

Chronic Toxicity Test Results for the Carlsbad Desalination Plant

❖ Sample ID: M-001 Sample Collection Date: June 30, 2017

Prepared for: IDE AMERICAS, Inc.

4590 Carlsbad Boulevard Carlsbad, CA 92008

Prepared by: Nautilus Environmental

Submitted: July 13, 2017

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 Department of Health Services Environmental Laboratory Accreditation Program (Certificate No. 1802) and the State of Washington Department of Ecology (Lab ID C552).
- 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.

California 4340 Vandever Avenue San Diego, California 92120

858.587.7333 fax: 858.587.3961 Results verified by: ______ adrienne Cibor

EXECUTIVE SUMMARY

CHRONIC TOXICITY TESTING

CARLSBAD DESALINATION PLANT - JUNE 2017

ORDER NO. R9-2006-0065; NPDES NO. CA0109223

Sampling Date: June 30, 2017

Test Date: June 30, 2017

Sample IDs: M-001 Brine Effluent, ERI Brine, Brine Pit, Train 4,

PT Filter EFF, and M-INF

M-001

Effluent Limitation: 16.5 TUc

Results Summary:

Bioassay Type:	M-001 Efflu Resu		Effluent Limitation Met? (Yes/No)
	<u>NOEC</u>	<u>TU</u> c	NI-
Echinoderm Fertilization	5.0	20	No

INTRODUCTION

A 24-hour composite discharge sample was collected in June 2017 for the Poseidon Resources (Channelside) LLC, Carlsbad Desalination Project (CDP) for chronic toxicity monitoring purposes. Due to effects observed in a sample collected and tested for monthly monitoring purposes on June 17, 2016 from the CDP discharge monitoring point (M-001), accelerated monitoring was triggered according to the permit that was adopted in 2006 (Order No. R9-2006-0065). Additional samples collected throughout the facility were also tested for comparison purposes. Bioassay testing was conducted at the Nautilus Environmental (Nautilus) laboratory in San Diego, California on June 30, 2017 using the purple urchin (*Strongylocentrotus purpuratus*) chronic fertilization test.

Client: IDE Americas, Inc.

Sample Collection Date: June 30, 2017

MATERIALS AND METHODS

The samples were collected on June 30, 2017. Sample collection was performed by IDE Americas, Inc. (IDE) personnel, and the samples were hand delivered to Nautilus the day of sample collection. Following arrival at Nautilus, an aliquot of the sample was poured off and the following water quality parameters were measured: pH, dissolved oxygen (DO), temperature, salinity, alkalinity, and total chlorine. A summary of the sample collection and receipt information is provided in Table 1, and water quality parameters measured upon receipt at Nautilus are presented in Table 2. Testing was conducted in accordance with the protocol described in USEPA 1995, and the methods are summarized in Table 3.

Table 1. Sample Information

Client/Project: IDE Americas, Inc./ Carlsbad Desalination Plant

Monitoring Period: June 2017

Sample ID: 1. M-001, desalination plant brine effluent

ERI Brine
 Brine Pit
 Train 4
 PT Filter EFF
 M-INF

Sample Collection Date, Time: 1. 6/30/17, 08:00

2. 6/30/17, 08:00 3. 6/30/17, 08:00 4. 6/30/17, 08:00 5. 6/30/17, 08:00 6. 6/30/17, 08:00

Sample Receipt Date, Time: 6/30/17, 11:48

Sampling Method: 24-hour Composite

Table 2. Water Quality Measurements upon Sample Receipt

Sample ID	рН	DO (mg/L)	Temp (°C)	Salinity (ppt)	Alkalinity (mg/L as CaCO₃)	Total Chlorine (mg/L)
M-001	7.72	6.7	6.5	62.6	190	0.03
ERI Brine	7.21	7.3	4.5	68.4	199	< 0.02
Brine Pit	8.00	7.0	6.5	30.2ª	100	0.03
Train 4	7.75	7.0	5.5	70.2	206	< 0.02
PT Filter EFF	7.97	7.2	5.5	33.9	107	0.02
M-INF	8.06	7.4	5.5	33.4	114	0.02

^a Salinity measured in the Brine Pit sample was 30.2 ppt, the value was double checked.

Statistical analyses were conducted using EPA flowchart specifications as outlined in the test guidance manual (USEPA 1995). Organism performance in each sample dilution series was compared to that observed in the control. For the purposes of this round of testing, the M-INF sample was used as control/dilution water for all other samples; Nautilus laboratory seawater collected from the Scripps Institution of Oceanography (SIO) inlet was used as control/dilution water only for the M-INF sample dilution series. Results were used to calculate the No Observed Effect Concentration (NOEC) and chronic toxic unit (TUc) values.

In addition to EPA flowchart statistical methods, the results were also analyzed using the **USEPA's Test of** Significant Toxicity (TST) approach specified in National Pollution Discharge Elimination System Test of Significant Toxicity Implementation Document (USEPA, 2010). Notably, the California State Water Resources Control Board (SWRCB) published a Draft Policy for Toxicity Assessment and Control (SWRCB, 2012), which includes the TST as an alternative method to evaluate toxicity data. This approach applies a modified t-test that takes into account both the statistical power of the test and the magnitude of biological effects in determining the presence of a response. For the M-001 effluent sample, the in-stream waste concentration (IWC) is 6.06 percent unadjusted effluent, and results are reported as "Pass" if a sample is considered non-toxic according to the TST calculation, or "Fail" if considered toxic according to the TST. As the TST statistical analysis is not in the 2006 CDP permit, the TST results are included for comparison purposes only.

Test ID: 1706-S203 to S209 Sample Collection Date: June 30, 2017

Table 3. Echinoderm Fertilization Chronic Bioassay Specifications

Test Period: 6/30/17, 17:50 through 18:30

Test Organism: Strongylocentrotus purpuratus (purple sea urchin)

Test Organism Source: Field-collected locally (off Point Loma in San Diego, CA)

Lab Control/Dilution Water: Natural seawater (source: Scripps Institution of Oceanography (SIO) inlet),

 34 ± 2 parts per thousand (ppt); 20- μ m filtered was used to prepare the M-INF dilution series. All other samples were diluted with the M-INF water

Client: IDE Americas, Inc.

itself.

Test Concentrations: 2.5, 5.0, 6.06, 10, and 15 percent unadjusted M-001 sample, lab control.

The same dilution series was also tested with the other samples and M-001 after adjustment to 40 ppt per request from Poseidon. This adjustment was performed to replicate sample adjustment allowable in the permit for acute testing to reflect maximum salinity concentrations in the effluent prior to discharge to the ocean (i.e., the maximum daily average salinity concentration limit for the combined Encina Power Station

Discharge (EPS) and CDP discharges).

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

Protocol Used: EPA/600/R-95/136, 1995 West Coast Marine Chronic

Test Type: Fertilization; 20-min sperm exposure to effluent followed by a 20-min egg

fertilization period

Acceptability Criteria: Mean fertilization ≥70% in the control, and percent minimum significant

difference (PMSD) value < 25%

Reference Toxicant Testing: Copper chloride

Statistical Analysis Software: CETIS™, version 1.8.7.20

RESULTS

There was a statistically significant decrease in fertilization rate observed in the 6.06, 10, and 15 percent concentrations of the unadjusted M-001 sample compared to the lab control, resulting in a NOEC of 5.0 percent effluent and a TU_c equal to 20. This exceeds the maximum daily permit effluent limitation of 16.5 TU_c . The 6.06 percent concentration (IWC) resulted in a 12 percent effect compared to the lab control, which was not significantly significant using to the TST calculation. The M-001 sample adjusted to 40 ppt prior to dilution preparation resulted in no statistically significant effects in any of the test concentrations and a TU_c less than 6.67.

Client: IDE Americas, Inc.

Sample Collection Date: June 30, 2017

The Brine Pit sample resulted in statistically significant decreases in fertilization rate for all but the 2.5 percent sample, resulting in a NOEC of 2.5 and a TU_c of 40. The percent effect in the 6.06 percent concentration was 17. The ERI Brine and Train 4 test resulted in a statistically significant decrease in fertilization rate in the 15 percent sample concentration compared to the lab control, resulting in a NOEC of 10 percent effluent and a TU_c equal to 10. The PT Filter and M-INF samples both resulted in no statistically significant effects for any of the concentrations tested, resulting in a NOEC of 15 percent and a TU_c less than 6.67. Two high salinity controls were tested for comparison purposes. One was run at 37.8 ppt with the unadjusted M-001 sample and the other at 39.0 ppt with the Train 4 sample. The high salinity controls for M-001 and Train 4 resulted in fertilization rates of 95.0 and 91.2, respectively. This indicates that decreases in fertilization rates observed in the samples were not likely attributable to elevated salinity.

Statistical results for urchin fertilization toxicity tests are summarized in Table 4, and detailed test results are summarized in Tables 5, 6, and 7. Raw test data and full statistical analyses can be found in Appendix A. Sample receipt information and a copy of the chain-of-custody form are in Appendices B and C, respectively.

Client: IDE Americas, Inc. Sample Collection Date: June 30, 2017

Table 4. Statistical Results for M-001 Purple Urchin Fertilization Testing

Sample ID	NOEC (% sample)	LOEC (% sample)	EC ₅₀ (% sample)	TUc value (toxic units)	TST Result (Pass/Fail)	Percent Effect at 6.06%
M-001 (unadjusted)	5.0	6.06	>15	20	Pass	12
M-001 (40 ppt adjusted)	15	>15	>15	<6.67	Pass	-4.2
ERI Brine	10	15	>15	10	Pass	0.22
Brine Pit	2.5	5.0	13.2	40	Pass	17
Train 4	10	15	>15	10	Pass	6.6
PT Filter EFF	15	>15	>15	<6.67	Pass	0.90
M-INF	15	>15	>15	<6.67	Pass	-0.64

NOEC = No Observed Effect Concentration

LOEC = Lowest Observed Effect Concentration

 EC_{50} = Concentration expected to cause an adverse effect to 50 percent of the test organisms

TU_c = Chronic Toxic Unit: 100÷NOEC

TST: Pass = sample is non-toxic at the 6.06% IWC according to the TST calculation; Fail = sample is toxic at the 6.06% IWC according to the TST calculation. The TST analysis is not in the existing CDP permit; TST analysis is included here for comparison purposes only.

Percent effect (PE) from control is calculated as: PE= ((mean response in control-mean response in the IWC)/mean response in control) *100. A negative PE results when organism performance in the sample is greater than that in the control.

Table 5. Detailed Results of Purple Urchin Fertilization Testing for the M-001 Sample

Test Concentration	M-001 L	Jnadjusted Sample	M-001 40 ppt Adjusted ^a		
(% Sample)	Salinity (ppt)			Mean Percent Fertilization	
Lab Control	34.0	88.0	33.9	86.0	
High Salinity Control	37.6	95.0			
2.5	34.5	92.6	34.2	93.2	
5.0	35.3	86.8	34.3	93.2	
6.06	35.5	77.4*	34.3	89.6	
10	36.5	66.6*	34.2	85.4	
15	37.8	68.0*	34.9	88.2	

^a For comparison to the M-001 unadjusted sample, the M-001 sample was adjusted with seawater to 40 ppt prior to preparing test concentrations.

^{*}An asterisk indicates a statistically significant decrease compared to the lab control using the standard USEPA flowchart statistical method (EPA 1995).

Table 6. Detailed Results of Purple Urchin Fertilization Testing for the ERI Brine and Brine Pit Samples

Test		ERI Brine	Brine Pit		
Concentration (% Sample)	Salinity (ppt)	Mean Percent Fertilization	Salinity (ppt)	Mean Percent Fertilization	
Lab Control	33.8	90.8	33.9	88.6	
2.5	34.8	91.8	33.9	88.4	
5.0	35.6	90.8	33.9	73.2*	
6.06	36.0	90.6	33.9	73.4*	
10	37.3	89.8	33.6	58.6*	
15	38.9	81.4*	33.5	36.4*	

^{*}An asterisk indicates a statistically significant decrease compared to the lab control using the standard USEPA flowchart statistical method (EPA 1995).

Table 7. Detailed Results of Purple Urchin Fertilization Testing for the Additional Facility Sample and Influent

Test	Т	Train 4		treatment ed Effluent	M-INF		
Concentration (% Sample)	Salinity (ppt)	Mean Percent Fertilization	Salinity (ppt)	Mean Percent Fertilization	Salinity (ppt)	Mean Percent Fertilization	
Lab Control	34.0	91.6	33.9	89.0	33.7	93.4	
High Salinity Control	39.0	91.2					
2.5	34.8	86.2	34.0	92.2	33.7	93.8	
5.0	35.7	86.2	33.9	89.8	33.8	92.4	
6.06	36.1	85.6	34.0	88.2	34.0	94.0	
10	37.2	86.4	34.1	87.2	33.9	95.0	
15	39.2	60.2*	34.0	90.4	33.9	93.8	

^{*}An asterisk indicates a statistically significant decrease compared to the lab control using the standard USEPA flowchart statistical method (EPA 1995).

QUALITY ASSURANCE

The samples were received the same day as collection and within the appropriate temperature range. All samples were tested within the allowable holding time of 36 hours. The laboratory controls met the minimum acceptability criteria as set by USEPA. The PMSD values, which are a measure of test variability, were within the acceptable range. Therefore, all test results were deemed valid for reporting purposes.

Client: IDE Americas, Inc.

Sample Collection Date: June 30, 2017

Statistical analyses followed USEPA flowchart selections and dose-response relationships were reviewed to evaluate reliability of the results. Additionally, appropriate alpha levels were used for statistical analyses according to the TST Implementation Document guidelines (USEPA 2010).

Results for the concurrent reference toxicant test used to monitor laboratory performance and test organism sensitivity are summarized in Table 8 and presented in full in Appendix D. The reference toxicant test met all test acceptability criteria. The median effect concentration (EC_{50} value) was within two standard deviations (SD) of the historical mean, indicating typical test organism sensitivity to copper. A list of qualifier codes used on bench datasheets can be found in Appendix E.

Table 8. Reference Toxicant Test Results

Test Species	Endpoint	EC ₅₀ (µg/L Copper)	Historical Mean EC ₅₀ ±2 SD (µg/L Copper)	CV (%)
Purple Urchin	Fertilization	39.4	45.1 ± 22.4	24.9

 EC_{50} = Concentration expected to cause an adverse effect to 50 percent of the test organisms Historical Mean $EC_{50} \pm 2$ SD = Mean of historical test results plus or minus two standard deviations CV = Coefficient of Variation

REFERENCES

California State Water Resources Control Board (SWRCB) 2012. Draft Policy for Toxicity Assessment and Control. June 2012. Sacramento, CA.

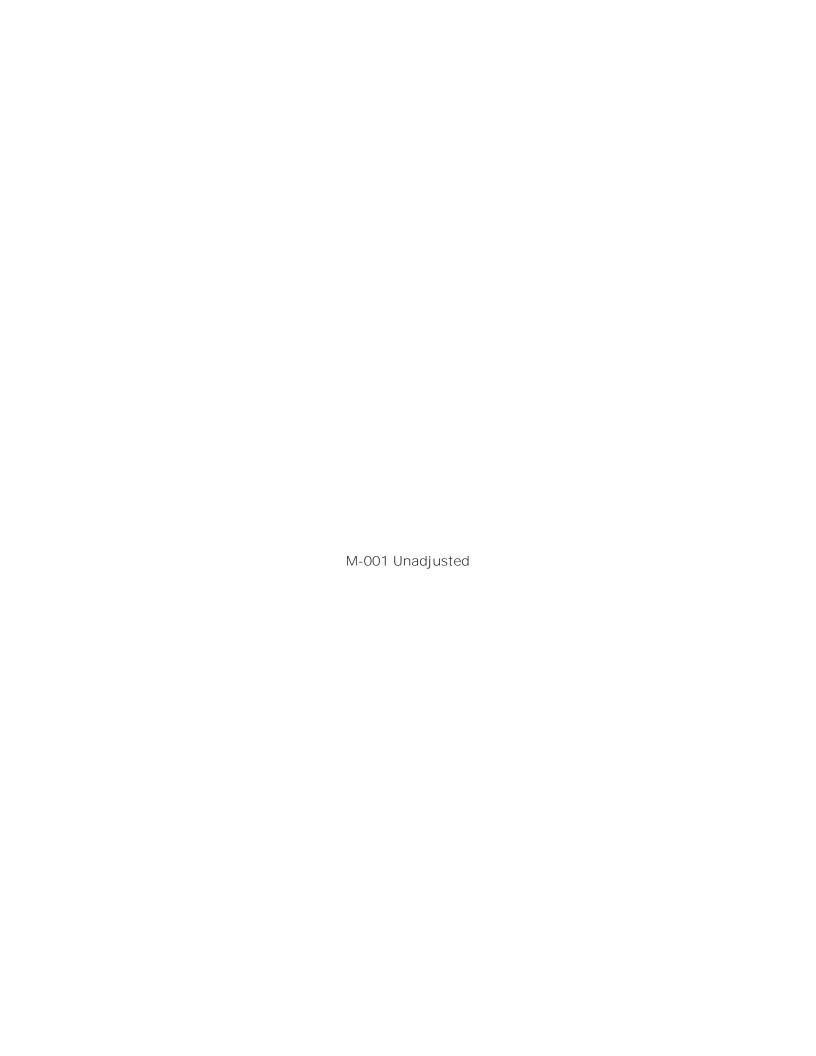
Client: IDE Americas, Inc.

Sample Collection Date: June 30, 2017

- 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 Granite Canyon.
- Tidepool Scientific Software. 2000-2013. **CETIS™ Com**prehensive Environmental Toxicity Information System Software, Version 1.8.7.20
- USEPA. 1991. Methods for Aquatic Toxicity Identification Evaluation Phase I Toxicity Characterization Procedures, 2nd Edition, EPA/600/6-91/003 February 1991.
- 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.
- USEPA. 2010. National Pollutant Discharge Elimination System Test of Significant Toxicity Implementation Document. EPA/833/R-10/003. June 2010.

Appendix A

Test Data and Statistical Analyses



CETIS Summary Report

Report Date:

07 Jul-17 15:28 (p 1 of 1)

Test Code:

1706-S203 | 12-4385-2719

Mary	Echinoid Spe	rm Cell Fertiliza	tion Test 1	5C		***************************************					Nautilu	Environm	nental (CA)
Project Pro	Start Date: Ending Date:	30 Jun-17 17:50 30 Jun-17 18:30	Pro Spe	tocol: cies:	EPA/600/R-95/ Strongylocentro	` '	tus		Brine:	. ivat	ural-Seawat		
Note	Sample Date: Receive Date:	30 Jun-17 08:00 30 Jun-17 11:48	Mat Sou	erial: rce:	Facility Effluent IDE Americas,	Inc.						Plant	
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Point Estimary Poi	Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	N	lethod			
Note	05-7230-3032	Fertilization Rat	е	5	6.06	5.505	8.86%	20	С	unnett N	/lultiple Com	parison Tes	it
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	Test Acceptal	oility											
14-6683-3143 Fertilization Raise	Analysis ID	Endpoint		Attribu	ute	Test Stat	TAC Limi	ts	C	verlap	Decision		
Fertilization Rate Summary PMSD 0.08862 NL - 0.25 No Passes Acceptability Criteria Fertilization Rate Summary C-% Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV% %Effect 0 High Salinity Co 5 0.95 0.952 0.9748 0.93 0.98 0.008944 0.02 2.11% 0.0% 0 Lab Control 5 0.88 0.8266 0.9343 0.83 0.93 0.01924 0.04201 4.89% 7.37% 2.5 5 0.868 0.8238 0.9122 0.81 0.9 0.01594 0.03564 4.11% 8.63% 6.06 0.674 0.8846 0.63 0.86 0.03982 0.08905 11.51% 18.53% 10 5 0.666 0.6042 0.7278 0.6 0.72 0.0498 7.48% 29.89% Fertilization Revisio	05-7230-3032	Fertilization Rat	е	Contro	l Resp	0.88	0.7 - NL		Y	es	Passes A	cceptability	Criteria
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5 0.9 0.88 0.86 0.81 0.89 6.06 0.76 0.63 0.79 0.83 0.86 10 0.72 0.68 0.7 0.6 0.63	0	Lab Control	0.86	0.83	0.92	0.86	0.93						
6.06 0.76 0.63 0.79 0.83 0.86 10 0.72 0.68 0.7 0.6 0.63	2.5		0.88	0.89	0.97	0.97	0.92						
6.06 0.76 0.63 0.79 0.83 0.86 10 0.72 0.68 0.7 0.6 0.63	5		0.9	0.88	0.86								
10 0.72 0.68 0.7 0.6 0.63	6.06												
	15		0.76	0.58	0.68	0.72	0.66						



Report Date:

07 Jul-17 15:28 (p 1 of 2)

Test Code:

1706-S203 | 12-4385-2719

							1031	Code:		0 0 2 0 0	2-4385-2719
Echinoid Sp	erm Cell Fertiliz	ation Test	15C						Nautilus	Environr	nental (CA)
Analysis ID:	05-7230-3032	Er	idpoint: Fer	tilization Ra	te		CET	IS Version:	CETISv1	.8.7	
Analyzed:	03 Jul-17 9:47	' Ar	ı alysis : Paı	ametric-Cor	trol vs Trea	tments	Offic	ial Results:	Yes		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C > T	NA	NA		8.86%	5	6.06	5.505	20
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(a:5%)		
Lab Control	2.5		-1.773	2.362	0.112 8	0.9984	CDF		ficant Effect		***************************************
	5		0.4291	2.362	0.112 8	0.6774	CDF		ficant Effect		
	6.06*		2.973	2.362	0.112 8	0.0136	CDF	Significant			
	10*		5.621	2.362	0.112 8	<0.0001	CDF	Significant			
	15*		5.288	2.362	0.112 8	<0.0001	CDF	Significant			
ANOVA Tabl	e							-			
Source	Sum Squ	uares	Mean Squ	ıare	DF	F Stat	P-Value	Decision(α:5%)		
Between	0.510281	9	0.1020564		5	18.21	<0.0001	Significant			
Error	0.134508	5	0.0056045	52	24						
Total	0.644790	4			29						
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision(α:1%)			
Variances	Bartlett B	Equality of V	/ariance	2.801	45.00				**		Carrie Company And Miles Are Company
Variances	• •		2.001	15.09	0.7307	Equal Var	iances				
Distribution				0.9675	0.9031	0.7307 0.4737	Equal Var Normal Di				
Distribution											
Distribution	Shapiro-								Std Err	CV%	%Effect
Distribution Fertilization	Shapiro- Rate Summary	Wilk W No	mality	0.9675	0.9031	0.4737	Normal Di	stribution	Std Err 0.01924	CV% 4.89%	%Effect
Distribution Fertilization C-%	Shapiro- Rate Summary Control Type	Wilk W No	mality Mean	0.9675 95% LCL	0.9031 95% UCL	0.4737 Median	Normal Di	stribution Max			
Distribution Fertilization C-% 0	Shapiro- Rate Summary Control Type	Wilk W Nor	Mean 0.88	0.9675 95% LCL 0.8266	0.9031 95% UCL 0.9334	0.4737 Median 0.86	Min 0.83	Max 0.93	0.01924	4.89%	0.0%
Distribution Fertilization C-% 0 2.5	Shapiro- Rate Summary Control Type	Count 5 5	Mean 0.88 0.926	0.9675 95% LCL 0.8266 0.8729	0.9031 95% UCL 0.9334 0.9791	0.4737 Median 0.86 0.92	Min 0.83 0.88	Max 0.93 0.97	0.01924 0.01913	4.89% 4.62%	0.0% -5.23% 1.36%
Fertilization C-% 0 2.5 5	Shapiro- Rate Summary Control Type	Count 5 5 5	Mean 0.88 0.926 0.868	0.9675 95% LCL 0.8266 0.8729 0.8238	0.9031 95% UCL 0.9334 0.9791 0.9122	0.4737 Median 0.86 0.92 0.88	Min 0.83 0.88 0.81	Max 0.93 0.97 0.9	0.01924 0.01913 0.01594	4.89% 4.62% 4.11%	0.0% -5.23%
Fertilization C-% 0 2.5 5 6.06	Shapiro- Rate Summary Control Type	Count 5 5 5 5	Mean 0.88 0.926 0.868 0.774	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634	95% UCL 0.9334 0.9791 0.9122 0.8846	0.4737 Median 0.86 0.92 0.88 0.79	Min 0.83 0.88 0.81 0.63	Max 0.93 0.97 0.9 0.86	0.01924 0.01913 0.01594 0.03982	4.89% 4.62% 4.11% 11.51%	0.0% -5.23% 1.36% 12.05%
Fertilization C-% 0 2.5 5 6.06 10 15	Shapiro- Rate Summary Control Type	Count 5 5 5 5 5	Mean 0.88 0.926 0.868 0.774 0.666 0.68	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634 0.6042	95% UCL 0.9334 0.9791 0.9122 0.8846 0.7278	0.4737 Median 0.86 0.92 0.88 0.79 0.68	Min 0.83 0.88 0.81 0.63 0.6	Max 0.93 0.97 0.9 0.86 0.72	0.01924 0.01913 0.01594 0.03982 0.02227	4.89% 4.62% 4.11% 11.51% 7.48%	0.0% -5.23% 1.36% 12.05% 24.32%
Fertilization C-% 0 2.5 5 6.06 10 15	Shapiro- Rate Summary Control Type Lab Control	Count 5 5 5 5 5	Mean 0.88 0.926 0.868 0.774 0.666 0.68	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634 0.6042	95% UCL 0.9334 0.9791 0.9122 0.8846 0.7278	0.4737 Median 0.86 0.92 0.88 0.79 0.68	Min 0.83 0.88 0.81 0.63 0.6	Max 0.93 0.97 0.9 0.86 0.72	0.01924 0.01913 0.01594 0.03982 0.02227	4.89% 4.62% 4.11% 11.51% 7.48%	0.0% -5.23% 1.36% 12.05% 24.32%
Pistribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Shapiro- Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 7 med Sumr	Mean 0.88 0.926 0.868 0.774 0.666 0.68	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634 0.6042 0.5958	0.9031 95% UCL 0.9334 0.9791 0.9122 0.8846 0.7278 0.7642	0.4737 Median 0.86 0.92 0.88 0.79 0.68 0.68	Min 0.83 0.88 0.81 0.63 0.6 0.58	Max 0.93 0.97 0.9 0.86 0.72 0.76	0.01924 0.01913 0.01594 0.03982 0.02227 0.03033	4.89% 4.62% 4.11% 11.51% 7.48% 9.97%	0.0% -5.23% 1.36% 12.05% 24.32% 22.73%
Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 5 cmed Sumr	Mean 0.88 0.926 0.868 0.774 0.666 0.68 mary Mean	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634 0.6042 0.5958	0.9031 95% UCL 0.9334 0.9791 0.9122 0.8846 0.7278 0.7642 95% UCL	0.4737 Median 0.86 0.92 0.88 0.79 0.68 0.68	Min 0.83 0.88 0.81 0.63 0.6 0.58	Max 0.93 0.97 0.9 0.86 0.72 0.76	0.01924 0.01913 0.01594 0.03982 0.02227 0.03033	4.89% 4.62% 4.11% 11.51% 7.48% 9.97%	0.0% -5.23% 1.36% 12.05% 24.32% 22.73%
Pistribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 cmed Sumr Count 5	Mean 0.88 0.926 0.868 0.774 0.666 0.68 mary Mean 1.221	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634 0.6042 0.5958 95% LCL 1.137	95% UCL 0.9334 0.9791 0.9122 0.8846 0.7278 0.7642 95% UCL 1.306	0.4737 Median 0.86 0.92 0.88 0.79 0.68 0.68 Median 1.187	Min 0.83 0.88 0.81 0.63 0.6 0.58	Max 0.93 0.97 0.9 0.86 0.72 0.76 Max 1.303	0.01924 0.01913 0.01594 0.03982 0.02227 0.03033 Std Err 0.03052	4.89% 4.62% 4.11% 11.51% 7.48% 9.97% CV% 5.59%	0.0% -5.23% 1.36% 12.05% 24.32% 22.73% Effect 0.0%
Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 Count Count Count 5 5 5 5 5 5 5 5 5 5 6 6 6 6 7 6 7 7 8 7 8 8 7 8 8 8 8 8 8 8	Mean 0.88 0.926 0.868 0.774 0.666 0.68 mary Mean 1.221 1.305	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634 0.6042 0.5958 95% LCL 1.137 1.198	95% UCL 0.9334 0.9791 0.9122 0.8846 0.7278 0.7642 95% UCL 1.306 1.413	0.4737 Median 0.86 0.92 0.88 0.79 0.68 0.68 Median 1.187 1.284	Min 0.83 0.88 0.81 0.63 0.6 0.58 Min 1.146 1.217	Max 0.93 0.97 0.9 0.86 0.72 0.76 Max 1.303 1.397	0.01924 0.01913 0.01594 0.03982 0.02227 0.03033 Std Err 0.03052 0.03887	4.89% 4.62% 4.11% 11.51% 7.48% 9.97% CV% 5.59% 6.66%	0.0% -5.23% 1.36% 12.05% 24.32% 22.73% %Effect 0.0% -6.87%
Pistribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5 5	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 Creed Sum Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.88 0.926 0.868 0.774 0.666 0.68 mary Mean 1.221 1.305 1.201	0.9675 95% LCL 0.8266 0.8729 0.8238 0.6634 0.6042 0.5958 95% LCL 1.137 1.198 1.138	95% UCL 0.9334 0.9791 0.9122 0.8846 0.7278 0.7642 95% UCL 1.306 1.413 1.264	0.4737 Median 0.86 0.92 0.88 0.79 0.68 0.68 Median 1.187 1.284 1.217	Min 0.83 0.88 0.81 0.63 0.6 0.58 Min 1.146 1.217 1.12	Max 0.93 0.97 0.9 0.86 0.72 0.76 Max 1.303 1.397 1.249	0.01924 0.01913 0.01594 0.03982 0.02227 0.03033 Std Err 0.03052 0.03887 0.02276	4.89% 4.62% 4.11% 11.51% 7.48% 9.97% CV% 5.59% 6.66% 4.24%	0.0% -5.23% 1.36% 12.05% 24.32% 22.73% %Effect 0.0% -6.87% 1.66%

Analyst: AC QA: P7/11/17

Report Date: Test Code: 07 Jul-17 15:28 (p 2 of 2) 1706-S203 | 12-4385-2719

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 05-7230-3032 Endpoint: Fertilization Rate CETISv1.8.7 **CETIS Version:** Analyzed: 03 Jul-17 9:47 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 1.0 0.15 0.9 0.10 Fertilization Rate 0.05 0.7 0.6 0.00 0.5 -0,05 0.4 -0.10 0.2 0.1 0.0 0 LC 2.5 6.06 10 15 -2.5 -1.5 -1.0 -2.0 -0.5 0.0 1.0 1.5 2.0 C-% Rankits

Report Date:

07 Jul-17 15:28 (p 1 of 1)

Test Code:

1706-S203 | 12-4385-2719

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: 14-6683-3143 Analyzed: 03 Jul-17 9:47

Linear

Endpoint: Fertilization Rate

1000

Analysis: Linear Interpolation (ICPIN)

CETIS Version: Official Results: Yes

Two-Point Interpolation

CETISv1.8.7

Linear Interpol	ation Options					
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method	

Yes

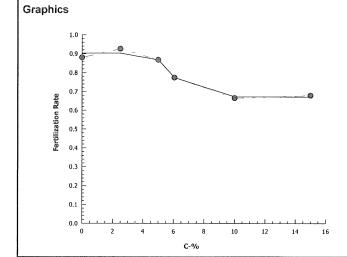
Point	Estimates
r Oilli	ESUIIIales

Linear

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
EC25	9.834	7.75	N/A	10.17	NA	12.9
EC50	>15	N/A	N/A	<6.667	NA	NA

753277

Fertiliza	tion Rate Summary				Cal	culated Varia	ite(A/B)				
C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.88	0.83	0.93	0.01924	0.04301	4.89%	0.0%	440	500
2.5		5	0.926	0.88	0.97	0.01913	0.04278	4.62%	-5.23%	463	500
5		5	0.868	0.81	0.9	0.01594	0.03564	4.11%	1.36%	434	500
6.06		5	0.774	0.63	0.86	0.03982	0.08905	11.51%	12.05%	387	500
10		5	0.666	0.6	0.72	0.02227	0.0498	7.48%	24.32%	333	500
15		5	0.68	0.58	0.76	0.03033	0.06782	9.97%	22.73%	340	500



Report Date:

07 Jul-17 15:28 (p 1 of 1)

CLIIS AII	aiyiicai Kep	JOIL					•	t Code:		06-S203 12	
Echinoid Sp	erm Cell Fertili:	zation Te	est 15C		TST					s Environr	
Analysis ID:	12-0184-579 ²		Endpoint: Fe	ertilization Ra	te		CET	'IS Version	n: CETISv	1.8.7	
Analyzed:	07 Jul-17 15:	28	Analysis: Pa	arametric Bio	equivalence	-Two Samp	le Offi	cial Result	s: Yes		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	rected)	NA	C*b < T	NA	NA	0.75	5.65%	6.06	10	7.785	16.5
TST-Welch's	t Test										
Control	vs C-%		Test Sta	t Critical	MSD DF	P-Value	P-Type	Decisio	n(α:5%)		
Lab Control	2.5*		8.631	1.943	0.088 6	<0.0001	CDF	Non-Sigi	nificant Effec	t	
	5*		8.831	1.895	0.061 7	<0.0001	CDF	Non-Sigr	nificant Effec	t	
	6.06*		3.18	2.015	0.104 5	0.0123	CDF	Non-Sigr	nificant Effec	t	
	10		1.195	1.895	0.062 7	0.1355	CDF	Significa			
	15		1.384	1.895	0.075 7	0.1044	CDF	Significa	nt Effect		
ANOVA Tabl	е										
Source	Sum Sq	uares	Mean So	luare	DF	F Stat	P-Value	Decision	n(α:5%)		
Between	0.51028	19	0.102056	64	5	18 21	<0.0001	Significa	nt Effect		***************************************
Error	0.13450	85	0.005604	52	24			-			
Total	0.64479	04			29	_					
Distributiona	al Tests										10x110x10x10x10x10x10x10x10x10x10x10x10x
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Bartlett	Equality	of Variance	2.801	15.09	0.7307	Equal Va	riances		***************************************	***************************************
Distribution	Shapiro	-Wilk W I	Normality	0.9675	0.9031	0.4737	Normal D	istribution			
Fertilization	Rate Summary										
C-%	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.88	0.8266	0.9334	0.86	0.83	0.93	0.01924	4.89%	0.0%
2.5		5	0.926	0.8729	0.9791	0.92	0.88	0.97	0.01913	4.62%	-5.23%
5		5	0.868	0.8238	0.9122	0.88	0.81	0.9	0.01594	4.11%	1.36%
6.06		5	0.774	0.6634	0.8846	0.79	0.63	0.86	0.03982	11.51%	12.05%
10		5	0.666	0.6042	0.7278	0.68	0.6	0.72	0.02227	7.48%	24.32%
15 		5	0.68	0.5958	0.7642	0.68	0.58	0.76	0.03033	9.97%	22.73%
Angular (Cor	rected) Transfo	rmed Su	ımmary								Mining the Annual Property Control of the Annual Property of the Ann
C-%	Control Type	Coun	t Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.221	1.137	1.306	1.187	1.146	1.303	0.03052	5.59%	0.0%
2.5		5	1.305	1.198	1.413	1.284	1.217	1.397	0.03887	6.66%	-6.87%
5		5	1.201	1.138	1.264	1.217	1.12	1.249	0.02276	4.24%	1.66%
6.06		5	1.081	0.9518	1.21	1.095	0.9169	1.187	0.04643	9.61%	11.52%
10		E	0.0554	0.0000	4.004	0.0005	0.0004	4.040	0.00050	E E00/	04.7004

Analyst: AC QA: VEP7/11/17

10

15

5

5

0.9554

0.9711

0.8899

0.8809

1.021

1.061

0.9695

0.9695

0.8861

0.8657

1.013

1.059

0.02356

0.03247

5.52%

7.48%

21.79%

20.5%

CETIS Test Data Worksheet

Report Date:

29 Jun-17 18:51 (p 1 of 1)

Test Code: 1706-5-224-12-4385-2719/4A23AFAF

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: 30 Jun-17 End Date: Sample Date: 30 Jun-17

30 Jun-17

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

Material: Facility Effluent

Sample Code: 17- 0738 Sample Source: IDE Americas, Inc. Sample Station: M-001 Unadjusted

C-%	Code	Rep	Pos	# Counted		(A) Notes
			31	100	94	ALS / 7/2/17
			32	100	89	
			33	100	81	
			34	100	®9768	
			35	100	97	
			36	100	36 76 83 96	
			37	100	76	
			38	100	83	
			39	100	96	
			40	100	38 49	
			41	100	68	
			42	100	68 72 86	
			43 44	100	96	
			45	100	88	
			46	100	76	
			47	100	98	
			48	100	92 66	
			49	100		
			50	100	79	
			51	100	86 92	
			52	100	63	
			53	100	90	
			54	100	97	
			55	100	86	
			56	100	83	
			57	100	89	
			58	100	88	
			59	100	70	
			60	100	72	
			61	100	58 93	
			62	100	93	
			63	100	93	
			64	100	60 63	
			65	100	63	

@ 618 7/2/17

(B)018 x-1/1/11

Report Date:

29 Jun-17 18:51 (p 1 of 1)

Test Code: 1706-S 20312-4385-2719/4A23AFAF

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date:	30 Jun-17	Species:	Strongylocentrotus purpuratus	Sample Code:	17-0738
End Date:	30 Jun-17	Protocol:	EPA/600/R-95/136 (1995)	Sample Source:	IDE Americas, Inc.
Sample Date:	30 Jun-17	Material:	Facility Effluent	Sample Station:	M-001 Unadjusted

C-%	Code	Rep	Pos		# Fertilized	Notes
0	HS	1	31	100	99	A0 7/1/17
0	HS	2	63	100		
0	HS	3	46			
0	HS	4	40			
0	HS	5	39			
0	LC	1	50	COI	91	
0	LC	2	38	100		
0	LC	3	51			
0	LC	4	55			
0	LC	5	62			
2.5		1	44			
2.5		2	57			
2.5		3	54			
2.5		4	35			
2.5		5	47			
5		1	53			
5		2	58			
5		3	36			
5		4	33			
. 5		5	32			
6.06		1	45	109	68	
6.06		2	65		9 0	
6.06		3	49			
6.06		4	56			
6,06		5	43			
10		1	42			
10		2	34			
10		3	59			
10		4	64			
10		5	52			·
15		1	37			
15		2	61			
15		3	41			
15		4	60			
15		5	48	100	63	

QC:CG

Water Quality Measurements

A 1	*		
7 -	ie	nt	•

IDE

Test Species: S. purpuratus

Sample ID:

M-001 (unadjusted)

Sample Log No.:

17-0738

End Date/Time: 6/30/2017 \

Dilutions made by:

DO)

Test No: _____1706-S203

			Analyst:	CH
			eadings	
Concentration	DO	рН	Salinity	Temperature
%	(mg/L)	(units)	(ppt)	(°C)
Lab Control	8.0	7.85	34.0	16.0
High Salinity Control	9.5	7.97	37.6	14.8
2.5	7.9	7.86	34.5	15.9
5.0	7.9	7.86	35.3	15.9
6.06	7.9	7.86	35.5	15.9
10	7.9	7.86	36.5	15.8
15	7.9	7.85	37.8	15.7

_				
\sim	om	m	010	60.
U				

DINKONS made W/M-INF

QC Check:

AC7/7/17

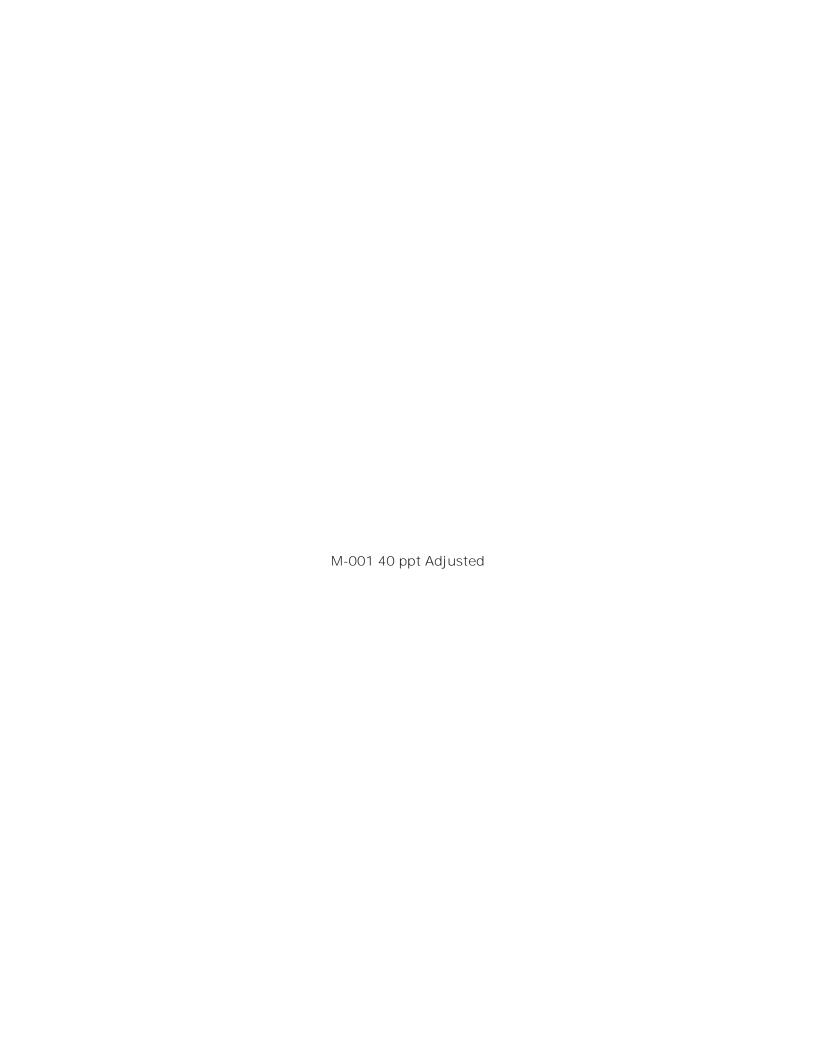
Final Review: MP 7 1117

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.:	1DE M-101 Unad 1706-S203	ju Steal		Start Date/Time End Date/Time Species		11830
Tech initials: Injection Time:	1650			Animal Source Date Collected		
Sperm Absorbance at	400 nm: <u>1, 009</u>	(target range of 0.8 -	1.0 for density of	4x10 ⁶ sperm/ml)	
Eggs Counted:	R8 Mea 16 17 (target Rafter	Dia ()	ertical pass on Sedo	ggs/ml	(Ō.
Initial density: Final density:	4000 eggs/ml	=dilution to =dilution to	stock	ig stock	ml	
Prepare the embryo sto existing stock (1 part) a	ock according to the calculand 125 ml of dilution wate	lated dilution factor. For r (1.25 parts).	example, if the di	lution factor is 2	25, use 100	O ml of
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1 1600 50 40 0.0 10	<u>1200:1</u> <u>800</u> 30	0 10	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 1710 1723 1733	Rangefinder Ratio:	Fert. Un: 34,83 14	fert.		
this range, choose the	m-to-egg ratio that results e ratio closest to 90 perc of reproductive season, si	cent unless professiona	30 and 90 percent. al judgment dictat	If more than or ses consideration	ne concentr on of other	ation is within factors (e.g.,
Definitive Test		Sperm:Egg Ratio Use	d: <u>1521</u>			
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time (B) 17450 1810 1830	QC1 QC2 Egg Control 1 Egg Control 2	Fert. Unf 97 3 94 0	ert.		
Comments:	Phodiuko Pho OS 4	70001V0	20		·	
QC Check:	AC 7/5/17			Final Review:	WP7/	11/17



CETIS Summary Report

Report Date: Test Code: 07 Jul-17 15:44 (p 1 of 1) 1706-S204 | 08-6882-6222

0 Lab Control 5 0.86 0.8111 0.9089 0.83 0.92 0.01761 0.03937 4.58% 0.0% 2.5 5 0.932 0.8769 0.9871 0.86 0.97 0.01985 0.04438 4.76% -8.37 5 0.932 0.8895 0.9745 0.88 0.97 0.0153 0.03421 3.67% -8.37 6.06 5 0.896 0.849 0.943 0.84 0.94 0.01691 0.03782 4.22% -4.19 10 5 0.854 0.7947 0.9133 0.8 0.93 0.02135 0.04775 5.59% 0.7%									Test Code:		170	6-S204 0	8-6882-6222
Start Date 30 Jun-17 17:15 Species Sp	Echinoid Spe	rm Cell Fertiliza	tion Test 1	15C							Nautilus	s Environr	nental (CA)
Sample Date 30 Jun-17 01:00 Material Source 10 E Americas Inc. Sample Age 10 (6.5 °c) Station Note Station Note Note	Start Date: Ending Date:	30 Jun-17 17:50 30 Jun-17 18:30	Pro Spe	otocol: ecies:	EPA/600/R-95 Strongylocentr	, ,	tus		Diluent: @ Brine:			er M-IN	٦
Analysis ID Endpoint NOEL LOEL TOEL PMSD TU Method 14-4818-0451 Fertilization Rate 15 >15 NA 9.08% € 6.667 Dunnett Multiple Comparison Test Point Estimates Summary Analysis ID Endpoint Level % 95% LCL 95% LCL 7U Method Test Sate Summary Analysis ID Endpoint EC25 >15 N/A N/A N/A 46.667 Linear Interpolation (ICPIN) Test Sate Summary Analysis ID Endpoint Attribute Test Stat TAC Limits Overlap Decision Test Stat Sate Summary Fertilization Rate Control Resp 0.86 0.7 - NL Yes Passes Acceptability Criteria Fertilization Rate Control Resp 0.86 0.7 - NL Yes Passes Acceptability Criteria Fertilization Rate Control Resp 0.86 0.7 - NL Yes Passes Acceptability Criteria <tr< td=""><td>Sample Date: Receive Date:</td><td>30 Jun-17 08:00 30 Jun-17 11:48</td><td>Ma So</td><td>terial: urce:</td><td>Facility Effluen</td><td></td><td></td><td></td><td></td><td></td><td>sbad Desal</td><td>Plant</td><td></td></tr<>	Sample Date: Receive Date:	30 Jun-17 08:00 30 Jun-17 11:48	Ma So	terial: urce:	Facility Effluen						sbad Desal	Plant	
Point Estimate Point Po	Comparison S	Summary											
Point Estimate Summary	Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Meti	nod			
Analysis ID	14-4818-0451	Fertilization Rat	е	15	>15	NA	9.08% «	6.66	7 Duni	nett M	lultiple Com	parison Te	st
13-2565-5640 Fertilization Rate	Point Estimate	e Summary											
Fest Acceptability	Analysis ID	Endpoint		Level	%	95% LCL	95% UCL	TU	Meth	nod			
Analysis ID Endpoint Attribute Test Stat TAC Limits Overlap Decision 13-2565-5640 Fertilization Rate Control Responses 0.86 0.7 - NL Yes Passes Acceptability Criteria 14-4818-0451 Fertilization Rate PMSD 0.09078 NL - 0.25 No Passes Acceptability Criteria Fertilization Rate Summary C-% Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV% %Eff 0 Lab Control 5 0.86 0.81111 0.9089 0.83 0.92 0.01761 0.03937 4.58% 0.0% 2.5 5 0.932 0.8769 0.9871 0.86 0.97 0.01685 0.04438 4.76% -8.37 6.06 - 5 0.932 0.8895 0.9745 0.88 0.97 0.0163 0.04438 4.76% -8.37 6.06 - 5 0.896 0.849	13-2565-5640	Fertilization Rat	е							ar Inte	erpolation (I	CPIN)	
13-2565-5640 Fertilization Rate	Test Acceptab	ility											N. N
14-4818-0451 14-4818-0451 Fertilization Rate Summary Control Respondents 0.86 0.7 - NL 0.09078 Yes Passes Acceptability Criterian Publish Publish Criterian Publish Criterian Publish Criterian Publish Criterian Publish Criterian Publish Criterian Publish Publ	Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Lim	its	Ove	rlap	Decision		
14-4818-0451 Fertilization Rate Possibility Control Responder Possibility Criteria Possibility Criteri	13-2565-5640	Fertilization Rat	е	Contr	ol Resp	0.86	0.7 - NL	***************************************	Yes	-	Passes A	cceptability	Criteria
Partilization Rate Summary State State	14-4818-0451	Fertilization Rat	е	Contr	ol Resp	0.86	0.7 - NL		Yes		Passes A	cceptability	/ Criteria
C-% Control Type Count Mean 95% LCL 95% UCL Min Max Std Err Std Dev CV% %Eff 0 Lab Control 5 0.86 0.8111 0.9089 0.83 0.92 0.01761 0.03937 4.58% 0.0% 2.5 5 0.932 0.8769 0.9871 0.86 0.97 0.01985 0.04438 4.76% -8.37 5 0.932 0.8895 0.9745 0.88 0.97 0.0153 0.03421 3.67% -8.37 6.06 - 5 0.896 0.849 0.943 0.84 0.94 0.01691 0.03782 4.22% -4.19 10 5 0.854 0.7947 0.9133 0.8 0.93 0.02135 0.04775 5.59% 0.7% 15 5 0.882 0.8191 0.944 0.8 0.93 0.02267 0.0507 5.75% -2.56 C-% Control Type Rep 1 Rep	14-4818-0451	Fertilization Rat	е	PMSE)	0.09078	NL - 0.25		No		Passes A	cceptability	/ Criteria
0 Lab Control 5 0.86 0.8111 0.9089 0.83 0.92 0.01761 0.03937 4.58% 0.0% 2.5 5 0.932 0.8769 0.9871 0.86 0.97 0.01985 0.04438 4.76% -8.37 5 0.932 0.8895 0.9745 0.88 0.97 0.0153 0.03421 3.67% -8.37 6.06 5 0.896 0.849 0.943 0.84 0.94 0.01691 0.03782 4.22% -4.19 10 5 0.854 0.7947 0.9133 0.8 0.93 0.02135 0.04775 5.59% 0.7% 15 5 0.882 0.8191 0.9449 0.8 0.93 0.02135 0.04775 5.59% 0.7% 15 5 0.882 0.8191 0.9449 0.8 0.93 0.02267 0.0507 5.75% -2.56 Fertilization Rate Detail C-% Control Type Rep	Fertilization R	ate Summary											
2.5	C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std	Err	Std Dev	CV%	%Effect
5 0.932 0.8895 0.9745 0.88 0.97 0.0153 0.03421 3.67% -8.37 6.06 5 0.896 0.849 0.943 0.84 0.94 0.01691 0.03782 4.22% -4.19 10 5 0.854 0.7947 0.9133 0.8 0.93 0.02135 0.04775 5.59% 0.7% 15 5 0.882 0.8191 0.9449 0.8 0.93 0.02267 0.0507 5.75% -2.56 Fertilization Rate Detail C-% Control Type Rep 1 Rep 2 Rep 3 Rep 4 Rep 5 0 Lab Control 0.83 0.88 0.92 0.83 0.84 2.5 0.92 0.86 0.96 0.97 0.95 5 0.88 0.92 0.95 0.94 0.97 6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93	0	Lab Control	5	0.86	0.8111	0.9089	0.83	0.92	0.01	761	0.03937	4.58%	0.0%
6.06			5	0.932	0.8769	0.9871	0.86	0.97	0.01	985	0.04438	4.76%	-8.37%
10						0.9745	0.88	0.97	0.01	53	0.03421	3.67%	-8.37%
15 5 0.882 0.8191 0.9449 0.8 0.93 0.02267 0.0507 5.75% -2.56 Fertilization Rate Detail C-% Control Type Rep 1 Rep 2 Rep 3 Rep 4 Rep 5 0 Lab Control 0.83 0.88 0.92 0.83 0.84 2.5 0.92 0.86 0.96 0.97 0.95 5 0.88 0.92 0.95 0.94 0.97 6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93					_								-4.19%
Fertilization Rate Detail C-% Control Type Rep 1 Rep 2 Rep 3 Rep 4 Rep 5 0 Lab Control 0.83 0.88 0.92 0.83 0.84 2.5 0.92 0.86 0.96 0.97 0.95 5 0.88 0.92 0.95 0.94 0.97 6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93													0.7%
C-% Control Type Rep 1 Rep 2 Rep 3 Rep 4 Rep 5 0 Lab Control 0.83 0.88 0.92 0.83 0.84 2.5 0.92 0.86 0.96 0.97 0.95 5 0.88 0.92 0.95 0.94 0.97 6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93	15		5	0.882	0.8191	0.9449	0.8	0.93	0.02	267	0.0507	5.75% 	-2.56%
0 Lab Control 0.83 0.88 0.92 0.83 0.84 2.5 0.92 0.86 0.96 0.97 0.95 5 0.88 0.92 0.95 0.94 0.97 6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93			_										
2.5 0.92 0.86 0.96 0.97 0.95 5 0.88 0.92 0.95 0.94 0.97 6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93													
5 0.88 0.92 0.95 0.94 0.97 6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93	_	Lab Control											
6.06 0.94 0.88 0.91 0.84 0.91 10 0.86 0.84 0.84 0.8 0.93													
10 0.86 0.84 0.84 0.8 0.93													
• • • • • • • • • • • • • • • • • • • •													
15 0.91 0.9 0.93 0.87 0.8													
	15		0.91	0.9	0.93	0.87	0.8						

@ @18AC7/10/17

Analyst: AC QA: MR 1 11

CETIS™ v1.8.7.20

Report Date:

07 Jul-17 15:43 (p 1 of 2) 2-6222

Test Code:	1706-S204	

							lest				8-6882-622
Echinoid Sp	erm Cell Fertiliza	ation Test 1	15C						Nautilu	s Environ	mental (CA)
Analysis ID: Analyzed:	14-4818-0451 07 Jul-17 15:4		dpoint: Fer alysis: Par	tilization Rai		tments		IS Version:		.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cori	ected)	NA	C > T	NA	NA		9.08%	15	>15	NA	6.667
Dunnett Mul	tiple Compariso	n Test						The second second			
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(a:5%)		
Lab Control	2.5	N	-2.825	2.362	0.106 8	1.0000	CDF		ificant Effec	+	
	5		-2.752	2.362	0.106 8	0.9999	CDF		ificant Effect		
	6.06		-1.253	2.362	0.106 8	0.9920	CDF	•	ificant Effect		
	10		0.161	2.362	0.106 8	0.7814	CDF	_	ificant Effect		
	15		-0.785	2.362	0.106 8	0.9702	CDF	_	ificant Effect		
		STATE OF THE PARTY	0.700	2.002	0.100 3	0.8702	CDF	Non-aign	mcant Enec		
ANOVA Tabl											
Source	Sum Squ	The state of the s	Mean Squ		DF	F Stat	P-Value	Decision			
Between	0.084808		0.0169617	9	5	3.402	0.0183	Significan	t Effect		
Error	0.119664		0.004986		24						
Total	0.204473				29			Who records the second continuous control		and a man on the same property of the same and	
Distributiona	ıl Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision(α:1%)			
Variances	Bartlett E	Equality of V	ariance	0.5809	15.09	0.9889	Equal Var	iances			
Distribution	Shapiro-\	Wilk W Norr	mality	0.974	0.9031	0.6538	Normal Di	stribution			
Fertilization	Rate Summary										
	reace Gainmany										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
C- %	-	Count 5	Mean 0.86	95% LCL 0.8111	95% UCL 0.9089	Median 0.84	Min 0.83	Max 0.92	Std Err 0.01761	CV% 4.58%	%Effect 0.0%
	Control Type										
0	Control Type	5	0.86	0.8111	0.9089	0.84	0.83	0.92	0.01761	4.58% 4.76%	0.0%
0 2.5	Control Type	5 5	0.86 0.932	0.8111 0.8769	0.9089 0.9871	0.84 0.95	0.83 0.86	0.92 0.97	0.01761 0.01985 0.0153	4.58% 4.76% 3.67%	0.0% -8.37% -8.37%
0 2.5 5	Control Type	5 5 5	0.86 0.932 0.932	0.8111 0.8769 0.8895	0.9089 0.9871 0.9745	0.84 0.95 0.94	0.83 0.86 0.88	0.92 0.97 0.97	0.01761 0.01985 0.0153 0.01691	4.58% 4.76% 3.67% 4.22%	0.0% -8.37% -8.37% -4.19%
0 2.5 5 6.06	Control Type	5 5 5 5	0.86 0.932 0.932 0.896	0.8111 0.8769 0.8895 0.849	0.9089 0.9871 0.9745 0.943	0.84 0.95 0.94 0.91	0.83 0.86 0.88 0.84	0.92 0.97 0.97 0.94	0.01761 0.01985 0.0153	4.58% 4.76% 3.67%	0.0% -8.37% -8.37%
0 2.5 5 6.06 10 15	Control Type	5 5 5 5 5 5	0.86 0.932 0.932 0.896 0.854 0.882	0.8111 0.8769 0.8895 0.849 0.7947	0.9089 0.9871 0.9745 0.943 0.9133	0.84 0.95 0.94 0.91 0.84	0.83 0.86 0.88 0.84 0.8	0.92 0.97 0.97 0.94 0.93	0.01761 0.01985 0.0153 0.01691 0.02135	4.58% 4.76% 3.67% 4.22% 5.59%	0.0% -8.37% -8.37% -4.19% 0.7%
0 2.5 5 6.06 10 15	Control Type Lab Control	5 5 5 5 5 5	0.86 0.932 0.932 0.896 0.854 0.882	0.8111 0.8769 0.8895 0.849 0.7947	0.9089 0.9871 0.9745 0.943 0.9133	0.84 0.95 0.94 0.91 0.84	0.83 0.86 0.88 0.84 0.8	0.92 0.97 0.97 0.94 0.93	0.01761 0.01985 0.0153 0.01691 0.02135	4.58% 4.76% 3.67% 4.22% 5.59%	0.0% -8.37% -8.37% -4.19% 0.7%
0 2.5 5 6.06 10 15 Angular (Cor	Control Type Lab Control rected) Transfor	5 5 5 5 5 5 5	0.86 0.932 0.932 0.896 0.854 0.882	0.8111 0.8769 0.8895 0.849 0.7947 0.8191	0.9089 0.9871 0.9745 0.943 0.9133 0.9449	0.84 0.95 0.94 0.91 0.84 0.9	0.83 0.86 0.88 0.84 0.8	0.92 0.97 0.97 0.94 0.93 0.93	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267	4.58% 4.76% 3.67% 4.22% 5.59% 5.75%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56%
0 2.5 5 6.06 10 15 Angular (Cor C-%	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 7med Summ	0.86 0.932 0.932 0.896 0.854 0.882	0.8111 0.8769 0.8895 0.849 0.7947 0.8191	0.9089 0.9871 0.9745 0.943 0.9133 0.9449	0.84 0.95 0.94 0.91 0.84 0.9	0.83 0.86 0.88 0.84 0.8 0.8	0.92 0.97 0.97 0.94 0.93 0.93	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267	4.58% 4.76% 3.67% 4.22% 5.59% 5.75%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% **Effect* 0.0%
0 2.5 5 6.06 10 15 Angular (Cor C- %	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 5 7med Summ Count	0.86 0.932 0.932 0.896 0.854 0.882 hary Mean 1.19	0.8111 0.8769 0.8895 0.849 0.7947 0.8191 95% LCL 1.116	0.9089 0.9871 0.9745 0.943 0.9133 0.9449 95% UCL 1.265	0.84 0.95 0.94 0.91 0.84 0.9 Median 1.159	0.83 0.86 0.88 0.84 0.8 0.8 0.8	0.92 0.97 0.97 0.94 0.93 0.93	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267 Std Err 0.02686 0.03729	4.58% 4.76% 3.67% 4.22% 5.59% 5.75% CV% 5.05% 6.33%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% **Effect* 0.0% -10.6%
0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 med Summ Count 5	0.86 0.932 0.932 0.896 0.854 0.882 mary Mean 1.19 1.317	0.8111 0.8769 0.8895 0.849 0.7947 0.8191 95% LCL 1.116 1.213 1.23	0.9089 0.9871 0.9745 0.943 0.9133 0.9449 95% UCL 1.265 1.42	0.84 0.95 0.94 0.91 0.84 0.9 Median 1.159 1.345 1.323	0.83 0.86 0.88 0.84 0.8 0.8 0.8 Min 1.146 1.187 1.217	0.92 0.97 0.97 0.94 0.93 0.93 Max 1.284 1.397 1.397	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267 Std Err 0.02686 0.03729 0.03017	4.58% 4.76% 3.67% 4.22% 5.59% 5.75% CV% 5.05% 6.33% 5.14%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% **Effect* 0.0% -10.6% -10.32%
0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5 5	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 med Summ Count 5 5	0.86 0.932 0.932 0.896 0.854 0.882 mary Mean 1.19 1.317 1.313	0.8111 0.8769 0.8895 0.849 0.7947 0.8191 95% LCL 1.116 1.213	0.9089 0.9871 0.9745 0.943 0.9133 0.9449 95% UCL 1.265 1.42 1.397	0.84 0.95 0.94 0.91 0.84 0.9 Median 1.159 1.345	0.83 0.86 0.88 0.84 0.8 0.8 0.8	0.92 0.97 0.97 0.94 0.93 0.93 Max 1.284 1.397	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267 Std Err 0.02686 0.03729	4.58% 4.76% 3.67% 4.22% 5.59% 5.75% CV% 5.05% 6.33%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% **Effect* 0.0% -10.6%

Analyst: AC QA: WFT III

C-%

Report Date: Test Code:

Rankits

07 Jul-17 15:43 (p 2 of 2) 1706-S204 | 08-6882-6222

2.0

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 14-4818-0451 Endpoint: Fertilization Rate CETISv1.8.7 **CETIS Version:** Analyzed: 07 Jul-17 15:43 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 1.0 F 0.12 0.10 0.9 0.08 0,06 Fertilization Rate 0.04 0.7 0.02 0.6 0.00 0.5 -0.02 0.04 0.4 -0.06 -0,08 -0.10 0.2 -0.12 0.1 -0.14 0.0 -0.16 0 LC 2.5 5 6.06 10 15

-2.5 -2,0 -1.5 -1.0 -0.5 0.0

Report Date:

07 Jul-17 15:43 (p 1 of 1)

Test Code:

1706-S204 | 08-6882-6222

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

Analysis ID: 13-2565-5640 Endpoint: Fertilization Rate CETIS Version: CETISv1.8.7

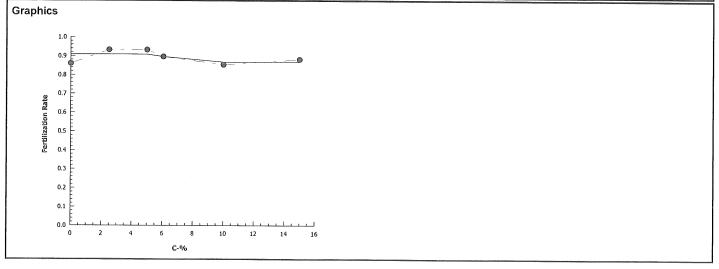
Analyzed: 07 Jul-17 15:43 Analysis: Linear Interpolation (ICPIN) Official Results: Yes

Linear Interpol	ation Options				
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	350119	1000	Yes	Two-Point Interpolation
Point Estimate	s				

Level % 95% LCL 95% UCL TU 95% LCL 95% UCL

EC25 >15 N/A N/A <6.667 NA NA EC50 >15 N/A N/A <6.667 NA NA

Fertilizat	tion Rate Summary		Calculated Variate(A/B)								
C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.86	0.83	0.92	0.01761	0.03937	4.58%	0.0%	430	500
2.5		5	0.932	0.86	0.97	0.01985	0.04438	4.76%	-8.37%	466	500
5		5	0.932	0.88	0.97	0.0153	0.03421	3.67%	-8.37%	466	500
6.06		5	0.896	0.84	0.94	0.01691	0.03782	4.22%	-4.19%	448	500
10		5	0.854	8.0	0.93	0.02135	0.04775	5.59%	0.7%	427	500
15		5	0.882	0.8	0.93	0.02267	0.0507	5.75%	-2.56%	441	500



Report Date:

07 Jul-17 15:44 (p 1 of 1)

Test Code:

1706-S204 | 08-6882-6222

							rest	Code:)6-S204 0	0-0002-022
Echinoid Sp	erm Cell Fertiliz	ation Test	15C	150					Nautilu	s Environ	mental (CA
Analysis ID:	16-7578-2505	Fr	ndpoint: Fer	tilization Ra	te .		CET	IS Version	: CETISv1	Q 7	
Analyzed:	07 Jul-17 15:4		•	rametric Bio		-Two Samp		ial Result		.0.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	rected)	NA	C*b < T	NA	NA	0.75	6.34%	15	>15	NA	6.667
TST-Welch's	t Test										
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	n(a:5%)		
Lab Control	2.5*		9.998	1.943	0.082 6	<0.0001	CDF		nificant Effec	f	
	5*		11.59	1.943	0.070 6	<0.0001	CDF	•	nificant Effec		
	6.06*		10.37	1.895	0.065 7	<0.0001	CDF	_	nificant Effec		
	10*		7.572	1.943	0.075 6	0.0001	CDF	•	nificant Effec		
	15*		8.464	1.943	0.076 6	<0.0001	CDF	_	nificant Effec		
ANOVA Table	e										
Source	Sum Sqւ	ıares	Mean Squ	ıare	DF	F Stat	P-Value	Decision	n(a:5%)		
Between	0.084808	94	0.0169617	79	5	3.402	0.0183	Significa			***************************************
Error	0.119664		0.004986		24			- 3			
Total	0.204473				29	_					
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision('α:1%)			
Attribute Variances		Equality of \		Test Stat 0.5809	Critical	P-Value 0.9889	Decision(9999 (M-030M) As a man as a magazinga.	
	Bartlett E	Equality of \					Decision(Equal Var Normal Di	iances		9999 1440 at 1 a a a a a a a a a a a a a a a a a	
Variances Distribution	Bartlett E			0.5809	15.09	0.9889	Equal Var	iances			
Variances Distribution	Bartlett E Shapiro-			0.5809	15.09	0.9889	Equal Var	iances	Std Err	CV%	%Effect
Variances Distribution Fertilization I	Bartlett E Shapiro- Rate Summary	Wilk W Noi	rmality	0.5809 0.974	15.09 0.9031	0.9889 0.6538	Equal Var Normal Di	iances stribution	Std Err 0.01761	CV% 4.58%	%Effect
Variances Distribution Fertilization I C-%	Bartlett E Shapiro- Rate Summary Control Type	Wilk W Nor	mality Mean	0.5809 0.974 95% LCL	15.09 0.9031 95% UCL	0.9889 0.6538 Median	Equal Var Normal Di	iances stribution	····	····	
Variances Distribution Fertilization I C-% 0 2.5	Bartlett E Shapiro- Rate Summary Control Type	Count 5	Mean 0.86	0.5809 0.974 95% LCL 0.8111	15.09 0.9031 95% UCL 0.9089	0.9889 0.6538 Median 0.84	Equal Var Normal Di Min 0.83	iances stribution Max 0.92	0.01761	4.58%	0.0%
Variances Distribution Fertilization I C-% 0 2.5	Bartlett E Shapiro- Rate Summary Control Type	Count 5 5	Mean 0.86 0.932	0.5809 0.974 95% LCL 0.8111 0.8769	15.09 0.9031 95% UCL 0.9089 0.9871	0.9889 0.6538 Median 0.84 0.95	Equal Var Normal Di Min 0.83 0.86	Max 0.92 0.97	0.01761 0.01985	4.58% 4.76% 3.67%	0.0% -8.37% -8.37%
Variances Distribution Fertilization I C-%	Bartlett E Shapiro- Rate Summary Control Type	Count 5 5 5	Mean 0.86 0.932 0.932	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895	15.09 0.9031 95% UCL 0.9089 0.9871 0.9745	0.9889 0.6538 Median 0.84 0.95 0.94	Equal Var Normal Di Min 0.83 0.86 0.88 0.84	Max 0.92 0.97 0.97	0.01761 0.01985 0.0153	4.58% 4.76% 3.67% 4.22%	0.0% -8.37% -8.37% -4.19%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10	Bartlett E Shapiro- Rate Summary Control Type	Count 5 5 5 5	Mean 0.86 0.932 0.932 0.896	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.849	15.09 0.9031 95% UCL 0.9089 0.9871 0.9745 0.943	0.9889 0.6538 Median 0.84 0.95 0.94 0.91	Min 0.83 0.86 0.88	Max 0.92 0.97 0.97 0.94	0.01761 0.01985 0.0153 0.01691	4.58% 4.76% 3.67%	0.0% -8.37% -8.37%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15	Bartlett E Shapiro- 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 0.86 0.932 0.932 0.896 0.854 0.882	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.849 0.7947	95% UCL 0.9089 0.9871 0.9745 0.943 0.9133	0.9889 0.6538 Median 0.84 0.95 0.94 0.91 0.84	Min 0.83 0.86 0.88 0.84 0.8	Max 0.92 0.97 0.97 0.94 0.93	0.01761 0.01985 0.0153 0.01691 0.02135	4.58% 4.76% 3.67% 4.22% 5.59%	0.0% -8.37% -8.37% -4.19% 0.7%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15	Bartlett E Shapiro- 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 0.86 0.932 0.932 0.896 0.854 0.882	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.849 0.7947	95% UCL 0.9089 0.9871 0.9745 0.943 0.9133	0.9889 0.6538 Median 0.84 0.95 0.94 0.91 0.84	Min 0.83 0.86 0.88 0.84 0.8	Max 0.92 0.97 0.97 0.94 0.93	0.01761 0.01985 0.0153 0.01691 0.02135	4.58% 4.76% 3.67% 4.22% 5.59%	0.0% -8.37% -8.37% -4.19% 0.7%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15 Angular (Core	Bartlett E Shapiro- Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 5 med Sumr	Mean 0.86 0.932 0.932 0.896 0.854 0.882	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.849 0.7947 0.8191	95% UCL 0.9089 0.9871 0.9745 0.943 0.9133 0.9449	0.9889 0.6538 Median 0.84 0.95 0.94 0.91 0.84 0.9	Equal Var Normal Di Min 0.83 0.86 0.88 0.84 0.8 0.8	Max 0.92 0.97 0.97 0.94 0.93 0.93	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267	4.58% 4.76% 3.67% 4.22% 5.59% 5.75%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15 Angular (Core	Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 5 cmed Sumr	Mean 0.86 0.932 0.932 0.896 0.854 0.882 mary Mean	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.849 0.7947 0.8191	95% UCL 0.9089 0.9871 0.9745 0.943 0.9133 0.9449	0.9889 0.6538 Median 0.84 0.95 0.94 0.91 0.84 0.9	Equal Var Normal Di Min 0.83 0.86 0.88 0.84 0.8 0.8	Max 0.92 0.97 0.97 0.94 0.93 0.93	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267	4.58% 4.76% 3.67% 4.22% 5.59% 5.75% CV% 5.05%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% %Effect 0.0%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15 Angular (Cort	Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 cmed Sumr Count 5	Mean 0.86 0.932 0.932 0.896 0.854 0.882 mary Mean 1.19	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.7947 0.8191 95% LCL 1.116	95% UCL 0.9089 0.9871 0.9745 0.943 0.9133 0.9449 95% UCL 1.265	0.9889 0.6538 Median 0.84 0.95 0.94 0.91 0.84 0.9 Median 1.159	Min 0.83 0.86 0.88 0.84 0.8 0.8 Min 1.146	Max 0.92 0.97 0.97 0.94 0.93 0.93	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267 Std Err 0.02686	4.58% 4.76% 3.67% 4.22% 5.59% 5.75%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% **Effect 0.0% -10.6%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15 Angular (Cort	Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 Count Count Count 5 5 5 5 5 5 5 5 5 6 6 6 6 6 7 6 7 6 7 7 7 7	Mean 0.86 0.932 0.932 0.896 0.854 0.882 mary Mean 1.19 1.317	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.7947 0.8191 95% LCL 1.116 1.213	95% UCL 0.9089 0.9871 0.9745 0.943 0.9133 0.9449 95% UCL 1.265 1.42	0.9889 0.6538 Median 0.84 0.95 0.94 0.91 0.84 0.9 Median 1.159 1.345	Min 0.83 0.86 0.88 0.84 0.8 0.8 Min 1.146 1.187	Max 0.92 0.97 0.97 0.94 0.93 0.93 Max 1.284 1.397	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267 Std Err 0.02686 0.03729 0.03017	4.58% 4.76% 3.67% 4.22% 5.59% 5.75% CV% 5.05% 6.33% 5.14%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% **Effect 0.0% -10.6% -10.32%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15 Angular (Cort C-% 0 2.5 5	Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 Count Count Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.86 0.932 0.932 0.896 0.854 0.882 mary Mean 1.19 1.317 1.313	0.5809 0.974 95% LCL 0.8111 0.8769 0.8895 0.849 0.7947 0.8191 95% LCL 1.116 1.213 1.23	95% UCL 0.9089 0.9871 0.9745 0.943 0.9133 0.9449 95% UCL 1.265 1.42 1.397	0.9889 0.6538 Median 0.84 0.95 0.94 0.91 0.84 0.9 Median 1.159 1.345 1.323	Min 0.83 0.86 0.88 0.84 0.8 0.8 1.146 1.187 1.217	Max 0.92 0.97 0.97 0.94 0.93 0.93 Max 1.284 1.397 1.397	0.01761 0.01985 0.0153 0.01691 0.02135 0.02267 Std Err 0.02686 0.03729	4.58% 4.76% 3.67% 4.22% 5.59% 5.75% CV% 5.05% 6.33%	0.0% -8.37% -8.37% -4.19% 0.7% -2.56% **Effect 0.0% -10.6%

CETIS Test Data Worksheet

Report Date:

29 Jun-17 18:52 (p 1 of 1)

Test Code: 1706-520403-8551-4552/16FA7C38

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: End Date:

30 Jun-17 30 Jun-17

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

Sample Code: 17- 0 73 € Sample Source: IDE Americas, Inc. Sample Statio NA 004 (40 --+ --+:)

ample Dat		Jun-17			al: Facility Ef	fluent	Sample Source: IDE Americas, Inc. Sample Station: M-001 (40 ppt adj)	
C-%	Code	Rep	Pos	# Counted	# Fertilized		Notes	
			66	100	93	ALS 7/3/17		
			67	100	95	710		
			68	100	90			
			69	100	91			
			70	100	83			
			71	100	94			
			72	(00	97			
			73	100	92			
			74	100	96			
			75	100	8b			
			76	100	86			
			77	100	95			
			78 79	100	89			
			80	100	90 92			
			81	100	92			
			82	100	84			
			83	100	97			
	-		84	100	94 84			
			85	100	88			
			86	100	83			
			87	100				
***************************************	+ 1		88	100	93 87			
			89	100	91			
3.0000.01			90	100	84			
			91	100	91			
			92	100	92			
			93	100				
			94	100	88 88			
			95	100	80			

Report Date:

29 Jun-17 18:52 (p 1 of 1)

Test Code: 705-5201 03-8551-4552/16FA7C38

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

17-0738 Start Date: 30 Jun-17 Species: Strongylocentrotus purpuratus Sample Code: End Date: Sample Source: IDE Americas, Inc. 30 Jun-17 Protocol: EPA/600/R-95/136 (1995) Sample Date: 30 Jun-17 Material: Facility Effluent Sample Station: M-001 (40 ppt adj)

C-%	Code	Rep	Pos	# Counted		Notes
0	LC	1	70	COI	88	to 7/1/11
0	LC	2	93			
0	LC	3	92			
0	LC	4	86			
0	LC	5	84			
2.5		1	73			
2.5		2	75			
2.5		3	74			
2.5		4	72			
2.5		5	67			
5		1	85			
5		2	80			
5		3	77			
5		4	83			
5		5	82			
6.06		1	71	100	94	
6.06		2	94	•	,	
6.06		3	89			
6.06		4	81			
6.06		5	91			
10		1	76			
10		2	90			
10		3	78			
10		4	95			
10		5	87			
15		1	69			
15		2	68			
15		3	66			
15		4	88			
15		5	79			

QU: CG

Water Quality Measurements

Client:

IDE

Test Species: S. purpuratus

Sample ID:

M-001 (40 ppt adjusted)

Start Date/Time: 6/30/2017 1750

Sample Log No.:

17- 0738

End Date/Time: 6/30/2017

Dilutions made by:

: 40V)

Test No: 1706-S204

			Analyst:	CH							
		Initial Readings									
Concentration %	DO (ma/l.)	pH (units)	Salinity	Temperature							
/0	(mg/L)	(units)	(ppt)	(°C)							
Lab Control	8,5	7.95	33.9	15.8							
2.5	8.1	7.85	34.2	15.9							
5.0	7.9	7.85	34.3	15.9							
6.06	7.9	7.86	34.3	16.0							
10	8.4	7.98	34.2	15.7							
15	8.4	7.97	34.9	15.4							

_					
Co	m	m	nt	0	

Diwhons made with M-INF

QC Check;

AC7/1/17

Final Review: 17 7 7 111 |

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

Brine Dilution Worksheet

Project:

IDE

Analyst: AD

Sample ID:

M-001 (40 ppt adjusted)

Test Date: 6/30/2017

Test No:

Test Type: Urchin Fertilization

Salinity of Effluent

62.6

Salinity of Seawater

33.5

Date of Brine used: NA

Target Salinity

40.0

Alk. of 40 ppt Adj. Sample: 219 124 mg/L as CaCO3

Effluent

Brine Control

Salinity Adjustment Factor: (TS

-SE)/(SB - TS) =

3.48

-6.15

TS = target salinity

SE = salinity of effluent

SB = salinity of brine

Concentration %	Effluent Salinity Volume Adjustment (ml) Factor		Seawater Volume (ml)	Final Volume (ml)
100	100	3.48	347.7	448

Comments:

Formula for amount of seawater to dilute sample to 40ppt

Use 40 ppt sample as 100% sample for testing.

NA = not applicable; sample not diluted with Nautilus brine.

Final Review: VFP

@AD Q18 7/10/17

Echinoderm Sperm-Cell Fertilization Worksheet

				-	
Client:	IDE			Stort Doto/Tiv	mai 0/00/0017 / 17173
Sample ID:	M-001 40 PP	t Adjusted			me: 6/30/2017 / 150 me: 6/30/2017 / 1830
Test No.:	1706-520		_		ies: S. purpuratus
	^ -				rce: Pt. Loma
Tech initials:	_10			Date Collect	ed: [p](p]
Injection Time:	1050				
Sperm Absorbance at	1400 nm: 1 000	(4			
Sperm Absorbance at	t 400 nm: 1, 009			ity of 4x10 ⁶ sperm/ı	ml)
Eggs Counted:	<i>8</i> 8	an: 30.8 x !	50 = <u>404</u>	O eaas/ml	
	76				
	(targ	et counts of 80 eggs p	er vertical pass on	Sedgwick-	
	Rafte	er slide for a final dens	ity of 4000 eggs/m	1)	
	- 00 82				
Initial density:	40410 eggs/ml	= dilut	ion factor		(6)
Final density:	4000 eggs/ml		egg stock	egg stock seawater	mI
•	9330////	t i	s seawater	Seawatei	ml
		-			
Prepare the embryo st	tock according to the calcu	lated dilution factor.	For example, if t	the dilution factor is	3 2.25, use 100 ml of
existing stock (1 part)	and 125 ml of dilution wate	r (1.25 parts).		~ :	
			_		
Rangefinder Test:	2000-4 4000	.4 4000.4	Sperm:Egg Rat		,
ml Sperm Stock	2000:1 1600 50 40	<u>1200:1</u> 30	800:1 400 20 10		<u>100:1</u> <u>50:1</u>
ml Seawater	0.0 10		30 40		2.5 1.25 47.5 48.75
					17.0
Contractor Adda d (400 D	Time	Rangefinder Ratio	E Fert.	<u>Unfert.</u>	
Sperm Added (100 μl): Eggs Added (0.5 ml):	1-1-7	50:1	- 34.83	16117	
Test Ended:	1123	100:1	910	4	
rest Ended.	112)		_		
		,			
NOTE: Choose a sper	m-to-egg ratio that results	in fertilization betwe	en 80 and 90 ner	rcent If more than	one concentration is within
this range, choose th	e ralio ciosest to 90 per	cent uniess profess	sional judgment	dictates considera	tion of other factors (e.g.,
organism health, stage	of reproductive season, si	te conditions).	, ,		
Dofinitive Teet		0 5 5	—	e	
<u>Definitive Test</u>		Sperm:Egg Ratio	Used: 15	n-	,
	Time (2)		Fort	l looks of	
Sperm Added (100 µl):	17450	QC1	Pert.	<u>Unfert.</u>	
Eggs Added (0.5 ml):	1810	QC2		$\frac{3}{10}$	
Test Ended:	1830	Egg Control 1	7/	100	
		Egg Control 2	-	100	
		_gg		100	
	•	ಬ್ಲ			
Comments:	@nodiluko	MYDOWY.	000		
	WAD ON IN	30/170			
		, .			
					,
QC Check:	AC 7/5/1				16-0-1-1-
	101/11	_		Final Review	N: PDP 7/11/17
Nautilus Environmental. 43	40 Vandever Avenue. San Die	go, CA 92120.			



CETIS Summary Report

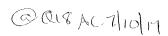
Report Date:

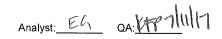
06 Jul-17 15:29 (p 1 of 1)

Test Code:

1706-S207 | 20-9818-9801

		.tomestoner#45-40*						rest oode.		11.	0 0201 2	0-9010-9001
Echinoid Spe	rm Cell Fertiliza	tion Te	est 15C					Actor Cold Color		Nautilu	s Environ	mental (CA)
Batch ID: Start Date: Ending Date: Duration:	12-4316-0758 30 Jun-17 17:5 30 Jun-17 18:3 40m		Test Type: Protocol: Species: Source:	Fertilization EPA/600/R-95 Strongylocenti Pt. Loma	. ,	itus		Analyst: Diluent: Brine: Age:		ıral Seawat Applicable	er M~1 N	1
· -	18-7927-7034 30 Jun-17 08:00 30 Jun-17 11:40 10h (4.5°C)		Code: Material: Source: Station:	17-0739 Facility Effluer IDE Americas ERI				Client: Project:	IDE Carl	sbad Desal	Plant	
Comparison S	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Meth	nod			
09-9212-2387	Fertilization Rat	te	10	15	12.25	6.11%	10		<u> </u>	lultiple Com	parison Te	st
Point Estimate	e Summary											
Analysis ID	Endpoint		Level	%	95% LCL	95% UCL	TU	Meth	nod			
12-0743-1815	Fertilization Rat	te	EC25 EC50	>15 >15	N/A N/A	N/A N/A	<6.66 <6.66		ar Inte	erpolation (l	CPIN)	
Test Acceptab	oility	***************************************										
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	its	Over	rlap	Decision		
09-9212-2387	Fertilization Rat	e	Contro	ol Resp	0.908	0.7 - NL		Yes	•	Passes A	cceptability	Criteria
ł	Fertilization Rat			ol Resp	0.908	0.7 - NL		Yes			cceptability	
09-9212-2387	Fertilization Rat	:e	PMSD		0.06111	NL - 0.25		No		Passes A	cceptability	r Criteria
Fertilization R	ate Summary											
C-%	Control Type	Coun	t Mean	95% LCL	95% UCL	Min	Max	Std I	Err	Std Dev	CV%	%Effect
	Lab Control	5	0.908	0.8944	0.9216	0.89	0.92	0.004	4898	0.01095	1.21%	0.0%
2.5		5	0.918	0.8941	0.9419	0.89	0.94	0.008	3602	0.01923	2.1%	-1.1%
5		5	0.908	0.8565	0.9595	0.87	0.97	0.018	355	0.04147	4.57%	0.0%
6.06		5	0.906	0.859	0.953	0.85	0.95	0.016		0.03782	4.17%	0.22%
10		5	0.898	0.8458	0.9502	0.85	0.96	0.018		0.04207	4.69%	1.1%
15		5	0.814	0.7696	0.8584	0.77	0.85	0.016	3	0.03578	4.4%	10.35%
Fertilization R	ate Detail											
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	0.89	0.91	0.91	0.91	0.92						
2.5		0.89	0.93	0.94	0.91	0.92						
5		0.97	0.89	0.88	0.87	0.93						
6.06		0.85	0.95	0.9	0.9	0.93						
10		0.85	0.91	0.87	0.96	0.9						
15		0.77	0.85	0.85	0.79	0.81						





Report Date: Test Code: 06 Jul-17 15:29 (p 1 of 2) 1706-S207 | 20-9818-9801

rest Code:	1700-3207 20-9
	The second secon
	Mautilus Environmo

Echinoid Sp	erm Cell Fertiliza	ation Test	15C						Nautilus	Environ	mental (CA)
Analysis ID: Analyzed:	09-9212-2387 06 Jul-17 14:4		•	Fertilization Rat Parametric-Cor		tments		IS Version		.8.7	
Data Transfo		Zeta			Seed	unents				TOF	7211
Angular (Corr		NA NA	Alt Hy	rp Trials NA	NA		PMSD 6.11%	NOEL 10	LOEL 15	TOEL 12.25	TU
			9 1	.,,	177		0.1170	10	10	12.20	10
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-%		Test S	tat Critical	MSD DF	P-Value	P-Type	Decision	n(α:5%)		
Lab Control	2.5		-0.5148	8 2.362	0.086 8	0.9421	CDF	Non-Sigr	ificant Effect		
	5		-0.1989	9 2.362	0.086 8	0.8854	CDF	Non-Sigr	ificant Effect		
	6.06		-0.024	18 2.362	0.086 8	0.8404	CDF	Non-Sigr	ificant Effect		
	10		0.3032	2.362	0.086 8	0.7287	CDF	Non-Sigr	ificant Effect		
	15*		3.738	2.362	0.086 8	0.0022	CDF	Significa	nt Effect		
ANOVA Tabl	е										
Source	Sum Squ	ıares	Mean	Square	DF	F Stat	P-Value	Decision	ι(α:5%)		
Between	0.083986	54	0.0167	9731	5	5.021	0.0027	Significal	nt Effect		
Error	0.080285	32	0.0033	45222	24			J			
Total	0.164271	9			29						
Distributiona	I Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Bartlett E	Equality of	Variance	8.431	15.09	0.1340	Equal Variances				
Distribution	Shapiro-	Wilk W No	rmality	0.9649	0.9031	0.4111	Normal Distribution				
Fertilization	Rate Summary						A TOTAL PROPERTY OF THE STATE OF				A CONTRACTOR OF THE CONTRACTOR
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.908	0.8944	0.9216	0.91	0.89	0.92	0.004898	1.21%	0.0%
2.5	•	5	0.918	0.8941	0.9419	0.92	0.89	0.94	0.008602	2.1%	-1.1%
5		5	0.908	0.8565	0.9595	0.89	0.87	0.97	0.01855	4.57%	0.0%
6.06		5	0.906	0.859	0.953	0.9	0.85	0.95	0.01691	4.17%	0.22%
10		5	0.898	0.8458	0.9502	0.9	0.85	0.96	0.01881	4.69%	1.1%
15		5	0.814	0.7696	0.8584	0.81	0.77	0.85	0.016	4.4%	10.35%
Angular (Cor	rected) Transfor	med Sum	mary								
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.263	1.24	1.286	1.266	1.233	1.284	0.00833	1.48%	0.0%
2.5		5	1.282	1.239	1.325	1.284	1.233	1.323	0.01555	2.71%	-1.49%
5		5	1.27	1.17	1.37	1.233	1.202	1.397	0.03604	6.34%	-0.58%
6.06		5	1.264	1.183	1.344	1.249	1.173	1.345	0.02902	5.13%	-0.07%
		_				·;- ·•	•				5.51 /0

10

15

5

5

1.252

1.126

1.158

1.069

1.346

1.184

1.249

1.12

1.173

1.071

1.369

1.173

0.03371

0.02064

6.02%

4.1%

0.88%

10.83%

Report Date: Test Code: 06 Jul-17 15:29 (p 2 of 2) 1706-S207 | 20-9818-9801

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 09-9212-2387 Endpoint: Fertilization Rate CETIS Version: CETISv1.8.7 Analyzed: 06 Jul-17 14:47 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 1.0 0.14 0.12 0.10 Fertilization Rate 0.7 0.06 0.6 0.04 0.5 0.4 0.00 -0.02 0.3 0.2 -0.06 0.1 -0.08 0.0 0 LC 2.5 6.06 10 15 -2.5 -2.0 -1.5 -1.0 -0.5 0.0 0.5 1.0 1.5 2.0 2.5 C-% Rankits

Report Date: Test Code:

06 Jul-17 15:29 (p 1 of 1)

1706-S207 | 20-9818-9801

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: 12-0743-1815 Endpoint: Fertilization Rate CETIS Version: CETISv1.8.7

Analyzed: 06 Jul-17 14:47 Analysis: Linear Interpolation (ICPIN) Official Results: Yes

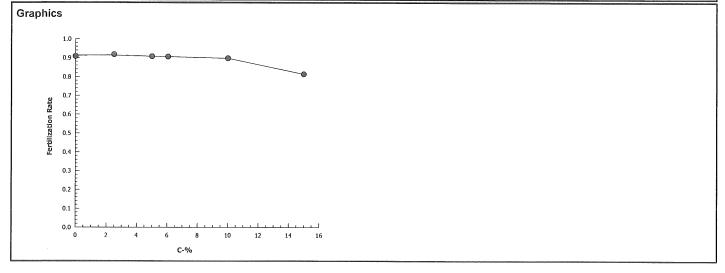
Linear Interpolation Options

X Transform Y Transform Seed Resamples Exp 95% CL Method

Linear Linear 741544 1000 Yes Two-Point Interpolation

Point Estimates Level % 95% LCL 95% UCL TU 95% UCL 95% LCL EC25 >15 N/A N/A <6.667 NA NA EC50 >15 N/A N/A <6.667 NA NA

Fertilizat	tion Rate Summary				Cal	culated Variat	te(A/B)				
C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.908	0.89	0.92	0.004898	0.01095	1.21%	0.0%	454	500
2.5		5	0.918	0.89	0.94	0.008602	0.01923	2.1%	-1.1%	459	500
5		5	0.908	0.87	0.97	0.01855	0.04147	4.57%	0.0%	454	500
6.06		5	0.906	0.85	0.95	0.01691	0.03782	4.17%	0.22%	453	500
10		5	0.898	0.85	0.96	0.01881	0.04207	4.69%	1.1%	449	500
15		5	0.814	0.77	0.85	0.016	0.03578	4.4%	10.35%	407	500



Report Date:

06 Jul-17 15:29 (p 1 of 1)

Test Code:

1706-S207 | 20-9818-9801

Echinoid Sp	erm Cell Fertiliz	ation Test	15C	TST	-				Nautilu	s Environ	mental (CA
Analysis ID:	02-7313-6603	En	dpoint: Fer	tilization Ra	te		CET	IS Version:	CETISv1	8.7	
Analyzed:	06 Jul-17 14:4	18 A n	alysis: Par	ametric Bio	equivalence	-Two Samp		ial Results		.0.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor.	rected)	NA	C*b < T	NA	NA	0.75	3.08%	15	>15	NA	6.667
TST-Welch's	t Test			40							
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(a:5%)		
Lab Control	2.5*		19.97	2.015	0.034 5	<0.0001	CDF		ificant Effect		
	5*		8.832	2.132	0.078 4	0.0005	CDF	J	ificant Effect		
	6.06*		10.67	2.132	0.063 4	0.0002	CDF	•	ificant Effect		
	10*		8.886	2.132	0.073 4	0.0004	CDF	-	ificant Effect		
	15*		8.302	2.132	0.046 4	0.0006	CDF		ficant Effect		
ANOVA Tabl	Δ			1				- Tron Oigin	THE ENGLE		
Source	Sum Squ	ıares	Mean Squ	are	DF	F Stat	P-Value	Decision	(a:E9/)		
Between	0.083986		0.0167973		5	5.021	0.0027		·		
Error	0.080285		0.0033452		24	5.021	0.0027	Significan	t Enect		
Total	0.164271		0.0000402		29	_					
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision((q·1%)			
Variances	Bartlett F	Equality of \	/a-ia-a-a				Decision	α. 1 /0/			
			ranance		15 114	በ 13//በ	Equal Var	iancec			
Distribution		Wilk W Nor		8.431 0.9649	15.09 0.9031	0.1340 0.4111	Equal Var Normal Di				
							•				
Fertilization	Shapiro-						•		Std Err	CV%	%Effect
Fertilization C-%	Shapiro- Rate Summary	Wilk W Nor	mality	0.9649	0.9031	0.4111 Median	Normal Di	stribution Max			
Fertilization C-%	Shapiro- Rate Summary Control Type	Wilk W Nor	mality Mean	0.9649 95% LC L	0.9031 95% UCL	0.4111	Normal Di	stribution	0.004898	1.21%	0.0%
Fertilization C-% 0 2.5	Shapiro- Rate Summary Control Type	Wilk W Nor Count 5	Mean 0.908	0.9649 95% LCL 0.8944	0.9031 95% UCL 0.9216	0.4111 Median 0.91	Min 0.89	Max 0.92 0.94	0.004898 0.008602	1.21% 2.1%	0.0% -1.1%
Fertilization C-% 0 2.5	Shapiro- Rate Summary Control Type	Count 5 5	Mean 0.908 0.918	0.9649 95% LCL 0.8944 0.8941	0.9031 95% UCL 0.9216 0.9419	0.4111 Median 0.91 0.92 0.89	Min 0.89 0.89 0.87	Max 0.92 0.94 0.97	0.004898 0.008602 0.01855	1.21% 2.1% 4.57%	0.0% -1.1% 0.0%
Fertilization C-% 0 2.5 5 6.06	Shapiro- Rate Summary Control Type	Count 5 5 5	Mean 0.908 0.918 0.908	0.9649 95% LCL 0.8944 0.8941 0.8565 0.859	95% UCL 0.9216 0.9419 0.9595 0.953	Median 0.91 0.92 0.89 0.9	Min 0.89 0.89 0.87 0.85	Max 0.92 0.94 0.97 0.95	0.004898 0.008602 0.01855 0.01691	1.21% 2.1% 4.57% 4.17%	0.0% -1.1% 0.0% 0.22%
Fertilization C-% 0 2.5 5 6.06	Shapiro- Rate Summary Control Type	Count 5 5 5 5	Mean 0.908 0.918 0.908 0.908	0.9649 95% LCL 0.8944 0.8941 0.8565	0.9031 95% UCL 0.9216 0.9419 0.9595	0.4111 Median 0.91 0.92 0.89	Min 0.89 0.89 0.87	Max 0.92 0.94 0.97	0.004898 0.008602 0.01855	1.21% 2.1% 4.57%	0.0% -1.1% 0.0% 0.22% 1.1%
Fertilization C-% 0 2.5 5 6.06 10	Shapiro- Rate Summary Control Type	Count 5 5 5 5 5 5	Mean 0.908 0.918 0.908 0.906 0.898 0.814	95% LCL 0.8944 0.8941 0.8565 0.859 0.8458	95% UCL 0.9216 0.9419 0.9595 0.953 0.9502	0.4111 Median 0.91 0.92 0.89 0.9 0.9	Min 0.89 0.89 0.87 0.85 0.85	Max 0.92 0.94 0.97 0.95 0.96	0.004898 0.008602 0.01855 0.01691 0.01881	1.21% 2.1% 4.57% 4.17% 4.69%	0.0% -1.1% 0.0% 0.22%
Fertilization C-% 0 2.5 5 6.06 10 15	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 5	Mean 0.908 0.918 0.908 0.906 0.898 0.814	95% LCL 0.8944 0.8941 0.8565 0.859 0.8458	95% UCL 0.9216 0.9419 0.9595 0.953 0.9502	0.4111 Median 0.91 0.92 0.89 0.9 0.9	Min 0.89 0.89 0.87 0.85 0.85	Max 0.92 0.94 0.97 0.95 0.96	0.004898 0.008602 0.01855 0.01691 0.01881	1.21% 2.1% 4.57% 4.17% 4.69%	0.0% -1.1% 0.0% 0.22% 1.1%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Shapiro- Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 7 med Summ	Mean 0.908 0.918 0.908 0.906 0.898 0.814	0.9649 95% LCL 0.8944 0.8941 0.8565 0.859 0.8458 0.7696	0.9031 95% UCL 0.9216 0.9419 0.9595 0.953 0.9502 0.8584	0.4111 Median 0.91 0.92 0.89 0.9 0.9	Min 0.89 0.89 0.87 0.85 0.85 0.77	Max 0.92 0.94 0.97 0.95 0.96 0.85	0.004898 0.008602 0.01855 0.01691 0.01881 0.016	1.21% 2.1% 4.57% 4.17% 4.69% 4.4%	0.0% -1.1% 0.0% 0.22% 1.1% 10.35%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 5 cmed Summ	Mean 0.908 0.918 0.908 0.906 0.898 0.814 mary Mean	95% LCL 0.8944 0.8941 0.8565 0.859 0.8458 0.7696	0.9031 95% UCL 0.9216 0.9419 0.9595 0.953 0.9502 0.8584 95% UCL	0.4111 Median 0.91 0.92 0.89 0.9 0.9 1.90 0.81 Median 1.266	Min 0.89 0.89 0.87 0.85 0.85 0.77	Max 0.92 0.94 0.97 0.95 0.96 0.85	0.004898 0.008602 0.01855 0.01691 0.01881 0.016 Std Err 0.00833	1.21% 2.1% 4.57% 4.17% 4.69% 4.4% CV% 1.48%	0.0% -1.1% 0.0% 0.22% 1.1% 10.35% %Effect 0.0%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 7 Treed Summ Count 5	Mean 0.908 0.918 0.908 0.906 0.898 0.814 mary Mean 1.263	95% LCL 0.8944 0.8941 0.8565 0.859 0.8458 0.7696 95% LCL 1.24	95% UCL 0.9216 0.9419 0.9595 0.953 0.9502 0.8584 95% UCL 1.286	0.4111 Median 0.91 0.92 0.89 0.9 0.9 Median	Min 0.89 0.89 0.87 0.85 0.85 0.77 Min 1.233 1.233	Max 0.92 0.94 0.97 0.95 0.96 0.85 Max 1.284 1.323	0.004898 0.008602 0.01855 0.01691 0.01881 0.016 Std Err 0.00833 0.01555	1.21% 2.1% 4.57% 4.17% 4.69% 4.4% CV% 1.48% 2.71%	0.0% -1.1% 0.0% 0.22% 1.1% 10.35% %Effect 0.0% -1.49%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5 5	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 7 med Summ Count 5 5 5	Mean 0.908 0.918 0.908 0.906 0.898 0.814 hary Mean 1.263 1.282	95% LCL 0.8944 0.8941 0.8565 0.859 0.8458 0.7696 95% LCL 1.24 1.239 1.17	95% UCL 0.9216 0.9419 0.9595 0.953 0.9502 0.8584 95% UCL 1.286 1.325 1.37	Median 0.91 0.92 0.89 0.9 0.81 Median 1.266 1.284 1.233	Min 0.89 0.89 0.87 0.85 0.85 0.77 Min 1.233 1.233 1.202	Max 0.92 0.94 0.97 0.95 0.96 0.85 Max 1.284 1.323 1.397	0.004898 0.008602 0.01855 0.01691 0.01881 0.016 Std Err 0.00833 0.01555 0.03604	1.21% 2.1% 4.57% 4.17% 4.69% 4.4% CV% 1.48% 2.71% 6.34%	0.0% -1.1% 0.0% 0.22% 1.1% 10.35% %Effect 0.0% -1.49% -0.58%
Fertilization C-% 0 2.5 5 6.06 10	Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 Count Count Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.908 0.918 0.908 0.906 0.898 0.814 nary Mean 1.263 1.282 1.27	95% LCL 0.8944 0.8941 0.8565 0.859 0.8458 0.7696 95% LCL 1.24 1.239	95% UCL 0.9216 0.9419 0.9595 0.953 0.9502 0.8584 95% UCL 1.286 1.325	0.4111 Median 0.91 0.92 0.89 0.9 0.81 Median 1.266 1.284	Min 0.89 0.89 0.87 0.85 0.85 0.77 Min 1.233 1.233	Max 0.92 0.94 0.97 0.95 0.96 0.85 Max 1.284 1.323	0.004898 0.008602 0.01855 0.01691 0.01881 0.016 Std Err 0.00833 0.01555	1.21% 2.1% 4.57% 4.17% 4.69% 4.4% CV% 1.48% 2.71%	0.0% -1.1% 0.0% 0.22% 1.1% 10.35% %Effect 0.0% -1.49%

CETIS Test Data Worksheet

Report Date:

29 Jun-17 18:55 (p 1 of 1)

Test Code: /706-5207 20-9818-9801/7D0FD5E9

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: End Date: 30 Jun-17 30 Jun-17 Species: Strongylocentrotus purpuratus Protocol: EPA/600/R-95/136 (1995)

Sample Code: 17- 0739
Sample Source: IDE Americas, Inc.

Sample Date: 30 Jun-17

Material: Facility Effluent

Sample Station: ERI

mple Dat				Materia	al: Facility Efflu	uent Sample Station: ERI
C-%	Code	Rep	Pos	# Counted	# Fertilized	Notes
			126	601	85	7/3/17
			127)	90	
			128		89	
			129		31	
			130		39	
			131		93	
			132		94	
			133		79	
			134		77	
			135		91	
			136		9(
			137		90	
			138		87	
			139		91	
			140		93	
			141		9(
			142		37 96	
			143		96	
			144		38	
			145		85	
			146		97	
			147		92	
			148		91	
			149		39	
			150		92	
			151		95	
			152		35	
			153		93 35	
			154		35	
			155	7	90	

Report Date:

29 Jun-17 18:55 (p 1 of 1)

Test Code: 1706-5 20720-9818-9801/7D0FD5E9

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Sample Code: 17- 57.39 Sample Source: IDE Americas, Inc. Start Date: 30 Jun-17 Species: Strongylocentrotus purpuratus End Date: 30 Jun-17 Protocol: EPA/600/R-95/136 (1995)

Sample Date: 30 Jun-17 Material: Facility Effluent Sample Station: ERI

imple Date					ii. Tacility Lillu	Ont	Samp	ile Station: ERI
C-%	Code	Rep	Pos		# Fertilized			Notes
0	LC	1	128	100	89	AVO	7/1/1	
0	LC	2	135	•				
0	LC	3	136					
0	LC	4	139					
0	LC	5	147					
2.5		1	130					
2.5		2	140					, AA-1-2-2
2.5		3	132					
2.5		4	148					
2.5	1	5	150					
5		1	146			AND AND DESCRIPTION OF THE PROPERTY OF THE PRO		
5		2	149					
5		3	144					
5		4	142					
5		5	131					
6.06		1	145	100	94			
6.06		2	151		1			
6.06		3	155					
6.06		4	127					
6.06		5	153					
10		1	154					
10		2	141					
10		3	138					
10		4	143					
10		5	137	,				
15		1	134					
15		2	152					
15		3	126					
15		4	133					
15		5	129					

QC: C6

Water Quality Measurements

Client: IDE Test Species: S. purpuratus

Sample ID: ERI Start Date/Time: 6/30/2017

Dilutions made by: Test No: 1706-S 207

			Analyst:	CH
Concentration	DO		Readings	
%	(mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	8.0	8.02	33.8	15.0
2.5	8,0	8.02	34.8	14.8
5.0	6.1	8.02	35.6	14.7
6.06	8.0	8,01	36.0	14.8
10	8.1	8.00	37.3	14.7
15	8.1	7.99	38.9	14.6

Comments:	Divions made with	M-INF	
QC Check:	A 7/7/17	Final Review: VTP 7/11/17	

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client:	IDE			_			
Sample ID:	ERI Brine					ne: 6/30/201	
Test No.:		57				ne: <u>6/30/201</u> es: <i>S. purpu</i>	
			_	A		ce: Pt. Loma	
Tech initials:	_A=				Date Collect		11-1
Injection Time:	1650						1
Sperm Absorbance at	400 nm: 1, 009	(target range of	0.8 - 1.0 for d	lensity of 4x	10 ⁶ sperm/r	nl)	
Eggs Counted:	<u>88</u>	an: <u>80.8</u> x	50 = 40	JUO eggs	s/ml		
	76						
	Reft	et counts of 80 eggs per slide for a final dens	per vertical pas sitv of 4000 eq	ss on Sedgwi as/ml)	ck-		
			,	5 ,			
	83						(T)
Initial density:	_UGUI [©] eggs/ml	= dilu	tion factor	ang.	stock	(I	3)
Final density:	4000 eggs/ml		t egg stock		vater	ml ml	
		1	ts seawater				
Prepare the embryo st	ock according to the calcu	lated dilution factor	For example	e if the dilut	ion factor is	2.25 400.4	00 ml of
existing stock (1 part) a	and 125 ml of dilution water	er (1.25 parts).	. Tor example	e, ii tile ullut	···	2.25, use 10	טט ווזו טו
Rangefinder Test:	2000-4 4600).4 4200.4	Sperm:Egg				
ml Sperm Stock	2000:1 1600 50 40		800:1 20	400:1 10	<u>200:1</u> 5.0	100:1 2.5	<u>50:1</u> 1.25
ml Seawater	0.0 10		30	40	45	47.5	48.75
	Time	Rangefinder Rati	o: Fert.	Linfor	4		
Sperm Added (100 µl):		50: 1	21.8	Unfer	<u>.</u> 17		
Eggs Added (0.5 ml):	1723	100:1	- <u> </u>	4) /		
Test Ended:	1733				*** **********************************		
		Timestaller.	Name (no.		_		
		·		-			
this range, choose the	m-to-egg ratio that results ratio closest to 90 per	in fertilization between	een 80 and 90	Dipercent. It	f more than	one concent	tration is within
organism health, stage	of reproductive season, s	ite conditions).	sional juugmi	ent dictates	considerai	tion of other	r factors (e.g.,
Definitive Test		Charmilas Dali-	U	- 0			
Deminive Test	_	Sperm:Egg Ratio	Usea:	<u> </u>	_		
	Time (B)		Fert.	Unfer	t.		
Sperm Added (100 μl):	17450	QC1	97	3	_		
Eggs Added (0.5 ml):	1810	QC2	94	<u>(</u>	_		
Test Ended:	<u> (830</u>	Egg Control 1	_0_	100	3		
		Egg Control 2	_0_	100	<u>)</u>		
		Cop.					
Comments:	@nodiluko	M YOOUY	000				
	BAD CAS L	30/70					
							,
	A = - 1 1					,	í
QC Check:	AC 7/5/17			i	Final Reviev	v: KAP 7/11	117
Nautilus Environmental. 43	. { 40 Vandever Avenue. San Di∈	∍go, CA 92120.					



CETIS Summary Report

Report Date:

07 Jul-17 16:57 (p 1 of 1)

Test Code:

1706-S209 | 19-6001-9623

Echinoid Spor	m Cell Fortiliza	tion Toot	450					. 501 50401	ALADONIA.	Manath	- F	
Echinola Sper	m Cell Fertiliza	uon rest	150				- Constant of the Constant of			Nautilu	s Environm	nental (CA
Batch ID: Start Date: Ending Date: Duration:	06-5489-2972 30 Jun-17 17:50 30 Jun-17 18:30 40m	O Pr O Sp	st Type: otocol: pecies: purce:	Fertilization EPA/600/R-95/ Strongylocentro Pt. Loma	` ,	itus		Analyst: Diluent. Brine: Age:		ıral Seawat Applicable	erM −1 1	JE
	18-2923-2635 30 Jun-17 08:00 30 Jun-17 11:48 10h (ららべ)	Ma B So	ode: aterial: ource: ation:	17-0741 Facility Effluent IDE Americas, Train 4				Client: Project:	IDE Carl	sbad Desal	Plant	
Comparison S	ummary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Meth	od			
03-6168-2472	Fertilization Rat	e	10	15	12.25	6.24%	10	Dunn	ett M	ultiple Com	parison Tes	st
Point Estimate	Summary						7					
Analysis ID	Endpoint		Level	%	95% LCL	95% UCL	TU	Meth	od			
05-4599-9515	Fertilization Rat	e	EC25 EC50	13.35	12.16 N/A	N/A N/A	7.48 <6.6	9 Linea		erpolation (I	CPIN)	
Test Acceptab	ility											
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	its	Over	lan	Decision		
03-6168-2472	Fertilization Rat	e		ol Resp	0.916	0.7 - NL		Yes			cceptability	Criteria
05-4599-9515	Fertilization Rat	е		ol Resp	0.916	0.7 - NL		Yes			cceptability	
03-6168-2472	Fertilization Rat	е	PMSD)	0.06236	NL - 0.25		No		Passes A	cceptability	Criteria
Fertilization Ra	ate Summary											
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std E	irr	Std Dev	CV%	%Effect
0	High Salinity Co	5	0.912	0.8405	0.9835	0.81	0.95	0.025	577	0.05762	6.32%	0.0%
0	Lab Control	5	0.916	0.8825	0.9495	0.89	0.95	0.012	809	0.02702	2.95%	-0.44%
2.5		5	0.862	0.8136	0.9104	8.0	0.9	0.017	44	0.03899	4.52%	5.48%
5		5	0.862	0.8016	0.9224	0.79	0.91	0.021	77	0.04868	5.65%	5.48%
6.06		5	0.856	0.8192	0.8928	0.81	0.89	0.013	327	0.02966	3.47%	6.14%
10		5	0.864	0.8398	0.8882	0.84	0.89	0.008	3718	0.01949	2.26%	5.26%
15		5	0.602	0.4805	0.7235	0.53	0.77	0.043	375	0.09783	16.25%	33.99%
Fertilization Ra	ate Detail											
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0 [High Salinity Co	0.94	0.93	0.93	0.81	0.95		A A STATE OF THE S				
	Lab Control	0.9	0.89	0.95	0.9	0.94						
2.5		0.8	0.88	0.9	0.88	0.85						
5		0.9	0.87	0.84	0.91	0.79						
6.06		0.85	0.87	0.86	0.89	0.81						
10		0.89	0.85	0.87	0.84	0.87						
15		0.54	0.57	0.57	0.6	0.87						
10		0.04	0.57	0.53	0.0	0.77						

@QIR ACTIOIN

Analyst: AC QA: WP 7/11/17

Analysis ID: 03-6168-2472

Report Date:

CETIS Version: CETISv1.8.7

07 Jul-17 16:57 (p 1 of 2)

	Test Code:	1706-S209 19-6001-9623
Echinoid Sperm Cell Fertilization Test 15C		Nautilus Environmental (CA)

Analyzed:	07 Jul-17 16:12	<u>Ar</u>	nalysis: Par	ametric-C	ontrol vs Treatments	Offic	cial Results	s: Yes			_
Data Transfor	m	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	ΤU	
Angular (Corre	cted)	NA	C > T	NA	NA	6.24%	10	15	12.25	10	

Endpoint: Fertilization Rate

Dunnett Multi	iple Co	omparison Test							
Control	vs	C-%	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(α:5%)
Lab Control		2.5	2.186	2.362	0.094	8	0.0704	CDF	Non-Significant Effect
		5	2.148	2.362	0.094	8	0.0758	CDF	Non-Significant Effect
		6.06*	2.43	2.362	0.094	8	0.0436	CDF	Significant Effect
		10	2.158	2.362	0.094	8	0.0742	CDF	Non-Significant Effect
		15*	9.759	2.362	0.094	8	<0.0001	CDF	Significant Effect

ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.4536385	0.09072769	5	22.81	<0.0001	Significant Effect
Error	0.09546186	0.003977577	24			
Total	0.5491003		29			

Distributional Tests											
Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)						
Variances	Bartlett Equality of Variance	7.005	15.09	0.2202	Equal Variances						
Distribution	Shapiro-Wilk W Normality	0.951	0.9031	0.1795	Normal Distribution						

Fertilization	on Rate Summary										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.916	0.8825	0.9495	0.9	0.89	0.95	0.01208	2.95%	0.0%
2.5		5	0.862	0.8136	0.9104	0.88	0.8	0.9	0.01744	4.52%	5.9%
5		5	0.862	0.8016	0.9224	0.87	0.79	0.91	0.02177	5.65%	5.9%
6.06		5	0.856	0.8192	0.8928	0.86	0.81	0.89	0.01327	3.47%	6.55%
10		5	0.864	0.8398	0.8882	0.87	0.84	0.89	0.008718	2.26%	5.68%
15		5	0.602	0.4805	0.7235	0.57	0.53	0.77	0.04375	16.25%	34.28%

Angular (0	Angular (Corrected) Transformed Summary													
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect			
0	Lab Control	5	1.28	1.217	1.343	1.249	1.233	1.345	0.02268	3.96%	0.0%			
2.5		5	1.193	1.124	1.261	1.217	1.107	1.249	0.02456	4.61%	6.81%			
5		5	1.194	1.108	1.281	1.202	1.095	1.266	0.0311	5.82%	6.69%			
6.06		5	1.183	1.131	1.235	1.187	1.12	1.233	0.01864	3.52%	7.57%			
10		5	1.194	1.158	1.229	1.202	1.159	1.233	0.01278	2.39%	6.73%			
15		5	0.8906	0.7611	1.02	0.8556	0.8154	1.071	0.04666	11.72%	30.41%			

Report Date: Test Code: 07 Jul-17 16:57 (p 2 of 2) 1706-S209 | 19-6001-9623

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) CETISv1.8.7 Analysis ID: 03-6168-2472 Endpoint: Fertilization Rate **CETIS Version:** Analyzed: 07 Jul-17 16:12 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 0.18 1.0 0.16 0.9 0.14 0.12 8.0 0.10 Fertilization Rate 0.7 0.08 0.06 0.6 0.04 0.02 0.5 0.00 0.4 -0.02 -0.04 0.3 -0.06 0.2 -0.08 -0.10 0.1 -0.12 0.0 0 LC 2.5 6.06 10 -2.5 -2.0 -1.5 -1.0 -0.5 0.0 0,5 1.0 1.5 2.0 C-% Rankits

Report Date: Test Code: 07 Jul-17 16:57 (p 1 of 1)

le:

1706-S209 | 19-6001-9623

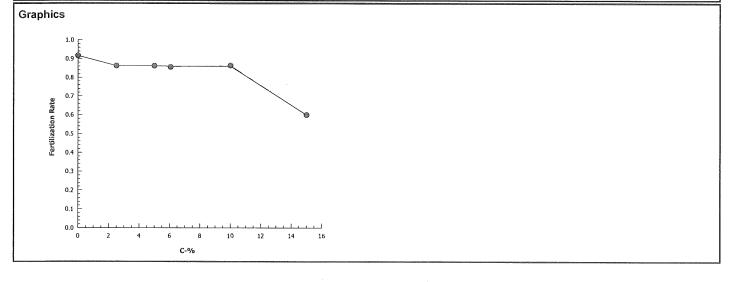
Echinoid Spe	rm Cell Fertilization	Test 15C		Nautilus Environmental (CA)	
Analysis ID:	05-4599-9515	Endpoint:	Fertilization Rate	CETIS Version:	CETISv1.8.7

Analyzed: 07 Jul-17 16:13 Analysis: Linear Interpolation (ICPIN) Official Results: Yes

Linear Interpolation Options									
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method				
Linear	Linear	1804808	1000	Yes	Two-Point Interpolation				

Point E	Point Estimates											
Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL						
EC25	13.35	12.16	N/A	7.489	NA	8.225						
EC50	>15	N/A	N/A	<6.667	NA	NA						

Fertiliza	tion Rate Summary	Calculated Variate(A/B)									
C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.916	0.89	0.95	0.01208	0.02702	2.95%	0.0%	458	500
2.5		5	0.862	8.0	0.9	0.01744	0.03899	4.52%	5.9%	431	500
5		5	0.862	0.79	0.91	0.02177	0.04868	5.65%	5.9%	431	500
6.06		5	0.856	0.81	0.89	0.01327	0.02966	3.47%	6.55%	428	500
10		5	0.864	0.84	0.89	0.008718	0.01949	2.26%	5.68%	432	500
15		5	0.602	0.53	0.77	0.04375	0.09783	16.25%	34.28%	300	500



Report Date:

07 Jul-17 16:58 (p 1 of 1)

Test Code:

1706-S209 | 19-6001-9623

	The second secon					_	1621	Code:	170)6-S209 1	3-0001-302
Echinoid Sp	erm Cell Fertiliz	ation Test	15C	İST					Nautilu	s Environ	mental (CA)
Analysis ID:	04-9502-9462	. Ei	ndpoint: Fe	rtilization Ra	ite		CFT	IS Version:	: CETISv1	8.7	
Analyzed:	07 Jul-17 16:	58 A ı	nalysis: Pa	rametric Bio	equivalence	-Two Samp		cial Results			
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	ΤU
Angular (Cori	rected)	NA	C*b < T	NA	NA	0.75	6.69%	10	15	12.25	10
TST-Welch's	t Test										
Control	vs C-%		Test Stat	Critical	MSD DE	P-Value	P-Type	Decision	(a:5%)		
Lab Control	2.5*		7.791	1.895	0.057 7	<0.0001	CDF		ificant Effect		
	5*		6.61	1.943	0.069 6	0.0003	CDF	_	ificant Effect		
	6.06*		8.84	1.895	0.048 7	<0.0001	CDF		ificant Effect		
	10*		10.99	1.895	0.040 7	<0.0001	CDF		ificant Effect		
	15		-1.395	2.015	0.100 5	0.8891	CDF	Significan			
ANOVA Table	e										
Source	Sum Sqt	uares	Mean Sq	uare	DF	F Stat	P-Value	Decision	(α:5%)		
Between	0.453638	5	0.0907276	5 9	5	22.81	<0.0001	Significan	t Effect		
Error	0.095461	86	0.003977	577	24			J			
Total	0.549100	3			29	omm.					
Distributiona	ıl Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision(α:1%)			
Attribute Variances		Equality of '	Variance	Test Stat 7.005	Critical 15.09	P-Value 0.2202	Decision(Equal Var				
	Bartlett E	Equality of ' Wilk W No						iances			
Variances Distribution	Bartlett E			7.005	15.09	0.2202	Equal Var	iances			
Variances Distribution Fertilization	Bartlett E Shapiro-			7.005	15.09	0.2202	Equal Var	iances	Std Err	CV%	%Effect
Variances Distribution Fertilization	Bartlett E Shapiro- Rate Summary	Wilk W No	rmality	7.005 0.951	15.09 0.9031	0.2202 0.1795	Equal Var Normal Di	iances stribution	Std Err 0.01208		
Variances Distribution Fertilization I C-% 0 2.5	Bartlett t Shapiro- Rate Summary Control Type	Wilk W No	rmality Mean	7.005 0.951 95% LCL	15.09 0.9031 95% UCL	0.2202 0.1795 Median	Equal Var Normal Di	iances stribution		CV% 2.95% 4.52%	0.0%
Variances Distribution Fertilization I C-% 0 2.5	Bartlett t Shapiro- Rate Summary Control Type	Count 5	Mean 0.916	7.005 0.951 95% LCL 0.8825	15.09 0.9031 95% UCL 0.9495	0.2202 0.1795 Median 0.9	Equal Var Normal Di Min 0.89	iances stribution Max 0.95	0.01208	2.95%	
Variances Distribution Fertilization C-% 0 2.5 5 6.06	Bartlett t Shapiro- Rate Summary Control Type	Count 5 5 5 5	Mean 0.916 0.862	7.005 0.951 95% LCL 0.8825 0.8136	15.09 0.9031 95% UCL 0.9495 0.9104	0.2202 0.1795 Median 0.9 0.88	Equal Var Normal Di Min 0.89 0.8	Max 0.95 0.9	0.01208 0.01744	2.95% 4.52%	0.0% 5.9%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10	Bartlett t Shapiro- Rate Summary Control Type	Count 5 5 5 5 5 5 5 5 5	Mean 0.916 0.862 0.862	7.005 0.951 95% LCL 0.8825 0.8136 0.8016	15.09 0.9031 95% UCL 0.9495 0.9104 0.9224	0.2202 0.1795 Median 0.9 0.88 0.87	Min 0.89 0.8 0.79	Max 0.95 0.9 0.91	0.01208 0.01744 0.02177 0.01327	2.95% 4.52% 5.65% 3.47%	0.0% 5.9% 5.9%
Variances Distribution Fertilization C-% 0 2.5 5 6.06	Bartlett t Shapiro- Rate Summary Control Type	Count 5 5 5 5	Mean 0.916 0.862 0.862 0.856	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192	95% UCL 0.9495 0.9104 0.9224 0.8928	0.2202 0.1795 Median 0.9 0.88 0.87 0.86	Equal Var Normal Di Min 0.89 0.8 0.79 0.81	Max 0.95 0.9 0.91 0.89	0.01208 0.01744 0.02177	2.95% 4.52% 5.65%	0.0% 5.9% 5.9% 6.55%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10	Bartlett t Shapiro- Rate Summary Control Type	Count 5 5 5 5 5 5 5 5 5	Mean 0.916 0.862 0.862 0.856 0.864 0.602	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192 0.8398	95% UCL 0.9495 0.9104 0.9224 0.8928 0.8882	0.2202 0.1795 Median 0.9 0.88 0.87 0.86 0.87	Min 0.89 0.79 0.81 0.84	Max 0.95 0.91 0.89 0.89	0.01208 0.01744 0.02177 0.01327 0.008718	2.95% 4.52% 5.65% 3.47% 2.26%	0.0% 5.9% 5.9% 6.55% 5.68%
Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Core	Bartlett B Shapiro- Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 cmed Sumi	Mean 0.916 0.862 0.862 0.856 0.864 0.602	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192 0.8398	95% UCL 0.9495 0.9104 0.9224 0.8928 0.8882	0.2202 0.1795 Median 0.9 0.88 0.87 0.86 0.87	Min 0.89 0.79 0.81 0.84	Max 0.95 0.91 0.89 0.89	0.01208 0.01744 0.02177 0.01327 0.008718	2.95% 4.52% 5.65% 3.47% 2.26%	0.0% 5.9% 5.9% 6.55% 5.68%
Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Core	Bartlett E Shapiro- Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 7 med Sumi	Mean 0.916 0.862 0.862 0.856 0.864 0.602	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192 0.8398 0.4805	95% UCL 0.9495 0.9104 0.9224 0.8928 0.8882 0.7235	0.2202 0.1795 Median 0.9 0.88 0.87 0.86 0.87 0.57	Min 0.89 0.8 0.79 0.81 0.84 0.53	Max 0.95 0.91 0.89 0.89 0.77	0.01208 0.01744 0.02177 0.01327 0.008718 0.04375	2.95% 4.52% 5.65% 3.47% 2.26% 16.25%	0.0% 5.9% 5.9% 6.55% 5.68% 34.28%
Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cort	Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 Count Count Count 5 5 5	Mean 0.916 0.862 0.862 0.856 0.864 0.602 mary Mean	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192 0.8398 0.4805	95% UCL 0.9495 0.9104 0.9224 0.8928 0.7235	0.2202 0.1795 Median 0.9 0.88 0.87 0.86 0.87 0.57	Equal Var Normal Di Min 0.89 0.8 0.79 0.81 0.84 0.53	Max 0.95 0.91 0.89 0.77	0.01208 0.01744 0.02177 0.01327 0.008718 0.04375	2.95% 4.52% 5.65% 3.47% 2.26% 16.25%	0.0% 5.9% 5.9% 6.55% 5.68% 34.28% %Effect 0.0%
Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cort C-% 0 2.5 5	Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 cmed Summed Su	Mean 0.916 0.862 0.862 0.856 0.864 0.602 mary Mean 1.28	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192 0.8398 0.4805 95% LCL 1.217	95% UCL 0.9495 0.9104 0.9224 0.8928 0.7235 95% UCL 1.343	0.2202 0.1795 Median 0.9 0.88 0.87 0.86 0.87 0.57 Median 1.249	Min 0.89 0.8 0.79 0.81 0.53 Min 1.233	Max 0.95 0.91 0.89 0.77 Max 1.345	0.01208 0.01744 0.02177 0.01327 0.008718 0.04375 Std Err 0.02268	2.95% 4.52% 5.65% 3.47% 2.26% 16.25% CV% 3.96%	0.0% 5.9% 5.9% 6.55% 5.68% 34.28% %Effect 0.0% 6.81%
Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Corr C-% 0 2.5 5	Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 Count Count Count 5 5 5	Mean 0.916 0.862 0.862 0.856 0.864 0.602 mary Mean 1.28 1.193	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192 0.8398 0.4805 95% LCL 1.217 1.124	95% UCL 0.9495 0.9104 0.9224 0.8928 0.8882 0.7235 95% UCL 1.343 1.261	0.2202 0.1795 Median 0.9 0.88 0.87 0.86 0.87 0.57 Median 1.249 1.217	Min 0.89 0.8 0.79 0.81 0.84 0.53 Min 1.233 1.107	Max 0.95 0.91 0.89 0.77 Max 1.345 1.249	0.01208 0.01744 0.02177 0.01327 0.008718 0.04375 Std Err 0.02268 0.02456	2.95% 4.52% 5.65% 3.47% 2.26% 16.25% CV% 3.96% 4.61% 5.82%	0.0% 5.9% 5.9% 6.55% 5.68% 34.28% %Effect 0.0% 6.81% 6.69%
Variances Distribution Fertilization I C-% 0 2.5 5 6.06 10 15 Angular (Core	Bartlett E Shapiro- Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 Count Count Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.916 0.862 0.862 0.856 0.864 0.602 mary Mean 1.28 1.193 1.194	7.005 0.951 95% LCL 0.8825 0.8136 0.8016 0.8192 0.8398 0.4805 95% LCL 1.217 1.124 1.108	95% UCL 0.9495 0.9104 0.9224 0.8928 0.8882 0.7235 95% UCL 1.343 1.261 1.281	0.2202 0.1795 Median 0.9 0.88 0.87 0.86 0.87 0.57 Median 1.249 1.217 1.202	Min 0.89 0.8 0.79 0.81 0.84 0.53 Min 1.233 1.107 1.095	Max 0.95 0.9 0.91 0.89 0.77 Max 1.345 1.249 1.266	0.01208 0.01744 0.02177 0.01327 0.008718 0.04375 Std Err 0.02268 0.02456 0.0311	2.95% 4.52% 5.65% 3.47% 2.26% 16.25% CV% 3.96% 4.61%	0.0% 5.9% 5.9% 6.55% 5.68% 34.28% %Effect 0.0% 6.81%

CETIS Test Data Worksheet

Report Date:

29 Jun-17 18:57 (p 1 of 1)

Test Code: 1706-52049-6001-9623/74D386A7

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

Start Date: End Date: Sample Date: 30 Jun-17

30 Jun-17 30 Jun-17 Species: Strongylocentrotus purpuratus **Protocol**: EPA/600/R-95/136 (1995)

Sample Code: 17- 074 Sample Source: IDE Americas, Inc.

	te: 30 J				al: Facility Eff	
Ç-%	Code	Rep	Pos	# Counted	# Fertilized	Notes (B)
			216	100	87	CG 7/3/17
			217	100	90	
			218	100	94	
			219	100	85	
			220	(00	89.	
			221	100	88	
			222	100	85	
			223	iou	86	
			224	i00	OS-057	
			225	100	87	
			226	100	80	
			227	100	91	
			228	100	77	
			229	100	81	
			230	100	90,	*
			231	100	90	
			232	100	54	
			233	(00	87	V
			234	OOi	53	(G 7/6/17)
			235	00)	87	
			236	100	95	
			237	100	60	
			238	100	79	
			239	100	84	
			240	100	89	
			241	100	89	
			242	100	88	
			243	100	90	
			244	100	9b 84	
			245	(00)	85	<u> </u>

A CGQ187/3/17 3) Q18 HK 7/7/17

> HSC A 100 94 B 100 93 C 100 93 D 100 18 E 100 95

CETIS Test Data Worksheet

Report Date:

29 Jun-17 18:57 (p 1 of 1)

Test Code: \706~526919-6001-9623/74D386A7

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: End Date:

Sample Date: 30 Jun-17

30 Jun-17 30 Jun-17

Species: Strongylocentrotus purpuratus

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

Material: Facility Effluent

Sample Code:

Sample Source: IDE Americas, Inc.

Sample Station: Train

C-%	Code	Rep	Pos	# Counted		Notes
0	LC	1	243	100	91	Pro 7/1/17
0	LC	2	220			
0	LC	3	236			
0	LC	4	231			
0	LC	5	218			
2.5		1	226			
2.5		2	242			
2,5		3	217			
2.5		4	221			
2.5		5	222			
5		1	230			
5		2	235			
5		3	244			
5		4	227			
5		5	238			
6.06		1	245	100	84	
6.06		2	225			
6.06		3	223			
6.06		4	240			
6.06		5	229			
10		1	241			
10		2	219			
10		3	216			
10		4	239			
10		5	233			
15		1	232			
15		2	224			
15		3	234			
15		4	237			
15		5	228			

QC: CG BAO ON8 6/30/17

Water Quality Measurements

-					
C	RI	Δ	E/B	Ŧ.	•

IDE

Test Species: S. purpuratus

Sample ID:

Train 9 4

Start Date/Time: 6/30/2017 \

Sample Log No.:

17-0741

End Date/Time: 6/30/2017

Dilutions made by:

AD

Test No: 1705-5209

			Analyst:	CH
			eadings	
Concentration %	DO (mg/L)	pH (units)	Salinity (ppt)	Temperature (°C)
Lab Control	7.7	7.87	34.0	15.3
2.5	7.9	7.87	34.8	15.2
5.0	8.0	7.87	35.7	15.2
6.06	7.9	7.87	36.1	15.2
10	\$ 08.2	@187.88	9405 37.2	15.7
15	8.0	7.88	39.2	15.2
HSC	8.7	7.98	39.0	14.4

Comments:

ACHORS 6/30/17 DIWHORS Made with M-INE

QC Check:

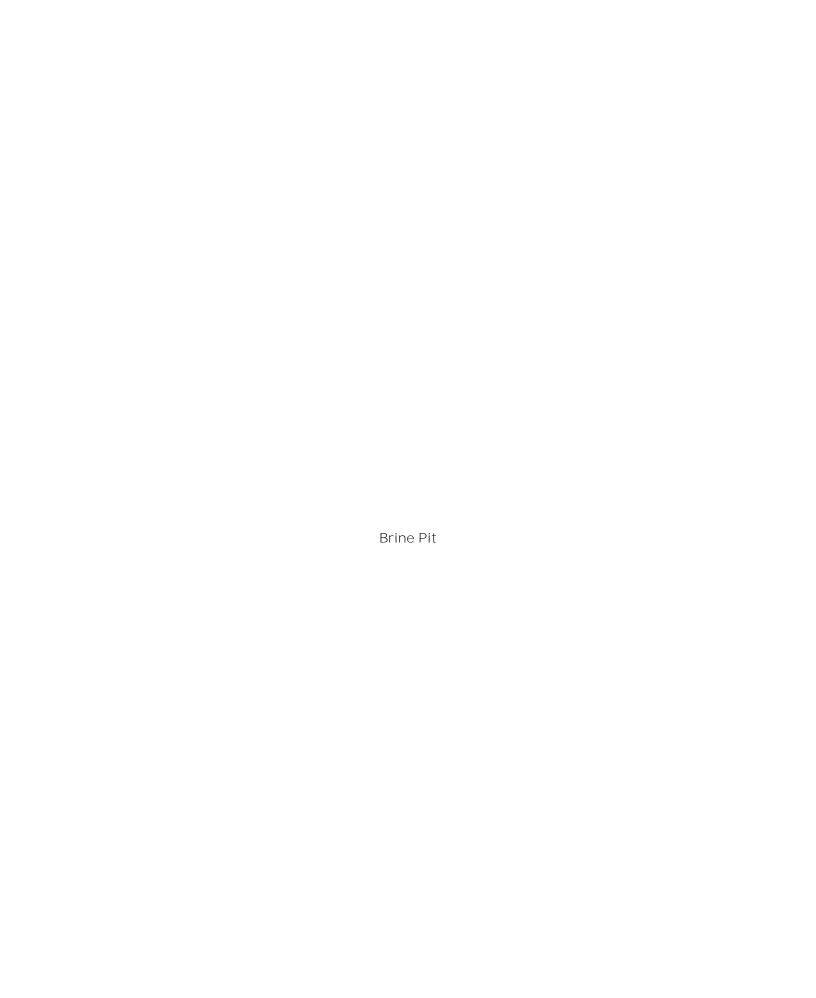
AC7/7/17

Final Review: PTP 7/11/17

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	1DE Train 4 _1706-520	9	Start Date/Time: 6/30/2017 / End Date/Time: 6/30/2017 / Species: S. purpuratus	1750 1830
Tech initials: Injection Time:	1650		Animal Source: Pt. Loma Date Collected: (0)(0)	1
Sperm Absorbance at	400 nm: <u> 1, 009</u>	(target range of 0.8 - 1.0 for der	sity of 4x10 ⁶ sperm/ml)	
Eggs Counted:	R8 Mea	Dec (1)	eggs/ml	
Initial density: Final density:	4000 eggs/ml	= dilution factor - 1.0 part egg stock parts seawater	egg stockml seawater ml	
Prepare the embryo sto existing stock (1 part) a	ock according to the calcu and 125 ml of dilution wate	lated dilution factor. For example, i er (1.25 parts).	f the dilution factor is 2.25, use 100 m	l of
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1 1600 50 40 0.0 10	30 20	$\frac{200:1}{10}$ $\frac{200:1}{5.0}$ $\frac{100:1}{2.5}$ $\frac{50}{1.0}$	0:1 .25 3.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1710 1723 1733	Rangefinder Ratio: Fert.	Unfert.	
this range, choose the	m-to-egg ratio that results e ratio closest to 90 per of reproductive season, s	cent unless professional judgmen	ercent. If more than one concentratio t dictates consideration of other fac	n is within tors (e.g.,
Definitive Test		Sperm:Egg Ratio Used:		
Sperm Added (100 μl): Eggs Added (0.5 ml): Test Ended:	Time (3) 17450 1810	QC1 QC2 Egg Control 1 Egg Control 2	Unfert. 3 100 100	
Comments:	Phodiuho	n voouve el	·	
QC Check: Nautilus Environmental. 43	AC 7/5/17 40 Vandever Avenue. San Die	 ego, CA 92120.	Final Review: VFP 7/11/	1



CETIS Summary Report

Report Date:

07 Jul-17 15:15 (p 1 of 1)

Test Code:

1706-S208 | 16-3408-8529

								Test Code.	RANGES SURV	171	0200 11	J-3400-0328
Echinoid Spe	erm Cell Fertiliza	ation T	est 15C		42.00					Nautilu	s Environn	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	16-4465-8123 30 Jun-17 17:5 30 Jun-17 18:3 40m		Test Type: Protocol: Species: Source:	Fertilization EPA/600/R-95 Strongylocentr Pt. Loma	` '	atus		Analyst: Diluent: Brine: Age:		ural Seawat Applicable	Œr (~)-	VF
· .	09-4841-4586 30 Jun-17 08:0 : 30 Jun-17 11:4 10h (6.5°C)		Code: Material: Source: Station:	17-0740 Facility Effluen IDE Americas, Brine Pit				Client: Project:	IDE Carl	sbad Desal	Plant	
Comparison	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	ΤU	Meth	od			
04-9634-7083	Fertilization Ra	ite	2.5	5	3.536	10.4%	40			lultiple Com	parison Tes	st
Point Estimat	e Summary											
Analysis ID	Endpoint		Level	%	95% LCL	95% UCL	TU	Meth	od			
03-0782-7528	Fertilization Ra	te	EC25 EC50	7.896 13.22	5.755 11.39	9.888 14.98	12.6 7.56		r Inte	erpolation (I	CPIN)	
Test Acceptal	oility											
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	its	Overl	lap	Decision		
03-0782-7528	Fertilization Ra	te	Contro	ol Resp	0.886	0.7 - NL		Yes	·	Passes A	cceptability	Criteria
04-9634-7083			Contro	ol Resp	0.886	0.7 - NL		Yes		Passes A	cceptability	Criteria
04-9634-7083	Fertilization Ra	te	PMSD		0.1043	NL - 0.25		No		Passes A	cceptability	Criteria
Fertilization F	tate Summary											
C-%	Control Type	Cour	nt Mean	95% LCL	95% UCL	Min	Max	Std E	rr	Std Dev	CV%	%Effect
0	Lab Control	5	0.886	0.8154	0.9566	0.81	0.96	0.025	42	0.05683	6.42%	0.0%
2.5		5	0.884	0.7968	0.9712	0.81	0.98	0.031	4	0.07021	7.94%	0.23%
5		5	0.732	0.6414	0.8226	0.65	0.83	0.032	62	0.07294	9.96%	17.38%
6.06		5	0.734	0.6668	0.8012	0.66	0.81	0.024	21	0.05413	7.38%	17.16%
10		5	0.586	0.4928	0.6792	0.48	0.67	0.033	56	0.07503	12.8%	33.86%
15		5	0.364	0.2577	0.4703	0.22	0.44	0.038	29	0.08562	23.52%	58.92%
Fertilization R	ate Detail											Ald Comment
C-%	Control Type	Rep '	1 Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	0.87	0.87	0.96	0.92	0.81						
2.5		0.98	0.82	0.9	0.91	0.81						
5		0.65	0.83	0.67	0.75	0.76						
6.06		0.81	0.73	0.66	0.72	0.75						
10		0.55	0.64	0.59	0.67	0.48						
15		0.44	0.39	0.36	0.22	0.41						



Analyst: AC QA: MP7/11/17

Report Date:

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

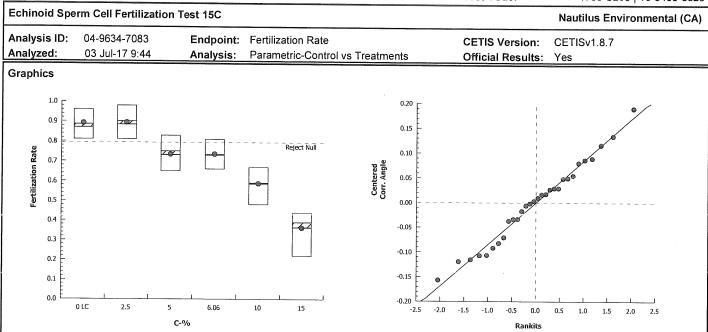
Test Code:

1706-S208 | 16-3408-8529

Echinoid Sp	erm Cell Fertiliz	ation Test	15C						Nautilus	Environn	nental (CA)
Analysis ID:	04-9634-7083	En	dpoint: Fer	tilization Ra	:e		CET	IS Version:	CETISv1	.8.7	
Analyzed:	03 Jul-17 9:44	. An	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		10.4%	2.5	5	3.536	40
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(α:5%)		
Lab Control	2.5		-0.06711	2.362	0.136 8	0.8524	CDF	Non-Signif	icant Effect	·····	
	5*		3.568	2.362	0.136 8	0.0034	CDF	Significant			
	6.06*		3.552	2.362	0.136 8	0.0035	CDF	Significant			
	10*		6.292	2.362	0.136 8	<0.0001	CDF	Significant			
	15*		10.23	2.362	0.136 8	<0.0001	CDF	Significant			
ANOVA Tabl	e							and the second s			
Source	Sum Squ	ıares	Mean Squ	ıare	DF	F Stat	P-Value	Decision(a:5%)		
Between	1.280516	1	0.2561031		5	30.81	<0.0001	Significant	Effect	AND THE REAL PROPERTY AND THE PROPERTY A	
Error	0.199524	5	0.0083135	522	24						
Total	1.48004		1		29	_					
Distributiona	al Tests										
Distributiona Attribute	al Tests Test			Test Stat	Critical	P-Value	Decision((α:1%)			
	Test	Equality of ∖	/ariance	Test Stat	Critical 15.09	P-Value 0.8457	Decision(Equal Var	·			
Attribute	Test Bartlett E	Equality of \						iances			
Attribute Variances Distribution	Test Bartlett E			2.025	15.09	0.8457	Equal Var	iances			
Attribute Variances Distribution Fertilization	Test Bartlett E Shapiro-			2.025	15.09	0.8457	Equal Var	iances	Std Err	CV%	%Effect
Attribute Variances Distribution	Test Bartlett I Shapiro-	Wilk W Nor	mality	2.025 0.9827	15.09 0.9031	0.8457 0.8920	Equal Var Normal Di	iances istribution	Std Err 0.02542	CV% 6.42%	%Effect 0.0%
Attribute Variances Distribution Fertilization C-%	Test Bartlett B Shapiro- Rate Summary Control Type	Wilk W Nor	mality Mean	2.025 0.9827 95% LCL	15.09 0.9031 95% UCL	0.8457 0.8920 Median	Equal Var Normal Di	iances stribution			***
Attribute Variances Distribution Fertilization C-% 0	Test Bartlett B Shapiro- Rate Summary Control Type	Count	Mean 0.886	2.025 0.9827 95% LCL 0.8154	15.09 0.9031 95% UCL 0.9566	0.8457 0.8920 Median 0.87	Equal Var Normal Di Min 0.81	iances istribution Max 0.96	0.02542	6.42%	0.0%
Attribute Variances Distribution Fertilization C-% 0 2.5	Test Bartlett B Shapiro- Rate Summary Control Type	Count 5 5	Mean 0.886 0.884	2.025 0.9827 95% LCL 0.8154 0.7968	15.09 0.9031 95% UCL 0.9566 0.9712	0.8457 0.8920 Median 0.87 0.9	Equal Var Normal Di Min 0.81 0.81	Max 0.96 0.98	0.02542 0.0314	6.42% 7.94%	0.0% 0.23%
Attribute Variances Distribution Fertilization C-% 0 2.5 5	Test Bartlett B Shapiro- Rate Summary Control Type	Count 5 5 5	Mean 0.886 0.884 0.732	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414	95% UCL 0.9566 0.9712 0.8226	0.8457 0.8920 Median 0.87 0.9 0.75	Min 0.81 0.65	Max 0.96 0.98 0.83	0.02542 0.0314 0.03262	6.42% 7.94% 9.96%	0.0% 0.23% 17.38%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06	Test Bartlett B Shapiro- Rate Summary Control Type	Count 5 5 5 5	Mean 0.886 0.884 0.732 0.734	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668	95% UCL 0.9566 0.9712 0.8226 0.8012	0.8457 0.8920 Median 0.87 0.9 0.75 0.73	Equal Var Normal Di Min 0.81 0.81 0.65 0.66	Max 0.96 0.98 0.83 0.81	0.02542 0.0314 0.03262 0.02421	6.42% 7.94% 9.96% 7.38%	0.0% 0.23% 17.38% 17.16%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15	Test Bartlett B Shapiro- Rate Summary Control Type	Count 5 5 5 5 5 5	Mean 0.886 0.884 0.732 0.734 0.586 0.364	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668 0.4928	95% UCL 0.9566 0.9712 0.8226 0.8012 0.6792	0.8457 0.8920 Median 0.87 0.9 0.75 0.73 0.59	Min 0.81 0.65 0.66 0.48	Max 0.96 0.83 0.81 0.67	0.02542 0.0314 0.03262 0.02421 0.03356	6.42% 7.94% 9.96% 7.38% 12.8%	0.0% 0.23% 17.38% 17.16% 33.86%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Core	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5	Mean 0.886 0.884 0.732 0.734 0.586 0.364	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668 0.4928	95% UCL 0.9566 0.9712 0.8226 0.8012 0.6792	0.8457 0.8920 Median 0.87 0.9 0.75 0.73 0.59	Min 0.81 0.65 0.66 0.48	Max 0.96 0.83 0.81 0.67	0.02542 0.0314 0.03262 0.02421 0.03356	6.42% 7.94% 9.96% 7.38% 12.8%	0.0% 0.23% 17.38% 17.16% 33.86%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Test Bartlett E Shapiro- 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 0.886 0.884 0.732 0.734 0.586 0.364	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668 0.4928 0.2577	95% UCL 0.9566 0.9712 0.8226 0.8012 0.6792 0.4703	0.8457 0.8920 Median 0.87 0.9 0.75 0.73 0.59 0.39	Min 0.81 0.65 0.66 0.48 0.22	Max 0.96 0.98 0.83 0.81 0.67 0.44	0.02542 0.0314 0.03262 0.02421 0.03356 0.03829	6.42% 7.94% 9.96% 7.38% 12.8% 23.52%	0.0% 0.23% 17.38% 17.16% 33.86% 58.92%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 5 rmed Sumr	Mean 0.886 0.884 0.732 0.734 0.586 0.364 mary Mean	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668 0.4928 0.2577	95% UCL 0.9566 0.9712 0.8226 0.8012 0.6792 0.4703	0.8457 0.8920 Median 0.87 0.9 0.75 0.73 0.59 0.39	Min 0.81 0.65 0.66 0.48 0.22	Max 0.96 0.98 0.83 0.81 0.67 0.44	0.02542 0.0314 0.03262 0.02421 0.03356 0.03829	6.42% 7.94% 9.96% 7.38% 12.8% 23.52%	0.0% 0.23% 17.38% 17.16% 33.86% 58.92%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 7 Trmed Sumr Count 5	Mean 0.886 0.884 0.732 0.734 0.586 0.364 mary Mean 1.235	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668 0.4928 0.2577 95% LCL 1.118	95% UCL 0.9566 0.9712 0.8226 0.8012 0.6792 0.4703 95% UCL 1.353	0.8457 0.8920 Median 0.87 0.9 0.75 0.73 0.59 0.39 Median 1.202	Min 0.81 0.65 0.66 0.48 0.22 Min 1.12	Max 0.96 0.98 0.83 0.81 0.67 0.44 Max 1.369	0.02542 0.0314 0.03262 0.02421 0.03356 0.03829 Std Err 0.04239	6.42% 7.94% 9.96% 7.38% 12.8% 23.52% CV% 7.67%	0.0% 0.23% 17.38% 17.16% 33.86% 58.92% %Effect 0.0%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 7 Trmed Sumr Count 5 5 5	Mean 0.886 0.884 0.732 0.734 0.586 0.364 nary Mean 1.235 1.239	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668 0.4928 0.2577 95% LCL 1.118 1.084	95% UCL 0.9566 0.9712 0.8226 0.8012 0.6792 0.4703 95% UCL 1.353 1.394	0.8457 0.8920 Median 0.87 0.9 0.75 0.73 0.59 0.39 Median 1.202 1.249	Min 0.81 0.65 0.66 0.48 0.22 Min 1.12 1.12	Max 0.96 0.98 0.83 0.81 0.67 0.44 Max 1.369 1.429	0.02542 0.0314 0.03262 0.02421 0.03356 0.03829 Std Err 0.04239 0.05587	6.42% 7.94% 9.96% 7.38% 12.8% 23.52% CV% 7.67% 10.08%	0.0% 0.23% 17.38% 17.16% 33.86% 58.92% %Effect 0.0% -0.31%
Attribute Variances Distribution Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5 5	Test Bartlett E Shapiro- Rate Summary Control Type Lab Control rrected) Transfor Control Type	Count 5 5 5 5 7 Trmed Sumr Count 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.886 0.884 0.732 0.734 0.586 0.364 mary Mean 1.235 1.239 1.03	2.025 0.9827 95% LCL 0.8154 0.7968 0.6414 0.6668 0.4928 0.2577 95% LCL 1.118 1.084 0.9256	95% UCL 0.9566 0.9712 0.8226 0.8012 0.6792 0.4703 95% UCL 1.353 1.394 1.134	0.8457 0.8920 Median 0.87 0.9 0.75 0.73 0.59 0.39 Median 1.202 1.249 1.047	Min 0.81 0.65 0.66 0.48 0.22 Min 1.12 1.12 0.9377	Max 0.96 0.98 0.83 0.81 0.67 0.44 Max 1.369 1.429 1.146	0.02542 0.0314 0.03262 0.02421 0.03356 0.03829 Std Err 0.04239 0.05587 0.03749	6.42% 7.94% 9.96% 7.38% 12.8% 23.52% CV% 7.67% 10.08% 8.14%	0.0% 0.23% 17.38% 17.16% 33.86% 58.92% %Effect 0.0% -0.31% 16.65%

Report Date: Test Code: 07 Jul-17 15:15 (p 2 of 2)

1706-S208 | 16-3408-8529



Report Date:

07 Jul-17 15:15 (p 1 of 1)

1706-S208 | 16-3408-8529

Test Code:

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

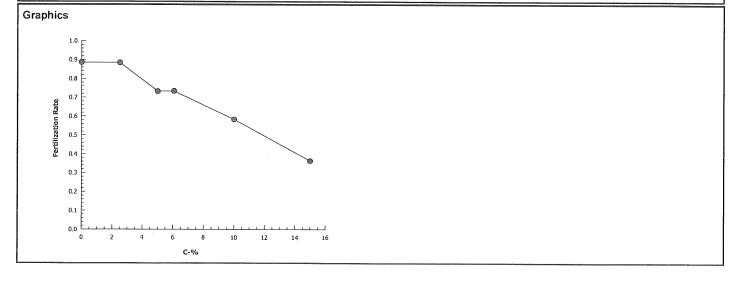
Analysis ID: 03-0782-7528 Endpoint: Fertilization Rate CETIS Version: CETISv1.8.7

Analyzed: 03 Jul-17 9:44 Analysis: Linear Interpolation (ICPIN) Official Results: Yes

Linear Interpola	Linear Interpolation Options										
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method						
Linear	Linear	1754982	1000	Yes	Two-Point Interpolation						

Point E	stimates			M-1888		
Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
EC25	7.896	5.755	9.888	12.66	10.11	17.38
EC50	13.22	11.39	14.98	7.564	6.676	8.78

Fertilizat	tion Rate Summary		Calculated Variate(A/B)									
C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В	
0	Lab Control	5	0.886	0.81	0.96	0.02542	0.05683	6.42%	0.0%	443	500	
2.5		5	0.884	0.81	0.98	0.0314	0.07021	7.94%	0.23%	442	500	
5		5	0.732	0.65	0.83	0.03262	0.07294	9.96%	17.38%	366	500	
6.06		5	0.734	0.66	0.81	0.02421	0.05413	7.38%	17.16%	367	500	
10		5	0.586	0.48	0.67	0.03356	0.07503	12.8%	33.86%	292	500	
15		5	0.364	0.22	0.44	0.03829	0.08562	23.52%	58.92%	182	500	



Report Date: Test Code: 07 Jul-17 15:16 (p 1 of 1)

1706-S208 | 16-3408-8529

								rest	Code:	1700	5-5208 16	-3408-8529
Echinoid Spe	erm Cell Fe	tilization Te	est 15C		5T					Nautilus	Environm	ental (CA)
Analysis ID:	15-3102-4		· -	Fertilization					S Version		8.7	
Analyzed:	07 Jul-17	15:16	Analysis:	Parametric	Bioequivale	nce-	Two Sampl	e Offic	ial Results	: Yes		
Data Transfo	orm	Zeta	Alt H	yp Trials	Seed		TST b	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	rected)	NA	C*b <	T NA	NA		0.75	7.12%	6.06	10	7.785	16.5
TST-Welch's	t Test					-						
Control	vs C-%	ı	Test	Stat Critica	I MSD	DF	P-Value	P-Type	Decision	(α:5%)		
Lab Control	2.5*		4.865	1.943	0.125	6	0.0014	CDF	Non-Sign	ificant Effect		
	5*		2.098	1.895	0.093	7	0.0371	CDF	Non-Sign	ificant Effect		
	6.06	*	2.466	1.895	0.08	7	0.0215	CDF	Non-Sign	ificant Effect		
	10		-1.15 ⁻	7 1.895	0.088	7	0.8574	CDF	Significa	nt Effect		
	15		-5.38	3 1.895	0.099	7	0.9995	CDF	Significa	nt Effect		
ANOVA Tabl	le											
Source	Sum	Squares	Mean	Square	DF		F Stat	P-Value	Decision	(α:5%)		
Between	1.28)516	0.256	1031	5		30.81	<0.0001	Significa	nt Effect		
Error	0.19	95245	0.008	313522	24							
Total	1.48	004			29		_					
Distributiona	al Tests											
Attribute	Tes	t		Test S	tat Critica	al	P-Value	Decision((α:1%)			
Variances	Bar	lett Equality	of Variance	2.025	15.09		0.8457	Equal Var	iances			
Distribution	Sha	piro-Wilk W	Normality	0.9827	0.9031	1	0.8920	Normal D	istribution			
Fertilization	Rate Summ	ary										,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
C-%	Control Ty	pe Cour	nt Mear	95% L	CL 95% L	JCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Contro	ol 5	0.886	0.8154	0.9566	3	0.87	0.81	0.96	0.02542	6.42%	0.0%
2.5		E		0.7000								0.23%
		5	0.884	0.7968	0.9712	2	0.9	0.81	0.98	0.0314	7.94%	0.2370
5		5 5	0.884 0.732				0.9 0.75	0.81 0.65	0.98 0.83	0.0314 0.03262	7.94% 9.96%	17.38%
				0.6414	0.8226	3						
5		5	0.732	0.6414 0.6668	0.8226 0.8012	5 2	0.75	0.65	0.83	0.03262	9.96%	17.38%
5 6.06		5 5	0.732 0.734	0.6414 0.6668 0.4928	0.8226 0.8012 0.6792	6 2 2	0.75 0.73	0.65 0.66	0.83 0.81	0.03262 0.02421	9.96% 7.38%	17.38% 17.16%
5 6.06 10	rrected) Trai	5 5 5 5	0.732 0.734 0.586 0.364	0.6414 0.6668 0.4928	0.8226 0.8012 0.6792	6 2 2	0.75 0.73 0.59	0.65 0.66 0.48	0.83 0.81 0.67	0.03262 0.02421 0.03356	9.96% 7.38% 12.8%	17.38% 17.16% 33.86%
5 6.06 10 15	rrected) Trai	5 5 5 5 nsformed S	0.732 0.734 0.586 0.364 ummary	0.6414 0.6668 0.4928 0.2577	0.8226 0.8012 0.6792 0.4703	6 2 2 3	0.75 0.73 0.59	0.65 0.66 0.48	0.83 0.81 0.67	0.03262 0.02421 0.03356	9.96% 7.38% 12.8%	17.38% 17.16% 33.86%
5 6.06 10 15 Angular (Cor	•	5 5 5 nsformed So	0.732 0.734 0.586 0.364 ummary	0.6414 0.6668 0.4928 0.2577	0.8226 0.8012 0.6792 0.4703	6 2 2 3	0.75 0.73 0.59 0.39	0.65 0.66 0.48 0.22	0.83 0.81 0.67 0.44	0.03262 0.02421 0.03356 0.03829	9.96% 7.38% 12.8% 23.52%	17.38% 17.16% 33.86% 58.92%
5 6.06 10 15 Angular (Cor	Control Ty	5 5 5 nsformed So pe Cour	0.732 0.734 0.586 0.364 ummary nt Mear	0.6414 0.6668 0.4928 0.2577 95% L 1.118	0.8226 0.8012 0.6792 0.4703	6 2 2 3	0.75 0.73 0.59 0.39	0.65 0.66 0.48 0.22	0.83 0.81 0.67 0.44	0.03262 0.02421 0.03356 0.03829 Std Err	9.96% 7.38% 12.8% 23.52%	17.38% 17.16% 33.86% 58.92%
5 6.06 10 15 Angular (Cor C-%	Control Ty	5 5 5 5 nsformed So pe Cour	0.732 0.734 0.586 0.364 ummary nt Mear 1.235	0.6414 0.6668 0.4928 0.2577 95% L	0.8226 0.8012 0.6792 0.4703 CL 95% L 1.353 1.394	6 2 2 3	0.75 0.73 0.59 0.39 Median	0.65 0.66 0.48 0.22 Min 1.12	0.83 0.81 0.67 0.44 Max 1.369	0.03262 0.02421 0.03356 0.03829 Std Err 0.04239	9.96% 7.38% 12.8% 23.52% CV% 7.67%	17.38% 17.16% 33.86% 58.92% %Effect 0.0%
5 6.06 10 15 Angular (Cor C-% 0 2.5	Control Ty	5 5 5 5 nsformed So vpe Cour ol 5 5	0.732 0.734 0.586 0.364 ummary nt Mear 1.235 1.239	0.6414 0.6668 0.4928 0.2577 95% L 1.118 1.084 0.9256	0.8226 0.8012 0.6792 0.4703 CL 95% L 1.353 1.394 1.134	6 2 2 3	0.75 0.73 0.59 0.39 Median 1.202 1.249	0.65 0.66 0.48 0.22 Min 1.12 1.12	0.83 0.81 0.67 0.44 Max 1.369 1.429	0.03262 0.02421 0.03356 0.03829 Std Err 0.04239 0.05587	9.96% 7.38% 12.8% 23.52% CV% 7.67% 10.08%	17.38% 17.16% 33.86% 58.92% %Effect 0.0% -0.31%
5 6.06 10 15 Angular (Cor C-% 0 2.5 5	Control Ty	5 5 5 nsformed So rpe Cour ol 5 5 5	0.732 0.734 0.586 0.364 ummary nt Mear 1.235 1.239	0.6414 0.6668 0.4928 0.2577 95% L 1.118 1.084 0.9256 0.9537	CL 95% L 1.353 1.394 1.107	5 2 2 3 JCL	0.75 0.73 0.59 0.39 Median 1.202 1.249 1.047	0.65 0.66 0.48 0.22 Min 1.12 1.12 0.9377	0.83 0.81 0.67 0.44 Max 1.369 1.429 1.146	0.03262 0.02421 0.03356 0.03829 Std Err 0.04239 0.05587 0.03749	9.96% 7.38% 12.8% 23.52% CV% 7.67% 10.08% 8.14%	17.38% 17.16% 33.86% 58.92% %Effect 0.0% -0.31% 16.65%

Report Date:

29 Jun-17 18:57 (p 1 of 1)

Test Code: 1706-508 16-3408-8529/61663651

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: End Date:

30 Jun-17 30 Jun-17 Species: Strongylocentrotus purpuratus Protocol: EPA/600/R-95/136 (1995)

Sample Code: 17- OT Y O Sample Source: IDE Americas, Inc.

	te: 30 c				al: Facility Ef	naciit		Sample Station: Brine Pit	
C-%	Code	Rep		# Counted	# Fertilized			Notes	
			186	100	44	ACS	7/2/17		
			187	100	76				
			188	100	72				
			189	100	82				
			190	100	72 82 67				
			191	100	73				
			192	100	87				
			193	100	81		The state of the s		
			194	100	41 75				
			195	100	92				
			196	100	39 55				
			197	100	55			1	
			198	100	36				
			199	100	22				
			200	100	81				
			201	100	98				
			202	100	67				
			203	100	66				
			204	100	59				
			205	100	48				
			206	100	65				
			207	100	75				
			208	100	83				
			209	100	41				
			210	00	91				
			211	100	64				
			212	100	96		77.		
			213	100	90		400-00		
			214	100	81				
			215	100	87				

Report Date:

29 Jun-17 18:56 (p 1 of 1)

Test Code: 170%-\$20\\$16-3408-8529/61663651

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date:30 Jun-17Species:Strongylocentrotus purpuratusSample Code:17- ΟΤΨοEnd Date:30 Jun-17Protocol:EPA/600/R-95/136 (1995)Sample Source:IDE Americas, Inc.Sample Source:IDE Americas, Inc.

Sample Date: 30 Jun-17 Material: Facility Effluent Sample Station: Brine Pit

C-%	Code	Rep	Pos	# Counted	# Fertilized	Notes
0	LC	1	215	100	81	AD 1/1/17
0	LC	2	192			
0	LC	3	212			
0	LC	4	195			
0	LC	5	214			
2.5		1	201			
2.5		2	189			
2.5		3	213			
2.5		4	210			
2.5		5	193			
5		1	206			
5		2	208			
5		3	190			
5		4	194			
5		5	187			
6.06		1	200	100	64	
6.06		2	191	1	<u> </u>	
6.06		3	203			·
6.06		4	188			
6.06		5	207			
10		1	197			
10		2	211			
10		3	204			
10		4	202		And the set of statement	
10		5	205			
15		1	186			
15		2	196			
15		3	198			
15		4	199			
15		5	209			

QC: CG

Water Quality Measurements

Client :	IDE	Test Species: S. purpuratus
Sample ID:	Brine Pit	Start Date/Time: 6/30/2017 \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \
Sample Log No.:	17-0740	End Date/Time: 6/30/2017 1830
Dilutions made by:	18VO	Test No: 1706-5208

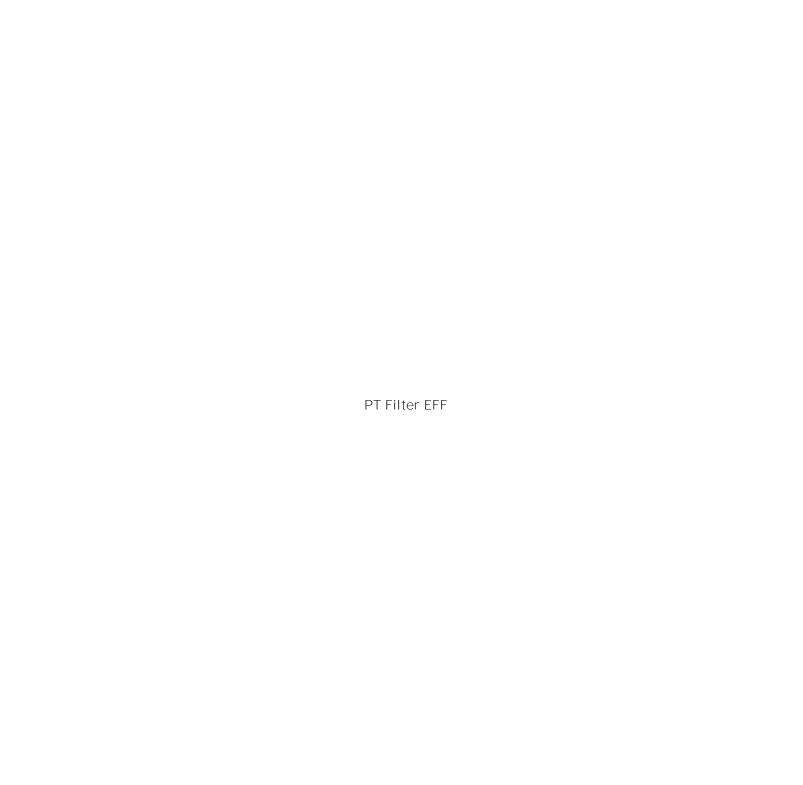
			Analyst:	CH
			eadings	
Concentration	DO (77.7/1)	pH	Salinity	Temperature
%	(mg/L)	(units)	(ppt)	(,C)
Lab Control	8.0	8.01	33.9	15.0
2.5	8.0	8.03	33.9	14.8
5.0	8,0	8.03	33.9	14.6
6.06	8.0	8.03	33.9	14.8
10	8,0	8.03	33.6	14.7
15	8.0	8.02	33.5	14.7

Comments:	Piluhons made	with M-INP
QC Check:	AC7/7/17	Final Review: VFP7/11/17

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	10E Binne Pit 1706-5208			Start Date/Time End Date/Time Species Animal Source	: 6/30/2017 / \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \
Tech initials: Injection Time:	1650			Date Collected	
Sperm Absorbance at	400 nm: 1,009	(target range of 0.8	- 1.0 for density	of 4x10 ⁶ sperm/ml)	
Eggs Counted:		(200	= <u>4040</u>	eggs/ml	-(D)
Initial density:	4040 eggs/ml	=dilution	factor	egg stock	ml
Final density:	4000 eggs/ml	- 1.0 part egg	g stock	seawater	ml
Prepare the embryo steexisting stock (1 part) a	ock according to the calcu and 125 ml of dilution wate	lated dilution factor. Fo		dilution factor is 2.	25, use 100 ml of
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1 1600 50 40 0.0 10	<u>1200:1</u> 80	berm:Egg Ratio 00:1 400:1 20 10 30 40	200:1 5.0 45	100:1 50:1 2.5 1.25 47.5 48.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1710 1723 1733	Rangefinder Ratio:	Fert. 1	Unfert.	
this range, choose the	m-to-egg ratio that results e ratio closest to 90 per of reproductive season, si	cent unless profession	80 and 90 perce nal judgment dic	nt. If more than on tates consideration	ne concentration is within nof other factors (e.g.,
<u>Definitive Test</u>		Sperm:Egg Ratio Use	ed: <u>15</u> ‡	<u> </u>	
Sperm Added (100 μl): Eggs Added (0.5 ml): Test Ended:	Time (3) 17450 1810 1830	QC1 QC2 Egg Control 1 Egg Control 2	Fert. U	Unfert. 3 (0 100	
Comments:	Phodiumo	700UV	00		
QC Check: Nautilus Environmental. 43-	AC 7/5/17 40 Vandever Avenue. San Die	 ego, CA 92120.		Final Review:	KAP 7/11/17



CETIS Summary Report

Report Date:

07 Jul-17 16:03 (p 1 of 1)

Test Code:

1706-S206 | 16-3657-6775

processor and the second								rest code.		170	J0-3200 T	0-3037-077
Echinoid Spe	rm Cell Fertiliza	ation Tes	st 15C			,				Nautilu	s Environr	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	19-0828-4416 30 Jun-17 17:5 30 Jun-17 18:3 40m	50 I 30 S	Test Type: Protocol: Species: Source:	Fertilization EPA/600/R-95, Strongylocentr Pt. Loma	, ,	itus		Analyst: Diluent: Brine: Age:	ivatt	ural Seawat Applicable	er M-1N	JE.
•	13-1009-4228 30 Jun-17 08:0 30 Jun-17 11:4 10h (5.5°C)	00 I 18 \$	Code: Material: Source: Station:	17-0742 Facility Effluen IDE Americas, Pre-Treatment	Inc.			Client: Project:	IDE Carl	sbad Desal	Plant	
Comparison S	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Meth	od			
21-1002-5944	Fertilization Ra	ite	15	>15	NA	7.01%	€ 6.66			lultiple Com	parison Te	st
Point Estimat	e Summary									1000		**************************************
Analysis ID	Endpoint		Level	%	95% LCL	95% UCL	. TU	Meth	od			
15-3250-9312	Fertilization Ra	ite	EC25 EC50	>15 >15	N/A N/A	N/A N/A	<6.6 <6.6		ar Inte	erpolation (I	CPIN)	
Test Acceptat	oility	· · · · · · · · · · · · · · · · · · ·										
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Lim	its	Over	lap	Decision		
15-3250-9312	Fertilization Ra	ite	Contro	ol Resp	0.89	0.7 - NL		Yes		Passes A	cceptability	Criteria
21-1002-5944	Fertilization Ra	ite	Contro	ol Resp	0.89	0.7 - NL		Yes			cceptability	
21-1002-5944	Fertilization Ra	ite	PMSE)	0.07012	NL - 0.25		No		Passes A	cceptability	Criteria
Fertilization R	ate Summary											
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std E	Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.89	0.8444	0.9356	0.83	0.92	0.016	643	0.03674	4.13%	0.0%
2.5		5	0.922	0.876	0.968	0.88	0.97	0.016	555	0.03701	4.01%	-3.6%
5		5	0.898	0.8574	0.9386	0.85	0.93	0.014	163	0.03271	3.64%	-0.9%
6.06		5	0.882	0.8255	0.9385	0.81	0.92	0.020)35	0.0455	5.16%	0.9%
10		5	0.872	0.826	0.918	0.81	0.9	0.016	355	0.03701	4.25%	2.02%
15		5	0.904	0.8554	0.9526	0.85	0.95	0.017	749	0.03912	4.33%	-1.57%
Fertilization R	ate Detail											
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	0.83	0.92	0.88	0.91	0.91						
2.5		0.91	0.9	0.95	0.97	0.88						
5		0.93	0.92	0.88	0.85	0.91						
6.06		0.81	0.92	0.89	0.87	0.92						
10		0.87	0.81	0.88	0.9	0.9						
15		0.95	0.88	0.92	0.92	0.85						
												

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Report Date:

07 Jul-17 16:02 (p 1 of 2)

Test Code:

1706-S206 | 16-3657-6775

							Test	Code:	170	6-S206 1	6-3657-6775
Echinoid Sp	erm Cell Fertiliza	ation Test 1	15C						Nautilus	Environ	mental (CA)
Analysis ID:	21-1002-5944	End	dpoint: Fer	tilization Rat	te		CET	S Version	: CETISv1	.8.7	
Analyzed:	07 Jul-17 16:0	2 A na	alysis: Par	ametric-Cor	trol vs Trea	ments	Offic	ial Results	s: Yes		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C > T	NA	NA		7.01%	15	>15	NA	6.667
Dunnett Mul	tiple Compariso	n Test									
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	ι(α:5%)		
Lab Control	2.5		-1.495	2.362	0.093 8	0.9962	CDF	Non-Sign	ificant Effect		
	5		-0.3254	2.362	0.093 8	0.9117	CDF	Non-Sign	ificant Effect		
	6.06		0.2868	2.362	0.093 8	0.7351	CDF	Non-Sign	ificant Effect		
	10		0.7209	2.362	0.093 8	0.5474	CDF	Non-Sign	ificant Effect		
	15		-0.6304	2.362	0.093 8	0.9561	CDF	Non-Sign	ificant Effect		
ANOVA Tabl	e										
Source	Sum Squ	ıares	Mean Squ	ıare	DF	F Stat	P-Value	Decision	ι(α:5%)		
Between	0.023319	27	0.0046638	355	5	1.198	0.3401	Non-Sign	ificant Effect		
Error	0.093460	31	0.0038941	8	24						
Total	0.116779	6			29	_					
Distribution	al Tests								· · · · · · · · · · · · · · · · · · ·		
Attribute	Test			Test Stat	Critical	P-Value	Decision	α:1%)			
Variances	Bartlett B	Equality of V	'ariance	0.7582	15.09	0.9796	Equal Var	iances			
Distribution	Shapiro-	Wilk W Nor	mality	0.9497	0.9031	0.1659	Normal Di	stribution			
Fertilization	Rate Summary										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.89	0.8444	0.9356	0.91	0.83	0.92	0.01643	4.13%	0.0%
2.5		5	0.922	0.876	0.968	0.91	0.88	0.97	0.01655	4.01%	-3.6%
5		5	0.898	0.8574	0.9386	0.91	0.85	0.93	0.01463	3.64%	-0.9%
6.06		5	0.882	0.8255	0.9385	0.89	0.81	0.92	0.02035	5.16%	0.9%
10		5	0.872	0.826	0.918	0.88	0.81	0.9	0.01655	4.25%	2.02%
15		5	0.904	0.8554	0.9526	0.92	0.85	0.95	0.01749	4.33%	-1.57%
Angular (Co	rrected) Transfo	med Sumn	nary			AMAKAN MANANGAN MANAN		***************************************			
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.236	1.166	1.306	1.266	1.146	1.284	0.02511	4.54%	0.0%
0		5	1.295	1.203	1.387	1.266	1.217	1.397	0.03309	5.71%	-4.78%
2.5		-				4 000	1 172	4 202	0.00000	4.040/	4.040/
		5	1.249	1.183	1.314	1.266	1.173	1.303	0.02369	4.24%	-1.04%
2.5			1.249 1.225	1.183 1.14	1.314 1.309	1.266	1.173	1.284	0.02369	4.24% 5.57%	0.92%
2.5 5		5									

Report Date: Test Code:

Rankits

07 Jul-17 16:03 (p 2 of 2)

1706-S206 | 16-3657-6775

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 21-1002-5944 Endpoint: Fertilization Rate **CETIS Version:** CETISv1.8.7 07 Jul-17 16:02 Analyzed: Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 1.0 0.12 0.9 0.08 0.8 Fertilization Rate Corr. Angle 0.04 0.6 0.5 -0.04 0.3 -0.08 0.1 0.0 -0.12 0 LC 2.5 5 6.06 10 15 -2.5 -2.0 -1.5 -1.0 1.0 1.5 2.0 C-%

Report Date: Test Code:

07 Jul-17 16:03 (p 1 of 1)

1706-S206 | 16-3657-6775

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: 15-3250-9312

Endpoint: Fertilization Rate

CETIS Version:

n: CETISv1.8.7

Analyzed: 07 Jul-17 16:02 Analysis: Linear Interpolation (ICPIN)

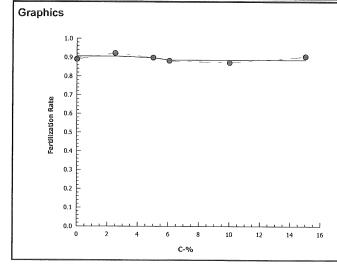
Official Results: Yes

Linear Interpol	ation Options					
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method	
Linear	Linear	130088	1000	Yes	Two-Point Interpolation	
Point Estimate	^					

Point Estimates

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
EC25	>15	N/A	N/A	<6.667	NA	NA
EC50	>15	N/A	N/A	<6.667	NA	NA

Fertiliza	tion Rate Summary	Calculated Variate(A/B)									
C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.89	0.83	0.92	0.01643	0.03674	4.13%	0.0%	445	500
2.5		5	0.922	0.88	0.97	0.01655	0.03701	4.01%	-3.6%	461	500
5		5	0.898	0.85	0.93	0.01463	0.03271	3.64%	-0.9%	449	500
6.06		5	0.882	0.81	0.92	0.02035	0.0455	5.16%	0.9%	441	500
10		5	0.872	0.81	0.9	0.01655	0.03701	4.25%	2.02%	436	500
15		5	0.904	0.85	0.95	0.01749	0.03912	4.33%	-1.57%	452	500



Report Date: Test Code: 07 Jul-17 16:03 (p 1 of 1) 1706-S206 | 16-3657-6775

							1631	Code:			6-3657-677
Echinoid Sp	erm Cell Fertiliz	ation Test	15C	TST					Nautilu	s Environi	mental (CA)
Analysis ID:	15-7034-8634	En	dpoint: Fer	tilization Ra	te		CET	IS Version:	CETISv1	.8.7	
Analyzed:	07 Jul-17 16:0	3 A n	ı alysis : Par	ametric Bio	equivalence	-Two Samp		ial Results			
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C*b < T	NA	NA	0.75	4.97%	15	>15	NA	6.667
TST-Welch's	t Test										
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(a:5%)		
Lab Control	2.5*		9.665	1.943	0.074 6	<0.0001	CDF	****	ificant Effect	<u> </u>	
	5*		10.63	1.895	0.057 7	<0.0001	CDF	-	ificant Effect		
	6.06*		8.298	1.943	0.07 6	<0.0001	CDF	•	ificant Effect		
	10*		9.256	1.895	0.057 7	<0.0001	CDF	_	ificant Effect		
	15*		9.458	1.943	0.069 6	<0.0001	CDF	•	ificant Effect		
ANOVA Tabl	е							***************************************			
Source	Sum Squ	ares	Mean Squ	ıare	DF	F Stat	P-Value	Decision	(α:5%)		
Between	0.023319	<u>2</u> 7	0.0046638	355	5	1.198	0.3401	Non-Signi	ficant Effect		
Error	0.093460	31	0.0038941	8	24			J			
Total	0.116779	6			29	_					
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision((α:1%)			
Variances	Bartlett E	quality of \	/ariance	0.7582	15.09	0.0700					
				0.7362	10.00	0.9796	Equal Var	iances			
Distribution	Shapiro-	Wilk W No		0.9497	0.9031	0.9796	Equal Var Normal Di				
	Shapiro- Rate Summary						•				
							•		Std Err	CV%	%Effect
Fertilization	Rate Summary	Wilk W Nor	mality	0.9497	0.9031	0.1659	Normal Di	stribution	Std Err 0.01643	CV% 4.13%	%Effect
Fertilization C-%	Rate Summary Control Type	Wilk W Nor	mality Mean	0.9497 95% LCL	0.9031 95% UCL	0.1659 Median	Normal Di	stribution Max			
Fertilization C-%	Rate Summary Control Type	Wilk W Nor Count 5	Mean 0.89	0.9497 95% LCL 0.8444	0.9031 95% UCL 0.9356	0.1659 Median 0.91	Min 0.83	Max 0.92	0.01643	4.13%	0.0%
Fertilization C-% 0 2.5	Rate Summary Control Type	Count 5 5	Mean 0.89 0.922	0.9497 95% LCL 0.8444 0.876	0.9031 95% UCL 0.9356 0.968	0.1659 Median 0.91 0.91	Min 0.83 0.88	Max 0.92 0.97	0.01643 0.01655	4.13% 4.01%	0.0% -3.6%
Fertilization C-% 0 2.5 5	Rate Summary Control Type	Count 5 5 5	Mean 0.89 0.922 0.898	0.9497 95% LCL 0.8444 0.876 0.8574	0.9031 95% UCL 0.9356 0.968 0.9386	0.1659 Median 0.91 0.91 0.91	Min 0.83 0.88 0.85	Max 0.92 0.97 0.93	0.01643 0.01655 0.01463	4.13% 4.01% 3.64%	0.0% -3.6% -0.9%
Fertilization C-% 0 2.5 5 6.06	Rate Summary Control Type	Count 5 5 5 5	Mean 0.89 0.922 0.898 0.882	0.9497 95% LCL 0.8444 0.876 0.8574 0.8255	95% UCL 0.9356 0.9386 0.9386 0.9385	0.1659 Median 0.91 0.91 0.91 0.91 0.89	Min 0.83 0.88 0.85 0.81	Max 0.92 0.97 0.93 0.92	0.01643 0.01655 0.01463 0.02035	4.13% 4.01% 3.64% 5.16%	0.0% -3.6% -0.9% 0.9%
Fertilization C-% 0 2.5 5 6.06 10	Rate Summary Control Type	Count 5 5 5 5 5 5	Mean 0.89 0.922 0.898 0.882 0.872 0.904	0.9497 95% LCL 0.8444 0.876 0.8574 0.8255 0.826	95% UCL 0.9356 0.9386 0.9386 0.9385 0.918	0.1659 Median 0.91 0.91 0.91 0.89 0.88	Min 0.83 0.88 0.85 0.81	Max 0.92 0.97 0.93 0.92 0.9	0.01643 0.01655 0.01463 0.02035 0.01655	4.13% 4.01% 3.64% 5.16% 4.25%	0.0% -3.6% -0.9% 0.9% 2.02%
Fertilization C-% 0 2.5 5 6.06 10	Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5	Mean 0.89 0.922 0.898 0.882 0.872 0.904	0.9497 95% LCL 0.8444 0.876 0.8574 0.8255 0.826	95% UCL 0.9356 0.9386 0.9386 0.9385 0.918	0.1659 Median 0.91 0.91 0.91 0.89 0.88	Min 0.83 0.88 0.85 0.81	Max 0.92 0.97 0.93 0.92 0.9	0.01643 0.01655 0.01463 0.02035 0.01655	4.13% 4.01% 3.64% 5.16% 4.25%	0.0% -3.6% -0.9% 0.9% 2.02%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Rate Summary Control Type Lab Control	Count 5 5 5 5 5 5 med Sumr	Mean 0.89 0.922 0.898 0.882 0.872 0.904	0.9497 95% LCL 0.8444 0.876 0.8574 0.8255 0.826 0.8554	0.9031 95% UCL 0.9356 0.968 0.9386 0.9385 0.918 0.9526	0.1659 Median 0.91 0.91 0.91 0.89 0.88 0.92	Min 0.83 0.88 0.85 0.81 0.81 0.85	Max 0.92 0.97 0.93 0.92 0.9 0.95	0.01643 0.01655 0.01463 0.02035 0.01655 0.01749	4.13% 4.01% 3.64% 5.16% 4.25% 4.33%	0.0% -3.6% -0.9% 0.9% 2.02% -1.57%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5	Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 5 count Count	Mean 0.89 0.922 0.898 0.882 0.872 0.904 mary Mean	0.9497 95% LCL 0.8444 0.876 0.8574 0.8255 0.826 0.8554 95% LCL	95% UCL 0.9356 0.9386 0.9385 0.918 0.9526	0.1659 Median 0.91 0.91 0.91 0.89 0.88 0.92 Median	Min 0.83 0.88 0.85 0.81 0.81	Max 0.92 0.97 0.93 0.92 0.9 0.95	0.01643 0.01655 0.01463 0.02035 0.01655 0.01749	4.13% 4.01% 3.64% 5.16% 4.25% 4.33%	0.0% -3.6% -0.9% 0.9% 2.02% -1.57%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5	Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 5 med Sumr Count 5	Mean 0.89 0.922 0.898 0.882 0.872 0.904 mary Mean 1.236	95% LCL 0.8444 0.876 0.8574 0.8255 0.826 0.8554 95% LCL 1.166	0.9031 95% UCL 0.9356 0.9386 0.9385 0.918 0.9526 95% UCL 1.306	0.1659 Median 0.91 0.91 0.91 0.89 0.88 0.92 Median 1.266	Min 0.83 0.88 0.85 0.81 0.81 0.85	Max 0.92 0.97 0.93 0.92 0.9 0.95 Max 1.284	0.01643 0.01655 0.01463 0.02035 0.01655 0.01749 Std Err 0.02511	4.13% 4.01% 3.64% 5.16% 4.25% 4.33% CV% 4.54%	0.0% -3.6% -0.9% 0.9% 2.02% -1.57% %Effect 0.0%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5 5	Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 med Sumr Count 5 5 5	Mean 0.89 0.922 0.898 0.882 0.872 0.904 mary Mean 1.236 1.295	95% LCL 0.8444 0.876 0.8574 0.8255 0.826 0.8554 95% LCL 1.166 1.203	95% UCL 0.9356 0.9386 0.9385 0.9385 0.918 0.9526 95% UCL 1.306 1.387	0.1659 Median 0.91 0.91 0.91 0.89 0.88 0.92 Median 1.266 1.266	Min 0.83 0.88 0.85 0.81 0.81 0.85 Min 1.146 1.217	Max 0.92 0.97 0.93 0.92 0.9 0.95 Max 1.284 1.397	0.01643 0.01655 0.01463 0.02035 0.01655 0.01749 Std Err 0.02511 0.03309	4.13% 4.01% 3.64% 5.16% 4.25% 4.33% CV% 4.54% 5.71%	0.0% -3.6% -0.9% 0.9% 2.02% -1.57% %Effect 0.0% -4.78%
Fertilization C-% 0 2.5 5 6.06 10 15 Angular (Cor	Rate Summary Control Type Lab Control rected) Transfor Control Type	Count 5 5 5 5 med Sumr Count 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Mean 0.89 0.922 0.898 0.882 0.872 0.904 mary Mean 1.236 1.295 1.249	95% LCL 0.8444 0.876 0.8574 0.8255 0.826 0.8554 95% LCL 1.166 1.203 1.183	95% UCL 0.9356 0.9386 0.9385 0.918 0.9526 95% UCL 1.306 1.387 1.314	0.1659 Median 0.91 0.91 0.91 0.89 0.88 0.92 Median 1.266 1.266 1.266	Min 0.83 0.88 0.85 0.81 0.81 0.85 Min 1.146 1.217 1.173	Max 0.92 0.97 0.93 0.92 0.9 0.95 Max 1.284 1.397 1.303	0.01643 0.01655 0.01463 0.02035 0.01655 0.01749 Std Err 0.02511 0.03309 0.02369	4.13% 4.01% 3.64% 5.16% 4.25% 4.33% CV% 4.54% 5.71% 4.24%	0.0% -3.6% -0.9% 0.9% 2.02% -1.57% %Effect 0.0% -4.78% -1.04%

Report Date:

29 Jun-17 18:56 (p 1 of 1)

Test Code: / 706-5) 06 16-3657-6775/618C2E07

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: 30 Jun-17 End Date: 30 Jun-17 Sample Date: 30 Jun-17

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

Sample Code: 17-0742 Sample Source: IDE Americas, Inc.

Material: Facility Effluent

Sample Station: Pre-Treatment Filtered

C-%	Code	Rep		# Counted	# Fertilized			Notes
			156	100	92	7/3/17	DM	
			157	1	97			
			158		83			
			159		O38 33			
			160		92			
			161		91			
			162		37			
			163		88 91			
			164		91			
			165		92			
			166		93			
			167		97			
			168		95			
			169		83			
			170		3 3			
			171		90			
			172		35 37			
			173		37			
			174		39 90 90			
			175		90	* 485		
			176		90			
			177		95			
			178		78 81			
			179		3(
			180		BRY 81			
			181		92 35 92			
			182		35			
			183		92			
			184	1	91			
			185	4	38			

@ Q18 PM07/03/17

Report Date:

29 Jun-17 18:56 (p 1 of 1)

Test Code: |706-520€16-3657-6775/618C2E07

Nautilus Environmental (CA)

Echinoid Sperm Cell Fertilization Test 15C

30 Jun-17

Species: Strongylocentrotus purpuratus

17-0742 Sample Code:

End Date: 30 Jun-17

Start Date:

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

Sample Source: IDE Americas, Inc.

Sample Date: 30 Jun-17 Material: Facility Effluent Sample Station: Pre-Treatment Filtered

C-%	Code	Rep	Pos	# Counted	# Fertilized	Notes
0	LC	1	170	100	94	BO 7/1/17
0	LC	2	156			111
0	LC	3	185			
0	LC	4	164			
0	LC	5	184			
2.5		1	169			
2.5		2	171			
2.5		3	177			· · · · · · · · · · · · · · · · · · ·
2.5		4	157			
2.5		5	178			
5		1	166			
5		2	160			
5		3	163			
5		4	182			
5		5	161			
6.06		1	179	100	87	
6.06		2	183	100		
6.06		3	174			
6.06		4	162			
6.06		5	167			
10		1	173			
10		2	180			
10		3	159			
10		4	175			
10		5	176			
15		1	168			
15		2	158			ALCO TO THE PROPERTY OF THE PR
15		3	165			
15		4	181			
15		5	172			

QC: (G

Water Quality Measurements

Client :	IDE	Test Species: S. purpuratus

Sample ID: Pre-Treatment Filtered Start Date/Time: 6/30/2017

Sample Log No.: 17-0742 End Date/Time: 6/30/2017

> Analyst: CH Initial Readings Concentration DO рΗ Salinity Temperature (units) (mg/L) (ppt) (°C) 15.3 Lab Control 7.9 801 33.9 8.03 15.4 7.9 34.0 2.5 8.02 3.0 5.0 15.3 33.9 8.03 34.0 7.9 6.06 15.3 8.02 34.1 15.4 7.9 10 7.97 8.0 34.0 15.2 15

Comments:	Dilutions made	WITH M-INF	
QC Check:	AC7/7/17	Final Review:	MP7/11/17

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

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	DE Pre-treatme 1706-S206	nt filtered	Į	End Date/Tim Specie	es: S. purpurat	11830
Tech initials:	A00			Animal Source Date Collecte		1
Injection Time:	1650				-GIV 1	
Sperm Absorbance at	400 nm: 1, 009	(target range of 0.	.8 - 1.0 for densit	y of 4x10 ⁶ sperm/m	ıl)	<u>~</u>
Eggs Counted:	Mean: 30.8 x 50 = 4040 eggs/ml (target counts of 80 eggs per vertical pass on Sedgwick- Refter slide for a final density of 4000 eggs/ml)					
	83	r slide for a final densit	y of 4000 eggs/ml)	(A) ₎
Initial density:	HOUV eggs/ml		on factor	egg stock	ml	
Final density:	4000 eggs/ml		egg stock seawater	seawater	ml	
Prepare the embryo stock according to the calculated dilution factor. For example, if the dilution factor is 2.25, use 100 ml of existing stock (1 part) and 125 ml of dilution water (1.25 parts).						
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1 1600 50 40 0.0 10	<u>1200:1</u> 30 20	Sperm:Egg Rate 800:1 400: 20 10 30 40	200:1 5.0	100:1 2.5 47.5	50:1 1.25 48.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time 1710 1723 1733	Rangefinder Ratio: 50:\ LUO:\	Fert. 91,83	Unfert.		
NOTE: Choose a sperm-to-egg ratio that results in fertilization between 80 and 90 percent. If more than one concentration is within this range, choose the ratio closest to 90 percent unless professional judgment dictates consideration of other factors (e.g., organism health, stage of reproductive season, site conditions).						
Definitive Test		Sperm:Egg Ratio U	Jsed:			
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time (2) 17450 1810 1830	QC1 QC2 Egg Control 1 Egg Control 2	Fert. 97 94 0	Unfert. 3 100 100		
Comments:	Phodiuko Pho OS V	70011V	1e0			
QC Check: Nautilus Environmental. 43-	AC 7/5/17 10 Vandever Avenue. San Die	– go, CA 92120.		Final Review	v: KAP7/11/	17



CETIS Summary Report

Report Date:

07 Jul-17 15:52 (p 1 of 1)

Test Code:

1706-S205 | 01-9508-5319

								rest Code:		~	·	1-9506-5318
Echinoid Sper	rm Cell Fertiliza	tion Test 1	5C		*****					Nautilus	Environr	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	09-2930-1447 30 Jun-17 17:50 30 Jun-17 18:30 40m	Pro Spe	t Type: tocol: cies: rce:	Fertilization EPA/600/R-95/ Strongylocentro Pt. Loma	, ,	tus		Analyst: Diluent: Brine: Age:		ıral Seawate Applicable	er/Naut stau	iilus Aten
-	03-5917-9333 30 Jun-17 08:00 30 Jun-17 11:40 10h (5.5 °C)	Sou	le: erial: rce: ion:	17-0743 Receiving Wate IDE Americas, M-INF				Client: Project:	IDE Carl	sbad Desal	Plant	
Comparison S	Summary											
Analysis ID	Endpoint		NOEL	. LOEL	TOEL	PMSD	TU	Meth	nod			
15-4355-0639	Fertilization Rat	:e	15	>15	NA	3.6%	∠6.66			ultiple Comp	parison T e	st
Point Estimate	e Summary										CONTRACTOR OF THE STATE OF THE	
Analysis ID	Endpoint		Level	%	95% LCL	95% UCL	TU	Meth	nod			
02-7881-1690	Fertilization Rat	e	EC25 EC50	>15 >15	N/A N/A	N/A N/A	<6.6 <6.6		ar Inte	erpolation (IC	CPIN)	
Test Acceptab	oility											
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Lim	its	Ove	rlap	Decision		
02-7881-1690	Fertilization Rat	e	Contro	ol Resp	0.934	0.7 - NL		Yes		Passes Ac	ceptability	Criteria
15-4355-0639	Fertilization Rat			ol Resp	0.934	0.7 - NL		Yes		Passes Ac	ceptability	Criteria
15-4355-0639	Fertilization Rat	e	PMSC)	0.03597	NL - 0.25		No		Passes Ac	ceptability	Criteria
Fertilization Ra	ate Summary											
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std	Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.934	0.9068	0.9612	0.92	0.97	0.00	9798	0.02191	2.35%	0.0%
2.5		5	0.938	0.9276	0.9484	0.93	0.95	0.00	3741	0.008366	0.89%	-0.43%
5		5	0.924	0.8941	0.9539	0.89	0.95	0.01	077	0.02408	2.61%	1.07%
6.06		5	0.94	0.9168	0.9632	0.92	0.97	0.00	8366	0.01871	1.99%	-0.64%
10		5	0.95	0.9237	0.9763	0.93	0.98	0.00	9487	0.02121	2.23%	-1.71%
15		5	0.938	0.9126	0.9634	0.92	0.96	0.00	9165	0.02049	2.19%	-0.43%
Fertilization Ra	ate Detail											, Alexander de la constantina della constantina
C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
•	Lab Control	0.94	0.92	0.97	0.92	0.92						
0				0.04	0.93	0.94						
0 2.5		0.95	0.93	0.94	0.93	0.54						
		0.95 0.89	0.93 0.94	0.94	0.93	0.95						
2.5				0.93	0.91	0.95						
2.5 5		0.89	0.94									

10

-1.296

2.362

Report Date:

Non-Significant Effect

07 Jul-17 15:52 (p 1 of 2) 1706-S205 | 01-9508-5319

			Test Code:	1706-S205 01-9508-5319
Echinoid Spe	rm Cell Fertilization	on Test 15C		Nautilus Environmental (CA)
Analysis ID:	15-4355-0639	Endpoint: Fertilization Rate	CETIS Version:	CETISv1.8.7

Analyzed:	07 Jul-17 15:50)	Analysis:	Parame	tric-Con	trol vs Treatments	Offic	cial Results	s: Yes		
Data Transfor	n	Zeta	Alt Hy	/p Tri	als	Seed	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corre	cted)	NA	C > T	NA	1	NA	3.6%	15	>15	NA	6.667

Dunnett Multi	ple C	omparison Test							
Control	vs	C-%	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(α:5%)
Lab Control		2.5	-0.1887	2.362	0.065	8	0.8830	CDF	Non-Significant Effect
		5	0.7422	2.362	0.065	8	0.5376	CDF	Non-Significant Effect
		6.06	-0.4254	2.362	0.065	8	0.9290	CDF	Non-Significant Effect

0.065 8 0.9929

CDF

	15	-0.2761 2.362	0.065 8	0.9021	CDF	Non-Significant Effect
ANOVA Table						
Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.008165863	0.001633173	5	0.8702	0.5155	Non-Significant Effect
Error	0.04504281	0.001876784	24			-
Total	0.05320867		29			

Distributional Te	ests				
Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance	3.976	15.09	0.5529	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9408	0.9031	0.0957	Normal Distribution

Fertilization	n Rate Summary										
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	0.934	0.9068	0.9612	0.92	0.92	0.97	0.009798	2.35%	0.0%
2.5		5	0.938	0.9276	0.9484	0.94	0.93	0.95	0.003741	0.89%	-0.43%
5		5	0.924	0.8941	0.9539	0.93	0.89	0.95	0.01077	2.61%	1.07%
6.06		5	0.94	0.9168	0.9632	0.94	0.92	0.97	0.008366	1.99%	-0.64%
10		5	0.95	0.9237	0.9763	0.95	0.93	0.98	0.009487	2.23%	-1.71%
15		5	0.938	0.9126	0.9634	0.93	0.92	0.96	0.009165	2.19%	-0.43%

Angular (Corrected) Transfor	med Sumr	mary								
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	5	1.314	1.254	1.375	1.284	1.284	1.397	0.02193	3.73%	0.0%
2.5		5	1.32	1.298	1.341	1.323	1.303	1.345	0.007862	1.33%	-0.39%
5		5	1.294	1.238	1.35	1.303	1.233	1.345	0.02013	3.48%	1.55%
6.06		5	1.326	1.273	1.379	1.323	1.284	1.397	0.01911	3.22%	-0.89%
10		5	1.35	1.285	1.415	1.345	1.303	1.429	0.02349	3.89%	-2.7%
15		5	1.322	1.267	1.377	1.303	1.284	1.369	0.01968	3.33%	-0.58%

Report Date: Test Code: 07 Jul-17 15:52 (p 2 of 2) 1706-S205 | 01-9508-5319

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Analysis ID: 15-4355-0639 Endpoint: Fertilization Rate **CETIS Version:** CETISv1.8.7 Analyzed: 07 Jul-17 15:50 Analysis: Parametric-Control vs Treatments Official Results: Yes Graphics 0.10 -0-80.0 0.06 Fertilization Rate 0.7 0.04 0.6 0.02 0.5 0.00 0.4 -0.02 0.3 -0.04 0.2 -0.06 0.1 -0.08 0.0 -0.10 0 LC 2.5 6.06 10 15 1.0 1.5 2.0 C-% Rankits

Report Date:

07 Jul-17 15:52 (p 1 of 1)

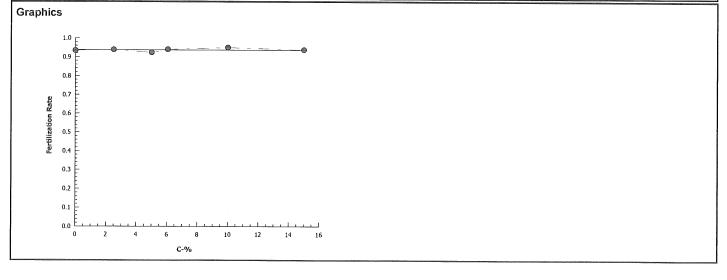
Test Code:

1706-S205 | 01-9508-5319

Echinoid Spe	rm Cell Fertilization	Test 15C			Nautilus Environmental (CA)
Analysis ID:	02-7881-1690	Endpoint:	Fertilization Rate	CETIS Version:	CETISv1.8.7
Analyzed:	07 Jul-17 15:50	Analysis:	Linear Interpolation (ICPIN)	Official Results:	Yes

Linear	Interpola	ition Options					
X Trans	form	Y Transform	Seed	ı t	Resamples	Exp 95% CL	Method
Linear		Linear	1617	'941 <i>'</i>	1000	Yes	Two-Point Interpolation
Point E	stimates	•					
Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL	
EC25	>15	N/A	N/A	<6.667	NA	NA	
EC50	>15	N/A	N/A	<6.667	NA	NA	

Fertilizat	tion Rate Summary										
C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В
0	Lab Control	5	0.934	0.92	0.97	0.009798	0.02191	2.35%	0.0%	467	500
2.5		5	0.938	0.93	0.95	0.003741	0.008366	0.89%	-0.43%	469	500
5		5	0.924	0.89	0.95	0.01077	0.02408	2.61%	1.07%	462	500
6.06		5	0.94	0.92	0.97	0.008366	0.01871	1.99%	-0.64%	470	500
10		5	0.95	0.93	0.98	0.009487	0.02121	2.23%	-1.71%	475	500
15		5	0.938	0.92	0.96	0.009165	0.02049	2.19%	-0.43%	469	500



Report Date: Test Code: 07 Jul-17 15:52 (p 1 of 1) 1706-S205 | 01-9508-5319

							Test	ooue.		0-3203 0	1-9508-531
Echinoid Sp	erm Cell Fertiliz	ation Test	15C						Nautilus	s Environ	mental (CA)
Analysis ID:	20-5871-2625	Er	ndpoint: Fe	tilization Ra	te		CET	IS Version	: CETISv1	.8.7	
Analyzed:	07 Jul-17 15:5	<u>i2 Ar</u>	nalysis: Pa	rametric Bio	equivalence	Two Samp	le Offic	ial Results	s: Yes		
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	rected)	NA	C*b < T	NA	NA	0.75	2.59%	15	>15	NA	6.667
TST-Welch's	t Test										
Control	vs C-%		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	ι(α:5%)		
Lab Control	2.5*		18.31	2.015	0.037 5	<0.0001	CDF	Non-Sign	ificant Effect		
	5*		11.86	1.895	0.049 7	<0.0001	CDF	•	ificant Effect		
	6.06*		13.5	1.895	0.048 7	<0.0001	CDF		ificant Effect		
	10*		12.7	1.895	0.054 7	<0.0001	CDF		ificant Effect		
	15*		13.11	1.895	0.049 7	<0.0001	CDF	•	ificant Effect		
ANOVA Table	е										
Source	Sum Squ	ıares	Mean Squ	ıare	DF	F Stat	P-Value	Decision	(α:5%)		
Between	0.008165	863	0.001633	73	5	0.8702	0.5155	Non-Sian	ificant Effect	A	
Error	0.045042	.81	0.0018767	'84	24			J			
Total	0.053208	67			29	_					
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision(α:1%)			
Variances	Bartlett E	Equality of '	Variance	3.976	15.09	0.5529	Equal Var	iances			
Distribution	Shapiro-	Wilk W No	rmality	0.9408	0.9031	0.0957	-	stribution			
							Nonnai Di				
Fertilization	Rate Summary						Normal Di				
Fertilization C-%	Rate Summary Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
	•	Count 5	Mean 0.934	95% LCL 0.9068	95% UCL 0.9612	Median 0.92		Max 0.97	Std Err 0.009798	CV% 2.35%	%Effect
C-%	Control Type					***************************************	Min				
C- %	Control Type	5	0.934	0.9068	0.9612	0.92	Min 0.92	0.97	0.009798	2.35%	0.0%
C- % 0 2.5	Control Type	5 5	0.934 0.938	0.9068 0.9276	0.9612 0.9484	0.92 0.94	Min 0.92 0.93	0.97 0.95	0.009798 0.003741	2.35% 0.89% 2.61%	0.0% -0.43%
C-% 0 2.5 5	Control Type	5 5 5	0.934 0.938 0.924	0.9068 0.9276 0.8941	0.9612 0.9484 0.9539	0.92 0.94 0.93	Min 0.92 0.93 0.89	0.97 0.95 0.95	0.009798 0.003741 0.01077	2.35% 0.89%	0.0% -0.43% 1.07%
C-% 0 2.5 5 6.06	Control Type	5 5 5 5	0.934 0.938 0.924 0.94	0.9068 0.9276 0.8941 0.9168	0.9612 0.9484 0.9539 0.9632	0.92 0.94 0.93 0.94	Min 0.92 0.93 0.89 0.92	0.97 0.95 0.95 0.97	0.009798 0.003741 0.01077 0.008366	2.35% 0.89% 2.61% 1.99%	0.0% -0.43% 1.07% -0.64%
C-% 0 2.5 5 6.06 10 15	Control Type	5 5 5 5 5 5	0.934 0.938 0.924 0.94 0.95 0.938	0.9068 0.9276 0.8941 0.9168 0.9237	0.9612 0.9484 0.9539 0.9632 0.9763	0.92 0.94 0.93 0.94 0.95	Min 0.92 0.93 0.89 0.92 0.93	0.97 0.95 0.95 0.97 0.98	0.009798 0.003741 0.01077 0.008366 0.009487	2.35% 0.89% 2.61% 1.99% 2.23%	0.0% -0.43% 1.07% -0.64% -1.71%
C-% 0 2.5 5 6.06 10 15 Angular (Cor	Control Type Lab Control	5 5 5 5 5 5	0.934 0.938 0.924 0.94 0.95 0.938	0.9068 0.9276 0.8941 0.9168 0.9237	0.9612 0.9484 0.9539 0.9632 0.9763	0.92 0.94 0.93 0.94 0.95	Min 0.92 0.93 0.89 0.92 0.93	0.97 0.95 0.95 0.97 0.98	0.009798 0.003741 0.01077 0.008366 0.009487	2.35% 0.89% 2.61% 1.99% 2.23%	0.0% -0.43% 1.07% -0.64% -1.71%
C-% 0 2.5 5 6.06 10 15 Angular (Cor	Control Type Lab Control rected) Transfor	5 5 5 5 5 5 5	0.934 0.938 0.924 0.94 0.95 0.938	0.9068 0.9276 0.8941 0.9168 0.9237 0.9126	0.9612 0.9484 0.9539 0.9632 0.9763 0.9634	0.92 0.94 0.93 0.94 0.95 0.93	Min 0.92 0.93 0.89 0.92 0.93 0.92	0.97 0.95 0.95 0.97 0.98 0.96	0.009798 0.003741 0.01077 0.008366 0.009487 0.009165	2.35% 0.89% 2.61% 1.99% 2.23% 2.19%	0.0% -0.43% 1.07% -0.64% -1.71% -0.43%
C-% 0 2.5 5 6.06 10 15 Angular (Cor	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 rmed Sumi	0.934 0.938 0.924 0.94 0.95 0.938 mary	0.9068 0.9276 0.8941 0.9168 0.9237 0.9126	0.9612 0.9484 0.9539 0.9632 0.9763 0.9634	0.92 0.94 0.93 0.94 0.95 0.93	Min 0.92 0.93 0.89 0.92 0.93 0.92	0.97 0.95 0.95 0.97 0.98 0.96	0.009798 0.003741 0.01077 0.008366 0.009487 0.009165	2.35% 0.89% 2.61% 1.99% 2.23% 2.19%	0.0% -0.43% 1.07% -0.64% -1.71% -0.43%
C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 mmed Sumi	0.934 0.938 0.924 0.94 0.95 0.938 mary Mean 1.314	0.9068 0.9276 0.8941 0.9168 0.9237 0.9126 95% LCL 1.254	0.9612 0.9484 0.9539 0.9632 0.9763 0.9634 95% UCL 1.375	0.92 0.94 0.93 0.94 0.95 0.93 Median	Min 0.92 0.93 0.89 0.92 0.93 0.92 Min 1.284	0.97 0.95 0.95 0.97 0.98 0.96	0.009798 0.003741 0.01077 0.008366 0.009487 0.009165 Std Err 0.02193	2.35% 0.89% 2.61% 1.99% 2.23% 2.19% CV% 3.73%	0.0% -0.43% 1.07% -0.64% -1.71% -0.43% %Effect 0.0% -0.39%
C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 5 Count 5	0.934 0.938 0.924 0.94 0.95 0.938 mary Mean 1.314 1.32	0.9068 0.9276 0.8941 0.9168 0.9237 0.9126 95% LCL 1.254 1.298	0.9612 0.9484 0.9539 0.9632 0.9763 0.9634 95% UCL 1.375 1.341	0.92 0.94 0.93 0.94 0.95 0.93 Median 1.284 1.323	Min 0.92 0.93 0.89 0.92 0.93 0.92 Min 1.284 1.303	0.97 0.95 0.95 0.97 0.98 0.96 Max 1.397 1.345	0.009798 0.003741 0.01077 0.008366 0.009487 0.009165 Std Err 0.02193 0.007862	2.35% 0.89% 2.61% 1.99% 2.23% 2.19% CV% 3.73% 1.33%	0.0% -0.43% 1.07% -0.64% -1.71% -0.43% %Effect 0.0% -0.39% 1.55%
C-% 0 2.5 5 6.06 10 15 Angular (Cor C-% 0 2.5 5	Control Type Lab Control rected) Transfor Control Type	5 5 5 5 5 5 crmed Sum Count 5 5	0.934 0.938 0.924 0.94 0.95 0.938 mary Mean 1.314 1.32 1.294	0.9068 0.9276 0.8941 0.9168 0.9237 0.9126 95% LCL 1.254 1.298 1.238	0.9612 0.9484 0.9539 0.9632 0.9763 0.9634 95% UCL 1.375 1.341 1.35	0.92 0.94 0.93 0.94 0.95 0.93 Median 1.284 1.323 1.303	Min 0.92 0.93 0.89 0.92 0.93 0.92 Min 1.284 1.303 1.233	0.97 0.95 0.95 0.97 0.98 0.96 Max 1.397 1.345 1.345	0.009798 0.003741 0.01077 0.008366 0.009487 0.009165 Std Err 0.02193 0.007862 0.02013	2.35% 0.89% 2.61% 1.99% 2.23% 2.19% CV% 3.73% 1.33% 3.48%	0.0% -0.43% 1.07% -0.64% -1.71% -0.43% %Effect 0.0% -0.39%

CETIS Test Data Worksheet

Report Date:

29 Jun-17 18:53 (p 1 of 1)

Test Code: 1706-S205 01-9508-5319/BA0C407

Echinoid Sperm Cell Fertilization Test 15C Nautilus Environmental (CA) Start Date: 30 Jun-17 Species: Strongylocentrotus purpuratus

End Date: 30 Jun-17 Sample Date: 30 Jun-17

Sample Code: 17- 0743 Protocol: EPA/600/R-95/136 (1995)

Sample Source: IDE Americas, Inc.

Material: Pacility Effluent Receiving Inc.

Sample Station: IPS M-INF

C-%	Code	Rep	Pos	# Counted	# Fertilized	Notes
			96	100	93	7/3/17
			97	100	94	
			98	100	95	
			99	100	95 92 93	
			100	100	93	
			101	100	94	
			102	∞ i	93	
			103	/00	89	
			104	/00	92	·
			105	700	94	
			106	100	94	
			107	100	92	
			108	00/	96	
			109	100	95	
			110	.100	97	
			111	100	96	
			112	100	97 95	
			113	100	<u>95</u>	
			114	100	92	·
	-		115	/00/	93	
			116 117	100	96 94	
			118	100	74	
			119	100	92	
			120	100	93	
			121	100	91	
			122	100	92 98	
			123		<u>プロ</u> 砂菜 93	
	1		124		ツ <u>キ フン</u> 93	
	1		125	100	94	

Report Date:

29 Jun-17 18:53 (p 1 of 1)

Test Code: \706\\$205 01-9508-5319/BA0C407

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date:	30 Jun-17	Species:	Strongylocentrotus purpuratus	Sample Code:	17-0743
End Date:	30 Jun-17	Protocol:	EPA/600/R-95/136 (1995)	Sample Source:	IDE Americas, Inc.
Sample Date:	30 Jun-17	Material:	EPA/600/R-95/136 (1995) Facility Effluent Receiving Water	Sample Station:	SPSM-INF

C-%		Rep			# Fertilized	Notes
0	LC	1	105	100	98	7/1/17 AD
0	LC	2	107		•	
0	LC	3	110			
0	LC	4	99			
0	LC	5	104			
2.5		1	109			
2.5		2	115			
2.5		3	106			
2.5		4	96			
2.5		5	117			
5		1	103			
5		2	125			
5		3	123			
5		4	120			
5		5	98			
6.06		1	112	100	94	
6.06		2	97	1		
6.06		3	121			
6.06		4	101			
6.06		5	124			
10		1	122			
10		2	116			
10		3	113			
10		4	119			
10		5	102			
15		1	114			
15		2	111			
15		3	108			
15		4	118			
15		5	100			

QC: C6 @018AC7[7]17

Marine Chronic Bioassay

Water Quality Measurements

Client : IDE

Test Species: S. purpuratus

Sample ID: N

CHES M-INF

Start Date/Time: 6/30/2017

Sample Log No.: 17- 0743

End Date/Time: 6/30/2017

1830

Dilutions made by:

Test No: 1706-5005

			Analyst:	CH				
		Initial Readings						
Concentration	DO	рН	Salinity	Temperature				
%	(mg/L)	(units)	(ppt)	(°C)				
Lab Control	8.5	7.91	33.7	15.2				
2.5	8.6	7.95	33.7	14.6				
5.0	8.6	7.97	33.8	14.4				
6.06	8.6	7.97	34.0	14.6				
10	8.6	7.98	33.9	14.3				
15	8.6	7.98	33.9	14.3				

Comments:	Dilvhons made with	NOUTHUS Slawater.	
QC Check:	AC 7/7/11	Final Review: VFP 7/11/17	

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

Marine Chronic Bioassay

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	DE M-1NF 1706-S20	25		End Date/	Time: 6/30/2017 / 150 Time: 6/30/2017 / 1830 ecies: S. purpuratus
Tech initials: Injection Time:	AD 1650			Animal So	purce: Pt. Loma ected: 10 (p)
Sperm Absorbance at	400 nm:	(target range of	0.8 - 1.0 for d	ensity of 4x10 ⁶ sperr	n/ml)
Eggs Counted:	76 (ta	ean: X X x x get counts of 80 eggs fter slide for a final der	50 = 40	eggs/ml	,
Initial density: Final density:	4000 eggs/m		ution factor rt egg stock rts seawater	egg stock seawater _	ml ml
Prepare the embryo steexisting stock (1 part) a	ock according to the cale and 125 ml of dilution wa	culated dilution factor lter (1.25 parts).	r. For example	e, if the dilution factor	is 2.25, use 100 ml of
Rangefinder Test: ml Sperm Stock ml Seawater	50 4	00:1 30 30 10 20	<u>Sperm:Egg</u> 800:1 20 30	Ratio 400:1 10 5.0 40 45	100:1 50:1 2.5 1.25 47.5 48.75
Sperm Added (100 μl): Eggs Added (0.5 ml): Test Ended:	Time 1710 1723 1733	Rangefinder Rat	io: Fert.	3 <u>Unfert.</u> 3 <u>U</u> 17	
this range, choose the	m-to-egg ratio that result e ratio closest to 90 po of reproductive season,	ercent unless profes	reen 80 and 90 ssional judgme	percent. If more the percent dictates conside	an one concentration is within eration of other factors (e.g.,
Definitive Test		Sperm:Egg Ratio	Used:	5 ~ 1	
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time(B) 17450 1810 1830	QC1 QC2 Egg Control 1 Egg Control 2	Fert. 97 94 0	Unfert. 3 (00 100	
Comments:	Phodium Phodis	37 YOUNY 130/170	000		·
QC Check: Nautilus Environmental. 43-	AC 7/5/17 40 Vandever Avenue. San I	 Diego, CA 92120.		Final Rev	riew: 1/11/17

Appendix B

Sample Receipt Information

Nautilus Environmental 4340 Vandever Avenue San Diego, CA 92120

Client:	1DE
Sample ID:	See below
Test ID No(s).:	1706-S203 to S209

Sample (A, B, C):	M-001	ERI BRINE	BRINE PIT	TRAIN 4
Log-in No. (17-хоох):	0738	0739	0740	0741
Sample Collection Date & Time:	6/30/170800	6/30/17 0800	680/170800	6/30/170800
Sample Receipt Date & Time:	6130/17 (14.8	6/30/17 1149	6/30/17 (148	6/30/17/148
Number of Containers & Container Type:	141 Whi	14Lwbi	14L Cubi	14Lasi
Арргох. Total Volume Received (L):	~ 46	-46	~46	~416
Check-in Temperature (°C)	6.5	4.5	6.5	5.5
Temperature OK? ¹	Ŋ N	Ŷ N	YN	Ý N
DO (mg/L)	6.67	7.3	7.0	7.0
pH (units)	7.72	7.21	8,00	7.75
Conductivity (μS/cm)				
Salinity (ppt)	62.6	68.4	30.2	70.2
Alkalinity (mg/L) ²	190	199	100	206
Hardness (mg/L) ^{2, 3}				
Total Chlorine (mg/L)	0.03	60.02	0.03	40.02
Technician Initials	BO	B0	30	BOLUH

Test Performed:	Urchin Ferfication	Control/Dilution Water: 8:2 / Lab SW Lab ART Other:
	Additional Control? Y N	= Alkalinity: Hardness or Salinity:
Test Performed:		Control/Dilution Water: 8:2 / Lab SW / Lab ART Other:
	Additional Control? Y N	Alkalinity: Hardness or Salinity:
Test Performed:		Control/Dilution Water: 8:2 / Lab SW / Lab ART Other:
	Additional Control? Y N	= Alkalinity: Hardness or Salinity:
Notes:		d be 0-6°C, if received more than 24 hours past collection time.
	² mg/L as CaCO3, ³ Measured	for freshwater samples only, NA = Not Applicable
ditional Comments:		

Sample Check-In Information

Sample Description:							
M-001: Colorless, ERIBRINE: Colorless	clear, ode	orless, no	debris				
ERIBRINE: COLONLOSS	, clear 100	lovless, no	s debvis				
BRINE PIT: Colorles	, clear,	odorles	s, no debi				
TRAIN 4: whorles	s, clear,	6dones	, no debr				
COC Complete (Y/N)?	· ·						
A_X B C							
Filtration? Y (N)							
Pore Size:		,					
Organisms	or	Debris					
			at adi				
Salinity Adjustment?) " W-	001 4011	n oeg				
Test: M-001	Source: 📞	awad Jarge	et ppt:VD				
Test:	Source:	Targe	et ppt:				
Test:	Source:	Targe	et ppt:				
pH Adjustment? Y	N						
	Α	В	С				
Initial pH:							
Amount of HCl added:							
Final pH:							
Cl₂ Adjustment? Y ((N						
·	A	В	c				
Initial Free Cl₂:							
STS added:							
Final Free Cl ₂ :			,				
	.~						
Sample Aeration? Y	(N)						
	A	В	C				
Initial D.O.							
Duration & Rate							
Final D.O.							
•	Subsamples for Additional Chemistry Required? Y (N						
	r						
Tech Initials A	/ R	- ·	/ 1 .				
	QC Ch	eck: AC	7/7/17				
-	Final Rev		7/11/17				

Nautilus Environmental 4340 Vandever Avenue San Diego, CA 92120

Client:	IDE		_
Sample ID:	Seebelow		
Test ID No(s).:	1706-203 to S209		

Sample (A, B, C):	PT FILETER EFF	M-INF		
Log-in No. (17-)οοοκ):	0742	0743		
Sample Collection Date & Time:	6(30/17 0800	6(30)7 0800		
Sample Receipt Date & Time:	6/30/17 1148	6130171148		
Number of Containers & Container Type:	141 Wb:	1 41 wbi		
Approx. Total Volume Received (L):	~46	~41		
Check-in Temperature (°C)	5.5	5.5		
Temperature OK? 1	Y N	◯ N	ΥN	Y N
DO (mg/L)	7.2	7.4		
pH (units)	7.97	8.06		
Conductivity (μS/cm)				
Salinity (ppt)	33.9	33.4		
Alkalinity (mg/L) ²	(07	114		
Hardness (mg/L) ^{2, 3}		_		
Total Chlorine (mg/L)	0.02	0.02		
Technician Initials	BO/CH	B0/01		

Test Performed:	Win tert Control/Dilution Water: 8:2 / Lab StV / Lab ART Other:
	Alkalinity: Hardness or Salinity: 34 00
	Additional Control? Y N =Alkalinity: Hardness or Salinity:
Test Performed:	Control/Dilution Water: 8:2 / Lab SW / Lab ART Other:
	Alkalinity: Hardness or Salinity:
	Additional Control? Y N = Alkalinity: Hardness or Salinity:
Test Performed:	Control/Dilution Water: 8:2 / Lab SW / Lab ART Other:
	Alkalinity: Hardness or Salinity:
	Additional Control? Y N =Alkalinity: Hardness or Salinity:
Notes:	¹ Temperature of sample should be 0-6°C, if received more than 24 hours past collection time.
	² mg/L as CaCO3, ³ Measured for freshwater samples only, NA = Not Applicable
tional Comments:	

Sample Check-In Information

Sample Description: PTFILEVEFF Color N-INFS ColorUS	635 ; ((lar,odor	less, mode	bis
n-(NFS Colorles	s, clea	1, odorle	15, no de	biis
COC Complete (Y/N)?	?			
A B C				
Filtration? Y (N)			
Pore Size:		_		
Organisms	or	Debris		
Salinity Adjustment?	v 🛍			
		Torac	at moto	
Test: Test:	Source:	_	et ppt: et ppt:	
	Source:	_	et ppt:	
Test:	<i>(</i> ~	rarye	er hhr.	
pH Adjustment? Y) _A	В	С	
initial nU				
Initial pH: Amount of HCI added:				
Final pH: Cl ₂ Adjustment? Y	4			
Ci ₂ Aujustinent:	(^N) _A	В	С	
Initial Free Cl ₂ :				
STS added:				
Final Free Cl ₂ :				
· · · · · · · · · · · · · · · · · · ·				
Sample Aeration? Y	(N)			
·	Α	В	С	
Initial D.O.				
Duration & Rate				
Final D.O.				
Subsamples for Addi	itional Chen	nistry Requir	ed? Y (N)	
Tech Initials			~	
			-10017	
-	QC Ch	eck:AC	(17/10)	
	Final Rev		1117 30	<u>.</u>

Appendix C

Chain-of-Custody Form



IDE Technologies



 CDP laoratory:
 Turn Around Time

 Entahlpy Laboratory:
 Normal:
 X

 WECK Laboratory:
 RUSH (24 hr):
 Nautilus:

 Nautilus:
 X
 3 Days:

 AIM:
 5 Days:
 Other:

 Other:
 ??? Days

										, Ott	ner:		the Section of the last	777 Days	
Project Name: Toxicity Screening			Project Manager:_			act Informa	ation:	(760)	201-77	77					
Special instruction: Samples collections of the collection of the	ted during normal pl se run unadjusted. St	ant operation. M-001 art: 6/29/17 @ 08:00,	. is to be run unadjusted a . End: 6/30/17 @ 08:00. KC	nd adjus	ted to 40 ppt for weekly				ANALY	YSES		a Beauting cons	27/4 mg 27/4	NOTES:	
						Chronic Fertilization									
		Glass=G Plastic	=P			ic Fe									
	Yes=Y No=N	Acid=A Base=B				hron			l						
Drinkin	g Water=DW Seawa	ter=SW Soil=S Brine=	=B	Pre		Urchin C									
Sample ID	Date	Tīme	Sample	Preservative	Container	Purple Urc									
			Туре	رب	Туре	Pu									0 C
₩ M-001 (17- 2188)	6/29-30/17	8:00	24 HR COMP -B	N	4L CUBIE	Х								TDS - 56.44 ppt EC - 80.12 mS/cm🧩	6.5
* ERI BRINE (17-2189)	6/29-30/17	8:00	24 HR COMP -B	N	4L CUBIE	х								TDS - 63.70 ppt EC - 88.20 mS/cm 🛧	4.5
* BRINE PIT (17-2190)	6/29-30/17	8:00	24 HR COMP -B	N	4L CUBIE	Х								TDS - 28.71 ppt EC - 45.25 mS/cm 🔏	6.5
TRAIN 4 (17-2191)	6/29-30/17	8:00	24 HR COMP -B	N	4L CUBIE	Х								TDS - 65.52 ppt EC - 90.13 mS/cm	5.5
PT FILTER EFF (17-2192)	6/29-30/17	8:00	24 HR COMP -B	N	4L CUBIE	Х								TDS - 32.18 ppt EC - 50.17 mS/cm	5.5
M-INF (17-2193)	6/29-30/17	8:00	24 HR COMP -B	N	4L CUBIE	Х								TDS - 32.18 ppt EC - 50.13 mS/cm	5.5
							1							-	
														-	
									\dashv					-	
Relinquished By:		Date:	Time:		Received By:	A	4	_ ^	Т	īme:			Sam	ple Condition Upon Receipt:	
Herry	~~~~~ <u>~</u>	6/30/17	11:00			6/	30/	17	1	1:4		ced		Ambient or°C	
		6/30/17	11.1		17000	- 6/	30 /)	14	1	1:48		ced		Ambient orOC	

*To be diluted with M-INF dilution water. Le

Nautilus IDs 17-0738 to-0743

Appendix D

Reference Toxicant Test Data and Statistical Analyses

CETIS Summary Report

Report Date:

05 Jul-17 15:41 (p 1 of 1)

Test Code:

170630sprt | 19-1859-0537

								rest Code:		170	63USPR 18	g-1009-U53,
Echinoid Spe	erm Cell Fertiliza	tion Tes	t 15C							Nautilu	s Environm	nental (CA)
Batch ID: Start Date: Ending Date: Duration:	06-7011-3891 30 Jun-17 17:50 30 Jun-17 18:30 40m	0 P	est Type: Protocol: Species: Source:	Fertilization EPA/600/R-95/136 (1995) Strongylocentrotus purpuratus Pt. Loma				Analyst: Diluent: Brine: Age:		ıral Seawat Applicable	er	
Sample ID: Sample Date: Receive Date Sample Age:	: 30 Jun-17	N S	code: faterial: cource: tation:	170630sprt Copper chlorid Reference Tox Copper Chlorid	icant			Client: Project:	Inter	rnal		
Comparison	Summary									MANAGEMENT AND ADDRESS OF THE PARTY OF THE P		
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	TU	Meth	od			
03-8133-6222	Fertilization Rat	te	10	20	14.14	6.35%	***************************************	Dunr	ett M	ultiple Com	parison Tes	st
Point Estima	te Summary											
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	TU	Meth	od			
20-9128-8719	Fertilization Rat	e	EC50	39.38	37.84	40.98		Trim	med S	Spearman-k	Kärber	
Test Accepta	bility								······································			•
Analysis ID	Endpoint		Attrib	ute	Test Stat	TAC Limi	its	Over	lap	Decision		
03-8133-6222	Fertilization Rat	:e	Contro	ol Resp	0.946	0.7 - NL	····	Yes		Passes A	cceptability	Criteria
20-9128-8719	Fertilization Rat	e	Contro	ol Resp	0.946	0.7 - NL		Yes		Passes A	cceptability	Criteria
03-8133-6222	Fertilization Rat	e	PMSE		0.06351	NL - 0.25		No		Passes A	cceptability	Criteria
Fertilization F	Rate Summary					- HAROSENIA CA						
C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std E	Err	Std Dev	CV%	%Effect
0	Lab Control	5	0.946	0.9148	0.9772	0.91	0.98	0.011	122	0.0251	2.65%	0.0%
10		5	0.934	0.9005	0.9675	0.9	0.96	0.012	208	0.02702	2.89%	1.27%
20		5	0.822	0.7489	0.8951	0.75	0.89	0.026	34	0.05891	7.17%	13.11%
40		5	0.546	0.4032	0.6888	0.44	0.72	0.051	144	0.115	21.07%	42.28%
80		5	0.034	0.009796	0.0582	0	0.05		3718	0.01949	57.33%	96.41%
160		5	0	0	0	0	0	0		0		100.0%
Fertilization F	Rate Detail											
C-μg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5						
0	Lab Control	0.95	0.98	0.91	0.95	0.94						
10		0.95	0.95	0.91	0.96	0.9						
20		0.87	0.75	0.78	0.82	0.89						
40		0.45	0.53	0.59	0.72	0.44						
80		0.05	0.04	0	0.04	0.04						
160		0	0	0	0	0						

Analyst: AC QA: VTP7 1417

Report Date:

05 Jul-17 15:41 (p 1 of 2)

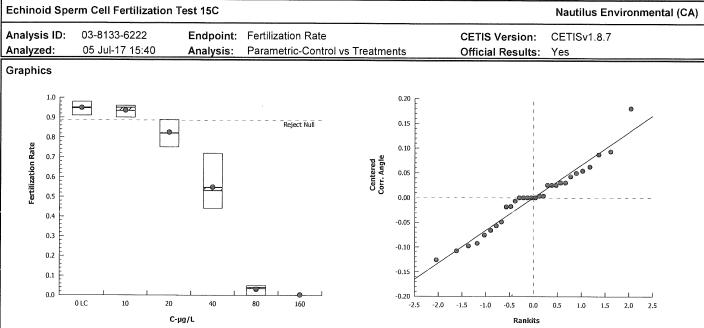
Test Code: 170630sprt | 19-1859-0537

0 Lab Control 5 1.342 1.269 1.414 1.345 1.266 1.429 0.02615 4.36% 0.0% 10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92%						~		1621	Code:	1700	osuspit i	9-1859-0537
Analyzed O5 Jul 17 15 to 10 J	Echinoid Sp	erm Cell Fertiliz	ation Test	t 15C						Nautilus	s Environr	nental (CA)
Pata Transform Pata Pat	Analysis ID:	03-8133-6222	E	ndpoint: Fer	tilization Ra	te		CET	IS Version:	CETISv1	.8.7	
May May	Analyzed:	05 Jul-17 15:4	10 A	nalysis: Par	ametric-Cor	ntrol vs Trea	itments	Offic	ial Results	: Yes		
Control Variance Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU	
Control Vs C-yg/L Control	Angular (Corr	rected)	NA	C > T	NA	NA		6.35%	10	20	14.14	
	Dunnett Mul	tiple Compariso	n Test					7.004				
10	Control	vs C-μg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(α:5%)		
40° 10.15 2.305 0.116 8 <0.0001 CDF Significant Effect	Lab Control	10	***************************************	0.5336	2.305	0.116 8	0.5887					
No		20*		4.037	2.305	0.116 8	0.0012	CDF	Significar	nt Effect		
No		40*		10.15	2.305	0.116 8	<0.0001	CDF	Significan	t Effect		
Source Sum Squ* Mean Squ* Sq		80*		23.26	2.305	0.116 8	<0.0001		-			
Between	ANOVA Tabl	е										
Error 0.006281928 20 Total 4.800103	Source	Sum Sqւ	uares	Mean Squ	ıare	DF	F Stat	P-Value	Decision	(α:5%)		
Distribution Test	Between	4.674465		1.168616		4	186	<0.0001	Significan	t Effect		
Distributional Tests Fest Test Stat Statical P-Value Decision(α:1%) Decision(α:1%) Decision(α:1%) Decision(α:	Error	0.125638	6	0.0062819	28	20			_			
Attribute Test Test Stat Critical P-Value Decision(α:1%) Variances Distribution Bartlett E∪ality of Variance Shapiro-Wik W Normality 3.034 13.28 0.5522 Equal Variances 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 0.946 0.9148 0.9772 0.95 0.91 0.98 0.01122 2.65% 0.0% 10 5 0.946 0.9148 0.9772 0.95 0.91 0.98 0.01122 2.65% 0.0% 10 5 0.934 0.9005 0.967 0.95 0.91 0.98 0.01122 2.65% 0.0% 10 5 0.934 0.9005 0.96 0.91 0.98 0.01208 2.89% 1.27% 40 5 0.546 0.4032 0.6888 0.53 0.44	Total	4.800103				24						
Variances Bartlett Equality of Variance Shapiro-Wilk W Normality 0.9701 0.8877 0.6469 Normal Distribution	Distributiona	al Tests								+		
Distribution Shapiro-Wilk W Normal Normal Distribution Normal Distribution	Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Fertilization Rate Summary Su	Variances	Bartlett B	Equality of	Variance	3.034	13.28	0.5522	Equal Var	iances			
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 0.91 0.98 0.01122 2.65% 0.0% 10 5 0.934 0.9005 0.9675 0.95 0.9 0.96 0.01208 2.89% 1.27% 20 5 0.822 0.7489 0.8951 0.82 0.75 0.89 0.02634 7.17% 13.11% 40 5 0.546 0.4032 0.6888 0.53 0.44 0.72 0.05144 21.07% 42.28% 80 5 0.034 0.009796 0.0582 0.04 0 0.05 0.008718 57.33% 96.41% 160 Template (Corrected) Transformers Very State (Corrected) Transformers 20 Lab Control 5 1.342 1.269 1.414 1.345 1.266 <td>Distribution</td> <td>Shapiro-</td> <td>Wilk W No</td> <td>ormality</td> <td>0.9701</td> <td>0.8877</td> <td>0.6469</td> <td>Normal Di</td> <td>stribution</td> <td></td> <td></td> <td></td>	Distribution	Shapiro-	Wilk W No	ormality	0.9701	0.8877	0.6469	Normal Di	stribution			
0 Lab Control 5 0.946 0.9148 0.9772 0.95 0.91 0.98 0.01122 2.65% 0.0% 10 5 0.934 0.9005 0.9675 0.95 0.9 0.96 0.01208 2.89% 1.27% 20 5 0.822 0.7489 0.8951 0.82 0.75 0.89 0.02634 7.17% 13.11% 40 5 0.546 0.4032 0.6888 0.53 0.44 0.72 0.05144 21.07% 42.28% 80 5 0.034 0.009796 0.0582 0.04 0 0.05 0.008718 57.33% 96.41% 160 5 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	Fertilization	Rate Summary										
10 5 0.934 0.9005 0.9675 0.95 0.9 0.96 0.01208 2.89% 1.27% 20 5 0.822 0.7489 0.8951 0.82 0.75 0.89 0.02634 7.17% 13.11% 40 5 0.546 0.4032 0.6888 0.53 0.44 0.72 0.05144 21.07% 42.28% 80 5 0.034 0.009796 0.0582 0.04 0 0.05 0.008718 57.33% 96.41% 160 5 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
20	-	Lab Control		0.946	0.9148	0.9772	0.95	0.91	0.98	0.01122	2.65%	0.0%
40 5 0.546 0.4032 0.6888 0.53 0.44 0.72 0.05144 21.07% 42.28% 80 5 0.034 0.009796 0.0582 0.04 0 0.05 0.008718 57.33% 96.41% 160 5 0 0 0 0 0 0 0 0 0 0 0 0 100.0% 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 1.342 1.269 1.414 1.345 1.266 1.429 0.02615 4.36% 0.0% 10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%						0.9675	0.95	0.9	0.96	0.01208	2.89%	1.27%
80 5 0.034 0.009796 0.0582 0.04 0 0.05 0.008718 57.33% 96.41% 160 5 0 0 0 0 0 0 0 0 0 0 0 100.0% 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 1.342 1.269 1.414 1.345 1.266 1.429 0.02615 4.36% 0.0% 10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%								0.75	0.89	0.02634	7.17%	13.11%
160 5 0 0 0 0 0 0 0 0 0 0 0 0 100.0% 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 1.342 1.269 1.414 1.345 1.266 1.429 0.02615 4.36% 0.0% 10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%								0.44	0.72	0.05144	21.07%	42.28%
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 1.342 1.269 1.414 1.345 1.266 1.429 0.02615 4.36% 0.0% 10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%									0.05	0.008718	57.33%	96.41%
C-μg/L Control Type Count Mean 95% LCL 95% UCL Median Min Max Std Err CV% %Effect 0 Lab Control 5 1.342 1.269 1.414 1.345 1.266 1.429 0.02615 4.36% 0.0% 10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%	160		5	0	0	0	0	0	0	0		100.0%
0 Lab Control 5 1.342 1.269 1.414 1.345 1.266 1.429 0.02615 4.36% 0.0% 10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%	Angular (Corrected) Transformed Summary											
10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%	C-μg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
10 5 1.315 1.248 1.382 1.345 1.249 1.369 0.02402 4.08% 1.99% 20 5 1.139 1.042 1.236 1.133 1.047 1.233 0.03493 6.85% 15.08% 40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%	0	Lab Control	5	1.342	1.269	1.414	1.345	1.266	1.429	0.02615	4.36%	0.0%
40 5 0.833 0.6864 0.9796 0.8154 0.7253 1.013 0.05281 14.18% 37.92% 80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%			5	1.315	1.248	1.382	1.345	1.249	1.369	0.02402	4.08%	
80 5 0.1759 0.08757 0.2643 0.2014 0.05002 0.2255 0.03182 40.45% 86.89%	20		5	1.139	1.042	1.236	1.133	1.047	1.233	0.03493	6.85%	15.08%
100	40		5	0.833	0.6864	0.9796	0.8154	0.7253	1.013	0.05281	14.18%	37.92%
<u>160</u> <u>5 0.05002 0.05001 0.05003 0.05002 0.05002 0</u> <u>0.05002 0</u> <u>0.0% 96.27%</u>	80		5	0.1759	0.08757	0.2643	0.2014	0.05002	0.2255	0.03182	40.45%	86.89%
	160		5	0.05002	0.05001	0.05003	0.05002	0.05002	0.05002	0	0.0%	96.27%

Report Date:

05 Jul-17 15:41 (p 2 of 2)

Test Code: 170630sprt | 19-1859-0537



Report Date:

05 Jul-17 15:41 (p 1 of 1)

Test Code:

170630sprt | 19-1859-0537

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Analysis ID: Analyzed:

20-9128-8719 05 Jul-17 15:40

Endpoint: Fertilization Rate Analysis:

Trimmed Spearman-Kärber

CETIS Version:

CETISv1.8.7

Official Results:

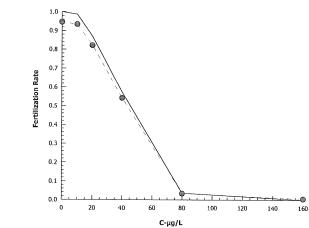
Yes

rimmed Spearman-Karber Estimates	3
----------------------------------	---

Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0.054	1.27%	1.595	0.008643	39.38	37.84	40.98

Fertilizati	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	0.946	0.91	0.98	0.01122	0.0251	2.65%	0.0%	473	500
10		5	0.934	0.9	0.96	0.01208	0.02702	2.89%	1.27%	467	500
20		5	0.822	0.75	0.89	0.02634	0.05891	7.17%	13.11%	411	500
40		5	0.546	0.44	0.72	0.05144	0.115	21.07%	42.28%	271	500
80		5	0.034	0	0.05	0.008718	0.01949	57.33%	96.41%	17	500
160		5	0	0	0	0	0		100.0%	0	500

Graphics



Report Date:

05 Jul-17 15:41 (1 of 1)

Echinoid Sperm Cell Fertilization Test 15C

Sigma:

11.22

CV:

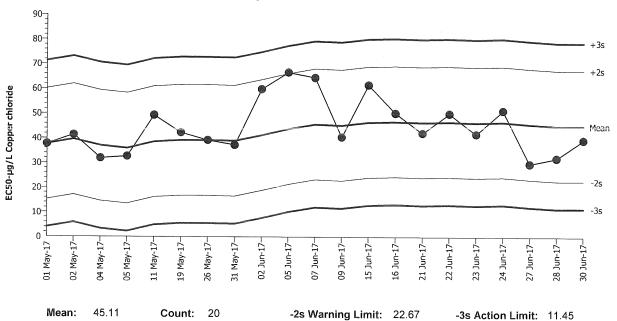
24.90%

Nautilus Environmental (CA)

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:

67.55

+3s Action Limit: 78.77

Quali	ty Con	trol Data	a								
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2017	May	1	17:50	37.72	-7.386	-0.6583			18-0409-9294	21-4338-2021
2			2	11:56	41.38	-3.729	-0.3324			15-1584-1378	12-5072-1723
3			4	16:15	31.87	-13.24	-1.18			08-0627-0095	01-9095-4567
4			5	15:10	32.62	-12.49	-1.113			16-0368-0463	05-0853-8226
5			11	15:35	49.31	4.197	0.3741			01-5463-5574	13-6197-3009
6			19	16:58	42.3	-2.814	-0.2508			01-3808-3529	10-6921-8357
7			26	16:55	39.23	-5.877	-0.5238			19-8209-6027	02-5970-9183
8			31	15:42	37.28	-7.829	-0.6978			01-0947-8219	10-1735-9410
9		Jun	2	17:00	59.87	14.76	1.315			07-9823-1222	14-2289-5480
10			5	16:33	66.68	21.57	1.922			12-3374-6038	12-3894-8538
11			7	17:28	64.53	19.42	1.731			14-9580-6318	06-4574-2541
12			9	17:15	40.49	-4.625	-0.4122			07-9177-2202	00-5029-9905
13			15	14:30	61.64	16.53	1.473			08-8166-8875	14-1352-0089
14			16	14:30	50.25	5.14	0.4581			02-2217-4481	09-2276-1445
15			21	14:17	42.15	-2.958	-0.2637			20-6379-6831	00-5386-2071
16			22	17:25	50.05	4.935	0.4399			10-9823-5082	04-6220-9409
17			23	16:55	41.8	-3.307	-0.2948			06-0771-4160	11-6079-2504
18			24	13:27	51.33	6.219	0.5543			01-7420-9579	03-5890-9605
19			27	13:13	29.77	-15.34	-1.367			11-6174-9094	14-8592-6950
20			28	14:40	32.02	-13.09	-1.167			06-0030-2581	03-5443-1685
21			30	17:50	39.38	-5.733	-0.511			19-1859-0537	20-9128-8719

CETIS Test Data Worksheet

Report Date: Test Code: 29 Jun-17 18:42 (p 1 of 1)

19-1859-0537/170630sprt

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: End Date:

30 Jun-17 30 Jun-17 Species: Strongylocentrotus purpuratus

Sample Code: 170630sprt
Sample Source: Reference Toxicant

Sample Date: 30 Jun-17

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

Sample Station: Copper Chloride

C-μg/L	Code	Rep	Pos	# Counted	il: Copper ch # Fertilized	loride Sample Station: Copper Chloride Notes
			1	100	96	7/3/17
			2	1	16	113117
			3		91	
			4		37	
			5			
			6		82 82	
			7	. }	0	
			8		53	
			9		Ý.	
			10		Ò	
			11		94	
			12		0	
			13		O	
			14		5	
			15		90	
			16		39	
			17		98	
			18		78	
			19		0_	
			20		75	
			21	To the second se	72	
	-		22		4	
			24	1000	0	
			25		44	
			26		45	
***************************************			27	1	95	·
			28			
			29	libility of the second	95	
			30	-	91	

Report Date:

29 Jun-17 18:42 (p 1 of 1)

Test Code:

19-1859-0537/170630sprt

Echinoid Sperm Cell Fertilization Test 15C

Nautilus Environmental (CA)

Start Date: 30 Jun-17 End Date: 30 Jun-17 Sample Date: 30 Jun-17 Material: Copper chloride

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

Sample Code: 170630sprt Sample Source: Reference Toxicant Sample Station: Copper Chloride

inple Date					ar: Copper chloride	Sample Station: Copper Chloride	
C-µg/L		Rep	Pos	# Counted	# Fertilized	Notes	
0	LC	1	28	100	93	AP 7/1/17	
0	LC	2	17				
0	LC	3	29				
0	LC	4	27				
0	LC	5	11				
10		1	26	100	91		
10		2	30	,			
10		3	3				
10		4	1				
10		5	15				
20		1	4	100	8-7		
20		2	20				
20		3	18				
20		4	6				
20		5	16				
40		1	25	100	29		
40		2	8				
40		3	5				
40		4	21				
40		5	24				
80		1	14	100	5		
80		2	22				
80		3	7				
80		4	2				
80		5	9				
160		1	10	100			
160		2	13	1			
160		3	19				******
160		4	23				
160		5	12				

QC: CG

Marine Chronic Bioassay

Water Quality Measurements

Client:

Internal

Test Species: S. purpuratus

Sample ID:

CuCl₂

Start Date/Time: 6/30/2017

1750

Test No:

170630sprt

End Date/Time: 6/30/2017

830

Dilutions made by:

AD

Cu stock concentration (µg/L):

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

160 7.8

Final Volume (mL):

500

Analyst:

CH Initial Readings Concentration DO рΗ Salinity Temperature (μg/L) (mg/L) (units) (ppt) (°C) **Lab Control** 7.95 8.4 14.8 7.90 10 20 8.1 33.9 40 7.94 14.5 7.94 80 14,5 7.94 160 14.3

Comments:		
	0 (- 1)	
QC Check:	AC 7/5/17	Final Review: KTP 7/4/17

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

Marine Chronic Bioassay

Echinoderm Sperm-Cell Fertilization Worksheet

Client: Sample ID: Test No.:	Internal CuCIZ 170630sprt		End Date/Tim	es: S. purpuratus
Tech initials: Injection Time:	AO		Date Collecte	
injection rime.	1050			
Sperm Absorbance at 400 nm: 1.009 (target range of 0.8 - 1.0 for density of 4x10 ⁶ sperm/ml)				
Eggs Counted:	Mean: X 50 = 4040 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:	4000 eggs/ml	= dilution factor - 1.0 part egg stoc parts seawate	k seawater	ml ml
Prepare the embryo stock according to the calculated dilution factor. For example, if the dilution factor is 2.25, use 100 ml of existing stock (1 part) and 125 ml of dilution water (1.25 parts).				
Rangefinder Test: ml Sperm Stock ml Seawater	2000:1 1600:1 50 40 0.0 10	Sperm: 1200:1 800:1 30 20 30 30	400:1 200:1 10 5.0 40 45	100:1 50:1 2.5 1.25 47.5 48.75
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:	Time Ra 1710 1723 1733	ngefinder Ratio: Fe	Unfert. 1,83 16,17	
NOTE: Choose a sperm-to-egg ratio that results in fertilization between 80 and 90 percent. If more than one concentration is within this range, choose the ratio closest to 90 percent unless professional judgment dictates consideration of other factors (e.g., organism health, stage of reproductive season, site conditions).				
Definitive Test	Sp	erm:Egg Ratio Used:	75:1	
Sperm Added (100 µl): Eggs Added (0.5 ml): Test Ended:		C1 <u>O</u>	O 100	
Comments: (enodiukon BAD ONS 432	10000 el		
QC Check: Nautilus Environmental. 434	AC 7/5/17 Vandever Avenue. San Diego, C	~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~	Final Reviev	N: KFP7/6/17

Appendix E

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 or ganisms r eceived at a <u>temperature</u> greater than 3°C ou tside the r ecommended t est 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. O rganisms 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 t o age -specific pr otocol r equirements and/ or s ample ho lding t ime 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