458
2		Oct	8	11:40	127.8	17.6	0.558			15-0209-2295	12-7369-3569
3			13	15:00	121.9	11.71	0.3714			07-9089-8433	21-1969-7792
4			21	11:45	116.2	5.985	0.1898			05-9690-1778	07-6459-5729
5		Nov	1	17:30	134.9	24.67	0.7821			01-6548-8876	19-8891-4456
6			5	12:10	147.1	36.91	1.17			16-4808-9223	03-9213-0972
7			19	11:45	69.84	-40.36	-1.28			19-5595-5174	10-2118-3209
8		Dec	1	14:25	155.2	44.97	1.426			00-5019-0818	01-5801-5093
9			10	11:45	75.77	-34.43	-1.091			20-1153-5143	06-3379-4906
10			12	16:00	113.5	3.325	0.1054			11-8986-9857	15-5507-8419
11	2015	Jan	7	11:50	70.09	-40.11	-1.272			05-1839-9516	16-0872-3144
12			13	15:20	112.1	1.942	0.06156			19-5672-8241	21-3328-2708
13		Feb	3	14:10	124.6	14.4	0.4565			02-0093-1321	09-6992-3779
14			10	13:45	69.37	-40.83	-1.295			15-5769-6471	18-0756-1200
15		Mar	2	19:25	94.56	-15.64	-0.4958			02-8587-0344	12-3520-6702
16			11	11:40	78.5	-31.7	-1.005			16-3863-7280	09-2265-0470
17			18	12:00	80.1	-30.1	-0.9544			00-4348-8755	17-4019-8643
18		Apr	8	13:20	97.16	-13.04	-0.4134			16-4963-4427	03-3491-3881
19			21	16:15	93.03	-17.17	-0.5445			05-9446-7141	00-0365-1165
20		May	9	14:25	172.4	62.22	1.973			11-5639-4173	20-3923-9025
21			13	12:20	118.1	7.857	0.2491			08-7733-4797	07-4758-4339

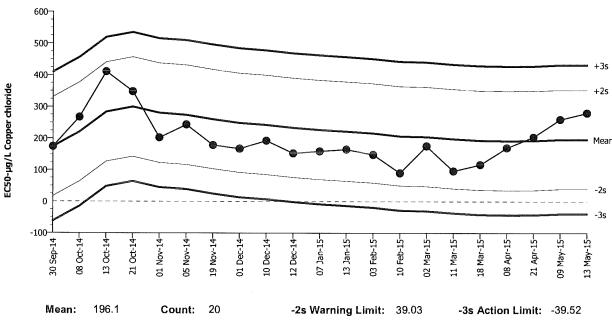
Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental (CA)

Test Type: Growth-Germination Organism: Macrocystis pyrifera (Giant Kelp) Material: Copper chloride

Protocol: EPA/600/R-95/136 (1995) Endpoint: Mean Length Source: Reference Toxicant-REF

Macrocystis Germination and Germ Tube Growth Test



o i giii a i	. 0.00	•••	10.1070	· 25 Warning Limit.	000,2	103 Action Ellinit.	701.0
Sigma:	78.55	CV:	40.10%	+2s Warning Limit:	353.2	+3s Action Limit:	431.8
weam.	190.1	Count:	20	-25 Warning Limit:	39.03	-35 Action Limit:	-39.52

Quali	ty Con	trol Data	a								
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Sep	30	12:15	173.8	-22.25	-0.2833			13-9787-7056	19-7873-0878
2		Oct	8	11:40	266.9	70.78	0.9011			15-0209-2295	14-5837-8655
3			13	15:00	410.8	214.7	2.734	(+)		07-9089-8433	18-0074-3690
4			21	11:45	348	151.9	1.934			05-9690-1778	02-4326-8088
5		Nov	1	17:30	202.1	6.005	0.07645			01-6548-8876	12-7526-4191
6			5	12:10	243.4	47.34	0.6026			16-4808-9223	00-4645-3717
7			19	11:45	178.4	-17.7	-0.2253			19-5595-5174	15-0041-2395
8		Dec	1	14:25	167.1	-29.02	-0.3695			00-5019-0818	01-4688-9594
9			10	11:45	192.7	-3.373	-0.04294			20-1153-5143	19-2658-2480
10			12	16:00	152.8	-43.27	-0.5509			11-8986-9857	14-7967-2457
11	2015	Jan	7	11:50	159.3	-36.79	-0.4684			05-1839-9516	20-5481-8709
12			13	15:20	165.1	-30.96	-0.3941			19-5672-8241	11-2312-3644
13		Feb	3	14:10	148.5	-47.62	-0.6062			02-0093-1321	00-8294-2622
14			10	13:45	90.23	-105.9	-1.348			15-5769-6471	18-5702-3882
15		Mar	2	19:25	175.9	-20.24	-0.2576			02-8587-0344	07-8323-3127
16			11	11:40	96.64	-99.46	-1.266			16-3863-7280	04-6998-1152
17			18	12:00	116.8	-79.35	-1.01			00-4348-8755	09-8096-6426
18		Apr	8	13:20	170	-26.14	-0.3328			16-4963-4427	08-2168-6694
19			21	16:15	204.1	7.963	0.1014			05-9446-7141	17-6422-9986
20		May	9	14:25	260	63.9	0.8135			11-5639-4173	11-0369-2164
21			13	12:20	280.1	83.99	1.069			08-7733-4797	14-5916-3987

Report Date:

02 Jun-15 17:35 (1 of 1)

Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental (CA)

Test Type: Growth-Germination **Protocol:** EPA/600/R-95/136 (1995)

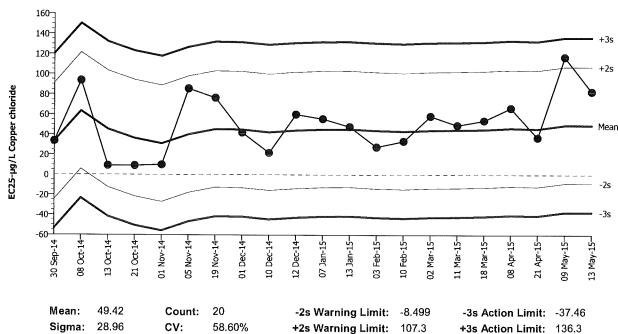
Organism: Macrocystis pyrifera (Giant Kelp)

Endpoint: Mean Length

Material: Copper chloride

Source: Reference Toxicant-REF

Macrocystis Germination and Germ Tube Growth Test



Qua	lity	Control	Data	
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Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Sep	30	12:15	33.48	-15.94	-0.5505			13-9787-7056	19-7873-0878
2		Oct	8	11:40	93.66	44.24	1.528			15-0209-2295	14-5837-8655
3			13	15:00	9.074	-40.35	-1.393			07-9089-8433	18-0074-3690
4			21	11:45	9.019	-40.4	-1.395			05-9690-1778	02-4326-8088
5		Nov	1	17:30	9.855	-39.56	-1.366			01-6548-8876	12-7526-4191
6			5	12:10	85.74	36.32	1.254			16-4808-9223	00-4645-3717
7			19	11:45	76.63	27.21	0.9394			19-5595-5174	15-0041-2395
8		Dec	1	14:25	42.05	-7.375	-0.2546			00-5019-0818	01-4688-9594
9			10	11:45	21.81	-27.61	-0.9535			20-1153-5143	19-2658-2480
10			12	16:00	60.17	10.75	0.3713			11-8986-9857	14-7967-2457
11	2015	Jan	7	11:50	55.64	6.221	0.2148			05-1839-9516	20-5481-8709
12			13	15:20	47.84	-1.579	-0.05453			19-5672-8241	11-2312-3644
13		Feb	3	14:10	27.6	-21.82	-0.7535			02-0093-1321	00-8294-2622
14			10	13:45	33.37	-16.05	-0.5543			15-5769-6471	18-5702-3882
15		Mar	2	19:25	58.41	8.991	0.3105			02-8587-0344	07-8323-3127
16			11	11:40	49.36	-0.06467	-0.00223			16-3863-7280	04-6998-1152
17			18	12:00	54.03	4.608	0.1591			00-4348-8755	09-8096-6426
18		Apr	8	13:20	66.52	17.1	0.5903			16-4963-4427	08-2168-6694
19			21	16:15	37.04	-12.38	-0.4276			05-9446-7141	17-6422-9986
20		May	9	14:25	117.1	67.72	2.338	(+)		11-5639-4173	11-0369-2164
21		,	13	12:20	82.9	33.48	1.156	. ,		08-7733-4797	14-5916-3987

Analyst: BK QA: Schol

Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental - San Diego

Start Date: 13-May-15

Species: Macrocystis pyrifera

Test ID: 150513mprt

End Date: 15-May-15

Protocol: EPA/600/R-95/136 (1995 West Coast Manual)

Sample Source: Internal

Sampled: 13-May-15

Sample Station: CuCl₂

Random	Number	Number			\								Calibration	Mean Tube
Number	Counted	Germinated				Tube Lengt	th Measuren	nents (micro	meter units)	l			Factor	Length (µm)
1	100	75	4	4	5	5	4	5	5	4	4	3	2.5	10.75
2	*		-										2.5	10.73
3	100	32	3	2	3	5	2	2	3	3	3	3	2.5	7.25
4	100	49	4	4	3	3	4	3	2	3	2	3	2.5	7.75
5	100	10	2	2	2	2	2	4	2	3	2	2	2.5	5.75
6	100	63	5	3	3	4	2	3	3	4	5	4	2.5	9.00
7	100	80	4	6	5	6	4	5	5	4	4	4	2.5	11.75
8	100	78	5	5	6	4	5	3	4	5	5	4	2.5	11.50
9	100	4	2	2	3	2	2	2	2	2			2.5	5.31
10	100	83	6	4	5	6	5	5	5	5	5	4	2.5	12.50
11	100	74	5	4	5	5	4	4	4	3	3	5	2.5	10.50
12	100	28	2	2	4	3	3	3	2	4	4	2	2.5	7.25
13	100	2	2	2	2	2	2	2					2.5	5.00
14	100	81	4	4	5	7	5	5	7	6	5	4	2.5	13.00
15	100	78	5	5	5	4	5	6	3	4	5	4	2.5	11.50
16	100	0	2	2									2.5	5.00
17	100	84	5	3	6	5	3	5	6	6	5	5	2.5	12.25
18	100	0	2	2	,					W			2.5	5.00
19	100	30	2	2	6	3	3	3	2	2	5	2	2.5	7.50
20	100	85	5	4	4	5	6	5	5	5	5	3	2.5	11.75
21	100	76	3	4	3	4	5	4	4	4	5	4	2.5	10.00
22	100	53	3	3	5	2	4	3	3	4	4	2	2.5	8.25
23	100	0	2	2									2.5	5.00
24	100	60	5	4	4	3	3	3	4	3	5	3	2.5	9.25
25	100	79	5	6	5	4	4	5	5	5	5	5	2.5	12.25
26	100	0	2	2	2								2.5	5.00
27	100	2	2	2	2	2							2.5	5.00
28	100	0	2	2									2.5	5.00
29	100	83	5	3	4	6	5	6	5	4	4	5	2.5	11.75
30	100	26	4	3	5	4	3	3	5	3	3	5	2.5	9.50
31	100	80	3	7	5	6	4	3	4	5	4	4	2.5	11.25
32	100	27	3	4	5	4	3	4	3	3	4	4	2.5	9.25
33	100	5	2	2	2	2							2.5	5.00
34	100	72	4	4	3	3	5	4	5	4	5	4	2.5	10.25
35	100	84	6	6	6	5	4	5	4	5	5	5	2.5	12.75

Technician Note: If there are any germinated spores in the replicate, scan the slide to measure 10 lengths if possible (regardless of number germinated).

* Slide not inoculated, technician error.

QC Check: BL 6/2/15

Final Review

De 6/4/15

Analyst:

BK

Macrocystis Germination and Germ Tube Growth Test

Nautilus Environmental - San Diego

Start Date: 13-May-15

Species: Macrocystis pyrifera

Test ID: 150513mprt

15-May-15 End Date:

Protocol: EPA/600/R-95/136 (1995 West Coast Manual)

Sample Source: Internal

13-May-15 Sampled:

Sample Station: CuCl₂

Random Number	Number Counted	Number Germinated				Tube Lengt	h Measuren	nents (micro	meter units)				Calibration Factor	Mean Tube Length (μm)
1	100	75	4	Ы	5	5	4	5	5	4	1 4	3	2.5	#DIV/0!
2	100	(4)	<u> </u>							/			1	#DIV/0!
3	100	32	3	2	3	5	2	2	3	3	3	3		#DIV/0!
4	100	49	<u> </u>	L)	3	13	4	3	2	3	2	3		#DIV/0!
5	100	10	-72	2	2	2	2	4	2	3	2	2		#DIV/0!
6	100	08363	5	3	3	4	2	3	3	4	5	4		#DIV/0!
7	100	80	4	6	5	6	4	5	S	Ĕ	4	4		#DIV/0!
8	100	18	5	5	6	4	5	3	4	5	.5	4		#DIV/0!
9	100	Ϋ́	2	2	3	2	2	2	2	2_		7		#DIV/0!
10	100	83	6	4	5	6	5	5	5	5	5	4		#DIV/0!
11	100	74	5	4	5	5	4	4	Lj.	3	3	1 5		#DIV/0!
12	100	@7528	7,	2	4	3	3	3	2	4	ef	2		#DIV/0!
13	100	2	2	2	2	2	2	1		1				#DIV/0!
14	100	81	4	4			5	5	7	6	5	C-f		#DIV/0!
15	100	18	5	5	5	LA	5	6	3	4	5	4		#DIV/0!
16	100	0	2	2				-						#DIV/0!
17	100	84	5	3	1.	5	25	5	6	6	S	5		#DIV/0!
18	100	0	2	2	- W									#DIV/0!
19	100	30	2	2	6	3	3	3	2	2_	5	2		#DIV/0!
20	100	85	5	4	1 4	É	1/2	5	5	5	5	3		#DIV/0!
21	100	76	3	if	3	4	5	11	Ŭ,	¥	5	4		#DIV/0!
22	100	53	3	3	5	2	4	3	3	¥	4	2		#DIV/0!
23	100	0	2	2										#DIV/0!
24	100	(eD	5	4	Ч	3	3	3	Ц	3	5	3		#DIV/0!
25	100	99	5	6	5	C.f.	4	.5	5	5	5	5		#DIV/0!
26	100	0	2	2	2	(1							#DIV/0!
27	100	2	2	2	2-	2								#DIV/0!
28	100	0	2-	2										#DIV/0!
29	100	83	5	3	4	6	5	6	5	4	¥	5		#DIV/0!
30	100	26	4	3	5	<i>U</i>	3	1 2	5	3	3	15		#DIV/0!
30	100	80	3	1 3	5	16	I I	3	4	5	4	4		#DIV/0!
	100	27	3	4	5	4	3	Ú	3	3	4	4		#DIV/0!
32 33	100	5	2	2	2	2						1		#DIV/0!
	100	72	4	1 G	3	3	5	4	5	4	5	Ч		#DIV/0!
34 35	100	84	6	10		5	4	5	4	5	5	4	1	#DIV/0!

Technician Note: If there are any germinated spores in the replicate, scan the slide to measure 10 lengths if possible (regardless of number germinated).

Analyst:

QC Check: 8+ 6/2/15

@ O18 ver 5/15/15 @ Stide not inoculated, technician error.

CETIS Test Data Worksheet

Report Date:

12 May-15 10:55 (p 1 of 1)

Test Code:

08-7733-4797/150513mprt

Macrocystis	Germi	natio	n and	Germ Tube	Growth Test				Nautilus Environmental (CA)
Start Date: End Date: Sample Date	15 N	Лау-1: Лау-1: Лау-1:	5		s: Macrocyst ol: EPA/600/F al: Copper ch	R-95/136 (1995)	Sample Code: Sample Source: Sample Station:	150513mprt Reference Toxicant Copper Chloride
C-μg/L	Code	Rep	Pos	# Counted	# Germinated	Mean Length	CalFactor		Notes
0	LC	1	25	100			1		
0	LC	2	29	100			1	2000	
0	LC	3	35	100			1		
0	LC	4	8	100			1		
0	LC	5	17	100			1		
10		1	10	100			1	,	
10		2	7	100			1		
10		3	14	100			1		
10		4	20	100			1		
10		5	31	100			1		
32		1	34	100			1		
32		2	1	100			1		
32		3	11	100			1		
32		4	15	100			1		
32		5	21	100			1		
100		1	6	100			1		
100		2	22	100			1		
100	-	3	2	100			1		
100		4	4	100			1		
100		5	24	100			1		
180		1	12	100			1		
180		2	30	100			1		
180		3	3	100			1		
180		4	32	100			1		
180		5	19	100			1		
320		1	33	100			1		
320		2	13	100		10 To	1		
320	-	3	9	100			1		
320		4	5	100			1		
320		5	27	100			1		
560		1	26	100			1		
560	 	2	28	100			1		
560		3	23	100			1	,	
560		4	16	100			1		
560		5	18	100			1	19.3011	

Analyst: CB QA: SCGY (5

Marine Chronic Bioassay

Cu Stock Conc. (μg/L)

Water Quality Measurements

Client :	Internal			- 1	Test Species:	Macrocystis	s pyrifera
Sample ID:	CuCl ₂			Sta	rt Date/Time:	5/13/2015	1220
Test No.:	150513mprt			_ Er	ıd Date/Time:	5/15/2015	11.15
Dilutions made by:	AG			_			
Dilution calcs. (final volun	ne 250 mL):						_
Conc. μg/L	10	32	100	180	320	560	
Val. Cu stack added (ml.):	24-	09	2.8	e 2	67	1.6	7

2,630

8,530

8,630

86,900

			Analyst:	AG	·		Analyst:	SC		
Concentration			eadings			Final Re				
(μg/L)	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)	DQ (mg/L)	pH (units)	Salinity (ppt)	(°C)		
Lab Control	8.2	8.03	33.1	15.6	7.4	7-96	33.Z	15.(
10	8-0	8.06	33.0	15.5	7.4	7-96	33.2	(5.)		
32	7.9	8.06	33,2	15.6	7.4	7.96	324	15.1		
100	7.0	8.07	331	15,5	7.4	7.96	33./	15.1		
180	7.9	8.07	32.9	15.7	7.4	7.96	329	15.		
320	7.9	8.09	32.5	15.7	7.4	7.95	32.5	15.1		
560	7-9	8.06	33,2	15.8	7.4	7.96	32,5	15.1		

QC C	heck:	BIL	61	2	15			
Nautilus Environmental.		dever Avenue.	Sá	n	Diego,	CA	92120	

Comments:

959

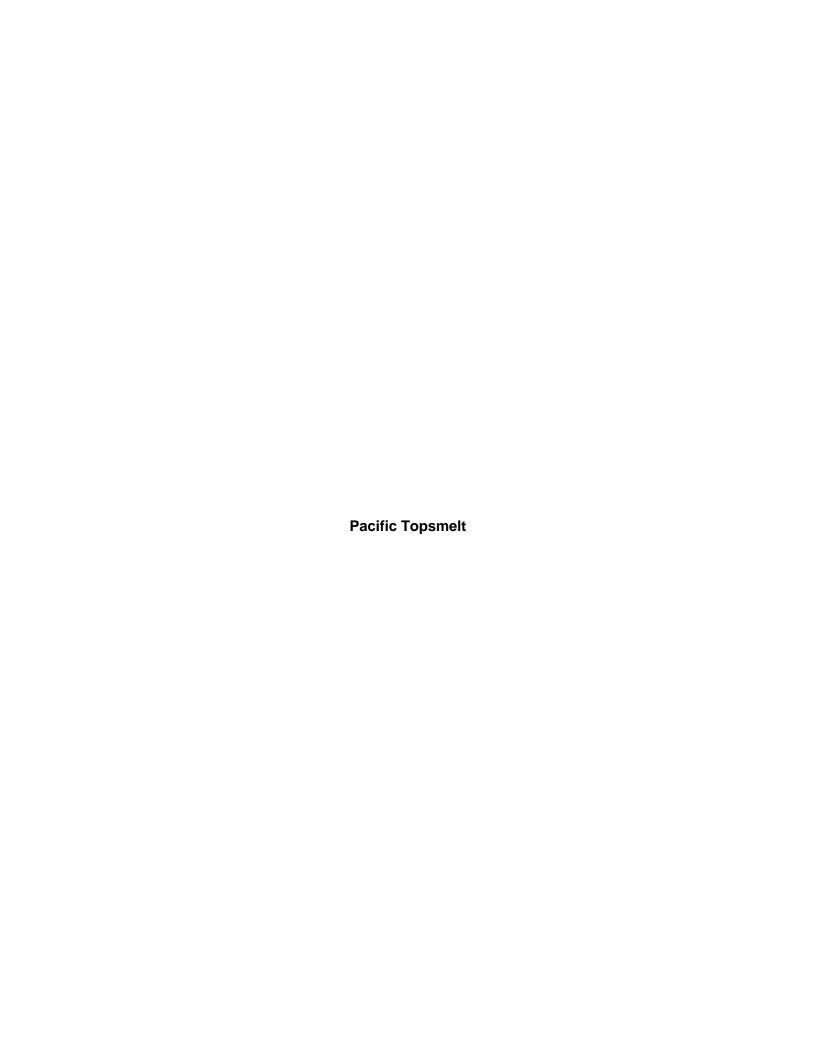
Final Review: Sc 6/4/15

Marine Chronic Bioassay

Kelp Spore Germination & Growth Worksheet

Client: Internal Test No.: 150513 mprt Tech. Initials: A		Start Date/Tin End Date/Tin Test Speci	me: 5/15/20	
Date Collected: Kelp Collectors: Collection Location:	5/12/15 OG La Jolla Cove			
Time of Initial Rinsing and Des Time of Rinsing and Transfer Conditions of Zoospore Densi Time of Blade Removal From	to Release Beaker: 3/	12/15 1645 13/15 1155 19/15 1210		
	128 130 132 1 1318,000 spores/ml (density		<u>1.8</u>	
If spore release = 900,000 s	pores/ml: Inoculate with 0.25 ml			
If spore release > 900,000 s	pores/ml: Calculate a dilution facto	r, x, create a new spore stock of 900,000	spores/ml and	inoculate with 0.25 ml.
If snore release < 900 000 s	1 container	$\frac{329,500}{\text{spores}} = \frac{1.0}{225,000}$ spores $\text{Id not exceed 0.5 ml. (This volume exceed 0.5 ml.)}$	eds the EPA a	dil.factor 1.46 dil.factor 1.0 part spore stock 75 ml 1.46 part(s) seawater 34.5 ml and MBP required volume
of no greater than 1% of the trackieve the desired spore del	otal test solution volume. However,	it may sometimes be necessary to excee	∍d the 0.3 ml re	equirement in order to
Time of inoculation:\2	20 Amount inc	culated: 0.25 mL		
	Chamber (All replicates in each tes	t must be on the same shelf;		
do not split up tests among sl Shelf number	Measured Light Intensity Range (must be between 160 and 240 ft-		24-hour germin	ation check
1			QC dish #	% germ.
2			3	81
3	164-239	1-25		
ROFTEN (4)		1-35 186-225 226-265		
6	161 - 224	726-265		
Timers Checked? /	Should be on 16:8 light:dark cyc			
Comments:				
		//	1 , ,	
QC Check: BK 6/2/15		Final Review:	26/4/17	<u>·</u>

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



CETIS Summary Report

Report Date:

01 Jun-15 09:05 (p 1 of 2)

Test Code:

150505aart | 20-1963-0951

Batch ID:	
Ending Date: 12 May-15 11:00 Species: Atherinops affinis Source: Aquatic Biosystems, CO Source: Reference Toxicant Source:	
Duration: 6d 21h Source: Aqualic Biosystems, CO Age: 15 d Sample ID: 17-1594-3816 Code: 150505aart Copper Chloride Project: Project: Sample Age: 15 d Sample Age: 14 h Sation: Copper Chloride Project: Sample Age: 14 h Sation: Copper Chloride Sample Age: Sation: Copper Chloride Sation: Copper Chloride	
Sample Date 05 May-15 Saurice Source Source Sample Age 14	
Sample Date 05 May-15 Sample Age 14	
Receive Date 15 May-15	
Sample Age	
Analysis D	
Analysis D	
03-0930-3044 96h Survival Rate	
03-0930-3044 96h Survival Rate	t
Point Estimate Summary Summar	
Note	st
13-6031-1332 7d Survival Rate	
02-8524-0195 96h Survival Rate EC50 69.03 58.59 81.34 Trimmed Spearman-Kärber 13-1227-8396 Mean Dry Biomas-mg IC25 49.96 40.85 61.5 Linear Interpolation (ICPIN)	
13-1227-8396	
Test Acceptability	
Test Acceptability Survival Rate Control Resp 1 0.8 - NL Yes Passes Acceptability 1.069 0.85 - NL Yes Passes Acceptability 1.069 0.85 - NL Yes Passes Acceptability 1.069 0.85 - NL Yes Passes Acceptability 1.066513-3526 Mean Dry Biomas-mg Control Resp 1.069 0.85 - NL Yes Passes Acceptability 1.066513-3526 Mean Dry Biomas-mg Control Resp 1.069 0.85 - NL Yes Passes Acceptability 1.066513-3526 Mean Dry Biomas-mg Control Resp 1.069 0.85 - NL Yes Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.2054 NL - 0.25 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 Mean Dry Biomas-mg PMSD 0.1287 NL - 0.5 No Passes Acceptability 1.0666513-3526 No Passes Acceptability 1.0696 N.0666513-3526 Mean Dry Biomas-mg PMSD NL - 0.25 No Passes Acceptability 1.06966513-3526 Mean Dry Biomas-mg PMSD NL - 0.25 No Passes Acceptability 1.0696 N.0666513-3526 N.0666513-3526 Mean Dry Biomas-mg PMSD NL - 0.25 No Passes Acceptability 1.0696 N.0666513-3526 N.06665	
Analysis ID Endpoint Attribute Test Stat TAC Limits Overlap Decision 13-6031-1332 7d Survival Rate Control Resp 1 0.8 - NL Yes Passes Acceptability 15-7845-5090 7d Survival Rate Control Resp 1 0.8 - NL Yes Passes Acceptability 06-6513-3-3526 Mean Dry Biomass-mg Control Resp 1.069 0.85 - NL Yes Passes Acceptability 15-7845-5090 7d Survival Rate PMSD 0.2054 NL - 0.25 No Passes Acceptability 15-7845-5090 7d Survival Rate PMSD 0.1287 NL - 0.5 No Passes Acceptability 15-7845-5090 7d Survival Rate PMSD 0.1287 NL - 0.5 No Passes Acceptability 15-7845-5090 7d Survival Rate PMSD 0.1287 NL - 0.5 No Passes Acceptability 15-7845-5090 7d Survival Rate PMSD 95% UCL Min Max Std Err Std Dev CV% C-µg/L Control Type	
13-6031-1332	
15-7845-5090 7d Survival Rate	
06-6513-3526 Mean Dry Biomass-mg Control Resp 1.069 0.85 - NL Yes Passes Acceptability 13-1227-8396 Mean Dry Biomass-mg Control Resp 1.069 0.85 - NL Yes Passes Acceptability 15-7845-5090 7d Survival Rate PMSD 0.2054 NL - 0.25 No Passes Acceptability C-μg/L Control Type PMSD 95% LCL 95% UCL Min Max Std Err Std Dev CV% 0 Lab Control 5 1 1 1 1 0 0 0.0% 37.5 5 0.96 0.8489 1 0.8 1 0.04 0.08944 9.32% 75 5 0.28 0 0.6132 0 0.6 0.12 0.2683 95.83% 150 5 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	Criteria
13-1227-8396	
15-7845-5090 7d Survival Rate	
Oc. Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV%	
7d Survival Rate Summary C-μg/L Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV% 0 Lab Control 5 1 1 1 1 0 0 0.0% 37.5 5 0.96 0.8489 1 0.8 1 0.04 0.08944 9.32% 75 5 0.28 0 0.6132 0 0.6 0.12 0.2683 95.83% 150 5 0 0 0 0 0 0 0 0 0 0 0 95.83% 0<	
C-μg/L Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV% 0 Lab Control 5 1 1 1 1 0 0 0.0% 37.5 5 0.96 0.8489 1 0.8 1 0.04 0.08944 9.32% 75 5 0.28 0 0.6132 0 0.6 0.12 0.2683 95.83% 150 5 0 <td>Criteria</td>	Criteria
O Lab Control 5 1 1 1 1 1 0 0 0.0% 37.5 5 0.96 0.8489 1 0.8 1 0.04 0.08944 9.32% 75 5 0.28 0 0.6132 0 0.6 0.12 0.2683 95.83% 150 5 0	
37.5 5 0.96 0.8489 1 0.8 1 0.04 0.08944 9.32% 75 5 0.28 0 0.6132 0 0.6 0.12 0.2683 95.83% 150 5 0 0 0 0 0 0 0 0 0 0 300 5 0 0 0 0 0 0 0 0 0 600 5 0 0 0 0 0 0 0 0 0 96h Survival Rate Summary C-μg/L Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV%	%Effect
75 5 0.28 0 0.6132 0 0.6 0.12 0.2683 95.83% 150 5 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	0.0%
150 5 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	4.0%
300 5 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	72.0%
600 5 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	100.0%
96h Survival Rate Summary С-µg/L Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV%	100.0%
C-µg/L Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV%	100.0%
	a
U LAN CONTROL 5 7 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	%Effect
	0.0%
37.5 5 0.96 0.8489 1 0.8 1 0.04 0.08944 9.32%	4.0%
75 5 0.36 0.03621 0.6838 0 0.6 0.1166 0.2608 72.44%	
150 5 0.08 0 0.216 0 0.2 0.04899 0.1095 136.9%	64.0%
300 5 0 0 0 0 0 0	92.0%
600 5 0 0 0 0 0 0 0	92.0% 100.0%
Mean Dry Biomass-mg Summary	92.0%
C-μg/L Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV%	92.0% 100.0% 100.0%
0 Lab Control 5 1.069 0.9786 1.159 0.99 1.162 0.03249 0.07264 6.8%	92.0% 100.0% 100.0% %Effect
37.5 5 1.047 0.8626 1.232 0.804 1.19 0.06647 0.1486 14.19%	92.0% 100.0% 100.0% %Effect 0.0%
75 5 0.308 -0.1115 0.7275 0 0.818 0.1511 0.3378 109.7%	92.0% 100.0% 100.0% %Effect 0.0% 2.02%
150 5 0 0 0 0 0 0 0	92.0% 100.0% 100.0% %Effect 0.0% 2.02% 71.18%
300 5 0 0 0 0 0 0	92.0% 100.0% 100.0% %Effect 0.0% 2.02% 71.18% 100.0%
600 5 0 0 0 0 0 0	92.0% 100.0% 100.0% %Effect 0.0% 2.02% 71.18%

Report Date: Test Code: 01 Jun-15 09:05 (p 2 of 2) 150505aart | 20-1963-0951

						rest code.	100000aart 20-1000-0001
psmelt 7-d Surviva	I and Grov	vth Test					Nautilus Environmental (CA)
I Rate Detail							
Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
Lab Control	1	1	1	1	1		Sec. 1990
	1	1	1	8.0	1		
	0	0.4	0	0.6	0.4		
	0	0	0	0	0		
	0	0	0	0	0		
	0	0	0	0	0		
al Rate Detail							
Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
Lab Control	1	1	1	1	1		
	1	1	1	0.8	1		
	0.2	0.4	0	0.6	0.6		
	0	0	0	0.2	0.2		
	0	0	0	0	0		
	0	0	0	0	0		
Biomass-mg Detail							
Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5		
Lab Control	1.032	0.99	1.128	1.032	1.162		
	1.074	1.032	1.19	0.804	1.136		
	0	0.384	0	0.818	0.338		
	0	0	0	0	0		
	0	0	0	0	0		
	0	0	0	0	0		
	I Rate Detail Control Type Lab Control al Rate Detail Control Type Lab Control Giomass-mg Detail Control Type	Control Type Rep 1	Control Type Rep 1 Rep 2	Rate Detail Control Type Rep 1 Rep 2 Rep 3	Rate Detail Control Type Rep 1 Rep 2 Rep 3 Rep 4	Rate Detail Control Type Rep 1 Rep 2 Rep 3 Rep 4 Rep 5	Rate Detail Control Type Rep 1 Rep 2 Rep 3 Rep 4 Rep 5

2.034508

0.4048181

2.439326

1.017254

0.03373484

Between

Error

Total

Report Date: Test Code: 01 Jun-15 09:04 (p 1 of 4) 150505aart | 20-1963-0951

											0000.		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	0 1000 000 1
Pacific Topsn	nelt 7	-d Survival	and G	Frowth Test								Nautilu	ıs Environr	nental (CA)
Analysis ID: Analyzed:		'845-5090 Jun-15 9:04				urvival Ra metric-Co		「rea	tments		S Version		1.8.7	
Data Transfor	m		Zeta	Alt H	ур	Trials	Seed			PMSD	NOEL	LOEL	TOEL	TU
Angular (Corre	cted)		NA	C > T		NA	NA			20.5%	37.5	75	53.03	
Dunnett Multi	ple C	omparison	Test											
Control	vs	C-µg/L		Test S	Stat	Critical	MSD	DF	P-Value	P-Type	Decision	n(<i>a</i> :5%)		
Lab Control		37.5		0.41		2.108	0.245	8	0.4965	CDF	Non-Sigr	nificant Effe	t .	
		75*		6.921		2.108	0.245	8	<0.0001	CDF	Significa	nt Effect		
ANOVA Table														
Source		Sum Squa	res	Mean	Saua	are	DF		F Stat	P-Value	Decision	n(a:5%)		

Distributional T	Distributional Tests									
Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)					
Variances	Mod Levene Equality of Variance	4.092	8.022	0.0545	Equal Variances					
Variances	Levene Equality of Variance	17.78	6.927	0.0003	Unequal Variances					
Distribution	Shapiro-Wilk W Normality	0.8802	0.8328	0.0478	Normal Distribution					

30.15

< 0.0001

Significant Effect

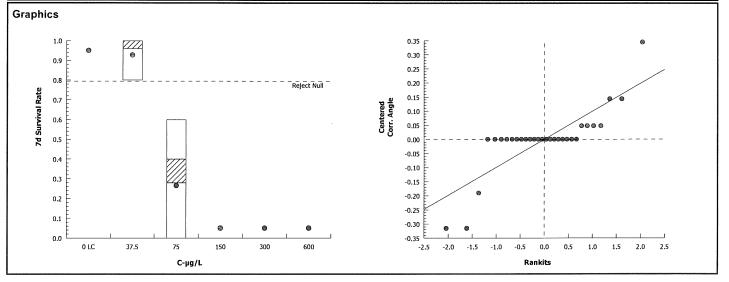
2

12

14

7d Surviva	d Survival Rate Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1	1	1	1	1	1	0	0.0%	0.0%
37.5		5	0.96	0.8489	1	1	8.0	1	0.04	9.32%	4.0%
75		5	0.28	0	0.6132	0.4	0	0.6	0.12	95.83%	72.0%
150		5	0	0	0	0	0	0	0		100.0%
300		5	0	0	0	0	0	0	0		100.0%
600		5	0	0	0	0	0	0	0		100.0%

Angular (C	Angular (Corrected) Transformed Summary										
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.345	1.345	1.346	1.345	1.345	1.345	0	0.0%	0.0%
37.5		5	1.298	1.165	1.43	1.345	1.107	1.345	0.04763	8.21%	3.54%
75		5	0.5413	0.1691	0.9135	0.6847	0.2255	0.8861	0.1341	55.38%	59.76%
150		5	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
300		5	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
600		5	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%



Report Date:

01 Jun-15 09:05 (p 2 of 4)

		70.1					Test	Code:	150	505aart 20	D-1963-0951	
Pacific Tops	melt 7-d Surviv	al and Gro	wth Test						Nautilus Environmental (CA)			
Analysis ID: Analyzed:	03-0930-3044 01 Jun-15 9:0		ndpoint: 96h nalysis: Par		ate ntrol vs Trea	tments		IS Version: cial Results	CETISv1 : Yes	.8.7		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU	
Angular (Corr	ected)	NA	C > T	NA	NA		19.6%	37.5	75	53.03		
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	37.5		0.4547	2.227	0.233 8	0.5647	CDF	Non-Signi	ficant Effect		***************************************	
	75*		6.837	2.227	0.233 8	<0.0001	CDF	Significant	t Effect			
***************************************	150*		9.781	2.227	0.233 8	<0.0001	CDF	Significan	t Effect			
ANOVA Table	е											
Source	Sum Sq	uares	Mean Squ	are	DF	F Stat	P-Value	Decision(a:5%)			
Between	3.826172	2	1.275391		3	46.5	<0.0001	Significant	t Effect			
Error	0.438840)4	0.0274275	2	16							
Total	4.265012	2			19							
Distributiona	l Tests											
Attribute	Test	Test		Test Stat	Critical	P-Value	Decision	(a:1%)				
Variances	Mod Lev	ene Equal	ity of Variance	4.418	5.953	0.0260	Equal Var	iances				
Variances	Levene	Equality of	Variance	8.445	5.292	0.0014	Unequal \	/ariances				
Distribution	Shapiro	-Wilk W No	ormality	0.9329	0.866	0.1757	Normal D	istribution				
96h Survival	Rate Summary										HE MATERIAL STATES OF THE STAT	
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	Lab Control	5	1	1	1	1	1	1	0	0.0%	0.0%	
37.5		5	0.96	0.8489	1	1	8.0	1	0.04	9.32%	4.0%	
75		5	0.36	0.03621	0.6838	0.4	0	0.6	0.1166	72.44%	64.0%	
150		5	0.08	0	0.216	0	0	0.2	0.04899	136.9%	92.0%	
300		5	0	0	0	0	0	0	0		100.0%	
600		5	0	0	0	0	0	0	0	181111111111111111111111111111111111111	100.0%	
Angular (Cor	rected) Transfo	rmed Sum	mary									
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	Lab Control	5	1.345	1.345	1.346	1.345	1.345	1.345	0	0.0%	0.0%	
37.5		5	1.298	1.165	1.43	1.345	1.107	1.345	0.04763	8.21%	3.54%	
75		5	0.6292	0.275	0.9834	0.6847	0.2255	0.8861	0.1276	45.33%	53.23%	
150		5	0.3208	0.1588	0.4827	0.2255	0.2255	0.4636	0.05833	40.66%	76.16%	
300		5	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%	
		_										

Analyst: CB QA: AC (a/S)/5

0.0%

83.24%

600

5

0.2255

0.2255

0.2256

0.2255

0.2255

0.2255

0

Report Date: Test Code: 01 Jun-15 09:05 (p 3 of 4) 150505aart | 20-1963-0951

Pacific Topsmelt 7-d Survival and Growth Test Nautilus Environmental (CA) Analysis ID: 03-0930-3044 Endpoint: 96h Survival Rate **CETIS Version:** CETISv1.8.7 Analyzed: 01 Jun-15 9:04 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 1.0 0.4 0,9 8.0 Reject Null 0.2 96h Survival Rate 0.7 0.1 0.5 -0.1 0.4 -0.2 -0.3 0.2 -0.4 0.1 0.0 0 LC 37.5 75 150 300 600 -1.5 -1.0 -0.5 0.0 C-µg/L Rankits

Report Date:

01 Jun-15 09:05 (p 4 of 4)

Test Code: 150505aart | 20-1963-0951

Pacific Topsmelt 7-d Surviva	l and Growth Test
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Nautilus Environmental (CA)

Analysis ID: 06-6513-3526 Endpoint: Mean Dry Biomass-mg CETIS Version: CETIS V1.8.7

Analyzed: 01 Jun-15 9:04 Analysis: Parametric-Two Sample Official Results: Yes

Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	Test Result
Untransformed	NA	C > T	NA	NA	12.9%	Passes mean dry biomass-mg

Equal Variance t Two-Sample Test

Control	vs	C-µg/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(a:5%)
Lab Control		37.5	0.2919	1.86	0.138	8	0.3889	CDF	Non-Significant Effect

ANOVA Table

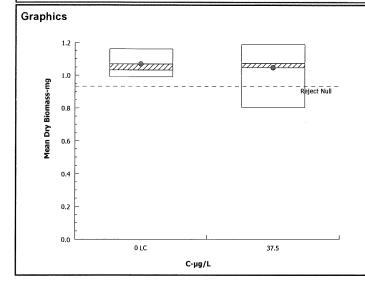
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(a:5%)
Between	0.001166405	0.001166405	1	0.08523	0.7778	Non-Significant Effect
Error	0.1094818	0.01368523	8			
Total	0.1106482		9			

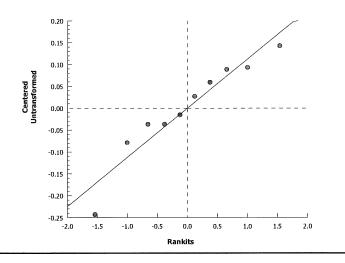
Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(a:1%)
Variances	Variance Ratio F	4.187	23.15	0.1944	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9262	0.7411	0.4112	Normal Distribution

Mean Dry Biomass-mg Summary

C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.069	0.9786	1.159	1.032	0.99	1.162	0.03249	6.8%	0.0%
37.5		5	1.047	0.8626	1.232	1.074	0.804	1.19	0.06647	14.19%	2.02%





Analyst: VB QA: ACGS 15

Report Date: **Test Code:**

01 Jun-15 09:05 (p 1 of 1) 150505aart | 20-1963-0951

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental (CA)

Analysis ID: 13-1227-8396 Analyzed: 01 Jun-15 9:04

Endpoint: Mean Dry Biomass-mg Analysis: Linear Interpolation (ICPIN)

CETISv1.8.7 **CETIS Version:** Official Results:

Yes

L	_inear	Interp	olation	Options
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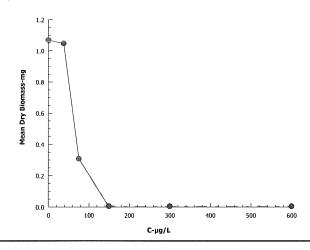
X Transform Y Transform Resamples Seed Exp 95% CL Method Linear 416493 1000 Linear Yes Two-Point Interpolation

Point Estimates

μg/L 95% LCL 95% UCL IC25 61.5 49.96 40.85 IC50 63.51 53.28 90.73

Mean Dry	Biomass-mg Sum	mary		Calculated Variate						
C-µg/L	Control Type	Count	Mean	Mean Min Max Std Err Std Dev CV% %Effect						
0	Lab Control	5	1.069	0.99	1.162	0.03249	0.07264	6.8%	0.0%	
37.5		5	1.047	0.804	1.19	0.06647	0.1486	14.19%	2.02%	
75		5	0.308	0	0.818	0.1511	0.3378	109.7%	71.18%	
150		5	0	0	0	0	0		100.0%	
300		5	0	0	0	0	0		100.0%	
600		5	0	0	0	0	0		100.0%	

Graphics



Analyst: KB QAACCEPS 15

000-089-180-4 CETIS™ v1.8.7.20

Report Date:

01 Jun-15 09:05 (p 1 of 2)

Test Code:

150505aart | 20-1963-0951

Pacific Topsme	lt 7-d Survival and	Growth Test
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Nautilus Environmental (CA)

Analysis ID: Analyzed:

13-6031-1332 01 Jun-15 9:04 Endpoint: 7d Survival Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version:

CETISv1.8.7

Trimmed Spearman-Kärber Estimates

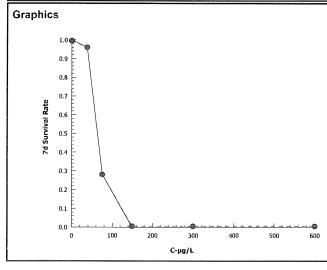
Official Results:

Yes

	Trimmea	3	pearman-Narber	Estimates
ı			_	

Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0	4.00%	1.797	0.02978	62.73	54.69	71.95

7d Surviv	al Rate Summary	Calculated Variate(A/B)									
C-μg/L	Control Type Count		Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	1	1	1	0	0	0.0%	0.0%	25	25
37.5		5	0.96	0.8	1	0.04	0.08944	9.32%	4.0%	24	25
75		5	0.28	0	0.6	0.12	0.2683	95.83%	72.0%	7	25
150		5	0	0	0	0	0		100.0%	0	25
300		5	0	0	0	0	0		100.0%	0	25
600		5	0	0	0	0	0		100.0%	0	25



Report Date:

01 Jun-15 09:05 (p 2 of 2)

Test Code: 150505aart | 20-1963-0951

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental (CA)

Analysis ID: 02-8524-0195 Analyzed: 01 Jun-15 9:04 Endpoint: 96h Survival Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version: Official Results:

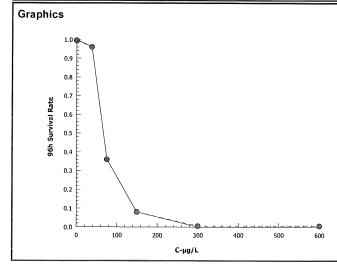
CETISv1.8.7

Yes

Trimmed	Spearman-Kärber	Estimates
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Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0	4.00%	1.839	0.03562	69.03	58.59	81.34

96h Survival Rate Summary			Calculated Variate(A/B)								
C-µg/L	Control Type Count		Mean	Mean Min Max Std Err Std Do		Std Dev	CV%	%Effect	Α	В	
0	Lab Control	5	1	1	1	0	0	0.0%	0.0%	25	25
37.5		5	0.96	8.0	1	0.04	0.08944	9.32%	4.0%	24	25
75		5	0.36	0	0.6	0.1166	0.2608	72.44%	64.0%	9	25
150		5	0.08	0	0.2	0.04899	0.1095	136.9%	92.0%	2	25
300		5	0	0	0	0	0		100.0%	0	25
600		5	0	0	0	0	0		100.0%	0	25

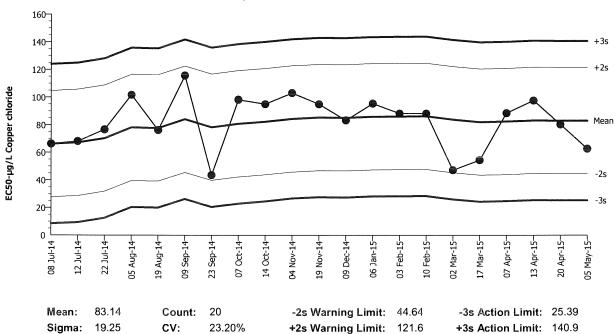


CETIS QC Plot

Pacific Topsmelt 7-d Survival and Growth Test Nautilus Environmental (CA)

Test Type:Growth-Survival (7d)Organism:Atherinops affinis (Topsmelt)Material:Copper chlorideProtocol:EPA/600/R-95/136 (1995)Endpoint:7d Survival RateSource:Reference Toxicant-REF

Pacific Topsmelt 7-d Survival and Growth Test



Quality Control Data											
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Jul	8	12:15	66.2	-16.94	-0.8799			11-1291-5570	01-3275-8518
2			12	15:30	68.06	-15.08	-0.7832			19-5679-7209	09-1848-2586
3			22	12:10	76.61	-6.532	-0.3393			08-9694-1010	04-9281-6630
4		Aug	5	12:30	101.7	18.52	0.962			02-3572-4318	04-1355-8832
5			19	12:50	76.14	-7.001	-0.3637			10-2220-5806	14-8107-1507
6		Sep	9	13:15	115.7	32.53	1.69			00-2260-7679	08-1265-9953
7			23	10:00	43.5	-39.64	-2.059	(-)		10-2715-0848	08-7935-5641
8		Oct	7	14:55	98.2	15.06	0.7825			03-4335-1097	08-4043-4062
9			14	14:55	94.93	11.79	0.6126			03-4389-3229	08-0313-6046
10		Nov	4	17:00	103.2	20.03	1.04			07-3336-0499	03-9882-1644
11			19	15:35	94.93	11.79	0.6126			01-4951-3716	16-3767-9666
12		Dec	9	13:30	83.34	0.2032	0.01056			18-0939-8222	20-7541-1411
13	2015	Jan	6	11:00	95.52	12.38	0.6429			09-8025-9732	14-3733-1272
14		Feb	3	14:00	88.25	5.109	0.2654			21-1065-0393	01-8543-8572
15			10	14:30	88.25	5.109	0.2654			02-7871-6683	00-0287-3888
16		Mar	2	15:30	47.25	-35.89	-1.865			16-8982-2124	03-5342-0079
17			17	10:15	54.52	-28.62	-1.487			18-0061-0968	11-1706-8798
18		Apr	7	12:35	88.59	5.445	0.2829			19-9342-6452	01-8109-7656
19			13	14:10	97.6	14.46	0.7512			21-1811-2763	06-4686-3592
20			20	13:50	80.38	-2.757	-0.1432			00-6349-8324	16-3484-4333
21		May	5	14:15	62.73	-20.41	-1.06			20-1963-0951	13-6031-1332

Analyst: VB QA: 15 6/3/15

000-089-180-4 CETIS™ v1.8.7.20

CETIS QC Plot Report Date: 01 Jun-15 09:05 (1 of 1)

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental (CA)

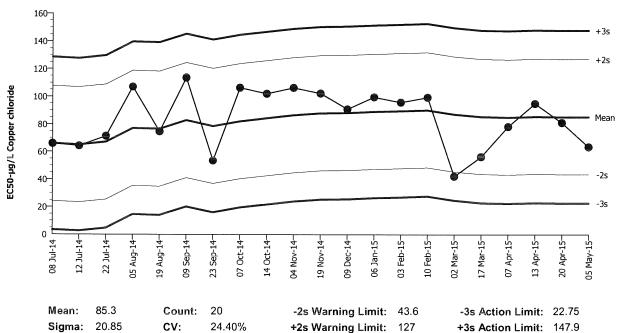
Test Type: Growth-Survival (7d)

Organism: Atherinops affinis (Topsmelt)

Material: Copper chloride

Protocol: EPA/600/R-95/136 (1995) Endpoint: Mean Dry Biomass-mg Source: Reference Toxicant-REF

Pacific Topsmelt 7-d Survival and Growth Test



Quality C	ontrol	Data
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Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Jul	8	12:15	65.94	-19.36	-0.9283			11-1291-5570	09-7900-1840
2			12	15:30	64.2	-21.1	-1.012			19-5679-7209	11-2284-5616
3			22	12:10	71.33	-13.97	-0.6701			08-9694-1010	11-8500-0101
4		Aug	5	12:30	107	21.7	1.041			02-3572-4318	06-4448-6794
5			19	12:50	74.65	-10.65	-0.5109			10-2220-5806	20-0116-6202
6		Sep	9	13:15	113.5	28.24	1.354			00-2260-7679	20-3452-2478
7			23	10:00	53.65	-31.65	-1.518			10-2715-0848	08-1906-9903
8		Oct	7	14:55	106.4	21.1	1.012			03-4335-1097	09-3471-6201
9			14	14:55	102.1	16.81	0.806			03-4389-3229	04-1492-4013
10		Nov	4	17:00	106.3	21.03	1.009			07-3336-0499	19-0008-4150
11			19	15:35	102.3	17	0.8156			01-4951-3716	05-1986-4487
12		Dec	9	13:30	90.88	5.578	0.2675			18-0939-8222	21-2156-2542
13	2015	Jan	6	11:00	99.63	14.33	0.6873			09-8025-9732	20-8969-8695
14		Feb	3	14:00	95.81	10.51	0.504			21-1065-0393	08-1230-3729
15			10	14:30	99.42	14.12	0.6771			02-7871-6683	01-6233-9739
16		Mar	2	15:30	42.15	-43.15	-2.07	(-)		16-8982-2124	13-6300-7842
17			17	10:15	56.41	-28.89	-1.386			18-0061-0968	00-1953-5316
18		Apr	7	12:35	78.22	-7.085	-0.3398			19-9342-6452	09-2081-8015
19			13	14:10	94.96	9.66	0.4633			21-1811-2763	05-5570-3525
20			20	13:50	81.16	-4.137	-0.1984			00-6349-8324	17-2159-2892
21		May	5	14:15	63.51	-21.79	-1.045			20-1963-0951	13-1227-8396

Analyst: CO QA: Su 6/3/15

000-089-180-4 CETIS™ v1.8.7.20

CETIS QC Plot Report Date: 01 Jun-15 09:05 (1 of 1)

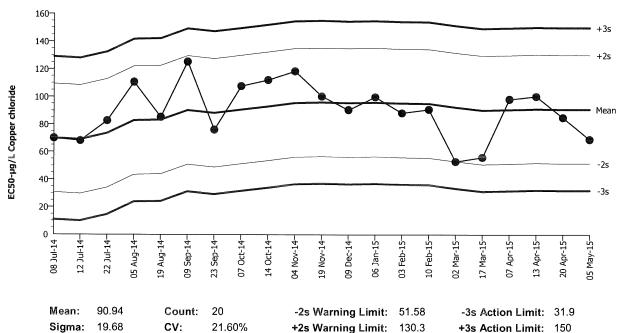
Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental (CA)

Test Type: Growth-Survival (7d) Organism: Atherinops affinis (Topsmelt) Material: Copper chloride

Protocol: EPA/600/R-95/136 (1995) Endpoint: 96h Survival Rate Source: Reference Toxicant-REF

Pacific Topsmelt 7-d Survival and Growth Test



Qual	ity Con	trol Data	а								
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2014	Jul	8	12:15	69.98	-20.96	-1.065			11-1291-5570	12-3485-8625
2			12	15:30	68.06	-22.88	-1.162			19-5679-7209	14-7190-8017
3			22	12:10	82.64	-8.297	-0.4216			08-9694-1010	03-0721-9902
4		Aug	5	12:30	110.7	19.72	1.002			02-3572-4318	17-8338-0224
5			19	12:50	85.25	-5.692	-0.2892			10-2220-5806	03-5586-7088
6		Sep	9	13:15	125.4	34.44	1.75			00-2260-7679	03-6498-2670
7			23	10:00	76.05	-14.89	-0.7568			10-2715-0848	05-4744-6984
8		Oct	7	14:55	107.7	16.77	0.8523			03-4335-1097	14-3670-0847
9			14	14:55	112.1	21.17	1.076			03-4389-3229	02-1869-3649
10		Nov	4	17:00	118.5	27.57	1.401			07-3336-0499	04-3369-9002
11			19	15:35	100.3	9.405	0.4779			01-4951-3716	01-3053-1585
12		Dec	9	13:30	90.54	-0.3953	-0.02009			18-0939-8222	16-6086-0113
13	2015	Jan	6	11:00	99.86	8.922	0.4533			09-8025-9732	16-4836-2900
14		Feb	3	14:00	88.25	-2.691	-0.1367			21-1065-0393	09-4908-9330
15			10	14:30	90.78	-0.1583	-0.00805			02-7871-6683	04-3347-7000
16		Mar	2	15:30	53.03	-37.91	-1.926			16-8982-2124	18-8086-0569
17			17	10:15	56.06	-34.88	-1.773			18-0061-0968	11-1889-0336
18		Apr	7	12:35	98.3	7.365	0.3742			19-9342-6452	04-7127-1266
19			13	14:10	100.3	9.405	0.4779			21-1811-2763	09-5535-4961
20			20	13:50	84.97	-5.974	-0.3035			00-6349-8324	11-3417-4341
21		May	5	14:15	69.03	-21.91	-1.113			20-1963-0951	02-8524-0195

Client: Internal	Test Species: A. affinis
Sample ID: CuCl ₂	Start Date/Time: 5/4/2015 5/5/15 1915
Test No.: 15050 Aaart	End Date/Time: 5/11/2015 5/12/15 1100

Conc.	Don	Rand			Test	Day / N	o. Orga	nisms /	Alive		Percent
(μg/L)	Rep.	#	0	1	2	3	4	5	6	7	Survival
Lab Control	а	3	5	5	5	5	5	5	5	5	100
	b	9	5	5	5	5	5	5	5	5	100
	С	1	5	5	5	S	5	5	- 5	5	100
	d	7	5	5	5	5	5	5	5	5	100
	е	6	5	5	5	ح	5	5	5	5	(00
37.5	a	27	5	5	5	5	5	5	5	5	100
	b	21	5	5	5	S	5	5	5	5	100
**************************************	С	2	5	5	5	ς	5	5	5	5	100
The state of the s	d	22	5	4	4	4	4	4	4	4	80
	е	30	5	5	5	-5	5	5	5	5	(00
75	a	17	5	4	3	i	1	l	0		0
	b	20	5	4	2	2_	2	2	2	2	40
	С	16	5	5	2	(0	i.ego-	~		0
	d	26	5	4	3	3	3	@B3	3	3 2	60
	е	29	5	5	3	3	3	3	3	2	40
150	a	15	5	4	3	i	0		, per	EP)	O
	b	25	5	4	Ø				- 0	er/	0
	С	13	5	3	2	0		-		/-	0
	d	24	5	3	2			0	PY		0
	е	8	5	ч	3	1	l	0			0
300	а	19	5	0							0
	b	28	5	0			a cod				0
	С	12	5	0		211	ead				0
	d	10	5	0							0
	е	23	5	0				************			0
600	a	11	5	0							0
M	b	14	5	0				1			0
	С	5	5	0		, \	Dea	9			Ö
	d	4	5	0	ļ	1211	V				0
	е	18	5	0							0
Rand # QC: FG Initial Count QC: PD	Tech	Initials Time	EG 1415	BY_ 1000	44	1203	VCR 1430	BIL	1450 CH	1/00 AD	

Initial Count QC:	AD		Time	1415	1000	1100	1703	1430	1200	CH	AD		
Time Fed (day):	0	1	2	3	4	5	6			D	rying Ov	en Info	
morning:	~	0815	0820	0930	0825	0830	0830		Tare v	vt. Initia	als/Date: 1	56 5/12/1	<u>5 o</u> 935
evening:	1540	1530	1655	1530	1730	1545	1700			Date/	Time in:	5/12/15	1335
										Date/T	ime out:	5/13/15	1430
										Te	mp (°C):	600	
	C	* ~ ~	121100	gara-									

Comments: @ 018 VCR 5/4/15

QC Check: KB5|2||5 Final Review: AC6/5/1

Client: _	Internal	Test Species: A. affinis
Sample ID: _	CuCl ₂	Start Date/Time: 5/5/2015 1415
Test No.:	150505aart	End Date/Time: 5/12/2015 1100

Conc.		pan weight	pan + fish weight	total organism
(μg/L)	Rep.	(mg)	(mg)	weight (mg)
Lab Control	а	22.87	28.03	5.16
	b	23.07	28.02	4.95
	С	20.23	25.87	5.64
	d	21.65	26.81	5.16
	е	24.4	30.21	5.81
	a	21.07	26.44	5.37
37.5	b	22.23	27.39	5.16
	С	20.3	26.25	5.95
	d	19.86	23.88	4.02
	е	23.07	28.75	5.68
	а	0	0	0
75	b	22.21	24.13	1.92
	С	0	0	0
	d	22.02	26.11	4.09
	е	22.86	24.55	1.69
	а	0	0	0
150	b	0	0	0
	С	0	0	0
	d	0	0	0
	е	0	0	0
	а	0	0	0
300	b	0	0	0
	С	0	0	0
	d	0	0	0
	e	0	0	0
	a	0	0	0
600	b	0	0	0
	С	0	0	0
	d	0	0	0
	е	0	0	0
	а			
	b			
	С			
	d			
	е			
<u>L</u>		66	66	

 Tech Initials:
 SG
 SG

 Date/Time:
 5/12/15 0935
 5/13/15 1430

QC Check: KB5|21|15
Final Review: AC 6/5/15

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

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

Client:	Internal	Test Species	: <u>A. affil</u>	nis						
Sample ID:	CuCl ₂	Start Date/Time	5/4/20	15 5)	5/15	1414)			
Test No:		End Date/Time:	() 5114191	D M=-5/	12/15	1100	_			
	ACQ18(6-5-15	End Date/Time	. 3/11/20	113 0/	12113	1100	ر			
Concentration	Lab Control	Concentration		·		150	μg/L		40.0.	
Day	0 1 2 3 4 5 6 7	Day	0	1	2	3	4	5	6	7
	Initial						itial			
pH	806 8.07 8.04 7.76 8.01 8.14 7.82	pН	8.06	8.04	8.04	7.98	8,02	8.14		
DO (mg/L)	7.9 7.8 7.2 6.6 7.7 7.6 7.3	DO (mg/L)	7.9	7-9	7.2	6.7	8.1	7.8	01	7)
Salinity (ppt)	30.2 70.4 30.5 30.4 30.0 29.8 29.8 19.1 20.1 19.9 70.0 19.5 19.3 20.2	Salinity (ppt)	30.2		30.4	30,4	29.4	29.9	100	e .
Temp (°C)	19.1 20.1 19.9 70.0 19.5 19.3 20.2 Final	Temp (°C)	19.1	20.1	120.0	19.8	19.3	19.3		N. V
рН	1.88 7.78 7.74 7.69 7.8117.78 7.78	pH	1	1 -1 a A	17 a .		nal			Links
DO (mg/L)	4.7 56 65 6.9 5.9 7.1 6.5	DO (mg/L)	-	7.94		7.82	7.85	7,87		1
Temp (°C)	20.1 20.1 19.8 20.1 20.2 20.1 20.2	Temp (°C)	-	20-1	20.2	6.0	7.0	6.9	7.3	
	- 120° 10°	remp (C)	<u> Printeininin</u>	1201	120.2	19.9	20,2	20.1	20.0	
Concentration	37.5 μg/L	Concentration	Τ			300	μg/L	~~~~		
Day	0 1 2 3 4 5 6 7	Day	0	1	2	3		- E		
	Initial	Day	1 0	16111111111111	1 2		4 tial	5	6	7
pН	8.07 8.08 8.03 798 8.02 8.14 781	рН	8.06	8.05	T.		uai	<u> </u>	1	
DO (mg/L)	79 78 7.2 68 7.9 7.7 7.2	DO (mg/L)	79	4-9				<u> </u>		
Salinity (ppt)	30.2 70.4 30.5 30.4 30.1 29.9 30.0	Salinity (ppt)	30.2	30-3			LOUR 1	1		
Temp (°C)	19.1 20-1 20.0 19.9 19.4 19.3 26.2	Temp (°C)	19.0	20.1			7	100		
	Final					Fi	nal	1	1	
pH	1.90 7.81 7.80 7.72 7.81 7.80 7.81	pH	_	7.95						
DO (mg/L)	v.7 6.0 59 6.5 6.2 7.3 6.4	DO (mg/L)		7.0						
Temp (°C)	20.0 -0.1 19.7 20.1 20.1 20.1 20.5	Temp (°C)		20.7						
Concentration	75 ug/l	Concentration				600	/1			
	75 μg/L	Concentration					μg/L	т		
Concentration Day	0 1 2 3 4 5 6 7	Day	0	1	2	3	4	5	6	7
	0 1 2 3 4 5 6 7	Day			2	3		5	6	7
Day	0 1 2 3 4 5 6 7 Initial	Day pH	8.04	8.05	2	3	4	5	6	7
Day pH	0 1 2 3 4 5 6 7 Initial:	Day pH DO (mg/L)	8.04 7.9	¥.05 1-1	2	3	4	5	6	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C)	0 1 2 3 4 5 6 7 S.07 8.00 8.02 1.96 8.02 8.14 7.89 30.2 30.3 30.5 30.4 30.0 29.9 39.1 19.0 20.1 20.0 19.4 19.3 19.4	Day pH	8.04	7.05 7-9 70.2	2	3	4	5	6	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C)	0 1 2 3 4 5 6 7 S.07 S.00 S.02 1.98 S.02 S.14 7.89 7.9 1.2 1.2 1.2 1.3 1.5 7.0 2.0 3.0 3.0 2.9 3.0 7.6 1.0 3.0 3.0 2.9 3.0 7.6 1.0 3.0 3.0 3.0 3.0 7.6 1.0 3.0 3.0 3.0 3.0 7.6 1.0 3.0 3.0 3.0 3.0 7.6 1.0 3.0 3.0 3.0 3.0 7.6 1.0 3.0 3.0 3.0 7.6 1.0 3.0 3.0 7.6 3.0 3.0 3.0 7.6 3.0 3.0 7.6 3.0 3.0 7.6 3.0 3.0 7.6 3.0	Day pH DO (mg/L) Salinity (ppt) Temp (°C)	30.1	¥.05 1-1	2	3	4	5	6	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH	0 1 2 3 4 5 6 7 S.07 8.00 8.02 1.98 8.02 8.14 7.89 7.9 7.2 8.0 7.8 7.5 7.9 7.2 8.0 7.8 7.5 7.8 7.8 7.8 7.8 7.8 7.8 7.8 7.8 7.8 7.9 7.8 7.8 7.8 7.8 7.8 7.9 7.8 7.8 7.8 7.8 7.8 7.9 7.8	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH	30.1	7.05 7-9 70.2	2	3	4 tial		6	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L)	0 1 2 3 4 5 6 7 S.07 8.00 8.02 4.96 8.02 8.14 7.89 79 79 70 8.00 7.8 7.5 70 70 70 70 70 70 70 70 70	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L)	30.1	8.05 7-9 70.2 20.2 7-94 7.2	2	3	4 tial		6	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH	0 1 2 3 4 5 6 7 S.07 8.00 8.02 1.98 8.02 8.14 7.89 7.9 7.2 8.0 7.8 7.5 7.9 7.2 8.0 7.8 7.5 7.8 7.8 7.8 7.8 7.8 7.8 7.8 7.8 7.8 7.9 7.8 7.8 7.8 7.8 7.8 7.9 7.8 7.8 7.8 7.8 7.8 7.9 7.8	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH	30.1	8.05 7-9 70.2 20.2	2	3	4 tial		6	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L)	0 1 2 3 4 5 6 7 S.07 8.00 5.02 1.96 5.02 5.14 7.59 7.9 1.2 1.2 1.6 8.0 7.8 7.5 30.2 70.2 70.5 30.4 30.0 29.9 30.4 16.10 20.1 70.0 19.4 19.3 19.4 17.18 7.82 7.83 7.81 7.91 7.62 7.89 10.4 6.2 6.8 6.8 6.5 7.2 7.0 20.7 20.0 19.7 20.0 20.1 20.0 20.5	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L)	8.04 7.9 30.1 19.0	8.05 7-9 70.2 20.2 7-94 7.2 20.1		3 Ini	4 tfal::::::::::::::::::::::::::::::::::::	\$ com		7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C)	0 1 2 3 4 5 6 7 S.07 8.00 5.02 1.96 5.02 5.14 7.89 T.9 1.7 7.2 6.6 8.0 7.8 7.5 30.2 70.3 30.5 30.4 30.0 29.9 30.4 T.8 11.0 20.1 20.0 30.4 30.0 29.9 30.4 T.8 7.82 7.63 7.81 7.91 7.62 7.89 10.4 6.2 6.0 6.8 6.5 7.2 7.0 20.7 20.0 19.7 20.0 20.1 20.0 20.5 10.2 6.1 1.5	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L)	8.04 7.9 30.1 19.0	8.05 7-9 70.2 20.2 7-94 7.2 20.1	2	3 Ini	4 tial and a second	5	6	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L)	0 1 2 3 4 5 6 7 S.07 8.00 5.02 1.96 5.02 5.14 7.69 T9	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L)	8.04 7.9 30.1 19.0	8.05 7-9 70.2 20.2 7-94 7.2 20.1		3 Ini	4 tfal::::::::::::::::::::::::::::::::::::	\$ com		7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C)	0 1 2 3 4 5 6 7 S.07 8.00 8.02 1.96 5.02 3.14 7.89 1.9	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial:	8.04 7.9 30.1 19.0	タ・0カ オ・9 カ・ス 20・2 干・94 モ・20 1 1 大しり	2 KS	3 Ini	4 tial and a distribution of the second of t	5 CH	6	
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F	0 1 2 3 4 5 6 7 S.07 8.00 5.02 1.96 5.02 5.14 7.69 T9	pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final:	3.09 7.9 3.0.1 19.0 NH	7.05 7.9 70.7 20.2 7.94 7.2 20.1 1 ALB	2 KB	3 3 NH NH	4 tial	5 CH CH	6 KB KD	7
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F	0 1 2 3 4 5 6 7 S.07 8.00 5.02 1.96 5.02 5.14 7.69 T9	pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final:	8:04 7:9 3:0:1 19:0 NH	1 ALB AWD EG	2 K3 k8 EG-	3 Ini 3 Nih Nh ALB	4 tial and a distribution of the second of t	5 CH	6 KB KB	
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F Animal Age at Initiation	0 1 2 3 4 5 6 7 S.07 8.00 5.02 1.96 5.02 5.14 7.59 1.9	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final: Dilutions made by: High conc. made (µg/L):	8:04 7:9 3:0:1 19:0 NH	7.05 1-9 20.2 20.2 1-94 1.2 20.1 1 ALB AWD EG 600	2 KB KB EG-150	3 NH NH ALB 150	4 ttal 4 ty Ye EG ISO	5 CH CH	6 KB KB T5	
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F	0 1 2 3 4 5 6 7 S.07 8.00 5.02 1.96 5.02 5.14 7.59 1.9	pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final:	8.04 7.9 30.1 19.0 NH	1 ALB AWD EG	2 KB KB EG-150	3 NH NH ALB 150	4 tial	5 CH CH	6 KB KB	
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F Animal Age at Initiation	0 1 2 3 4 5 6 7 \$.07 \$.00 \$.02 \$.96 \$.02 \$.14 7.69 \$.94 \$.4 \$.4 \$.0 \$.0 \$.0 \$.0 \$.0 \$.07 \$.00 \$.02 \$.04 \$.0 \$.0 \$.0 \$.0 \$.07 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.0	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final: Dilutions made by: High conc. made (µg/L):	8:04 7:9 3:0:1 19:0 NH	7.05 1-9 20.2 20.2 1-94 1.2 20.1 1 ALB AWD EG 600	2 KB KB EG-150	3 NH NH ALB 150	4 ttal 4 ty Ye EG ISO	5 CH CH	6 KB KB T5	
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F Animal Age at Initiation	0 1 2 3 4 5 6 7 S.07 8.00 8.02 1.96 8.02 8.14 7.89 30.2 70.3 70.5 30.4 30.0 7.8 7.5 30.2 70.3 70.5 30.4 30.0 7.8 7.5 16 17.0 20.1 20.0 19.4 19.4 19.3 19.4 17.18 7.82 2.83 7.81 7.91 7.82 7.89 10.4 6.2 6.0 6.8 6.5 7.2 7.0 20.7 20.0 19.7 20.0 20.1 20.0 20.5 Received: ABS S 1/15 15 0 Added to the control of the	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final: Dilutions made by: High conc. made (μg/L): /ol. Cu stock added (mL):	8:04 7:9 3:0:1 19:0 NH	7.05 1-9 20.2 20.2 1-94 1.2 20.1 1 ALB AWD EG 600	2 KB KB EG-150	3 NH NH ALB 150	4 ttal 4 ty Ye EG ISO	5 CH CH	6 KB KB T5	
Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F Animal Age at Initiation	0 1 2 3 4 5 6 7 \$.07 \$.00 \$.02 \$.96 \$.02 \$.14 7.69 \$.94 \$.4 \$.4 \$.0 \$.0 \$.0 \$.0 \$.0 \$.07 \$.00 \$.02 \$.04 \$.0 \$.0 \$.0 \$.0 \$.07 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.00 \$.0	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final: Dilutions made by: High conc. made (μg/L): /ol. Cu stock added (mL):	8:04 7:9 3:0:1 19:0 NH	7.05 1-9 20.2 20.2 1-94 1.2 20.1 1 ALB AWD EG 600	2 KB KB EG-150	3 NH NH ALB 150	4 ttal 4 ty Ye EG ISO	5 CH CH	6 KB KB T5	
pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F Animal Age at Initiation	0 1 2 3 4 5 6 7 S.07 8.00 8.02 1.96 8.02 8.14 7.89 30.2 70.3 70.5 30.4 30.0 7.8 7.5 30.2 70.3 70.5 30.4 30.0 7.8 7.5 16 17.0 20.1 20.0 19.4 19.4 19.3 19.4 17.18 7.82 2.83 7.81 7.91 7.82 7.89 10.4 6.2 6.0 6.8 6.5 7.2 7.0 20.7 20.0 19.7 20.0 20.1 20.0 20.5 Received: ABS S 1/15 15 0 Added to the control of the	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final: Dilutions made by: High conc. made (μg/L): /ol. Cu stock added (mL):	8:04 7:9 3:0:1 19:0 NH	1 ALB AVB EG 600 17.3	2 KB KB EG-150	3 Nit NIT ALB 150 4.325	4 ttal 4	5 CH CH	6 KB KD EG 75 2.163	
pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Animal Source/Date F Animal Age at Initiation	0 1 2 3 4 5 6 7 S.07 8.00 8.02 1.96 5.02 8.14 7.89 1.9 1.2 1.96 8.0 7.8 7.5 30.2 70.3 30.5 30.4 30.0 29.9 30.1 1.9 1.9 1.9 1.9 1.9 1.9 1.9 1.9 1.9 1.8 1.8 7.8 7.9 7.8 7.9 1.9 1.9 1.8 1.8 7.8 7.9 7.8 7.2 7.0 1.9 1.9 1.9 1.9 1.9 1.0 20.1 20.0 20.5 Received: ABS 6 1.15 1.5 1.5 1.15 1.5 1.15 1.5 1.15 1.5 1.15 2.5 1.15 3.6 1.15 3.6 1.15 3.7 1.15 3.8 1.15 3.8 1.15 3.9 1.15 3.9 1.15 3.9 1.15 3.9 1.15 3.9 1.15 3.9 1.9 3.9 1.9 3.9 1.9 3.9 1.9 3.9 1.9 3.9 1.9 3.9 1.9 3.9 1.9 3.9 1.9 3.9 1.9 4.9 1.9 5.0 1.9 5.0 1.9 6.0 1.9 7.0	Day pH DO (mg/L) Salinity (ppt) Temp (°C) pH DO (mg/L) Temp (°C) Analysts: Initial: Final: Dilutions made by: High conc. made (µg/L): o Final Volume = 2500 mL	8:04 7:9 3:0:1 19:0 NH	1 ALB AVB EG 600 17.3	2 KB KB FG- 150 4.325	3 Nit NIT ALB 150 4.325	4 ttal 4	5 CH CH NH 150 4:325	6 KB KD EG 75 2.163	