

Appendix G Acute Toxicity Study

Renewal of NPDES CA0109223 Carlsbad Desalination Project



Poseidon Acute Salinity Tolerance Toxicity Test Results

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Data Quality Assurance:

- Nautilus Environmental is accredited in accordance with NELAP by the State of Oregon Environmental Laboratory Accreditation Program (Certificate No. 4053-001). It is also certified by the State of California Water Resources Control Board Environmental Laboratory Accreditation Program (Certificate No. 1802) and the State of Washington Department of Ecology (Lab ID C552). Specific fields of testing applicable to each accreditation are available upon request.
- o All data have been reviewed and verified.
- All test results have met minimum test acceptability criteria under their respective EPA protocols, unless otherwise noted in this report.
- o All test results have met internal Quality Assurance Program requirements.

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Results verified by: __

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INTRODUCTION

Acute survival bioassay screening tests were conducted to determine the sensitivity of two marine organisms to salinity increases above ambient seawater levels. Testing was performed to satisfy acute salinity tolerance requirements described in the Poseidon Resources [Channelside] LP (Poseidon), Carlsbad Desalination Project permit that was adopted in 2006 (Order No. R9-2006-0065).

Poseidon has contracted with Nautilus Environmental (Nautilus) to conduct the salinity studies. Acute testing was conducted using the mysid shrimp (*Americamysis bahia*), and Pacific topsmelt (*Atherinops affinis*) at Nautilus in San Diego, California in February and March, 2015. This round of testing was performed as confirmation of results from previous salinity-related toxicity threshold studies conducted by Weston Solutions, Inc. in 2007.

MATERIALS AND METHODS Test Material

Test material consisted of hypersaline brine (HSB) prepared at Nautilus using methods described in United States Environmental Protection Agency (USEPA) protocols. Briefly, natural seawater collected from the intake at Scripps Institution of Oceanography (SIO) was filtered through a 20- μ m in-line filter. Filtered seawater was then partially frozen in a -20 degrees Celsius (°C) freezer overnight. The remaining liquid (now concentrated in salinity) was decanted from the ice, which is composed mainly of fresh water. The HSB used for this study was approximately twice the salinity of ambient seawater. HSB was added to natural seawater to achieve the desired salinity test concentrations.

Acute Screening Bioassay

The study was performed in accordance with the United States Environmental Protection Agency (USEPA) protocol "Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms" (EPA/821/R-02/012, October 2002). Test methods are summarized in Table 1. All test parameters were equal for both species unless otherwise specified in the table.

Test Organisms, Age:	<i>Americamysis bahia</i> (mysid shrimp), 4-5 days old at initiation <i>Atherinops affinis</i> (Pacific topsmelt), 12-15 days old at initiation
Test Period:	Mysid Tests: 02/05/2015, 15:45 to 02/09/2015, 13:50
	03/03/2015, 15:45 to 03/07/2015, 14:30
	Topsmelt Tests: 02/10/2015, 13:40 to 02/14/2015, 11:50
	03/05/2015, 15:50 to 03/09/2015, 14:40
Test Organism Source:	Aquatic Biosystems, Inc. (Fort Collins, Colorado)
Organism Acclimation:	Acclimated to laboratory seawater (source: Scripps Institution of Oceanography intake, held at Nautilus in a laboratory flow-through system with a 20-µm in-line fiber filter and a chiller unit). Salinity approximately 33 parts per thousand (ppt).
Protocol:	USEPA Acute Manual (EPA/821/R-02/012, 2002) 96-hr static-renewal test; 80% test solution renewal at 48 hours
Control Water:	Lab Control: laboratory seawater
	Brine Control: hypersaline brine and de-ionized (DI) water were added to natural seawater at the highest brine volume tested
Aeration:	None
Test Concentrations:	February: 38, 40, 42, and 44 ppt, plus lab and brine controls March: 40, 42, 43, and 44 ppt, plus lab and brine controls
Number of Replicates, Organisms per Replicate:	6 replicates, 5 animals per replicate
Test Temperature:	Mysid Tests: 25 ±1 °C Topsmelt Tests: 20 ±1 °C
Feeding:	Mysid Tests: Artemia nauplii, twice daily
	Topsmelt Tests: Artemia nauplii, once daily
Test Acceptability:	Mean survival must be 90% or greater in the control
Statistical Analysis Software:	GraphPad Prism, v. 4.02 and CETIS version 1.8.7.20

Table 1. Acute Test Specifications

Data Analysis and Reporting

Toxicity test responses were statistically evaluated using the Comprehensive Environmental Toxicity Information System[™] (CETIS) program by Tidepool Scientific Software according to flowchart specifications provided in method guidance (USEPA 2002). Results were used to calculate the No Observed Effect Concentration (NOEC) and Lowest Observed Effect Concentration (LOEC) values. Since we are interested in effects relative to ambient seawater salinity, organism performance in the test concentrations was compared to that in the lab control rather than the brine control. In cases where the brine control result was less than lab control, a statistical comparison was made between the two controls to ensure no adverse effects were produced due to the addition of HSB.

Additionally, for a more robust analysis, data for each salinity concentration were analyzed using the Test of Significant Toxicity (TST) t-test approach specified in the National Pollutant

Discharge Elimination System Test of Significant Toxicity Implementation Document (USEPA 2010). The TST applies a modified t-test that takes into account both the statistical power of the test and magnitude of biological effects in determining the presence of a response. 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.

RESULTS

There were no statistically significant effects observed for Pacific topsmelt survival at any of the salinity concentrations tested during both rounds of testing. A NOEC of 44 ppt and a LOEC of >44 ppt is reported for this species for both rounds of testing.

For the test initiated February 5, 2015 survival of mysid shrimp in the highest test concentration (44 ppt) was 86.7 percent; this result was significantly decreased relative to the lab control according to the TST but was not statistically significant using statistical methods outlined in the statistical flowchart of the EPA 2002 test protocol. The TST resulted in a NOEC of 42 and a LOEC of 44, while the EPA 2002 flowchart method resulted in a NOEC of 44 and a LOEC of >44. During the March 2015 testing, no statistically significant effects were observed for mysid shrimp survival at any of the salinity concentrations tested, resulting in a NOEC of 44 and a LOEC of >44.

Mean survival results for the February and March 2015 tests for both species are presented in Figure 1. Tabular summaries of mean test results for the mysid shrimp and Pacific topsmelt toxicity tests are provided in Tables 2 and 3. A summary of the statistical analysis results for both species is provided in Table 4. Detailed toxicity test results and statistical summaries are provided in Appendix A.

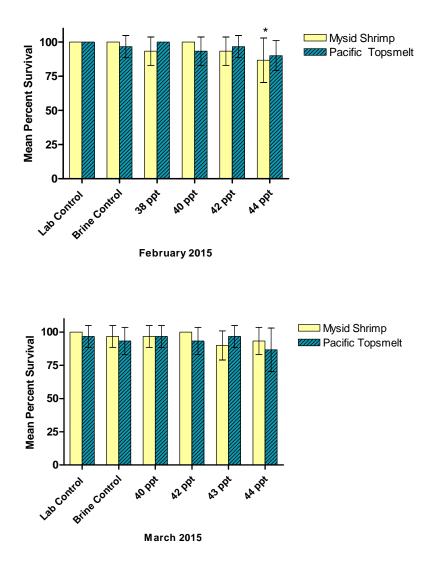


Figure 1. Summary of acute survival results for mysid and topsmelt tests conducted in February and March 2015 (mean ± SD). *The 44 ppt concentration in the February 2015 mysid test was statistically significant according to the TST, but was not significant using the USEPA 2002 flowchart statistical method (Steel Many-One Rank Sum Test).

Test		Mysid Shrimp		Pacific Topsmelt					
Concentration (ppt)	Mean Survival (%)	Standard Deviation	Percent Effect ^ª	Mean Survival (%)	Standard Deviation	Percent Effect ^ª			
Brine Control	100	0.0	0.0	97	8.2	0.0			
Lab Control	100	0.0	0.0	100	0.0	-3.5			
38	93	10	6.7	100	0.0	-3.5			
40	100	0.0	0.0	93	10	3.5			
42	42 93		6.7	97	8.2	0.0			
44	87*	16	13	90	11	6.9			

Table 2. Summary of 96-hr Acute Survival Toxicity Test Results – February 2015

n=6

^a Percent Effect = ((Mean Lab Control Response – Mean Test Concentration Response)/Mean Lab Control Response)*100. A negative value indicates better organism performance in the test concentrations than in the lab control.

* **Bold** values indicate a statistically significant effect compared to the lab control using the Test of Significant Toxicity calculation. This value was not found to be statistically significant using the traditional EPA flowchart statistical approach.

Test		Mysid Shrimp	,	Pacific Topsmelt				
Concentration (ppt)	Mean Survival (%)	Standard Deviation	Percent Effect ^a	Mean Survival (%)	Standard Deviation	Percent Effect ^a		
Brine Control	97	8.2	0.0	93	10	0.0		
Lab Control	100	0.0	-3.5	97	8.2	-3.6		
40	97	8.2	0.0	97	8.2	-3.6		
42	100	0.0	-3.5	93	10	0.0		
43	43 90		6.9	97	8.2	-3.6		
44	93	10	3.5	87	16	7.1		

Table 3. Summary of 96-hr Acute Survival Toxicity Test Results – March 2015

n=6

^a Percent Effect = ((Mean Lab Control Response – Mean Test Concentration Response)/Mean Lab Control Response)*100. A negative value indicates better organism performance in the test concentrations than in the lab control.

Table 4. Summary of Statistical Results for 96-hr Acute Mysid Shrimp and Pac	cific
Topsmelt Toxicity Tests	

Test Start Date	Statistical Test Used	NOEC (ppt)	LOEC (ppt)		
	Mysid Shrimp				
02/05/2015	Steel Many-One Rank Sum Test	44	>44		
02/05/2015	Test of Significant Toxicity	42	44		
03/03/2015	Steel Many-One Rank Sum Test	44	>44		
03/03/2015	Test of Significant Toxicity	44	>44		
	Pacific Topsmelt				
02/10/2015	Steel Many-One Rank Sum Test	44	>44		
02/10/2015	Test of Significant Toxicity	44	>44		
03/05/2015	Steel Many-One Rank Sum Test	44	>44		
03/03/2015	Test of Significant Toxicity	44	>44		

NOEC = No Observed Effect Level; the highest test concentration resulting in no observed effect

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

QUALITY ASSURANCE

Laboratory and brine controls met the minimum test acceptability criterion of 90 percent mean survival. Variability among replicates was minimal, and the ability to detect a statistical difference was deemed appropriate. Additionally, appropriate alpha levels were used to calculate TST results for this test per the 2010 EPA TST Implementation Guidance.

The formula for the brine control on the dilution preparation worksheet was incorrect for both February acute tests. In error, the technician adjusted the salinity of the brine control with seawater instead of DI water. Therefore, the volume of brine in the brine control was below that in the highest test concentration and the addition of brine was not properly controlled for. However, dilutions for all test concentrations were prepared correctly and there were no effects in any of the topsmelt concentrations, and only in the highest test concentration for mysid shrimp. The statistically significant decrease in the 44 ppt concentration of the mysid test is similar to results from previous studies using this species (Philips, et al. 2012). Additionally, all test concentrations were compared to the seawater lab control for statistical analysis; therefore, this deviation should not affect the interpretation of results.

Although there were no deviations in temperature from internal standard operating procedures for these tests, some evaporation occurred in the test chambers, particularly in the mysid test which is conducted at 25°C. This resulted in the final salinity levels approximately 0.5 to 1.0 ppt

higher at the end of the test than at initiation in some concentrations. Regardless, no effects were observed except for the highest test concentration in the mysid test.

Reference Toxicant Test

The reference toxicant tests met the acceptability criterion for survival. Additionally, the median lethal concentrations (LC_{50}) calculated for all reference toxicant tests were within two standard deviations of the internal control chart means, indicating typical test organism sensitivity to copper. Reference toxicant results are summarized in Table 5, and provided in full in Appendix B. A glossary of qualifier codes used on raw datasheets is available in Appendix C.

Test Endpoint	Test Start Date			LC₅₀ value (µg/L Cu)	Mean LC₅₀ ± 2 SD (µg/L Cu)
Mysid Shrimp	02/05/2015	100	200	258	212 ± 79.7
96-hour Survival	03/03/2015	50	100	150	207 ± 80.6
Pacific Topsmelt	02/10/2015	100	200	91.0	121 ± 60.2
96-hour Survival	03/05/2015	50	100	128	120 ± 61.7

 Table 5. Summary of Reference Toxicant Statistical Results

NOEC = No Observed Effect Level; the highest test concentration resulting in no observed effect

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

 LC_{50} = Lethal concentration 50, concentration expected to cause mortality to 50 percent of test organisms

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

REFERENCES

- Phillips, B.M., B.S. Anderson, K. Siegler, J.P. Voorhees, S. Katz, L. Jennings and R.S. Tjeerdema. 2012. Hyper-Saline Toxicity Thresholds for Nine California Ocean Plan Toxicity Test Protocols. Final Report. University of California, Davis, Department of Environmental Toxicology at Grand Canyon.
- Tidepool Scientific Software. 2000-2013. CETIS Comprehensive Environmental Toxicity Information System Software, Version 1.8.7.20.
- USEPA 2002. Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms. EPA/821/R-02/012, October 2002.
- USEPA. 2010. National Pollutant Discharge Elimination System Test of Significant Toxicity Implementation Document. EPA/833/R-10/003. June 2010.
- Weston Solutions. 2007. Toxicity Testing Results Test Substance RO Concentrate Comp. January 17, 2007.

APPENDIX A

Raw Data and Statistical Analyses

Mysid Acute Salinity Tolerance Test February 2015

CETIS Summary Report									Report Date: Test Code:			13 Mar-15 17:10 (p 1 of 1) 1502-S025 08-9077-1636		
Mysid 96-h Ac	ute Survival Te	st											ental (CA)	
Batch ID: Start Date: Ending Date: Duration:	14-3042-9926 05 Feb-15 15:4 09 Feb-15 13:5 94h	-	Test Type: Protocol: Species: Source:	EPA/82 America	1/R-02- imysis l	012 (2002) bahia tems, CO			Analyst: Diluent: Brine: Age:	Nat	ural Seawate zen Seawate			
Sample ID: Sample Date: Receive Date: Sample Age:			Code: Material: Source: Station:	Salinity Natural Poseido Nautilus	Seawat n	ler			Client: Project:	Pos	seidon			
Comparison S	Summary													
Analysis ID	Endpoint		NOEI	_ LO	EL	TOEL	PMSD	τu	М	ethod				
05-4638-4426	96h Survival R	ate	44	>4	4	NA	13.5%		St	teel Ma	ny-One Rank	Sum Test		
17-1981-1937	96h Survival R	ate	42	44		42.99	11.1%		т:	ST-Wel	ch's t Test			
96h Survival F	Rate Summary													
C-ppt	Control Type	Cou	nt Mear	95	% LCL	95% UCL	Min	Max	c St	td Err	Std Dev	CV%	%Effect	
0	Brine Control	6	1	1		1	1	1	0		0	0.0%	0.0%	
0	Lab Control	6	1	1		1	1	1	0		0	0.0%	0.0%	
38		6	0.933	3 0.8	249	1	0.8	1	0.	04216	0.1033	11.07%	6.67%	
40		6	1	1		1	1	1	0		0	0.0%	0.0%	
42		6	0.933	3 0.8	249	1 '	0.8	1	0.	04216	0.1033 '	11.07%	6.67%	
44		6	0.866	7 0.6	953	1	0.6	1	0.	06667	0.1633	18.84%	13.33%	
96h Survival F	Rate Detail													
C-ppt	Control Type	Rep	1 Rep 2	2 Re	р 3	Rep 4	Rep 5	Rep	o 6					
0	Brine Control	1	1	1		1	1	1						
0	Lab Control	1	1	1		1	1	1						
38		0.8	1	0.8		1	1	1						
40		1	1	1		1	1	1						
42		0.8	0.8	1		1	1	1						
44		1	0.6	0.8		1	0.8	1						

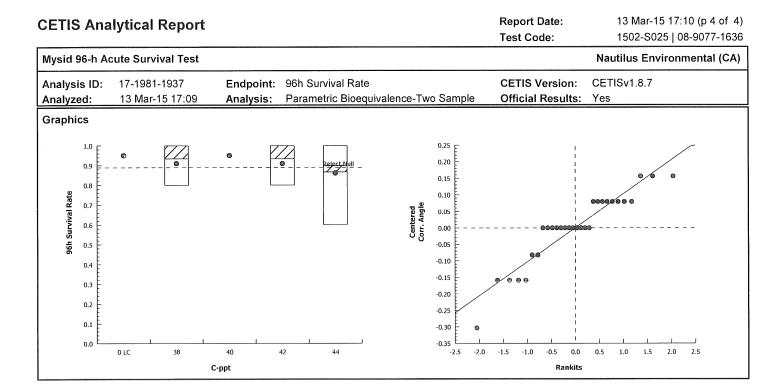
Report Date:

13 Mar-15 17:10 (p 1 of 1) 36

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Analyst: AC_____ QA: 🖄 3/ 11/ 16

CETIS Analytical Report							•	ort Date: Code:	13 Mar-15 17:09 (p 3 of _4) 1502-S025 08-9077-1636					
Mysid 96-h A	cute Survival Te	st							Nautilus	s Environm	ental (CA)			
Analysis ID: Analyzed:	17-1981-1937 13 Mar-15 17:0		dpoint : 96h alysis: Para			CETIS Version: uivalence-Two Sample Official Results:			CETISv1.8.7 Yes					
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	τυ			
Angular (Corre		NA	C*b < T	NA	NA	0.8	11.1%	42	44	42.99				
TST-Welch's	t Test													
Control	vs C-ppt		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(x:10%)					
Lab Control	38*		3.778	1.476	0.074 5	0.0065	CDF	Non-Signif	icant Effect					
	40*		0.2691	1.476	5	<0.1	CDF	Non-Signif	1					
	42*		3.778	1.476	0.074 5	0.0065	CDF	Non-Signif						
	44		1.467	1.476	0.114 5	0.1011	CDF	Significant	Effect					
ANOVA Table	9													
Source	Sum Squ	ares	Mean Squ		DF	F Stat	P-Value	Decision(
Between	0.102643		0.0256607		4	1.946	0.1340	Non-Signif	icant Effect					
Error	0.3296548		0.0131861	9	25									
Total	0.4322978	3			29									
Distributiona	l Tests													
Attribute	Test	· · · · · · · · · · · · · · · · · · ·		Test Stat		P-Value	Decision	<u>`````````````````````````````````````</u>						
Variances			y of Variance		4.177	0.0253	Equal Var							
Variances		Equality of \		14.95	4.177	<0.0001	Unequal \		m					
Distribution	Shapiro-	Wilk W Nor	mality	0.8889	0.9031	0.0046	Non-norm	al Distributio		6) 46 6 60 60 60 60 60 60 60 60 60 60 60 60 60				
96h Survival	Rate Summary													
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Мах	Std Err	CV%	%Effect			
0	Lab Control	6	1	1	1	1	1	1	0	0.0%	0.0%			
38		6	0.9333	0.8249	1	1	0.8	1	0.04216	11.07%	6.67%			
40		6	1	1	1	1	1	1	0	0.0%	0.0%			
42		6	0.9333	0.8249	1	1	0.8	1	0.04216	11.07%	6.67%			
44		6	0.8667	0.6953	1	0.9	0.6	1	0.06667	18.84%	13.33%			
Angular (Cor	rected) Transfor		mary											
C-ppt	Control Type	Count	Mean		95% UCL		Min	Max	Std Err	CV%	%Effect			
0	Lab Control	6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%			
38		6	1.266	1.137	1.395	1.345	1.107	1.345	0.0502	9.71%	5.9%			
40		6	1.345	1.345	1.345 1.395	1.345 1.345	1.345 1.107	1.345 1.345	0 0.0502	0.0% 9.71%	0.0% 5.9%			
42 44		6 6	1.266 1.189	1.137 0.9911	1.395	1.345	0.8861	1.345	0.0502	9.71% 15.88%	5.9% 11.59%			
96h Survival	Poto Dotail													
		Don 4	Don 3	Don 2	Pon A	Pon F	Pon 6							
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6							
0	Lab Control	1	1	1 0.8	1 1	1	1							
38		0.8 1	1 1	0.8 1	1	1	י 1							
40 42		0.8	і 0.8	1	1	1	1							
42		0.8 1	0.6	0.8	1	0.8	1							
				0.0		U.U	•							
•	rected) Transfor			Rep 3	Rep 4	Rep 5	Rep 6							
C-ppt 0	Control Type Lab Control	Rep 1	Rep 2	<u>кер з</u> 1.345	1.345	1.345	1.345							
0 38		1.345 1.107	1.345 1.345	1.345	1.345 1.345	1.345	1.345							
38 40		1.345	1.345	1.345	1.345	1.345	1.345							
		1.345	1.345	1.345	1.345 1.345	1.345	1.345							
42				1.345	1.345 1.345	1.345	1.345							
44		1.345	0.8861	1.107	1.040	1.107	1.040							

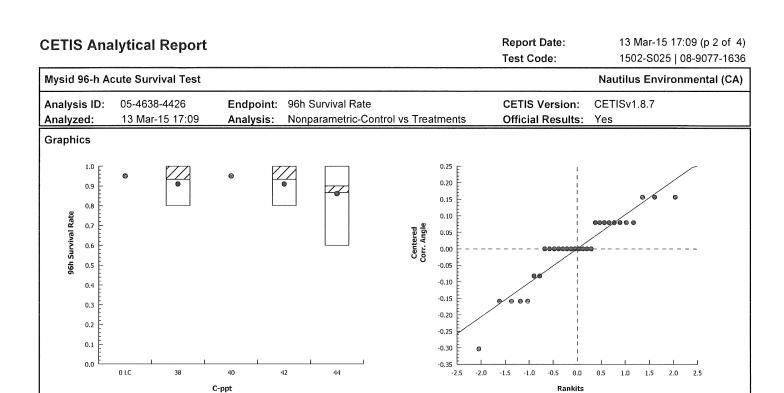


000-089-170-2

Analyst: AC QA: 5-3116115

CETIS Analytical Report								-	ort Date: Code:	13 Mar-15 17:09 (p 1 of 4) 1502-S025 08-9077-1636				
Mysid 96-h A	Acute Survival Te	st								Nautilus	Environn	nental (CA)		
Analysis ID: Analyzed:	05-4638-4426 13 Mar-15 17:0		n dpoint: 96h nalysis: Nor	Survival Ra		vs T	reatments		IS Version: al Results:	CETISv1. Yes	8.7			
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed			PMSD	NOEL	LOEL	TOEL	τυ		
Angular (Corr	rected)	NA	C > T	NA	NA			13.5%	44	>44	NA			
Steel Many-C	One Rank Sum T	est												
Control	vs C-ppt		Test Stat	Critical	Ties	DF	P-Value	P-Type	Decision(a:5%)				
Lab Control	38		33	25	1	10	0.3906	Asymp	Non-Signif	icant Effect				
	40		39	25	1	10	0.8000	Asymp	Non-Signif	icant Effect				
	42		33	25	1	10	0.3906	Asymp	Non-Signif	icant Effect				
	44		30	25	1	10	0.2033	Asymp	Non-Signif	icant Effect				
ANOVA Table	e													
Source	Sum Squ	ares	Mean Squ	are	DF		F Stat	P-Value	Decision(a:5%)				
Between	0.102643		0.0256607	5	4		1.946	0.1340	Non-Signif	icant Effect				
Error	0.3296548		0.0131861	9	25		-							
Total	0.4322978	3			29			An and a second s						
Distributiona	al Tests													
Attribute	Test			Test Stat	Critica		P-Value	Decision	(α:1%)					
Variances	Mod Leve	ene Equali	ty of Variance	3.342	4.177		0.0253	Equal Var	iances					
Variances	Levene E	Equality of	Variance	14.95	4.177		<0.0001	Unequal \	/ariances					
Distribution	Shapiro-	Nilk W No	rmality	0.8889	0.9031		0.0046	Non-normal Distribution						
96h Survival	Rate Summary													
C-ppt	Control Type	Count	Mean	95% LCL	95% U	CL	Median	Min	Max	Std Err	CV%	%Effect		
0	Lab Control	6	1	1	1		1	1	1	0	0.0%	0.0%		
38		6	0.9333	0.8249	1		1	0.8	1	0.04216	11.07%	6.67%		
40		6	1	1	1		1	1	1	0	0.0%	0.0%		
42		6	0.9333	0.8249	1		1	0.8	1	0.04216	11.07%	6.67%		
44		6	0.8667	0.6953	1		0.9	0.6	1	0.06667	18.84%	13.33%		
Angular (Cor	rected) Transfor	med Sum	mary											
C-ppt	Control Type	Count	Mean	95% LCL	95% U	CL	Median	Min	Max	Std Err	CV%	%Effect		
0	Lab Control	6	1.345	1.345	1.345		1.345	1.345	1.345	0	0.0%	0.0%		
38		6	1.266	1.137	1.395		1.345	1.107	1.345	0.0502	9.71%	5.9%		
40		6	1.345	1.345	1.345		1.345	1.345	1.345	0	0.0%	0.0%		
42		6	1.266	1.137	1.395		1.345	1.107	1.345	0.0502	9.71%	5.9%		
44		6	1.189	0.9911	1.388		1.226	0.8861	1.345	0.07712	15.88%	11.59%		
96h Survival														
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4		Rep 5	Rep 6						
0	Lab Control	1	1	1	1		1	1						
38		0.8	1	0.8	1		1	1						
40		1	1	1	1		1	1						
42		0.8	0.8	1	1		1	1						
44		1	0.6	0.8	1		0.8	1						
Angular (Cor	rected) Transfor	med Detai	il											
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4		Rep 5	Rep 6						
0	Lab Control	1.345	1.345	1.345	1.345		1.345	1.345						
38		1.107	1.345	1.107	1.345		1.345	1.345						
40		1.345	1.345	1.345	1.345		1.345	1.345						
42		1.107	1.107	1.345 1.107	1.345 1.345		1.345	1.345						

Analyst: <u>AC</u>, QA: <u>13-3/16/</u>16



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AC QA: 5-3/16/15

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Mysid 96-h A	cute Survival Te	est							Nautilu	s Environ	mental (CA
Analysis ID: Analyzed:	08-2120-2616 13 Mar-15 17:′		•	Survival Ra	ite Two Sample	e		IS Version: al Results		.8.7	
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed			Test Res	ult		
Angular (Corre	ected)	NA	C > T	NA	NA			Passes 9	6h survival r	ate	
Wilcoxon Rai	nk Sum Two-Sa	mple Test									
Control	vs Control		Test Stat	Critical	Ties DF	P-Value	Р-Туре	Decision	(α:5%)		
Lab Control	Brine Co	ntrol	39	NA	1 10	1.0000	Exact	Non-Sign	ificant Effect	t	
ANOVA Table	9										
Source	Sum Squ	ares	Mean Squ	are	DF	F Stat	P-Value	Decision	(α:5%)		
Between	0		0		1	65540	<0.0001	Significar	nt Effect		
Error	0		0		10	-					
Total	0				11						
96h Survival	Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1	1	1	1	1	1	0	0.0%	0.0%
0	Brine Control	6	1	1	1	1	1	1	0	0.0%	0.0%
Angular (Cor	rected) Transfor	med Sumr	mary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL		Min	Max	Std Err	CV%	%Effect
0 0	Lab Control Brine Control	6 6	1.345 1.345	1.345 1.345	1.345 1.345	1.345 1.345	1.345 1.345	1.345 1.345	0 0	0.0% 0.0%	0.0% 0.0%
		0	1.345	1.545	1.040	1.040	1.040	1.545	0	0.078	0.078
96h Survival		5 4		D	D	D	D 0				
C-ppt	Control Type Brine Control	Rep 1 1	Rep 2	Rep 3 1	Rep 4 1	Rep 5	Rep 6 1				
0 0	Lab Control	1	1	1.	1	1	1				
				•	•	•					
	rected) Transfor			Don 2	Bon 4	Bop 5	Bonf				
C-ppt 0	Control Type Brine Control	Rep 1 1.345	Rep 2 1.345	Rep 3 1.345	Rep 4 1.345	Rep 5 1.345	Rep 6 1.345		99999494994949494949494949494944444444		
0	Lab Control	1.345	1.345	1.345	1.345	1.345	1.345				
Graphics											
Graphics											
1.0 E	۵		•			1.0E+00	•				
0.9						-			1		
9.8 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1						7.5E-01					
22 0.7 E					per	ngle			1		
96h Survival Rate					Cente	Corr. Angle			1		
496 0.5						5.0E-01	-		1		
0.4 E						-			1		
0.3						2.5E-01			1		
0.2									1		
0.1						[1		
0.0 È	0 BC	l	0 LC		Ĺ	0.0E+00 -2.0	0 -1.5 -1	.0 -0.5	0.0 0.5	1.0 1.5	J 2.0
	0.00	C-ppt	510			-2.0	- 1.5 -1	Ranki		1.5 1.5	2.0

Analyst: <u>AC</u> QA: <u>93/16/1</u>5

Water Quality Measurements & Test Organism Survival

Client:	Client: Poseidon Test Species: <u>A. bahia</u>							
Sample ID:	Nautilus Frozen Brine	Start Date/Time: 2/5/2015 1545		0	24	48	72	96
Test No.:	1502-5025	End Date/Time: 2/9/2015 1350	Counts:	NH	EG	NH	CH	146
			Readings:	NH	nt	нч	BK	BK

Nit Dilutions made by: Number of Live Salinity Temperature **Dissolved Oxygen** pН Concentration Organisms (ppt) (°C) (units) (mg/L)Rep ppt 0 24 48 72 96 0 24 48 72 96 0 24 72 96 0 72 48 24 48 96 0 24 48 72 96 5 334 800 7.96 7.97 7.94 5 5 33.1 33.0 33.6 34.1 257 25.6 21325.2 25,1 6.2 797 Α 72 62 Lab Control 5 6.1 5.8 ß 5 5 5 В 23.9 5 24.8 6.1 7.95 6 5 С 5 5 1 5 Ś 5 5 D 5 5 Ε 5 ς 5 5 5 F 5 S 6 5 5 5 34. 124.3 34.1 343 34.8 21.6 25.4 24,2 25,2 25.3 А ς 7.460 6.861 5.6 7.94 7.99 798 8.00 7.97 5 **Brine Control** 5 В 5 5 5 5 34.8 as 1 0 7.99 Ś 5 Š С ς 5 5 5 5 D ξ 5 5 5 E 5 S 5 5 1 F 5 5 5 Α 5 5 4 370 37.1 25 5 25.6 24.1 25.3 25.4 199 800 17.95 8,02 7.98 5 38.0 38.4 38.8 1258 6759 5.6 38.0 ppt В 6 5 5 5 5 2 25.7 201 5 4 5 Ц С 5 5 S D 5 5 5 5 5 6 S Ε 5 5 \$ 5 F 5 5 40.0 40.1 40.0 40.4 40.7 25 5 25.8 24.2 15 ς 25.2 255 7.2 5.8 6.76.1 55 792600 795 803 8.00 А ĥ 5 40.0 ppt 5 5 S 5 В 5 5 40.8 252 6.6 5.07 5 Ĉ 5 ĥ 5 5 D 5 5 55 5 ς 5 Ε 5 5 5 5 5 F 5 S ъ 4 42.0 42.2 42.0 423 429 4 4 25425.821125.22557.2 790 400 794 803 802 А 5 4 5.8 686.0 5.9 42.0 ppt в 4 Ч U 4 5 ษาต 252 8.03 1 ê S С 5 5 5 5 5 5 5 D ς 5

Initial Counts

QC'd by:_<u>A</u>₩∕

Е

F

Animal Source/Date Received: <u>14/15/ABS</u> Age at Initiation: <u>5days</u>

5

5

5 ς 5 5

5 S 5

Feeding Times 0 24 48 72 96 0845 0130 6920 0815 AM: PM: 1000 1900 537 1530 5000

Comments:

QC Check:

i = initial reading in fresh test solution, f = final reading in test chamber prior to renewal Organisms fed prior to initiation, circle one (() n) Huch Sonston 5 Salarly Metry

Final Review: AC-4/29/15

KO3/12/15 Nautilus Environmental. 4340 Vandever Avenue. San Diego, CA 92120.

Water Quality Measurements & Test Organism Survival

Client:	Pose	eidor	n							Те	st Spe	cies:	A. ba	ahia									Teo	ch Initi	ials	
Sample ID:	Na	Nt	ilu	SF	VOZ	zen	Bri	÷ ~l.		Start	Date/	/Time: 2/5/2015 1545								0	24	48	72	96		
Test No.:	15	202	2-<	50G	25		<u> </u>				Date/					350	,			C	ounts:	NH	EG	NY	СН	P6
		<u> </u>											-						•	Rea	dings:	hΑ	HY	NH	BK	BIC
																		I	Dilutio			Lit/AL	,	wh		
Concentration	Rep			ber of ganis					alinity (ppt)	/			Ten	nperat (°C)	ture				lved C (mg/L)))	I			pH (units)	
PP*		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
44.0 ppt	Α	5	5	5	5	5	44.0	412	1440	44.2	.44.9	टर्ड्	26.6	241	25.1	25,5	7.2	5.8	6.7	5.9	5.8	7.87	Z.00	7,94	8,06	802
(\mathfrak{d})	в	5	É	5	5	3			440					r 251					f 5,8					f 8,63		
	С	5	5	5	5	el																				
	D	5	5	5	5	5																				
	Е	5	5	r L	4	Ц																				
	F	5	5	2	5	5																				
	Α	5						******	i					I					i					i		
			1	1									1	e				1	6	1		Π	0.0000000	5	1-2010-001	10000000000

	~	5	5	<u> </u>	2	4								ļ	ĺ											
	D	5	5	5	5	5																				
	Е	5	5	21	4	Ц																				
	F	5	5	5	5	5																				
	Α	5	Ĺ						i					1					1							
	в	5							f					f					f					f		
	С	5																								
	D	Б																								
	E	5																								
	F	5																								
	Α	5							i					I					i					1		
	в	5							f					f					f					f		
	С	Б																								
	D	5																								
	E	\$																								
	F	5																								
	Α	\$							I					i					1					1		
	в	Б							f					f					f					f		
	С	Б																								
	D	Б																								
	Е	Б																								
	F	\$																								
	Α	5							I					1					i 					ľ		
	в	5							f					f					f					f		
	С	5																								
	D	5																								
	Е	5																								
	F	5	211	¥Β																						
Initial Counts QC'd by:	A	. /	211	2/15	>																					
			-																							
Animal Source/[Date F	Recei	ived:	214	115/	<u>AB5</u>	>	_		Age	at Init	iation:	50	day	5		-						Feed	ding T	imes	
					l									1								0	24	48	72	96
Comments:		i = in	nitial re	eading	g in fr	esh te	st solu	ution, f	= fina	l read	ing in	test ch	ambei	r prior	to ren	ewal					AM:		C&F5	0830	6926	0815
		Orga	anism	s fed	prior t	o initia	ation, d	circle o	one (<u>у</u>) г	ı)	Hud	<u>n S</u>	ensl	on!	5 50	lint	1 h	r efer		PM:	160	1920	163	P1530	-
								Ò	Sab	samy	ite i	1Na/	had	w/1	o I C	+ 1	11	1471	ea f	ь				1	и.	
QC Check:	K	56	<u>əh</u> a	<u>>\</u> [5				0 6 H	sm 's	altw	Hud 111a 4 n	neusi	hv hm	1 fo	va	$\ln du$	15.		Fi	nal Re	eview:		KL	<u> 21 </u>	15

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

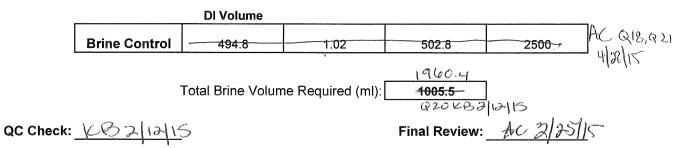
Brine Dilution Worksheet

Marine	Chronic	Bioassay
--------	---------	----------

Project:	Poseidon		Analyst: NH
Sample ID:	frozen seawater		Test Date: 2/5/2015 ((mitiation))
Test No:	1502-502	5	Test Type: My-a (Acute My sid)
Salinity of Sea	awater	33.1	
Salinity of Bri	ne	87.3	Date of Brine used: <u>117/15</u>
Test Dilution	/olume	2500	Alkalinity of Brine Control: _) 이 너 mg/L as CaCO3

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

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
Lab Control	100.0	250	NA	NA	2500
38.0	91.0	2274.0	0.10	226.0	2500
40.0	87.3	2181.7	0.15	318.3	2500
42.0	83.6	2089.5	0.20	410.5	2500
44.0	79.9	1997.2	0.25	502.8	2500



Q21: The bine control formula was incorrect and the resulting solinity was high (44 ppt). The technician reduced Salinity with seawater instead of DI. Therefore, the mine control did not accurately control for the maximum addition of brine. See QA section of report.

Brine Dilution Worksheet

Marine	Chronic	Bioassay
--------	---------	----------

Project:	Poseidon		Analyst: NH	
Sample ID:	frozen seawater		Test Date: 2/5/2015 (PC	newal)
Test No:	1502-502	6	Test Type: My-a (ຼີຼ່ Aເປີຍ	2 mysid)
Salinity of Se	awater	33.0	_	
Salinity of Br	ine	90.9	Date of Brine used: 12/8/14	
Test Dilution	Volume	1500	Alkalinity of Brine Control: 106	mg/L as CaCO3
TS = targo	t calinity			

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

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
Lab Control	100.0	250	NA	NA	1500
38.0	91.4	1370.5	0.09	129.5	1500
40.0	87.9	1318.7	0.14	181.3	1500
42.0	84.5	1266.8	0.18	233.2	1500
44.0	81.0	1215.0	0.23	285.0	1500

DI Volume AC-	
Brine Control 303.8 0.94 285.0 1500 QI&Q'2.1	
インゴロS Total Brine Volume Required (ml): 1114.0	
QC Check: $1000000000000000000000000000000000000$	
Q21: The brine control formula was incorrect and the resulting Sal was high. The technician reduced salinity with seawater instead of DI. Morefore, the brine control did not accurately control for the maximum addition of brin There was limited volume of the brine vsed Set	inity e.
the initiation. Q21:	

Topsmelt Acute Salinity Tolerance Test February 2015

CETIS Summary Report

Report Date: Test Code:

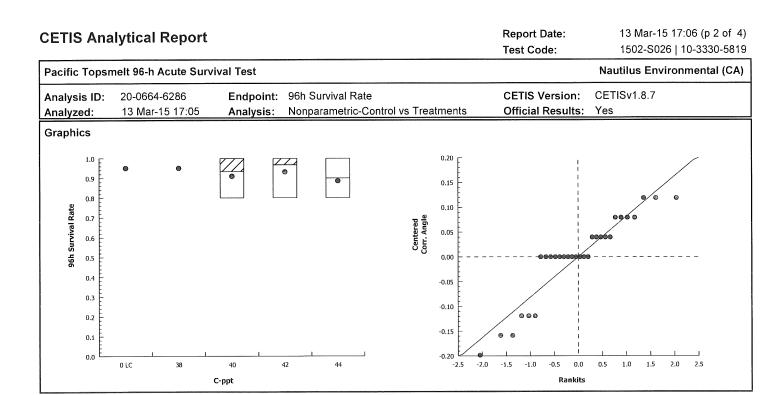
13 Mar-15 17:07 (p 1 of 1) 1502-S026 | 10-3330-5819

							165	st coue.	100	2-3020 10	-000-001
Pacific Topsm	nelt 96-h Acute	Surviva	al Test						Nautilu	s Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	09-5191-8844 10 Feb-15 13:4 14 Feb-15 11:5 94h		Test Type: Protocol: Species: Source:	Survival (96h) EPA/821/R-02 Atherinops aff Aquatic Biosys	inis			ne: Fro	ural Seawat zen Seawate d		
Sample ID: Sample Date: Receive Date: Sample Age:	: 10 Feb-15	Feb-15 Feb-15 mary		Code:Salinity StudyMaterial:Natural SeawaterSource:PoseidonStation:Nautilus Brine				ent: Pos bject:	seidon		
Comparison S	Summary										
Analysis ID	Endpoint		NOEI	LOEL	TOEL	PMSD	τu	Method			
20-0664-6286	96h Survival Ra 96h Survival Ra		44 44	>44 >44	NA NA	11.5% 8.97%	Benanchus na State ann an State an Anna Cardon e State a		ny-One Ranl ch's t Test	< Sum Test	
96h Survival F	Rate Summary										
C-ppt	Control Type	Coun	nt Mean	95% LCL	. 95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Brine Control	6	0.966	7 0.881	1	0.8	1	0.03333	0.08165	8.45%	0.0%
0	Lab Control	6	1	1	1	1	1	0	0	0.0%	-3.45%
38		6	1	1	1	1	1	0	0	0.0%	-3.45%
40		6	0.933	3 0.8249	1	0.8	1	0.04216	0.1033	11.07%	3.45%
42		6	0.966	7 0.881	1	0.8	1	0.03333	0.08165	8.45%	0.0%
44		6	0.9	0.785	1	0.8	1	0.04472	0.1095	12.17%	6.9%
96h Survival F	Rate Detail										
C-ppt	Control Type	Rep ′	i Rep 2	2 Rep 3	Rep 4	Rep 5	Rep 6				
0	Brine Control	1	1	1	0.8	1	1				
0	Lab Control	1	1	1	1	1	1				
38		1	1	1	1	1	1				
		1	1	1	0.8	0.8	1				
40											
40 42		1	0.8	1	1	1	1				

000-089-180-4

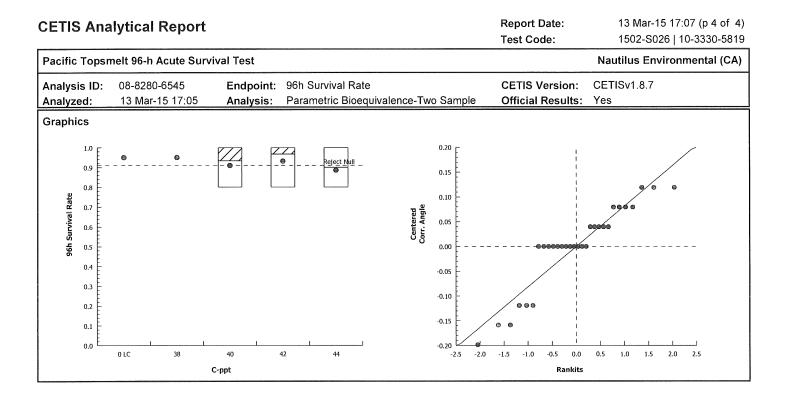
Analyst: <u>AC</u> QA: <u>31/1/</u>16

CETIS Ana	alytical Repo	ort					•	ort Date: Code:		/lar-15 17:0 2-S026 10	06 (p 1 of 0-3330-58´
Pacific Tops	melt 96-h Acute	Survival T	est						Nautilus	Environn	nental (CA
Analysis ID: Analyzed:	20-0664-6286 13 Mar-15 17:0		dpoint : 96h alysis: Nor		ate Control vs T	reatments		S Version: ial Results		8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	тυ
Angular (Corr	ected)	NA	C > T	NA	NA		11.5%	44	>44	NA	
Steel Many-C	Dne Rank Sum To	est									
Control	vs C-ppt		Test Stat	Critical	Ties DF	P-Value	P-Type	Decision	(a:5%)		
Lab Control	38		39	25		0.8000	Asymp		ificant Effect		
	40		33	25		0.3906	Asymp	-	ificant Effect		
	42		36	25		0.6101	Asymp	0	ificant Effect		
	44		30	25		0.2033	Asymp	-	ificant Effect		
ANOVA Table	A										
Source	Sum Squ	ares	Mean Squ	are	DF	F Stat	P-Value	Decision	(a:5%)		
Between	0.0642689		0.0160672	~~~~~	4	1.932	0.1363		ificant Effect		
Error	0.207929		0.0083171		4 25	1.352	0.1000	Ron-oign			
Total	0.207929	9	0.0003171		29	-					
		-									
Distributiona Attribute	al Tests Test			Test Stat	Critical	P-Value	Decision	a:1%)			
		ono Equalit	v of Vorianco		4.177	0.0275	Equal Var				
Variances			y of Variance	3.269 16.34	4.177		•				
Variances Distribution		Equality of Wilk W Nor		0.8773	4.177 0.9031	<0.0001 0.0025	Unequal \	al Distributi	on		
			manty	0.0775	0.3031	0.0025					Rector (1994)
	Rate Summary	• •	8.8	0.5%/ 1.01	05% 1101	8.0 I'			0445	0.4%	0/ E.c.
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1	1	1	1	1	1	0	0.0%	0.0%
38		6	1	1	1	1	1	1	0	0.0%	0.0%
40		6	0.9333	0.8249	1	1	0.8	1	0.04216	11.07%	6.67%
42		6	0.9667	0.881	1	1	0.8	1	0.03333	8.45%	3.33%
44		6	0.9	0.785	1	0.9	0.8	1	0.04472	12.17%	10.0%
Angular (Cor	rected) Transfor	med Sum	nary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Мах	Std Err	CV%	%Effect
0	Lab Control	6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%
38		6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%
40		6	1.266	1.137	1.395	1.345	1.107	1.345	0.0502	9.71%	5.9%
42		6	1.306	1.204	1.408	1.345	1.107	1.345	0.03969	7.45%	2.95%
44		6	1.226	1.089	1.363	1.226	1.107	1.345	0.05325	10.64%	8.85%
96h Survival	Rate Detail										
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6			-	
0	Lab Control	1	1	1	1	1	1				
38		1	1	1	1	1	1				
40		1	1	1	0.8	0.8	1				
42		1	0.8	1	1	1	1				
44		1	0.8	0.8	1	0.8	1				
	rected) Transfor	med Detai									
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	Lab Control	1.345	1.345	1.345	1.345	1.345	1.345				
		1.345	1.345								
		1.040	1.349	1.345	1.345	1.345	1.345				
38				1 245	1 107	1 107	1 9 4 5				
38 40		1.345	1.345	1.345	1.107	1.107	1.345				
38				1.345 1.345 1.107	1.107 1.345 1.345	1.107 1.345 1.107	1.345 1.345 1.345				



lyst: AC QA: 8-3/16/15

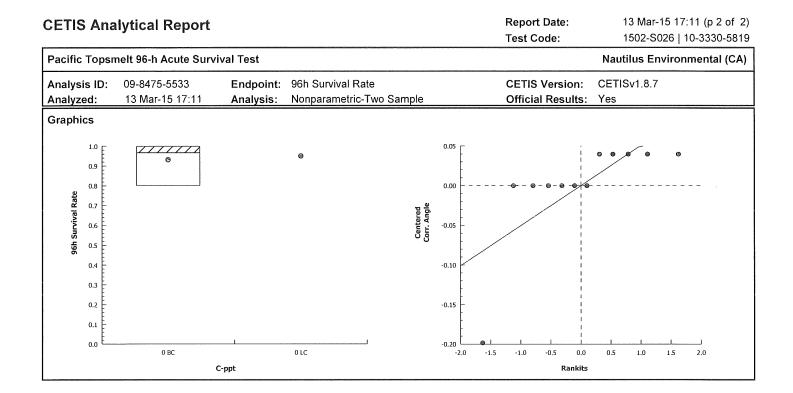
CETIS Ana	alytical Repo	ort	2000 (111)				•	ort Date: Code:		/lar-15 17:0 2-S026 10	
Pacific Tops	melt 96-h Acute \$	Survival T	est		2010-001-000-0				Nautilus	s Environm	nental (CA)
Analysis ID: Analyzed:	08-8280-6545 13 Mar-15 17:0		idpoint: 96h alysis: Para			Two Sample		S Version:	CETISv1 Yes	.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	тυ
Angular (Corr	rected)	NA	C*b < T	NA	NA	0.8	8.97%	44	>44	NA	
TST-Welch's	t Test										
Control	vs C-ppt		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(α:10%)		
Lab Control	38*		0.2691	NA		<0.1		Non-Signi	ficant Effect		
	40*		3.778	1.476	0.074 5	0.0065	CDF	Non-Signi	ficant Effect		
	42*		5.779	1.476	0.059 5	0.0011	CDF		ficant Effect		
	44*		2.817	1.476	0.079 5	0.0186	CDF	Non-Signi	ficant Effect		
ANOVA Table	e				*****						
Source	Sum Squ	ares	Mean Squ	are	DF	F Stat	P-Value	Decision(α:5%)		
Between	0.0642689	95	0.0160672		4	1.932	0.1363	Non-Signi	ficant Effect		
Error	0.207929		0.0083171	59	25	_					
Total	0.2721979	9			29						
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Mod Leve	ene Equali	ty of Variance	3.269	4.177	0.0275	Equal Var				
Variances	Levene E	Equality of	Variance	16.34	4.177	<0.0001	Unequal \				
Distribution	Shapiro-\	Wilk W No	rmality	0.8773	0.9031	0.0025	Non-norm	al Distributio	on		
96h Survival	Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1	1	1	1	1	1	0	0.0%	0.0%
38		6	1	1	1	1	1	1	0	0.0%	0.0%
40		6	0.9333	0.8249	1	1	0.8	1	0.04216	11.07%	6.67%
42		6	0.9667	0.881	1	1	0.8	1	0.03333	8.45%	3.33%
44		6	0.9	0.785	1	0.9	0.8	1	0.04472	12.17%	10.0%
Angular (Cor	rrected) Transfor	med Sum	mary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%
38		6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%
40		6	1.266	1.137	1.395	1.345	1.107	1.345	0.0502	9.71%	5.9%
42		6	1.306	1.204	1.408	1.345	1.107	1.345	0.03969	7.45%	2.95%
44		6	1.226	1.089	1.363	1.226	1.107	1.345	0.05325	10.64%	8.85%
96h Survival	Rate Detail										
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	Lab Control	1	1	1	1	1	1				
38		1	1	1	1	1	1				
40		1	1	1	0.8	0.8	1				
42		1	0.8	1	1	1	1				
44		1	0.8	0.8	1	0.8	1				
Angular (Cor	rrected) Transfor	med Deta	il								
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	Lab Control	1.345	1.345	1.345	1.345	1.345	1.345				
		1.345	1.345	1.345	1.345	1.345	1.345				
38		1.040									
38 40		1.345	1.345	1.345	1.107	1.107	1.345				
					1.107 1.345	1.107 1.345	1.345 1.345				



Analyst: AC QA: & 3/16/15

CETIS An	alytical Repo	ort						ort Date: Code:			11 (p 1 of 2) D-3330-5819
Pacific Tops	melt 96-h Acute	Survival T	est						Nautilus	Environr	nental (CA)
Analysis ID: Analyzed:	09-8475-5533 13 Mar-15 17:1			Survival Ra		Э		IS Version: ial Results	CETISv1 : Yes	.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	Test Res	ult		
Angular (Cor	rected)	NA	C > T	NA	NA		8.59%	Passes 96	Sh survival r	ate	
Wilcoxon Ra	ank Sum Two-Sa	mple Test									
Control	vs Control		Test Stat	Critical	Ties DF	P-Value	P-Type	Decision	α:5%)		
Lab Control	Brine Co	ntrol	36	NA	1 10	0.5000	Exact	Non-Signi	ficant Effect		
ANOVA Tabl	le										
Source	Sum Squ	ares	Mean Squ	are	DF	F Stat	P-Value	Decision(α:5%)		
Between	0.004725	658	0.0047256	58	1	1	0.3409	Non-Signi	ficant Effect		
Error	0.047256	58	0.0047256	58	10						
Total	0.051982	24			11						
Distribution	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Mod Lev	ene Equalit	y of Variance	1	10.04	0.3409	Equal Var	iances			
Variances	Levene E	Equality of V	/ariance	6.25	10.04	0.0314	Equal Var	iances			
Distribution	Shapiro-	Wilk W Nor	mality	0.5612	0.8025	<0.0001	Non-norm	al Distributi	on		
96h Surviva	I Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1	1	1	1	1	1	0	0.0%	0.0%
0	Brine Control	6	0.9667	0.881	1	1	0.8	1	0.03333	8.45%	3.33%
Angular (Co	rrected) Transfor	med Sumi	mary								
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%
0	Brine Control	6	1.306	1.204	1.408	1.345	1.107	1.345	0.03969	7.45%	2.95%
96h Surviva	I Rate Detail										
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	Brine Control	1	1	1	0.8	1	1				
0	Lab Control	1	1	1	1	1	1				
Angular (Co	rrected) Transfor	med Detai	il								
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	Brine Control	1.345	1.345	1.345	1.107	1.345	1.345				
0	Lab Control	1.345	1.345	1.345	1.345	1.345	1.345				

Analyst:<u>AC</u> QA:<u>& 3/16/1</u>5



Analyst: AC QA: 8 3/11/15

Water Quality Measurements & Test Organism Survival

NA

14 H

Dilutions made by:

Client: Poseidon	Test Species: A. affinis			Teo	:h Init	als	
Sample ID: MOZEN			0	24	48	72	96
Test No.: 1506	End Date/Time: 2/14/2015 1150 Cd	ounts: A	P	μH	VA	NH	Ar
	Rea	lings: N	H	NH	NB	NH	AP

Concentration	Rep			ber o ganis				5	Salinit (ppt)	у			Ten	nperat (°C)	ure				lved C (mg/L		1			pH (units))	2000 ék ék 20
AC		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
Lab Control	Α	5	15	5	5	5	33.0	33 1	h,p.0	33.4	33.9	20,0	20,8	19.0	70.L	30.6	7.4	66	81	63	6.4	716	7.80		7.86	7.84
	в	5	5	5	5	ŝ			33.4					1 20,7					f(p. 3					17.89		
	С	5	3	5	5	5																				
	D	5	2	5	5	ŝ																				
	Е	5	5	S	5	5																				
	F	5	S	5	5	5																				
Brine Control	Α	5	5	5	S	5	34.0	H2	54.1	34.3	34,8	Ð.Ö	20,8	191.1	20:5	20.7	14	6.3		6.3	6.4	7.76	1:51			7.88
	в	5	5	5	5	5			53.1					f ZB/5					fφ.z,	-				4-38		
	С	5	5	5	5	5																				
	D	5	5	5	5	4																				
	Е	5	5	5	S	5																				
	F	5	5	5	3	5																				
38.0 ppt	Α	5	5	5	5	5	3G,O	78.	380	36,2	38.6	70.1	7.05		20,4	20.8	7.4	6.1	80	6.3	6.3	7.14	7.82		JAU	790
	В	5	5	5	5	5			38,4					¹ 20.7					18.2					7.91		ļ
	С	5	S	5	3	5																				
	D	5	5	5	5	5																				
	Е	5	5	5	5	5											-							_		
	F	5	5	5	201	5	d. c		40.0	lita				1			2 >	ŀ	1.0			17.00	1.		-+	
40.0 ppt	Α	5	5	5	5	5	40.0	46,2	34.4		48.6	19.9			29:4	20.9	7.3	61	8.0	6.3	6.3	4.73	7:83		4.97	7.90
	В	5	5	5	5	5			40.4	121.X				f20.V					fu / 1					4-91		
	C	5	5	5	5	5				2/12/	5															
	D	5	5	7	2	Ч Ц																				
	E	5	r r	15	1	3-15																				
ang kapa pangang ang ang ang ang ang ang ang ang a		5	2	5		5	100	1125	1	812 -		100	201	LA a	20.6		7)	1.5	6 0	1 5	c 0	791	747	Li x.	791	7.0
42.0 ppt	A	5	5	> u	5	2 4	10			46.5	43.6	117	0.5	19-2	20.6	20.4	1.5	00	15.9	6.2	3.6	7.91	14C	<u>+.91</u> 4.90	<u>, , , ,</u>	1.10
	B C	5	5	5	5				42.4					20.7					17.1					7-70		
	D	5	>	25	2	5																				
i	E	5	5	5	2 5	» 5																		,		
	F	5	5	2	2	2																				
	Г	5	-2	\geq	5	2																	1			

Initial Counts QC'd by: _Ŋ\\

Animal Source/Date Received: 2/0/5/ ABS

Age at Initiation: 15d

Comments:

<u>i</u> = initial reading in fresh test solution, f = final reading in test chamber prior to renewal Organisms fed prior to initiation, circle one ((y)/n) Huch Soussian 5 solitory Weefer
 Feeding Times

 0
 24
 48
 72
 96

 AM:
 0820
 0805
 9115
 0900

 PM:
 1005

QC Check:

KBZILGIIS

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

Final Review: <u>& 3/16/15</u>

Water Quality Measurements & Test Organism Survival

Client:	Poseidon	Test Species: A. affinis				Te	ch Initi	als	
Sample ID:	fozen seawater brine	Start Date/Time: 2/10/2015	1340		0	24	48	72	96
Test No.:	1502-5026	End Date/Time: 2/14/2015	1150	Counts:	AÐ	NIŁ	NH	MA	AB
				Readings:	414	NH	AUD	NH	AB

Dilutions made by: NH

Concentration P <u>Pt-%</u> *1らてio	Rep			ber o ganis	f Live sms	9		:	Salinit (ppt)				Ter	npera (°C)	ture				lved C (mg/L		n			pH (units)	
×18240		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
44.0 ppt	Α	5	5	5	5	5	ЩC	44.2			45.4	149	70,5	19.2	20,4	20.2	7.3	61		62	6.1	7.91	784	4.96	792	793
Ø	в	5	5	5	5	4			49.2					20.5					5.9					7.90		
	C	5	4	4	Ч	4																				
	D	5	5	3	5	5																				
	E	5	5	5	5	4						<u> </u>	ļ													
	F	5	5	5	5	5			ļ																	
	Α	5							6					6					6					5		
	В	5							ľ					ľ					ľ							
	С	5		ļ			ļ							ļ		ļ										
	D	5										ļ	<u> </u>				 		 		ļ					
	E	5																						ļ		
	F	5		-										i					li					1		
	A	5							f					f					f.					f		
	B	5																								
	C	5																								
	D	5																								
	E	5																								
		5							1					i					1					1		
	A B	5							f					f					f					f		
	в С	5																								
	D	5																								
	E	5																								
	F	5 5																								
	A								i					i					i					1		
	В	5 5							f					f					f					f		
	c	5																								<u></u>
	D	5																								
	E	5																								
	F	5																								
Initial Counts QC'd by:		er.				, ,	L					L		ñ		L			L	1					I	
Animal Source/E	Date F	Recei	ved:	2/6	115	AB	5			Age a	at Initia	ation:	15	L			-						Feed	ding Ti	mes	
					1																	0	24	48	72	96
Comments:	-	i = in	itial re	eading	j in fre	esh te	st solu	tion, f	= fina	l readi	ng in t	est ch	amber	prior	to ren	ewal	7 a	A	10. 1		AM:		180	()NG	0915	udbi
	-	Orga	nisms	s fed p	prior to	o initia	ation, c		one (Ly	/]/ n)	Macl	~ 7	ens	100	5 2	alm	sy	met	ev^	PM:	1505	-		-	
		n	- 10				Ø.	Julo MIN	samy.	the 6	ritutt desta	سل ۵۷ سلہ ۵	Ting	615	a du	.k.	100 00.		2 fau	nall o	lugin		1	s. 1		
QC Check:	V	02	<u>110</u>	45			(NĮU	1 0.4	1.1	* U TV	* 90	VOH	urv	7a'M	$\sqrt{\gamma}$	0.01.60%	juren	F 40"	Fi	nál Re	view:	0	2/16	115	
Nautilus Environme	ental. 🗸	4340 V	andev	er Ave	enue.	San Di	iego, C	4 9212	0.																	

Brine Dilution Worksheet

Poseidon		Analyst: _	NH	
ozen seawater – in	itiations	Test Date:	2/10/2015	
502-5026		Test Type:	Aa-a Ctop	shelt)
ater	33.0	-		
_	86.1	Date of Brine used:	12/2/14	
ume	1500	Alkalinity of Brine Control:	107	mg/L as CaCO3
	rozen seawater – in 502 - 5026 ater	ater <u>33.0</u> 86.1	Solution Test Type: ater 33.0 86.1 Date of Brine used:	ちのスーちのうし Test Type: Aa-a C+op ater 33.0

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

Marine Chronic Bioassay

Target Salinity ppt	Concentration % seawater	Seawater Volume (mi)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
34.0	100.0	250	NA	NA	1500
38.0	90.6	1358.8	0.10	141.2	1500
40.0	86.8	1302.3	0.15	197.7	1500
42.0	83.1	1245.8	0.20	254.2	1500
44.0	79.3	1189.3	0.26	310.7	1500

	DI Volume				
Brine Control	-297.3	1.05	310.7	1500	AC Q18,02
Q20					. (5
	Fotal Brine Volum	e Required (ml):	1214.7	7	
		, ,			

QC Check: <u>vBalialis</u> Q20-Brine control calculation incorrect. See QA section of report.

Marine Chronic Bioassay

Brine Dilution Worksheet

Project:	Poseidon		Analyst: NH
Sample ID:	frozen seawater - r	eneusal	Test Date: 2/10/2015
Test No:	1502-502	6	Test Type: <u>Aa-a</u> (Toponelt)
Salinity of Sea	water	33.0	
Salinity of Brir	10	85.2	Date of Brine used: 12(2/12)
Test Dilution V	/olume	1500	Alkalinity of Brine Control: mg/L as CaCO3

TS = target salinity SE = salinity of effluent

SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
34.0	100.0	250	NA	NA	1500
38.0	90.4	1356.3	0.11	143.7	1500
40.0	86.6	1298.9	0.15	201.1	1500
42.0	82.8	1241.4	0.21	258.6	1500
44.0	78.9	1183.9	0.27	316.1	1500

	DI Volume				
Brine Control	296.0	1.07	316.1	1500	AC Q18,921
					- 1 ⁻ 1 ⁻

Total Brine Volume Required (ml): 1235.6

QC Check: 632/19/15

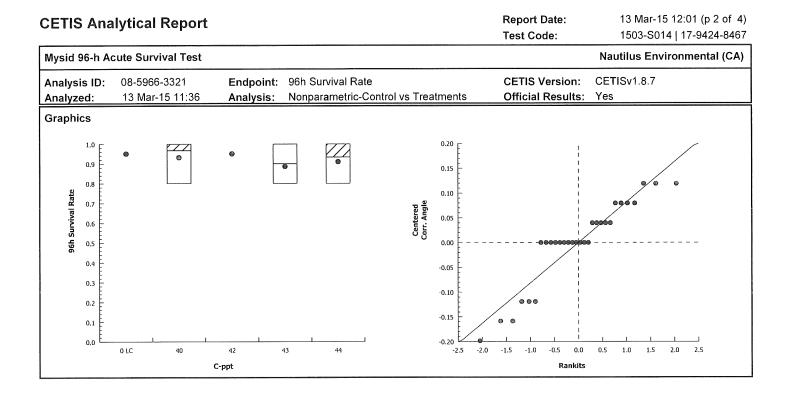
Final Review: AC 3/19/16

Q21: Brine control formula was incorrect and satindy was high (244ppt). Dre The technician reduced satinity with seawater instead of DI. Merebore, the brine control did not accurately control for the maximum addition of brine. See QA section of report. Mysid Acute Salinity Tolerance Test March 2015

CETIS Sun	nmary Repo	ort							•	rt Date: Code:	•			2 (p 1 of 1) -9424-8467
Mysid 96-h Ad	cute Survival Te	st										Nautilus	s Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	18-6763-0526 03 Mar-15 15:4 07 Mar-15 14:3 95h	-	Test Type: Protocol: Species: Source:	EPA Ame	ival (96h) /821/R-02-(ricamysis b atic Biosyst	bahia			Analy Dilue Brine Age:	nt: e:		iral Seawate en Seawate		
Sample ID: Sample Date: Receive Date: Sample Age:	: 03 Mar-15		Code: Material: Source: Station:	Natu Pose	8-S014 Iral Seawate eidon tilus Brine	er			Clien Proje		Pose	eidon 		
Comparison	Summary										stantoffilituared			
Analysis ID	Endpoint		NOEL	-	LOEL	TOEL	PMSD	τu		Metho	bd			
08-5966-3321 03-1884-4572	96h Survival Ra 96h Survival Ra		44 44		>44 >44	NA NA	11.5% 8.71%					y-One Rank h's t Test	Sum Test	
96h Survival	Rate Summary													
C-ppt	Control Type	Cou	nt Mean	ı	95% LCL	95% UCL	Min	Max	ĸ	Std E	rr	Std Dev	CV%	%Effect
0	Brine Control	6	0.966	7	0.881	1	0.8	1		0.0333	33	0.08165	8.45%	0.0%
0	Lab Control	6	1		1	1	1	1		0		0	0.0%	-3.45%
40		6	0.966	7	0.881	1	0.8	1		0.0333	33	0.08165	8.45%	0.0%
42		6	1		1	1	1	1		0		0	0.0%	-3.45%
43		6	0.9		0.785	1	0.8	1		0.0447		0.1095	12.17%	6.9%
44		6	0.933	3	0.8249	1	0.8	1		0.042	16	0.1033	11.07%	3.45%
96h Survival	Rate Detail					······································								
C-ppt	Control Type	Rep	1 Rep 2	2	Rep 3	Rep 4	Rep 5	Rep	06					
0	Brine Control	1	1		1	0.8	1	1						
0	Lab Control	1	1		1	1	1	1						
40		1	1		0.8	1	1	1 ·						
42		1	1		1	1	1	1						
43		0.8	0.8		1	1	1	0.8						
44		1	1		1	0.8	1	0.8						

Analyst: AC QA: 13/16/15

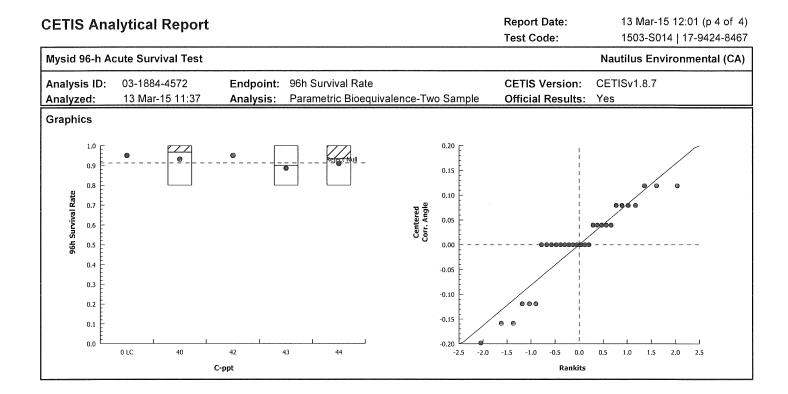
CETIS Analytical Report									Report Date: Test Code:		13 Mar-15 12:01 (p 1 of 4) 1503-S014 17-9424-8467		
Mysid 96-h A	cute Survival Te	st								Nautilus	s Environn	nental (CA)	
,			•	•					IS Version: ial Results		.8.7		
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed			PMSD	NOEL	LOEL	TOEL	TU	
Angular (Corre	ected)	NA	C > T	NA	NA			11.5%	44	>44	NA		
Steel Many-C) ne Rank Sum Te	est											
Control	vs C-ppt		Test Stat	Critical	Ties	DF	P-Value	P-Type	Decision	(α:5%)			
Lab Control	40		36	25	1		0.6101	Asymp		ificant Effect			
	42		39	25	1		0.8000	Asymp	-	ificant Effect			
	43		30	25	1	10	0.2033	Asymp	-	ificant Effect			
	44		33	25	25 1 10		0.3906	Asymp	Non-Significant Effect				
ANOVA Table	9												
Source	Sum Squ	Mean Sq	Mean Square		DF		P-Value	Decision(α:5%)					
Between	0.06426895		0.016067	0.01606724		4		0.1363	Non-Significant Effect				
Error	0.207929		0.008317	0.008317159			25						
Total	0.2721979	9			29								
Distributiona	I Tests												
Attribute	Test			Test Stat	Test Stat Critical		P-Value	Decision	(α:1%)				
Variances	Mod Levene Equality		lity of Varianc	e 3.269	3.269 4.177		0.0275	Equal Va					
Variances	Levene Equalit y of Va			16.34	4.177		<0.0001		equal Variances				
Distribution	Shapiro-Wilk W Norm		ormality	0.8773	3 0.9031		0.0025	Non-norm	al Distributi	on			
96h Survival	Rate Summary												
C-ppt	Control Type	Count	Mean	95% LCL	95% U	CL	Median	Min	Мах	Std Err	CV%	%Effect	
0	Lab Control	6	1	1	1		1	1	1	0	0.0%	0.0%	
40		6	0.9667	0.881	1		1	0.8	1	0.03333	8.45%	3.33%	
42		6	1	1	1		1	1	1	0	0.0%	0.0%	
43		6	0.9	0.785	1		0.9	0.8	1	0.04472	12.17%	10.0%	
44		6	0.9333	0.8249	1		1	0.8	1	0.04216	11.07%	6.67%	
Angular (Cor	rected) Transfor	med Sun	nmary										
C-ppt	Control Type	Count	Mean	95% LCL	95% U	CL	Median	Min	Max	Std Err	CV%	%Effect	
0	Lab Control	6	1.345	1.345	1.345		1.345	1.345	1.345	0	0.0%	0.0%	
40		6	1.306	1.204	1.408		1.345	1.107	1.345	0.03969	7.45%	2.95%	
42		6	1.345	1.345	1.345		1.345	1.345	1.345	0	0.0%	0.0%	
43		6	1.226	1.089	1.363		1.226	1.107	1.345	0.05325	10.64%	8.85%	
44		6	1.266	1.137	1.395		1.345	1.107	1.345	0.0502	9.71%	5.9%	
96h Survival													
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4		Rep 5	Rep 6					
0	Lab Control	1	1	1	1		1	1					
40		1	1	0.8	1		1	1					
42		1	1	1	1		1	1					
43		0.8	0.8	1	1		1	0.8					
44		1	1	1	0.8		1	0.8					
Angular (Cor	rected) Transfor	rmed Det	ail										
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4		Rep 5	Rep 6					
0	Lab Control	1.345	1.345	1.345	1.345		1.345	1.345					
40		1.345	1.345	1.107	1.345		1.345	1.345					
42		1.345	1.345	1.345	1.345		1.345	1.345					
43		1.107	1.107	1.345	1.345		1.345	1.107					
		1.345	1.345	1.345	1.107		1.345	1.107					



Analyst: _____ QA: <u>19-3/11/15</u>

CETIS Ana	alytical Repo	ort					•	ort Date: Code:	13 Mar-15 12:01 (p 3 of 4) 1503-S014 17-9424-8467				
Mysid 96-h A	cute Survival Te	st							Nautilus	Environm	nental (CA)		
Analysis ID: Analyzed:	03-1884-4572 13 Mar-15 11:3		i dpoint : 96h i alysis : Para			-Two Sample		S Version: ial Results:	CE T ISv1. Yes	8.7			
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	τU		
Angular (Corre	ected)	NA	C*b < T	NA	NA	0.8	8.71%	44	>44	NA			
TST-Welch's	t Test												
Control	vs C-ppt		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(a:10%)				
Lab Control	40*		5.779	1.476	0.059 5	0.0011	CDF	Non-Signif	icant Effect				
	42*		0.2691	1.476	5	<0.1	CDF	-	icant Effect				
	43*		2.817	1.476	0.079 5	0.0186	CDF	Non-Signif	icant Effect				
	44*		3.778	1.476	0.074 5	0.0065	CDF	Non-Signif	icant Effect				
ANOVA Table	9												
Source	Sum Squ	ares	Mean Squ	are	DF	F Stat	P-Value	Decision(a:5%)		wa 4100 4		
Between	0.064268	95	0.0160672	4	4	1.932	0.1363	Non-Signif	icant Effect				
Error	0.207929		0.0083171	59	25	_							
Total	0.2721979	9			29								
Distributiona	l Tests												
Attribute	Test			Test Stat	Critical	P-Value	Decision	α:1%)					
Variances	Mod Leve	ene Equali	ty of Variance	3.269	4.177	0.0275	Equal Var						
Variances		Equality of V		16.34	4.177	<0.0001	Unequal \						
Distribution	Shapiro-	Wilk W No	rmality	0.8773	0.9031	0.0025	Non-norm	al Distributio	n				
96h Survival	Rate Summary												
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect		
0	Lab Control	6	1	1	1	1	1	1	0	0.0%	0.0%		
40		6	0.9667	0.881	1	1	0.8	1	0.03333	8.45%	3.33%		
42		6	1	1	1	1	1	1	0	0.0%	0.0%		
43		6	0.9	0.785	1	0.9	0.8	1	0.04472	12.17%	10.0%		
44		6	0.9333	0.8249	1	1	0.8	1	0.04216	11.07%	6.67%		
Angular (Cor	rected) Transfor	med Sum	mary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect		
0	Lab Control	6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%		
40		6	1.306	1.204	1.408	1.345	1.107	1.345	0.03969	7.45%	2.95%		
42		6	1.345	1.345	1.345	1.345	1.345	1.345	0	0.0%	0.0%		
43		6	1.226	1.089	1.363	1.226	1.107	1.345	0.05325	10.64%	8.85%		
44		6	1.266	1.137	1.395	1.345	1.107	1.345	0.0502	9.71%	5.9%		
96h Survival	Rate Detail			_			_	_	_				
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6						
0	Lab Control	1	1	1	1	1	1						
40		1	1	0.8	1	1	1						
42		1	1	1	1	1	1						
43		0.8	0.8	1	1	1	0.8						
44		1	1	1	0.8	1	0.8						
Angular (Cor	rected) Transfor	med Deta	il										
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6						
0	Lab Control	1.345	1.345	1.345	1.345	1.345	1.345						
40		1.345	1.345	1.107	1.345	1.345	1.345						
42		1.345	1.345	1.345	1.345	1.345	1.345						
74													
43		1.107	1.107	1.345	1.345	1.345	1.107						

Analyst: <u>A</u>C QA: <u>& 3/11/15</u>



lyst: <u>AC</u> QA: <u>97 3/16/15</u>

TST Summary Sheet

Lab Name	Naut	Client Name	Pos
Test ID	1503-S015	Test Species	A. bahia (mysid shrimp)
Test Date	3/3/2015	Test Type	Acute
Test Duration	96h	Endpoint	Survival
Critical Conc.	43		

Statistic	Control	Critical Concentration	
Percent Mean of Raw Data	1.00	0.90	
Mean used in Calcuation (transformed)	1.35	1.23	
Variance used in Calcuation (transformed)	0.000	0.017	
Standard Deviation of Transformed Data	0.000	0.130	
CV of Transformed Data	0.000	0.106	
n	6	6	

Mean % Effect at Critical Conc.

10.00

Calculated t-value	Degrees of Freedom	Table t-value	Percent Difference
2.8168	5	1.4759	
Results			

Sample is Non-toxic

Results

Pass

Raw Data

Contro	ol Data	Critical Concentration Data
No. of Organisms	Response (Final	No. of Organisms Response (Final
Exposed or	Count, Weight,	Exposed or Count, Weight,
Counted	Length, etc.)	Counted Length, etc.)
5	5	5 4
5	5	5 4
5	5	5 5
5	5	5 5
5	5	5 5
5	5	5 4

Marine Acute Bioassay Static-Renewal Conditions

Water Quality Measurements & Test Organism Survival

Client:	Pos	eido	n						_	Te	est Sp	ecies:	A. ba	ahia					_				Те	ch Init	ials	
Sample ID:	brin	e - N	lautilu	us fro	ozen	seav	/ater		-	Star	t Date/	Time:	3/3/2	2015		545			-			0	24	48	72	96
Test No.:	1Fe	208	5-8	30	14				_	Enc	Date/	Time:	3/7/2	2015	<u> </u>	130	2		-		ounts:			EG		AD
																					dings:	1	145	AG	*	AB
																			Dilutio	ons ma	de by:	40		EG CH	W 31	5115 91
Concentration	Rep		Num Org	ber o ganis)		5	Salinit (ppt)			Temperature (°C)				Dissolved Oxygen (mg/L)				pH (units)						
PP*		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
Lab	Α	5	5	5	5	5	333	33.6			353	<u>26.</u> c	25.0			24.4	69	6.3	6.6	5.7	6.6	1.90	7.98		739	7.96
Control	B	5	5	5	5	50			¹ 342					237	Q1				f <u>5,8</u>					7.93		
	C D	5	5	55	5	25	-																			
	E	5 5	5	-) 6	5	É																<u> </u>				
	F	5	5	5	6	5																				
Brine	A	5	5	5	5	5	33:2	35.7	34.1	346	35.4	26.0	25,5	24.7	24.8	247	7.1	6.4	6.7	5.7	6,1	7.91	7,99	18.03	7.92	8.03
Control	в	5	5	5	5	5			¹ 34.0					¹ 24.0					f5.6					f 7.94		
	С	5	5	5	6	5																				
	D	5	5	5	4	4																				
	E	5	5	5	5	5																				
liant	F A	5	5	3	2	5	420	40.0	lines	i.a (1	146	457	bec .	200	247	1-	11.2	1/ 0		63	194	799	401	7,94	R 63
40 ppt	B	5 5	2	5	5	5			f 40.3	40.6	113	11.0	27,1	1342	17.1	124.1	0,1	6.2	6.5	6.1	6-	1.50	1.10	7.97	100000000000000000000000000000000000000	0.05
	С	5	5	4	4	4													0.5							
	D	5	5	5	5	5																				
	Е	5	5	6	5	5																				
	F	5	5	5	5	5																				
42ppt	Α	5	5	5	5	5	420	42.1	¹ Ψ,0	42.6	42,2	25.8	25.8		24.8	25.0	φ .	6.4			5.8	7.89	7,98		7.94	7.99
	В	5	5	5	5	2			f42.4					f74.2					5.7			<u> </u>		7.95		
	C D	5	5	55	5	5											-	-								
	E	5 5	5 6	$\frac{1}{6}$	5	2														-						-
·····	F	5	5	5	6	5																				
43ppt	Α	5	5	Б	5	4	430	43.0	430	94.2	912	259	25.9	25.5	25.0	25.4	20	6.4	6.6	5.2	5.6	190	7.97	7.93	7.94	7.99
÷ 0	в	5	5	Ý	4	4			43.6					f245					^f 5.5					¹ 7.98	2.00000000	
	С	5	5	5	5	5																				
	D	5	5	5	5	5							N								0.000					
	E F	5	5	5	5	5																				
Initial Counts		5	.))	/	4																				
QC'd by:	VV.	R																								
Animal Source/D)ate F	lecei	ved:	Pro:	5	3/3	115			Age	at Initi	ation:		Чd			-					0	Fee 24	ding T 48	imes 72	96
Comments:		i = in	itial re	ading	ı in fr	ash te	st solu	ition f	= fina	Iread	ina in t	est ch	amhei	nrior	to ren	ewal					۵M·	STATISTICS.				
	-	Orga	nisms	fed r	prior to	o initia	ation. d	circle o	ne (h) / r	n)	14	2/1	5	en	511	n5			-	PM:	1415	1619	5190	6830 1550 1550	
	-	@ \$	ubsa	mple	- 1.	luted	1 2	DI. 1	inte	-14 -14	,], , ,	5	ili	<u>n</u>	th.	me	te	٢	1	- alder		L	11.5 10		ل ـــــــ ـــــــــــــــــــــــــــــ] ,
QC Check:	$\leq \epsilon$	<u>5 C</u>	5/10	115							ט שיז	o tai	n s	ali.	14	y m	EUSULIA	emen	77 ¥5	יאוטיי Fi	nal Re	view:	ĸ	3/11	:115	
Nautilus Environme																									$\langle \rangle$	

Marine Acu Static-Rene																				Wa						ents vival
Client:	Pos	eido	n							т	est Sr	ecies	A.b	ahia									Te	ch Init	tials	
Sample ID:				us fre	ozen	seav	vater					/Time:			15	45			-			0	24	48	72	96
Test No.:									_			/Time:				130	>		-	c	ounts	: EG	CH	EG	KB	ALO
																			-	Rea	adings	:40	EH			AB
																			Dilutic		ade by			СН		
Concentration	Rep			ber c ganis	of Live sms	9			Salini (ppt)				Те	mpera (°C)				Dissolved Oxygen (mg/L)				pH (units)				
		0	24	48	72	96	Q	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
1400+	Α	5	5	5	5	5	144.	44.0	44,0	944.0	44.0	25.8	259	250	25.0	96	7.1		10.000		5.6	-	11100000000000	1.	7.95	and the state of t
@ " ''	в	5	5	5	5	5			44.7	2				Q4 <u><</u>					¹ 5.8					7.99		
	С	5	5	5	5	5																				
·	D	5	5	5	5	4																				
	Ε	5	5	5	6	5																				
	F	5	4	4	Ú	4								<u> </u>												
	<u>A</u>	5			ļ									1					 	8.000.000000				<u> </u>		
	B	þ			<u> </u>		ļ	-					ļ	ľ					1			ļ		ľ		
	C	5										 		ļ												
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	E F	5								<u> </u>	_					-						 				
	r A	5			<u> </u>	<u> </u>								i					i					1		
	B	5 5							ſ					f					f					f		
	c	ວ 5																								<u> </u>
	D	5																-				<u> </u>				
	E	5																								
	F	5																				<u> </u>				
	A	5							i					i					I					i		
	в	5			· ·				f					f					f					f		
	С	5																								
	D	5																								
	E	5																								
	F	5																								
	Α	5							I					i					i					I		
	в	5							f					f					f					f		
	С	5																								
	D	5																								
	E	5		-																						
Initial Counts	F	5		12	211.0																					
Initial Counts	10	~~ ^c	v∩ `	-12	710	1>																				

QC'd by:

Animal Source/Date Received: _____________

Feeding Times 24 0 48 72 96 (SA AM: 190 16151501 PM: 145 150 Final Review: 8 3/16/15

Comments:

i = initial reading in fresh test solution, f = final reading in test chamber prior to renewal

3/3/15

Organisms fed prior to initiation, circle one (1)/n) @ subsample diluted w(PI water 1:1 to obtain salinity measurement 453/4/15

QC Check: VB 211015 Nautilus Environmental. 4340 Vandever Avenue. San Diego, CA 92120.

Marine Chronic Bioassay

Brine Dilution Worksheet

Project:	Poseidon	*****	Analyst: <u>CH</u>
Sample ID:	frozen seawater		Test Date: 3315
Test No:	1503-5014		Test Date: <u>3/3/15</u> test initiation and Test Type: <u>mysid acute/48hr renewal</u>
Salinity of Se	awater	33.1	() an 3/12/15(2/5
Salinity of Bri	ine _	93.0	Date of Brine used: 2/2015 12/24/14, 12/31/14, 1/2/15, 1/15/15,
Test Dilution	Volume _	1500	Alkalinity of Brine Control:

TS = target salinity

SE = salinity of effluent

SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
33.1	100.0	250	NA	NA	1500
40.0	88.5	1327.2	0.13	172.8	1500
42.0	85.1	1277.1	0.17	222.9	1500
43.0	83.5	1252.1	0.20	247.9	1500
					·
44.0	81.8	1227.0	0.22	273.0	1500

	DI Volume _ୁହ			
Brine Control	-444.6 AC &	0.61 0.55	273.0	1500
	494.0	ACQ18 4729		

Total Brine Volume Required (ml): 5/6/13 545.9 1169.6

QC Check: <u>VB 3/10/15</u> Brine was mixed the day prior to test initiation from brine made on multiple dates in order to have enough volume for test initiation + 48hr renewal

Topsmelt Acute Salinity Tolerance Test March 2015

CETIS Summary Report

Report Date: Test Code:

13 Mar-15 12:03 (p 1 of 1) 1503-S015 | 07-4920-2856

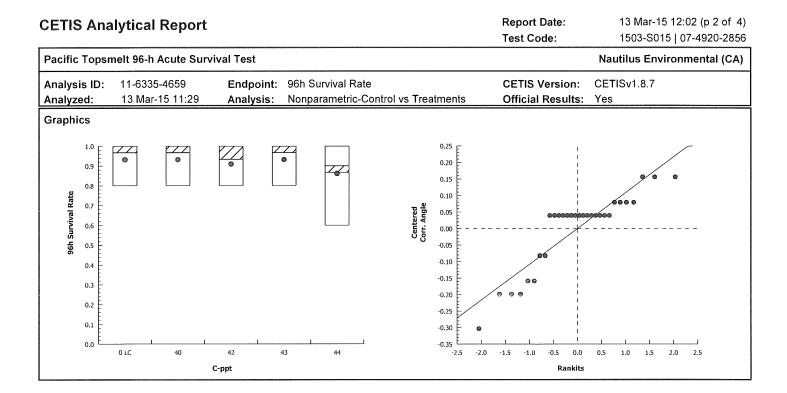
									est coue.				4520-2050
Pacific Topsm	nelt 96-h Acute \$	Surviva	al Test								Nautilus	Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	01-9359-5620 05 Mar-15 15:5 09 Mar-15 14:4 95h		Test Type: Protocol: Species: Source:	EPA Athe	vival (96h) \/821/R-02-(erinops affin atic Biosyst	is		D B			ural Seawate en Seawate		
Sample ID: Sample Date: Receive Date: Sample Age:	05 Mar-15		Code: Material: Source: Station:	Nati Pos	3-S015 ural Seawate eidon itilus Brine	er			lient: roject:	Pose	eidon		
Comparison S	Summary												
Analysis ID	Endpoint		NOE	L	LOEL	TOEL	PMSD	τu	Meth	od			
11-6335-4659	96h Survival Ra 96h Survival Ra		44 44		>44 >44	NA NA	14.6% 11.2%				y-One Rank h's t Test	Sum Test	
96h Survival F	Rate Summary												
C-ppt	Control Type	Cou	nt Mear	n	95% LCL	95% UCL	Min	Max	Std E	Irr	Std Dev	CV%	%Effect
0	Brine Control	6	0.933	33	0.8249	1	0.8	1	0.042	16	0.1033	11.07%	0.0%
0	Lab Control	6	0.966	67	0.881	1	0.8	1	0.033	33	0.08165	8.45%	-3.57%
40		6	0.966	67	0.881	1	0.8	1	0.033	33	0.08165	8.45%	-3.57%
42		6	0.933	33	0.8249	1	0.8	1	0.042	216	0.1033	11.07%	0.0%
43		6	0.966	67	0.881	1	0.8	1	0.033	33	0.08165	8.45%	-3.57%
44		6	0.866	67	0.6953	1	0.6	1	0.066	67	0.1633	18.84%	7.14%
96h Survival I	Rate Detail												
C-ppt	Control Type	Rep	1 Rep	2	Rep 3	Rep 4	Rep 5	Rep 6				20100-0010-00-00-00-00-00-00-00-00-00-00-	
0	Brine Control	1	1		0.8	1	1	0.8					
0	Lab Control	0.8	1		1	1	1	1					
40		1	1		1	0.8	1	1					
42		0.8	1		0.8	1	1	1					
43		1	1		1	0.8	1	1					
44		1	0.8		1	1	0.6	0.8					

Analyst A QA: 43 3/16/16

CETIS Ana	alytical Repo	ort		•	ort Date: Code:	13 Mar-15 12:02 (p 1 of 4) 1503-S015 07-4920-2856						
Pacific Tops	melt 96-h Acute	Survival	Test							Nautilus	5 Environm	ental (CA)
Analysis ID: Analyzed:	11-6335-4659 13 Mar-15 11:2			n Survival Ra nparametric-		s Tr	eatments		S Version: ial Results:	CETISv1 Yes	.8.7	
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed		, , , , , , , , , , , , , , , , ,	PMSD	NOEL	LOEL	TOEL	TU
Angular (Corr	ected)	NA	C > T	NA	NA			14.6%	44	>44	NA	
Steel Many-C)ne Rank Sum Te	est										
Control	vs C-ppt		Test Stat	Critical	Ties	DF	P-Value	P-Type	Decision(x:5%)		
Lab Control	40		39	25	2	10	0.8000	Asymp	Non-Signifi			
	42		36	25			0.6101	Asymp	Non-Signifi			
	43		39	25			0.8000	Asymp	Non-Signifi			
	44		32.5	25	2	10	0.3556	Asymp	Non-Signifi	cant Effect		
ANOVA Table	e											
Source	Sum Squ		Mean Squ		DF		F Stat	P-Value	Decision(
Between	0.0613280	09	0.0153320		4		0.9684	0.4422	Non-Signifi	cant Effect		
Error	0.395814	1	0.0158325	00	25 29							
Total	0.457142	I			29							
Distributiona												
Attribute	Test			Test Stat			P-Value	Decision				
Variances			Variance	3.615	13.28		0.4605	Equal Var				
Distribution	Shapiro-	Nilk W N	ormality	0.8174	0.9031		0.0001	Non-norm	al Distributio	n		
96h Survival	Rate Summary											
C-ppt	Control Type	Count	Mean	95% LCL	95% UC	CL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	0.9667	0.881	1		1	0.8	1	0.03333	8.45%	0.0%
40		6	0.9667	0.881	1		1	0.8	1	0.03333	8.45%	0.0%
42		6	0.9333	0.8249	1 1		1	0.8 0.8	1 1	0.04216 0.03333	11.07% 8.45%	3.45% 0.0%
43 44		6 6	0.9667 0.8667	0.881 0.6953	1		0.9	0.8 0.6	1	0.06667	8.45% 18.84%	0.0 <i>%</i> 10.34%
					1		0.0		•	0.00007	10.0170	
Angular (Cor	rected) Transfor	med Sun	nmary									
C-ppt	Control Type	Count	Mean	95% LCL	95% UC)L	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1.306	1.204	1.408		1.345	1.107	1.345	0.03969	7.45%	0.0%
40		6	1.306	1.204 1.137	1.408		1.345 1.345	1.107 1.107	1.345 1.345	0.03969 0.0502	7.45% 9.71%	0.0% 3.04%
42 43		6 6	1.266 1.306	1.137	1.395 1.408		1.345	1.107	1.345	0.0302	9.71% 7.45%	0.0%
43		6	1.189	0.9911	1.388		1.226	0.8861	1.345	0.07712	15.88%	8.9%
96h Survival	Rate Detail											
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4		Rep 5	Rep 6				
0	Lab Control	0.8	1	1	1		1	1			an a	
40		1	1	1	0.8		1	1				
42		0.8	1	0.8	1		1	1				
43		1	1	1	0.8		1	1				
44		1	0.8	1	1		0.6	0.8				
Angular (Cor	rected) Transfor	med Det	ail									
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4		Rep 5	Rep 6				
0	Lab Control	1.107	1.345	1.345	1.345		1.345	1.345				
40		1.345	1.345	1.345	1.107		1.345	1.345				
42		1.107	1.345	1.107	1.345		1.345	1.345				
43		1.345	1.345	1.345	1.107		1.345	1.345				
			1.107	1.345	1.345			1.107				

000-089-170-2

Analyst: AC____ QA: 53/16/16

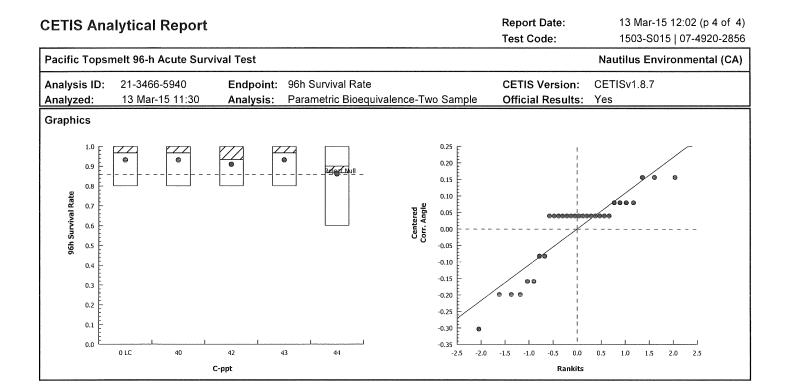


Analyst: AC QA: 5 3/16/16

CETIS An	alytical Rep	ort						ort Date: Code:			02 (p 3 of 4 7-4920-2856
Pacific Tops	melt 96-h Acute	Survival Te	est	T	ST				Nautilu	s Environn	nental (CA)
Analysis ID: Analyzed:	21-3466-5940 13 Mar-15 11:3		•	n Survival Ra rametric Biog		-Two Samp		IS Version: al Results:	CETISv1 Yes	.8.7	
Data Transfo	orm	Zeta	Alt Hyp	Trials	Seed	TST b	PMSD	NOEL	LOEL	TOEL	τu
Angular (Corr	rected)	NA	C*b < T	NA	NA	0.8	11.2%	44	>44	NA	
TST-Welch's	t Test										
Control	vs C-ppt		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(α:10%)		
Lab Control	40*		5.137	1.383	0.070 9	0.0003	CDF	-	ficant Effect		
	42*		3.728	1.397	0.083 8	0.0029	CDF	-	ficant Effect		
	43* 44*		5.137 1.737	1.383 1.44	0.070 9 0.120 6	0.0003 0.0665	CDF CDF	-	ficant Effect ficant Effect		
ANOVA Tabl			1.101		0.120 0	0.0000	001				
Source	Sum Squ	ares	Mean Sq	uare	DF	F Stat	P-Value	Decision(α:5%)		
Between	0.061328		0.015332		4	0.9684	0.4422		ficant Effect	t	*******
Error	0.395814		0.015832	56	25			-			
Total	0.457142	1			29						
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances		Equality of V		3.615	13.28	0.4605	Equal Va				
Distribution	Shapiro-	Wilk W Nor	mality	0.8174	0.9031	0.0001	Non-norm	al Distributic	on		
96h Survival	Rate Summary										
C-ppt	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	0.9667	0.881	1	1	0.8	1	0.03333	8.45%	0.0%
40 42		6 6	0.9667 0.9333	0.881 0.8249	1	1	0.8 0.8	1 1	0.03333 0.04216	8.45% 11.07%	0.0% 3.45%
43		6	0.9667	0.881	1	1	0.8	1	0.03333	8.45%	0.0%
44		6	0.8667	0.6953	1	0.9	0.6	1	0.06667	18.84%	10.34%
Angular (Co	rrected) Transfor	med Sumr	nary								
C-ppt	, Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	6	1.306	1.204	1.408	1.345	1.107	1.345	0.03969	7.45%	0.0%
40		6	1.306	1.204	1.408	1.345	1.107	1.345	0.03969	7.45%	0.0%
42		6	1.266	1.137	1.395	1.345	1.107	1.345	0.0502	9.71%	3.04%
43		6	1.306	1.204	1.408	1.345	1.107	1.345	0.03969	7.45%	0.0%
44		6	1.189	0.9911	1.388	1.226	0.8861	1.345	0.07712	15.88%	8.9%
96h Survival		D 1	Da A	Dallé		Dec 6					
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0 40	Lab Control	0.8 1	1 1	1	1 0.8	1	1 1				
40		0.8	1	0.8	0.8 1	1	1				
43		1	1	1	0.8	1	1				
44		1	0.8	1	1	0.6	0.8				
Angular (Co	rrected) Transfor	med Detai									
C-ppt	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	Lab Control	1.107	1.345	1.345	1.345	1.345	1.345				
40		1.345	1.345	1.345	1.107	1.345	1.345				
42		1.107	1.345	1.107	1.345	1.345	1.345				
43		1.345	1.345	1.345	1.107	1.345	1.345				
44		1.345	1.107	1.345	1.345	0.8861	1.107				

000-089-170-2

Analyst: AC QA: 63/16/15



CETIS™ v1.8.7.20

Analyst: AC QA: 3/16/15

Marine Acute Bioassay Static-Renewal Conditions

Water Quality Measurements & Test Organism Survival

.

Client:	Pose	aidon								Те	st Spe	cies:	A. af	finis									Tec	h Initia	als	
Sample ID:				is fro	zen s	seaw	ater				Date/				19	550	>					0	24	48	72	96
Test No.:											Date/					-140						CH			AB	
																								AD/AB	AB	AG
																		I	Dilutio	ns ma	de by:	4		NH		
													A.M				1	21000000								
Concentration				ber of ganis				S	alinit (ppt)	Y			Ten	nperat (°C)	ure		l	Dissol	ved O (mg/L)		וו		(pH (units)		
ppt	Rep									-	0.0				70	06	0	24	48	72	96	0	24	48	72	96
	A	0	24	48 4	72 4	96 니	0 32.9	24 22 D	48 1820	72	96 33 _, 2	0 199	24	48 10.2	72 10 5	96 19 S	7.6		40		4.8			804		
Lab Control	В	5 5	<u>4</u> 5	3	5	5	JL= 1	<u>,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,</u>	53.0	00.1	<i></i>	11.1		20.7	11.0	115		0,0	1.0 1 6.4	5.1		0.00	.,	1.89		
	c	5	5	Ś.	5_	5			<u></u>					~ <u>,</u>												
	D	5	5	5	5	5																				
	Е	5	5	5	5	5																				
	F	5	5	5	5	5																				
Brine	Α	5	5	5	5		32.9			33.4	3 3, 4	19.9			19.6	19.6	7.6	6.8	7.8	6.7	6.8	8.09		7.06	2.00	199
Control	В	5	5	5	5	5			f 33.1					10:7					63					7.89		
	С	5	4	4	ч	4																				
	D	5	5	5	5	5											ļ	-								
	E	5	5	>	5	5																				
	F	5	7	5	<u> </u>		40.0	100.0	ian n	ino.	40.2	19 9	ha a	ho V	101	19 <	77	65	47.7	/ 7	E G	z ())	7.94	1803	201	801
40 ppt	A B	5 5	55	5	5	55	-10.0	90.0	f 400	10.2	70.6			17.0 20.15	11.0		1.6	10	f 6.2	10.1	0. 1	0.01		17.92	0.01	
	c	5	5	5	5	5			700					10.15												
	D	5	Ü	4	4	4																				
	E	5	5	5	5	5																				
	F	5	5	5	5	5																				
42 ppt	Α	5	Ц	4	4	4	42.0	42.0	400	423	42.3	19.7	19.9	19:1	19.6	19.S	7.2	6.5	3 e -	-6.7	7,0	8.01	7.94	8.02	8.07	802
	в	5	9	5	5	5			<u>420</u>					507		<u> </u>			62			 		794		
	C	5	4	4	4	4	<u> </u>													-						1
		5	5	5	5	5						<u> </u>					<u> </u>									
	E F	5	55			5			-	Q.8	sislis								·							
40	A	5	9	5	5		42 ()	1320	hne		43.2		19.5	10 5	210,5	194	7.1	6.5	172	67	70	75.01	7.93	802	8 01	801
43 ppt	в	5	5	5	5	5	10.0	17		43.2			1	20.6					f 6.3					1 7.96		
	c	5	5	5	5	5																				
	D	5	4	4	4	4																				
	E	5	5	5	5	5				<u> </u>							<u> </u>				<u> </u>	<u> </u>	L	<u> </u>		
	F	5	5	5	5	6																				
Initial Counts QC'd by	: C	H																								
-			-	~	26	1 -	. 1 - 1	12					۱	71									Fee	ding T	imes	
Animal Source	/Date	Rece	ived:	A	67	1	5151	15		Age	at Initi	ation		20	۱ <u> </u>							0	24	48	72	96
									f fi	-1	ling in		amba	- nrior	to ron	owel					AM:		100	1083	1	0815
Comments:							ation,			~	ling in n)			· prior		5 <i>S</i> .	1: .:	f. M	le de v	-		1550	1673) - (1000	-	-
		Orga	anish	is ied	PHO		auon,	GIGE		<u>י ע</u>	<u></u>	<u>чп</u> (Ф)	AD	ers Ørg	31	7/15	<u>li ni</u>	7	<u>, , , , , , , , , , , , , , , , , , , </u>			Le la	1,	· · ·	ا	/
QC Check:	V	B	311	0/1	5							<u> </u>			r					F	inal R	eview	3	- 3/1	6/15	
Nautilus Environi			ŀ			San L	Diego, C	CA 921	20.																	
																	4									

Water Quality Measurements & Test Organism Survival

Client:	Pos	eido	n			_			_	Те	est Sp	ecies:	A. at	finis									Te	ch Init	ials	
Sample ID:	brine	e - N	lautil	us fro	ozen	seav	vater		_	Star	t Date	Time:	3/5/2	2015	15	50			-			0	24	48	72	96
Test No.:	15	707	3-	St	<u>ک</u> اد	>			_	Enc	Date	Time:	3/9/2	2015	14	140			-	c	ounts	CH	68	NO	AB	8K
																			-		dings	1 - 5 3			AB	
																			Dilutic		-	CH		NH		
ncentration ppt	Rep			ber o ganis		9		Salinity (ppt)										pH (units)							
		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
44 ppt	Α	5	5	5	5	5	44.0	44.0					C.C.C. (0.0.0000000000000000000000000000	North Contraction Contraction	5			6.7		1		8.01	· · · · · · · · · · · · · · · · · · ·			
	в	5	Ч	4	4	4			f 44.0					120.4		11.2		0.1	6.3		12			f	Diej	
	С	5	5	5	5	5			11.0					2011										796		
	D	5	5	5	5	5																				
	Е	5	R.	3	3	3																				
	F	5	5	4	4	4																				
	A				<u>-</u>				i					1					i					i		
	в					<u> </u>			f					f					f					f		
	с																						- 62			
	D					<u> </u>																				

	D																							<u> </u>
	E																							
	F																							
	A						i					li	T				1					i		
	В						f					f					f					f		
	C																							
	D																							
	E																							
	F																							
	Α						i				1	i					i					I		
	в						f					f					f					f		
	С																							
	D																							
	E																							
	F																							
	A						i					i					I					1		
	В						f					f					f					f		
	С																							
	D																							
	Е																							
	F																							
	r:			i							. /	o 1											<u> </u>	```
Animal Source	/Date	Received:	<u>AB5</u>	1313	115			Age a	at Initia	ation:	1	La									Feed	ling Ti	mes	
Comments:		i = initial re Organisms	eading i s fed pri ⊳∝mple	n fresh te or to initia	st solu ation, c	ition, f circle o	= final ne ((y	l readi	ng in t)	est ch	amber	prior t		ewal	Lave M	164			AM: PM:	0 (F	24 085	48 097 01570	0 ⁷²	96 0815 —
QC Check:	$\begin{array}{c c c c c c c c c c c c c c c c c c c $														-	Fin	al Rev	view:	Y	3/1	6/15			

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

Brine Dilution Worksheet

Project:	Poseidon		Analyst: _CH
Sample ID:	frozen seawater		Test Date: 3515
Test No:	1303-5015	>	Test Type: topsmelt acute / test inchation
Salinity of Se	awater _	33.1	-
Salinity of Br	ine _	84.0	Date of Brine used: 315415 115114 , 111314 , 112614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614 , 12614
Test Dilution	Volume _	1500	_ Alkalinity of Brine Control: <u>۱</u> 6 mg/L as CaCO3 3/5/15 عامار الح مد هاه عام عام ح
TS = targe SE = salin	t salinity ity of effluent		36365

SB = salinity of brine

Marine Chronic Bioassay

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
33.1	100.0	250	NA	NA	1500
40.0	86.4	1296.7	0.16	203.3	1500
42.0	82.5	1237.7	0.21	262.3	1500
43.0	80.6	1208.3	0.24	291.7	1500
		Strange Constant State			
44.0	78.6	1178.8	0.27	321.2	1500

	DI Volume			
Brine Cont	trol #DIV/07	«18 <u>3/57/5</u> 1.10	, 321.2	1500
5	23 477.2ª			_
	Total Brine Vol	ume Required (ml): 31615 642.4]
	~		1399,7	
QC Check: AC 3/13/1	5		Final Review:	43/16/15

Obrine was mixed the day prior to initiation from brine made op multiple dates to have enough volume for test in itation & renewal.

Brine Dilution Worksheet

Marine Chronic Bioassay

Project:	Poseidon		Analyst:	NH
Sample ID:	frozen seawater		Test Date:	3/5/2015
Test No:	1503-501S		Test Type:	48 hr Topsmelt Acute / Renewal
Salinity of Se	awater _	33.0	-	
Salinity of Br	ine _	84.0	Date of Brine used:	3/5/15/14,11/18/14, 11/25/14,12/11/14,
Test Dilution	Volume _	1500	Alkalinity of Brine Control:	-319+14
TS = targe	et salinity			AC Q18 3/13

SE = salinity of effluent SB = salinity of brine

Target Salinity ppt	Concentration % seawater	Seawater Volume (ml)	Salinity Adjustment Factor	Brine Volume (ml)	Dilute to: (ml)
33.1 3/11/5	100.0	250	NA	NA	1500
40.0	86.3	1294.1	0.16	205.9	1500
42.0	82.4	1235.3	0.21	264.7	1500
43.0	80.4	1205.9	0.24	294.1	1500
44.0	78.4	1176.5	0.28	323.5	1500

		DI Volume				
	Brine Control	3/16/16 294.1-	218 5716/15 1:40	323.5	1500	
		500.1	0,65			
		Total Brine Vol	ume Required (ml):	1411.8		
QC Check:	Ac 3/13/15	-		Final Review:	1204/2	1/15
@ Brine Made Íniti	uas mixe on mult ation & rev	d the date	ing prior to t	est initiat nough volu	ion from Ime for t	brine est

`

APPENDIX B

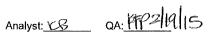
Reference Toxicant Test Data

Mysid Acute Reference Toxicant Test February 2015

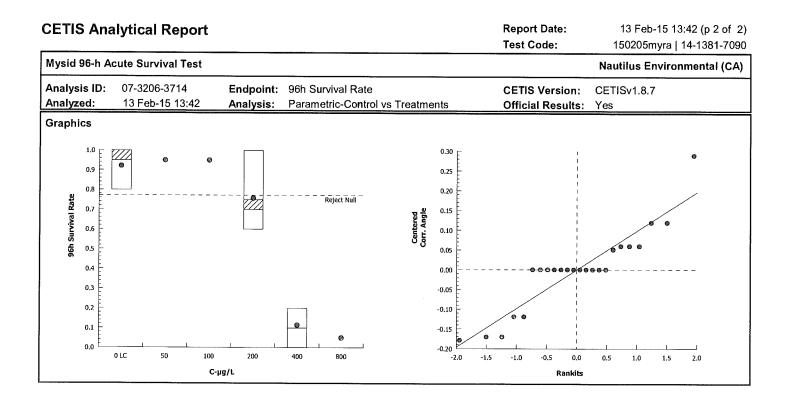
CETIS Sun	nmary Repo	ort						Report Dat Test Code				
Mysid 96-h Ac	ute Survival Te	st								Nautilu	s Environm	iental (CA)
Batch ID: Start Date: Ending Date: Duration:	04-3347-7787 05 Feb-15 15:5 09 Feb-15 14:0 94h	5 0	Test Type: Protocol: Species: Source:	Survival (96h) EPA/821/R-02- Americamysis Aquatic Biosys	bahia		·	Analyst: Diluent: Brine: Age:		ited Natural Applicable	Seawater	
Sample ID; Sample Date: Receive Date: Sample Age:	05 Feb-15		Code: Material: Source: Station:	150205myra Copper chlorida Reference Toxi Copper Chlorid	cant			Client: Project:	Inte	rnal	<u> </u>	
Comparison S	Summary											
Analysis ID	Endpoint		NOEL	LOEL	TOEL	PMSD	τu	Met	hod			
07-3206-3714	96h Survival Ra	ate	100	200	141.4	18.7%		Dun	nett N	Iultiple Com	parison Tes	st
Point Estimate	e Summary	With the design of the second										
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	τu	Met	hod			
20-9020-4206	96h Survival Ra	ate	EC50	257.5	219	302.7		Spea	Spearman-Kärber			
96h Survival F	Rate Summary											
C-µg/L	Control Type	Coun	t Mean	95% LCL	95% UCL	Min	Мах	std	Err	Std Dev	CV%	%Effect
0	Lab Control	4	0.95	0.7909	1	0.8	1	0.05		0.1	10.53%	0.0%
50		4	1	1	1	1	1	0		0	0.0%	-5.26%
100		4	1	1	1	1	1	0		0	0.0%	-5.26%
200		4	0.75	0.4453	1	0.6	1	0.09	574	0.1915	25.53%	21.05%
400		4	0.1	0	0.2837	0	0.2	0.05	774	0.1155	115.5%	89.47%
800	······	4	0	0	0	0	0	0		0		100.0%
96h Survival F	Rate Detail											
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4							
0	Lab Control	1	0.8	1	1							
50		1	1	1	1							
100		1	1	1	1							1
200		0.6	1	0.8	0.6							
400		0.2	0	0.2	0							

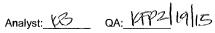
Analyst: KB QA: KTP 2/19/15

CETIS An	alytical Re	oort					-	ort Date: Code:			12 (p 1 of 2 1-1381-7090
Mysid 96-h /	Acute Survival	Fest							Nautilus	Environn	nental (CA)
Analysis ID: Analyzed:	07-3206-371 13 Feb-15 13		•	Survival Ra ametric-Cor		tments		IS Version: cial Results:	CETISv1. Yes	8.7	
Data Transf	orm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Cor	rected)	NA	C > T	NA	NA		18.7%	100	200	141.4	
Dunnett Mu	Itiple Comparis	on Test	ennimeter en								
Control	vs C-µg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision(a:5%)		
Lab Control	50		-0.6611	2.356	0.212 6	0.9445	CDF		, ficant Effect		
	100		-0.6611	2.356	0.212 6	0.9445	CDF	0	ficant Effect		
	200*		2.55	2.356	0.212 6	0.0350	CDF	Significant			
	400*		10.45	2.356	0.212 6	<0.0001	CDF	Significant			
ANOVA Tab	le										
Source	Sum Sq	uares	Mean Squ	are	DF	F Stat	P-Value	Decision(a:5%)		
Between	2.89755	3	0.7243882		4	44.66	< 0.0001	Significant			
Error	0.24328		0.0162191		15	11.00	0.0001	olgriniouni	Enool		
Total	3.14084			<u> </u>	19	_					
Distribution	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(a:1%)			
Variances	Mod Le	vene Equali	3.938	4.893	0.0222	Equal Var	iances				
Variances		Equality of		8.631	4,893	0.0008	Unequal \				
Distribution		-Wilk W No		0.9034	0.866	0.0478	Normal D				
96h Survival	Rate Summary	1									
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Мах	Std Err	CV%	%Effect
0	Lab Control	4	0.95	0.7909	1	1	0.8	1	0.05	10.53%	0.0%
50		4	1	1	1	1	1	1	0	0.0%	-5.26%
100		4	1	1	1	1	1	1	0	0.0%	-5.26%
200		4	0.75	0.4453	1	0.7	0.6	1	0.09574	25.53%	21.05%
400		4	0.1	0	0.2837	0.1	0	0.2	0.05774	115.5%	89.47%
800		4	0	0	0	0	0	0	0		100.0%
Angular (Co	rrected) Transfo	ormed Sum	mary								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Мах	Std Err	CV%	%Effect
0 µ9/m		4	1.286	1.096	1.475	1.345	1.107	1.345	0.05953	9.26%	0.0%
	Lab Control										
0	Lab Control	4	1.345	1.345	1.346	1.345	1.345	1.345	0	0.0%	-4.63%
0 50 100	Lab Control		1.345 1.345	1.345 1.345	1.346 1.346	1.345 1.345	1.345	1.345	0	0.0% 0.0%	-4.63% -4.63%
0 50	Lab Control	4									-4.63%
0 50 100	Lab Control	4 4	1.345	1.345	1.346	1.345	1.345	1.345	0	0.0%	



,





CETIS A	Analytical Re	eport					•	ort Date: Code:			3:42 (p 1 of 1 14-1381-709
Mysid 96-	h Acute Survival	Test							Nautilu	s Enviro	nmental (CA
Analysis I Analyzed:			•	6h Survival F ntrimmed Sp	Rate bearman-Kär	ber		S Version: ial Results:	CETISv1 Yes	.8.7	
Spearmar	n-Kärber Estimat	es			,						
Threshold	d Option	Threshold	Trim	Mu	Sigma		EC50	95% LCL	95% UCL		
Control Th	ireshold	0.05	0.00%	2.411	0.03513		257.5	219	302.7		
96h Survi	val Rate Summa	ry			Calcu	ulated Varia	te(A/B)				
C-µg/L	Control Type	Count	Mean	Min	Мах	Std Err	Std Dev	CV%	%Effect	А	в
0	Lab Control	4	0.95	0.8	1	0.05	0.1	10.53%	0.0%	19	20
50		4	1	1	1	0	0	0.0%	-5.26%	20	20
100		4	1	1	1	0	0	0.0%	-5.26%	20	20
200		4	0.75	0.6	1	0.09574	0.1915	25.53%	21.05%	15	20
400		4	0.1	0	0.2	0.05774	0.1155	115.5%	89.47%	2	20
800		4	0	0	0	0	0		100.0%	0	20
96h Survival Rate 0 0 0 0 0	.0 99 18 17 17 15 15 15 15 15 15 15 15 15 15 15 15 15										
0	0 100 200	300 400 С-µg/L	500 600	700 800							

alyst: UB QA: 14P2/19/15

CETIS QC Plot

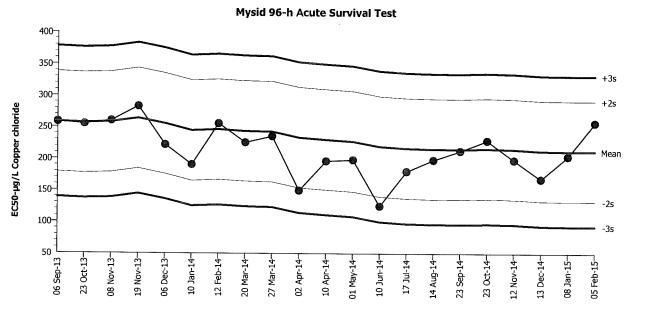
Mysid 96-h Acute Survival Test Nautilus Environmental (CA) Test Type: Survival (96h) Organism: Americamysis bahia (Opossum Shri Copper chloride Material:

Protocol: EPA/821/R-02-012 (2002)

Endpoint: 96h Survival Rate

Source:

Reference Toxicant-REF



Mean: Sigma:	212.3 39.84	Count: CV:	20 18.80%	-2s Warning Limit: +2s Warning Limit:	-3s Action Limit: +3s Action Limit:	
Quality Control Data					 	

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2013	Sep	6	15:05	258.5	46.24	1.161			13-4471-4704	00-6973-4292
2		Oct	23	13:30	254.9	42.61	1.07			16-6624-9217	07-4356-3093
3		Nov	8	14:45	260.1	47.84	1.201			01-3268-3774	14-6511-6184
4			19	18:00	282.8	70.54	1.771			18-1597-6213	02-6078-0521
5		Dec	6	16:15	221.9	9.614	0.2413			15-9430-7362	18-2355-9881
6	2014	Jan	10	14:40	190.3	-21.96	-0.5512			09-8692-0569	06-8811-0955
7		Feb	12	17:15	255.8	43.46	1.091			12-4495-5726	14-4047-7239
8		Mar	20	17:05	225.7	13.39	0.3361			04-4343-4707	19-2134-2594
9			27	17:15	235.7	23.38	0.5869			18-1249-5190	11-5455-8999
10		Apr	2	15:45	149.6	-62.7	-1.574			20-2330-3139	08-0863-3116
11			10	17:10	196.5	-15.77	-0.3958			16-5864-1833	17-8475-7680
12		May	1	15:15	198.3	-14.03	-0.352			13-9584-4772	21-4495-8860
13		Jun	10	16:15	124.9	-87.42	-2.194	(-)		00-0683-8894	06-8677-9461
14		Jul	17	16:45	180.3	-32.05	-0.8045			21-4298-8152	14-5683-3222
15		Aug	14	17:30	198.2	-14.07	-0.3531			18-8849-8580	16-2876-9748
16		Sep	23	16:35	212.8	0.5362	0.01346			12-4500-2750	04-3355-5833
17		Oct	23	15:15	229.4	17.1	0.4291			14-7022-2326	19-3230-6931
18		Nov	12	15:05	198.2	-14.07	-0.3531			15-3145-0944	01-6220-6073
19		Dec	13	16:30	168.2	-44.12	-1.107			11-0699-8383	10-8975-0476
20	2015	Jan	8	13:15	204.2	-8.055	-0.2022			16-0054-0041	01-6807-2003
21		Feb	5	15:55	257.5	45.17	1.134			14-1381-7090	20-9020-4206

Marine Acute Bioassay Static-Renewal Conditions

Water Quality Measurements & Test Organism Survival

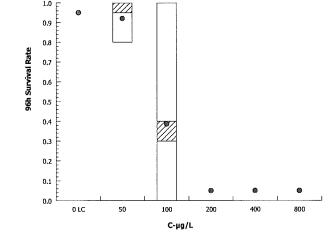
Client:	Inter	nal								Τe	est Sp	ecies;	A. b	ahia									Те	ch Init	ials	
Sample ID:									-		t Date/				15	15			-			0	24	48	72	96
Test No.:			yra						-	End	Date/	Time:	2/9/2	2015		160			-	С	ounts:	Ba	E67	AD	ĊĤ	AG
									-										•			ÅВ	Nt	AB,	BK	BK
																			Dilutio	ns ma	de by:	Ka		NH		
																		High	conc.	made	(µg/L):	800	-	\$00		
																	V	ol. Cu	stock	addec	l (mL):	18.4		18.4	1	
									Cu st	ock co	ncentr	ation	(µg/L):	56	90	0					(mL):			2000		
												n			1		1					r				
Concentration	Rand			ber o ganis		9		9	Salinit (ppt)	-			Ter	npera (°C)	ture		-	Disso	lved C (mg/L		n			pH (units)	Ň	
<u>(µg/L)</u>	#				Third Parts					200200														dedada;		
	8	0	24	48 5	72	96 S	0	24	48	72	96	0	24		72	96	0	24 50	48	72	96	0 7 av	24	48 i	72	96
Lab Control	。 7	5	5	25	5	4	29.8	<i>P.</i> (29.3	29.8	<i>3</i> 0,0	258	and the second sec		1220	25,3	U.T	29	6.8	6.3	5.9	7-98	194	7.96	<i>t:</i> 99	7.91
	, 15	5	5	1	7	<u> </u>			307					34.8					5.7					7.86		
	4	5 5	5	25	5	5																				
50	- - 11	5	5	5	5	ر S	79 9	101	hon	792	200	711	259	511-2	252	25.6	10 4	54	6.7	6 1	55	8 11	796	1,000	707	7.91
50	12	5	5	5	5	ر کا	-1.1	<u>90.1</u>	30.4	61.1	C1.8	25.0		74.7 1 72.2	23.3	210	4.1		5.3	0, 1	0,0	9.00	9.10	7.86	<u> 777</u>	
	2	5	Ś	5	5	5			10017										5.5					100		
	21	5	ら	6	5	5																				
100	10	5	5	5	5	5	29.9	20,2	ban	29.7	30,2	26.0	25.9	641	253	25.5	6.4	5.6	6.8	5.9	5.5	7.99	7.96	4.99	797	793
	13	5	Ś	5	5	5		1999/02/200	f 30.4					25.3					5.3					7.87		
	1	5	5	5	5	\$																				
	23	5	5	5	5	s							Ŕ,						505 100 100 100 100 100 100 100 100 100							
200	20	5	5	4	4		29.9	30,0	3.2	29.8	30,3	210.0	261	2412	25.3	25,4	64	5.9	6.7	6.1	5,5	7.9p	<i>1</i> :45	7.99	7.97	7.97
	14	5	5	5	5	Ч			f 30.5					25.4					! 5.5					7.93		
	6	5	5	5	5	^ب ų									Contraction Contraction											
	22	5	5	4	4	3																				
400	3	5	4	2		1 0	29.7	30,0	29.1	29.7	363					25,4	6.5	<u>(</u> .0	6.7	6,2	5,9	7.98	1.11.11.11.11.11.11	f i	794	7,98
	19	5	4	2		-			30.2			2	98 913 715	200					5.6					7.91		
	16	5	4	3	22	1							7/15	72.9												
	9	5	5	2		0		5.1	100													4	101	1.	Salara	
800	5	5	1	0	-0	n ² /	29.6	Accession.	6	-	0339333	24.4	25,7	f	-	-	ų.S	6,	6.7			7.94	4.76	f	1	`
	24 18	5	$\frac{0}{1}$	0	KY	$\left \right $			30.7					25.5					3.6					7.91		
	17	5 5	1 i	B									COLORES -													
Rand # QC:				<u> </u>	٢ <u>ــــــــــــــــــــــــــــــــــــ</u>		12220273	1999/1999	- 0404004069		202002000	10(0305135	- VIII (MILLIN		1000000000	(ALCOURSE)	3665555	1104000000	- ALCONTRA	0000000	Stewers	- (CC3+S(1))	9990000	100310000	- Superfeature	
Initial Count QC:	HO)				I																				
Animal Source/Da	te Rec	eived	I:	R	BS	2	14/1	5		_	Age a	at Initi	ation:	_5	d_								Fee	ding Ti	imes	
					/					-					•							0	24	48	72	96
Comments:		i = in	itial re	eading	g in fr	esh te	st solu	ition, f	= fina	l readi	ing in t	est ch	ambe	r prior	to ren	ewal					AM:		0846	6830		0815
		Orga	nism	s fed p	orior t	o initia	ation, c	circle o	one (<u>y</u> n)										PM:	140	193	1530	1530	-
		_							C													i	_ 1	, _}		
QC Check:	VP	2	21	12	<u>h5</u>	7													Fir	nal Re	view:	17F	2	1915	2	
Nautilus Environment	al. 4340) Vano	۱ lever A	Avenue	ə. San	n Diego	, CA 92	120.																		

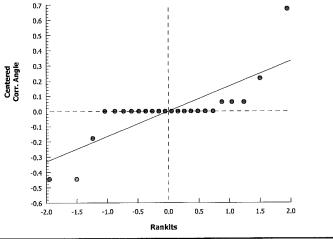
Topsmelt Acute Reference Toxicant Test February 2015

CETIS Sun	nmary Repo	rt						Report Dat Test Code:			Feb-15 12:0 210aara 05	
Pacific Topsm	nelt 96-h Acute S	Surviva	nl Test			÷				Nautilu	s Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	11-5441-5565 10 Feb-15 14:4 14 Feb-15 12:4 94h		Test Type: Protocol: Species: Source:	Survival (96h) EPA/821/R-02 Atherinops affi Aquatic Biosys	nis			Analyst: Diluent: Brine: Age:		Natural plicable	Seawater	
Sample ID: Sample Date: Receive Date: Sample Age:	10 Feb-15		Code: Material: Source: Station:	150210aara Copper chlorid Reference Tox Copper Chlorid	cicant			Client: Project:	Interna	I		
Comparison S	Summary											
Analysis ID	Endpoint		NOEL	. LOEL	TOEL	PMSD	τu	Met	hod			
04-7551-7551	96h Survival Ra	ite	100	200	141.4	43.9%		Stee	el Many-O	One Ran	< Sum Test	<u></u>
Point Estimate	e Summary											
Analysis ID	Endpoint		Level	μg/L	95% LCL	95% UCL	τU	Met	hod			
21-3305-5409	96h Survival Ra	ite	EC50		76.69	108.1		Trim	med Sp	earman-ł	Kärber	
96h Survival F	Rate Summary											
C-µg/L	Control Type	Coun	t Mean	95% LCL	95% UCL	Min	Max	Std	Err S	Std Dev	CV%	%Effect
0	Lab Control	4	1	1	1	1	1	0	C)	0.0%	0.0%
50		4	0.95	0.7909	1	0.8	1	0.05	C	.1	10.53%	5.0%
100		4	0.4	0	1	0	1	0.24	49 C	.4899	122.5%	60.0%
200		4	0	0	0	0	0	0	C	l		100.0%
400		4	0	0	0	0	0	0	C	I		100.0%
800		4	0	0	0	0	0	0	C			100.0%
96h Survival F	Rate Detail											
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4							
0	Lab Control	1	1	1	1							
50		1	1	0.8	1							
100		0.6	0	0	1							
200		0	0	0	0							
400		0	0	0	0							
100												



CETIS An	alytical Rep	ort					-	ort Date: Code:			6 (p 1 of 1) -4867-3928
Pacific Tops	melt 96-h Acute	Survival T	est						Nautilus	Environm	nental (CA)
Analysis ID: Analyzed:	04-7551-7551 20 Feb-15 12:			Survival Ra	ite Control vs T	reatments		IS Version: al Results:	CETISv1. Yes	8.7	
Data Transfo	ərm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	τU
Angular (Corr	ected)	NA	C > T	NA	NA		43.9%	100	200	141.4	
Steel Many-C	One Rank Sum T	est									
Control	vs C-µg/L		Test Stat	Critical	Ties DF	P-Value	P-Type	Decision(a	a:5%)		
Lab Control	50		16	11	1 6	0.4206	Asymp	Non-Signif	icant Effect		
	100		12	11	1 6	0.0738	Asymp	Non-Signif	icant Effect		
ANOVA Tabl	e										
Source	Sum Squ	ares	Mean Squ	are	DF	F Stat	P-Value	Decision(a	a:5%)		
Between	1.116211		0.5581056		2	5.342	0.0296	Significant	Effect		
Error	0.940362	5	0.1044847		9						
Total	2.056574				11						
Distributiona	al Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision	(a:1%)			
Variances	Mod Lev	ene Equali	ty of Variance	10.47	8.022	0.0045	Unequal	/ariances			
Variances	Levene E	Equality of	Variance	17.2	8.022	0.0008	Unequal	/ariances			
Distribution	Shapiro-	Wilk W No	rmality	0.8681	0.8025	0.0618	Normal D	istribution			
96h Survival	Rate Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	4	1	1	1	1	1	1	0	0.0%	0.0%
50		4	0.95	0.7909	1	1	0.8	1	0.05	10.53%	5.0%
100		4	0.4	0	1	0.3	0	1	0.2449	122.5%	60.0%
200		4	0	0	0	0	0	0	0		100.0%
400		4	0	0	0	0	0	0	0		100.0%
800		4	0	0	0	0	0	0	0		100.0%
Angular (Cor	rected) Transfor	med Sum	mary								
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	4	1.345	1.345	1.346	1.345	1.345	1.345	0	0.0%	0.0%
50		4	1.286	1.096	1.475	1.345	1.107	1.345	0.05953	9.26%	4.43%
100		4	0.6706	-0.1999	1.541	0.5558	0.2255	1.345	0.2735	81.58%	50.15%
200		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
400		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
800		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
Graphics											
1.0 E	17772					0.7 E		1			
	0 <u>22</u>					0.6					





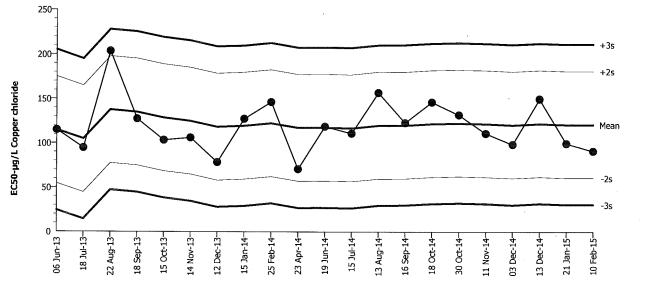
	Analytical Re	port					•	ort Date: Code:			2:06 (p 1 of 1 05-4867-392
Pacific To	opsmelt 96-h Acut	e Survival T	est						Nautilu	s Enviro	nmental (CA)
Analysis i Analyzed:			•	Sh Survival I immed Spe		ber		IS Version: al Results:	CETISv1 Yes	.8.7	
Trimmed	Spearman-Kärber	- Estimates									
Threshold	d Option	Threshold	Trim	Mu	Sigma		EC50	95% LCL	95% UCL		
Control Th	reshold	0	5.00%	1.959	0.03726		91.04	76.69	108.1		
96h Survi	val Rate Summar	y			Cal	culated Varia	ite(A/B)				
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	А	в
0	Lab Control	4	1	1	1	0	0	0.0%	0.0%	20	20
50		4	0.95	0.8	1	0.05	0.1	10.53%	5.0%	19	20
100		4	0.4	0	1	0.2449	0.4899	122.5%	60.0%	8	20
200		4	0	0	0	0	0		100.0%	0	20
400		4	0	0	0	0	0		100.0%	0	20
800		4	0	0	0	0	0		100.0%	0	20
9 6th Survival Rate 0 0 0 0											
	0 100 200	300 400 С-µg/L	500 600	700 800							

Analyst: KB QA: KTP2/21/0/15

CETIS QC Plot

Pacific Topsmelt 96-h Acute Survival Tes	st	Nautilus Environmental (CA)
Test Type: Survival (96h)	Organism: Atherinops affinis (Topsmelt)	Material: Copper chloride
Protocol: EPA/821/R-02-012 (2002)	Endpoint: 96h Survival Rate	Source: Reference Toxicant-REF

Pacific Topsmelt 96-h Acute Survival Test



Mean: Sigma:	121 30.12	Count: CV:	-2s Warning Limit: +2s Warning Limit:	-3s Action Limit: +3s Action Limit:	
Quality Control Data			 		

Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2013	Jun	6	14:25	114.9	-6.13	-0.2035			12-0033-0929	20-0252-9346
2		Jul	18	12:50	94.59	-26.41	-0.8768			19-2632-6339	04-8526-8990
3		Aug	22	14:40	203.7	82.68	2.745	(+)		16-8357-2725	11-7110-5550
4		Sep	18	13:50	127.5	6.456	0.2143			09-9085-4812	11-5673-1751
5		Oct	15	15:20	103.5	-17.47	-0.5801			00-5901-5898	17-6384-6991
6		Nov	14	9:40	106.1	-14.88	-0.4941			06-5418-8921	10-2371-6330
7		Dec	12	13:20	78.46	-42.54	-1.412			12-4998-2305	03-2148-1441
8	2014	Jan	15	15:25	127.5	6.456	0.2143			13-3854-5258	05-1070-1044
9		Feb	25	13:55	146.4	25.41	0.8436			20-0325-5939	07-6658-0335
10		Apr	23	16:00	70.71	-50.29	-1.67			11-8272-9093	14-5541-7971
11		Jun	19	15:35	118.9	-2.079	-0.06903			11-2944-5183	19-5384-3170
12		Jul	15	14:15	111	-10.04	-0.3334			00-8730-8108	10-9428-5566
13		Aug	13	14:10	156,9	35.92	1.192			12-9208-7415	02-1974-5349
14		Sep	16	13:10	123.1	2.114	0.0702			05-7478-8365	11-8140-9628
15		Oct	18	10:45	146.4	25.41	0.8436			18-6908-7115	01-0100-8379
16			30	11:50	132	10.95	0.3636			17-2734-9303	12-4790-6162
17		Nov	11	10:50	111	-10.04	-0.3334			19-6246-9477	14-2586-1124
18		Dec	3	16:45	98.53	-22.47	-0.746			07-3639-8754	16-6449-6521
19			13	15:30	150.1	29.08	0.9654			03-6652-6590	12-5127-9321
20	2015	Jan	21	15:35	99.71	-21.29	-0.7068			16-8270-8063	13-6137-3732
21		Feb	10	14:45	91.04	-29.96	-0.9947			05-4867-3928	21-3305-5409

Water Quality Measurements & Test Organism Survival

Client:	Intern	al								Τe	st Sp	ecies:	A. ai	finis									Tee	ch Initi	als	
Sample ID:									-	Star	Date/	Time.	2/9/2	2015	2/10	3/15	- 14	40	/			0	24	48	72	96
Test No.:	1502(16 99aa	ra						•	End	Date/	Time:	2/13	/2015	2	14/1	51	241	5	C	ounts:	ЛLВ	чH	144	NIT	N#
	6	Ð							•										-		dings:	<u>~</u> .	hit	 化 B	NN}	AN3
																			Dilutio	ns ma	de by:	Àl.ħ	1	$\beta $	ł	~
																		High	сопс. і	nade ((μg/L):	800		200		
														-			٧	/ol. Cu	stock	added	(mL):	18.4		4.6		
									Cu ste	ock co	ncentr	ation ((μg/L):	<u> </u>	2,90	0		F	inal V	olume	(mL):	2000		2000		
							a					1			,		1					1				
Concentration (µg/L)	Rand #		Num Or	ber o ganis				5	Salinit (ppt)	у			Ter	nperat (°C)		92			lved O (mg/L)		ו		(pH (units))	
		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
Lab Control	22	5	5	5	5	5	299.9	30.1		30.B	32.0	203	70.9		20.4	30.5	7,4	6.4	4 9	64	6.7	7,98	7,50	17.11	748	7.70
	2	5	5	5	5	5			€7£:V					20.9					f رو ۱					474		
	10	5	5	5	5	5																				
	15	5	5	5	5	5			1																el	
50	24	5	5	5	5	5	29.8	30. Z.		30.2	30.4	20,3	20:8		W:C	21.1	7,3	6.3		6.2	6.2	7.96	177		783	7.72
	8	5	5	5	5	5			¹ 19.7					E20.7					14.2					7 . 84	- individual Individual Individuality	
	17	5	4	4	4	7																				
	11	5	5	5	G	5			1.	a.) #			- 1	1	1990 1995 1995				i and a second s							
100	12	5	5	ય	4	3	29.8	71,7	-	30.5	318	20,3	70.6	201	O.U	20.4	1.3	69		65	6.4	798	411		3.41	7.78
	4	5	3	}					30.4					20.3					10,4					*7.Se		
	1 19	5	5	15	15	05																				
	23	5	0	-	2	2	200	241				74.0	201	120.0	-	-	70	63	1. a			7.96	170	4 14		0330-0-
200	13	5 5		0		1	29.8	Cr (29.8 530-1			20.5	100	20.7		(hailand)	14:3	6)	4.3		dargebedarg Statistics Statistics Statistics	<i>f</i> .16	4.40	1:11 4-87		
	14	5 5	0	-		-			()) ()			visisiois Nissois		20.7					8.7					T-01		
	18	5	0	~		/																				
400	9	5	0			$\overline{\nabla}$	29.7	79.45	i ,			20.4	70%	i			JE	63	i	-		7.97	7m	i		-
400	16	5	0		59	y	<u>~ 1,1</u>		1_					r			1,0		f			1.17		f		
	7	5	0	K'																						
	5	5	0	7		~																				
800	3	5	σ		0	Ň	29.6	246	I	-	_	20,3	70.7	- 1		-	7.3	6.3	۰	-	-	7.98	7.71	- 1	-	-
	21	5	0		X				f					f		Compositor			f					1-		
	6	5	Q	1×																						
	20	5	\mathcal{O}																	1992/19 21/1971 21/1971			anata Anata			
Rand # QC: Initial Count QC:	ALI	3				1																				
Animal Source/[Date Re	eceiv	ed:	Ą	BC	/ '	21	ψh	9			Age a	at Initi	ation:	1	<u> (</u>	114	\$				0	Fee 24	ding Ti 48	mes 72	96
Commontos		1 - 1-	itictra	adia	a in fr	och in	et col·	tion 4	- fina	Irood	ing in f	act ab	ambo	nnior	to ran	owol					A 84-				5915	
Comments:							st solu										 				AM:	ian			<u>, , , , , , , , , , , , , , , , , , , </u>	<u>u-100</u>
		La	6 2	nti	H a	and	5 100	$\frac{1}{5}$	alin	the	' র	ight	lur	light	ra	Ř. 9	6 hou	us.			r ivi;	,		L	[]	ang tao
QC Check:	VA	37	212	10	15		V	alm	25 U	nfi	mið	y w	NHV.	Oas	secu	olic A 9 Nd 3	salin	ity	yeta Fir	າal Re	view:	11	P 2	124	115	
Nautilus Environme	ental. 43	340 Va	ndeve	r Aver	nue. S	an Die	go, CA	92120.																		

Mysid Acute Reference Toxicant Test March 2015

CETIS Sum	imary Repo	rt						Report Dat Test Code:			/lar-15 11:10)3myra 16	
Mysid 96-h Ac	ute Survival Tes	st								Nautilus	Environm	ental (CA)
Batch ID: Start Date: Ending Date: Duration:	17-5434-7167 03 Mar-15 15:10 07 Mar-15 13:55 95h	D 5	Test Type: Protocol: Species: Source:	Survival (96h) EPA/821/R-02- Americamysis b Aquatic Biosyst	bahia			Analyst: Diluent: Brine: Age:		ed Natural S Applicable	Seawater	
Sample ID: Sample Date: Receive Date: Sample Age:			Code: Material: Source: Station:	150303myra Copper chloride Reference Toxi Copper Chlorid	cant			Client: Project:	Inter	nal		
Comparison S	Summary						-					
Analysis ID	Endpoint		NOEL		TOEL	PMSD	τu	Met			ria an Taa	.1
00-4701-4628	96h Survival Ra	ate	50	100	70.71	12.9%		Dun		lultiple Com	parison res	l
Point Estimate	e Summary											
Analysis ID	Endpoint		Level	µg/L	95% LCL	95% UCL	ΤU	Met	hod			
10-4165-9670	96h Survival Ra	ate	EC50	150.3	121.7	185.7		Trin	nmed	Spearman-k	(ärber	
96h Survival F	Rate Summary											
C-µg/L	Control Type	Cour	t Mean	95% LCL	95% UCL	Min	Max	std	Err	Std Dev	CV%	%Effect
0	Lab Control	4	1	1	1	1	1	0		0	0.0%	0.0%
50		4	0.95	0.7909	1	0.8	1	0.05	5	0.1	10.53%	5.0%
100		4	0.8	0.8	0.8	0.8	0.8	0		0	0.0%	20.0%
200		4	0.3	0.1163	0.4837	0.2	0.4	0.05	5774	0.1155	38.49%	70.0%
400		4	0	0	0	0	0	0		0		100.0%
800		4	0	0	0	0	0	0		0		100.0%
96h Survival F	Rate Detail			9999	<u>giterenn of the start</u>							
C-µg/L	Control Type	Rep	1 Rep 2	2 Rep 3	Rep 4							
0	Lab Control	1	1	1	1							
50		1	1	1	0.8							
100		0.8	0.8	0.8	0.8							
200		0.4	0.2	0.2	0.4							
			0	0	0							
400		0	0	0	0							

Analyst: AC QA: 177-3/21/15

CETIS Ana	lytical Rep	ort						ort Date: Code:		lar-15 11:16 3myra 16-	
Mysid 96-h A	cute Survival Te	est							Nautilus	Environm	ental (CA)
Analysis ID: Analyzed:	00-4701-4628 13 Mar-15 11:		•	Survival Ra metric-Con	te trol vs Treat	ments		S Version: ial Results	CETISv1. : Yes	8.7	
Data Transfo	rm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	τU
Angular (Corre		NA	C > T	NA	NA		12.9%	50	100	70.71	
Dunnett Mult	iple Compariso	n Test									
Control	vs C-µg/L		Test Stat	Critical	MSD DF	P-Value	P-Type	Decision	(α:5%)		
Lab Control	50		0.9647	2.287	0.141 6	0.3485	CDF	Non-Sign	ificant Effect		
	100*		3.859	2.287	0.141 6	0.0030	CDF	Significan			
	200*		12.49	2.287	0.141 6	<0.0001	CDF	Significar	t Effect		
ANOVA Table	9										
Source	Sum Sq	uares	Mean Squ	are	DF	F Stat	P-Value	Decision	(α:5%)		
Between	1.477123	3	0.4923743		3	64.64	<0.0001	Significar	nt Effect		
Error	0.091403	857	0.0076169	65	12	_					
Total	1.568527	7			15						
Distributiona	I Tests				<u>an</u>						
Attribute	Test			Test Stat	Critical	P-Value	Decision	(α:1%)			
Variances	Mod Lev	vene Equa	ality of Variance	3.21	5.953	0.0618	Equal Va				
Variances	Levene	Equality c	f Variance	15.36	5.953	0.0002		Variances			
Distribution	Shapiro	-Wilk W N	lormality	0.8697	0.8408	0.0269	Normal D	istribution			
96h Survival	Rate Summary										
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	4	1	1	1	1	1	1	0	0.0%	0.0%
50		4	0.95	0.7909	1	1	0.8	1	0.05	10.53%	5.0%
100		4	0.8	0.7997	0.8003	0.8	0.8	0.8	0 0.05774	0.0% 38.49%	20.0% 70.0%
200		4	0.3	0.1163	0.4837	0.3	0.2	0.4 0	0.05774	30.4970	100.0%
400		4	0	0 0	0 0	0 0	0 0	0	0		100.0%
800		4	0	U	U	0	0	U	.		
Angular (Cor	rrected) Transfo	ormed Su	mmary							C) (0/	%Effect
C-µg/L	Control Type	Count			95% UCL		Min	Max	Std Err	CV%	
0	Lab Control	4	1.345	1.345	1.346	1.345	1.345	1.345 1.345	0 0.05953	0.0% 9.26%	0.0% 4.43%
50		4	1.286	1.096	1.475 1.108	1.345 1.107	1.107 1.107	1.345	0.05955	0.0%	17.7%
100		4	1.107	1.107 0.3711	0,7773	0.5742	0.4636	0.6847	0.06382	22.23%	57.32%
200		4	0.5742 0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
400 800		4 4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
			0.2200	<u></u>							
96h Survival		Dan 4	Rep 2	Rep 3	Rep 4						
C-µg/L	Control Type	Rep 1	1	<u>1</u>	1						
0	Lab Control	1	1	' 1	0.8						
50		۱ 0.8	0.8	0.8	0.8						
100		0.8	0.8	0.2	0.4						
200		0.4 0	0.2	0.2	0.4						
400				0	0						
800		0	0		<u> </u>						

CETIS Ana	alytical Repo	ort						Report Date: Test Code:	13 Mar-15 11:16 (p 2 of 2) 150303myra 16-0363-1107
Mysid 96-h A	cute Survival Te	st							Nautilus Environmental (CA)
Analysis ID: Analyzed:	00-4701-4628 13 Mar-15 11:1			6h Survival Ra arametric-Cor		atme	nts	CETIS Version: Official Results:	CETISv1.8.7 Yes
	rected) Transfor								
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4				
0	Lab Control	1.345	1.345	1.345	1.345				
50		1.345	1.345	1.345	1.107				
100		1.107	1.107	1.107	1.107				
200		0.6847	0.4636	0.4636	0.6847				
400		0.2255	0.2255	0.2255	0.2255				
800		0.2255	0.2255	0.2255	0.2255				
Graphics 1.0 0.9 0.8 20 20 20 20 20 20 20 20 20 20	•	Θ		Reject Null	-	Centered Corr. Angle	0.15 0.10 0.05 0.00		0 0 0 0 0 0
496 0.5							-0.05		

0

400

100

C-µg/L

200

6

800

-0.05

-0,10

-0.15

-0.20 L... -2.0

9 0

-1,5

-1.0

-0,5

0,0

Rankits

Analyst: AC_____QA:4773/26/15

1.5

1.0

0.5

2.0

0.4

0.3

0.2

0.1

0,0

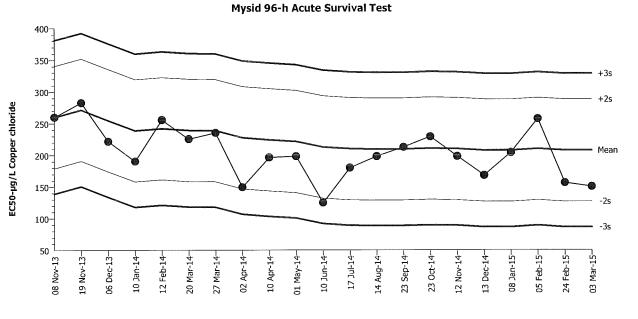
0 LC

50

ETIS Analytical R	eport			-	ort Date: Code:	13 Mar-15 11:16 (p 1 of 1 150303myra 16-0363-110					
Mysid 96-h Acute Surviva	l Test							Nautilus	s Enviroi	nmental (CA	
Analysis ID: 10-4165-96 Analyzed: 13 Mar-15			ih Survival F immed Spea	Rate arman-Kärbe	er		S Version: ial Results:	CETISv1.8.7 Yes			
Trimmed Spearman-Kärb	er Estimates		<u>,,,,</u>								
Threshold Option	Threshold	Trim	Mu	Sigma		EC50	95% LCL	95% UCL			
Control Threshold	0	5.00%	2.177	0,04587		150.3	121.7	185.7			
96h Survival Rate Summa	ary			Calc	ulated Varia	te(A/B)					
C-µg/L Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	А	В	
) Lab Control	4	1	1	1	0	0	0.0%	0.0%	20	20	
50	4	0.95	0.8	1	0.05	0.1	10.53%	5.0%	19	20	
100	4	0.8	0.8	0.8	0	0	0.0%	20.0%	16	20	
200	4	0.3	0.2	0.4	0.05774	0.1155	38.49%	70.0%	6	20	
400	4	0	0	0	0	0		100.0%	0	20	
800	4	0	0	0	0	0		100.0%	0	20	
96h Survival Rate Detail											
C-µg/L Control Type	e Rep 1	Rep 2	Rep 3	Rep 4							
0 Lab Control	1	1	1	1							
50	1	1	1	0.8							
100	0.8	0.8	0.8	0.8							
200	0.4	0.2	0.2	0.4							
400	0	0	0	0							
800	0	0	0	0							
Graphics	0 300 400			0							

CETIS QC Plot

Mysid 96-h Acute Survival Test			Nautilus Environmental (CA)
Test Type: Survival (96h)	Organism: Americamysis bahia (Opossum Shri	Material:	Copper chloride
Protocol: EPA/821/R-02-012 (2002)	Endpoint: 96h Survival Rate	Source:	Reference Toxicant-REF



Mean:	207.3	Count:	20	-2s Warning Limit:	126.7	-3s Action Limit:	86.38
Sigma:	40.32	CV:	19.50%	+2s Warning Limit:	288	+3s Action Limit:	328.3

Quali	ty Con	trol Data	a								
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2013	Νον	8	14:45	260.1	52.84	1.311			01-3268-3774	14-6511-6184
2			19	18:00	282.8	75.54	1.874			18-1597-6213	02-6078-0521
3		Dec	6	16:15	221.9	14.61	0.3624			15-9430-7362	18-2355-9881
4	2014	Jan	10	14:40	190.3	-16.96	-0.4207			09-8692-0569	06-8811-0955
5		Feb	12	17:15	255.8	48.46	1.202			12-4495-5726	14-4047-7239
6		Mar	20	17:05	225.7	18.39	0.4561			04-4343-4707	19-2134-2594
7			27	17:15	235.7	28.38	0.7039			18-1249-5190	11-5455-8999
8		Apr	2	15:45	149.6	-57.7	-1.431			20-2330-3139	08-0863-3116
9			10	17:10	196.5	-10.77	-0.2671			16-5864-1833	17-8475-7680
10		May	1	15:15	198.3	-9.025	-0.2238			13-9584-4772	21-4495-8860
11		Jun	10	16:15	124.9	-82.42	-2.044	(-)		00-0683-8894	06-8677-9461
12		Jul	17	16:45	180.3	-27.05	-0.6709			21-4298-8152	14-5683-3222
13		Aug	14	17:30	198.2	-9,069	-0.2249			18-8849-8580	16-2876-9748
14		Sep	23	16:35	212.8	5.536	0.1373			12-4500-2750	04-3355-5833
15		Oct	23	15:15	229.4	22.1	0.548			14-7022-2326	19-3230-6931
16		Nov	12	15:05	198.2	-9.069	-0.2249			15-3145-0944	01-6220-6073
17		Dec	13	16:30	168.2	-39.12	-0.9703			11-0699-8383	10-8975-0476
18	2015	Jan	8	13:15	204.2	-3.055	-0.07576			16-0054-0041	01-6807-2003
19		Feb	5	15:55	257.5	50.17	1.244			14-1381-7090	20-9020-4206
20			24	14:45	156.3	-50.99	-1.265			00-7545-3960	18-6061-5275
21		Mar	3	15:10	150.3	-56.99	-1.413			16-0363-1107	10-4165-9670

Analyst: AC QA: 4783/240/15

Marine Acute Bioassay Static-Renewal Conditions

Water Quality Measurements & Test Organism Survival

Client:	Interr	nal								Te	st Spe	cies:	A. ba	nhia									Тес	h Initi	als	
Sample ID:												Time:			15	0						0	24	48	72	96
Test No.:												Time:			1-	350	5			Co	ounts:	EG	CH	ĒG	KB	AD
1651110	10000	<u>, on y</u>														4.	/			Read	lings:	AĊ	V ~S		K	AB
																		[Dilutio	ns mac	de by:	AC		E6		
																		High (conc. r	nade (μg/L):	800	1999 - 1999 - 1999 - 1999 - 1999 - 1999 - 1999	800	-	-
																	v	ol. Cu	stock	added	(mL):	(8,4	ан -	182.4	1	
									Cu sto	ock co	ncentr	ation (μg/L):	86	,90	06		F	inal Ve	olume	(mL):	2000		2000		
												-														
Concentration	Rand			ber o ganis	f Live sms				Salinit (ppt)	у			Ten ৫২।	nperat (°C)	ure			Dissol	ved O (mg/L)		•		1	рН (units)		
<u>(µg/L)</u>	#	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
Lab Control	8	5	5	5	5	5		30,8		1111111111	29.8	24.3	25,8	124.(25.4	25.5	6,6	6,3	5,1	53	5.7	7.92	7,90	803	7.82	7.80
Lub Control	7	5	5	<u>بر</u> ۲	5	5	Shiredi		f 31.0	1994			alan.	f24.91	N.N.A.			elitere de la comparte Comparte de la comparte de la comparte Comparte de la comparte	5.8				egan Anne E	7.81		1.1.1
	15	5	B	5	5	5																				
	4	5	5	5	5	5																				
50	11	5	5	5	S	5	30,2	30,3	30.1	309	31.1	24.2	26,3	247	25.1	25.7	6.7	6.0	7.0	5,7	5.5	7.94	7.90	804	7.84	7.80
	12	5	5	5	5	5			305				ineria jerena	f 24.6	n de la constante Esta de la constante Esta de la constante de la const Esta de la constante de la const Esta de la constante de la const	i de la composition de la composition			5.8					180		
	2	5	5	5	5	5																				
	21	.5	5	4	4	4																				
100	10	5	5	5	ÿ	4	30.2	30,3	30.1	302	31.1	242	26,2	24.4	25,5	25.0	6.1	6.0	7.1	5,5	5.7	795	7,91	8.05	7.84	786
	13	5	5	4	4	4			30.6					24.5					5.9					283		
	1	5	5	5	4	4																				
	23	5	5	4	4	Ц	30.1								1338									1 (2010) 2 (2010) 1 L		
200	20	5	4	2	2	2	342	30,4	1.		30.6	242	26.3	24.7	25,4	25.2	67	6.1	7.0	5.7	5.7	7.96	7,92	8.04	7,87	7.96
	14	5	3	2	2	1	815		30.5					24.4					5.8					7.81		
	6	5	1		1		313																			
	22	5	5	4	4	2			1.30 7					1	<u>1999</u>				1			-105		6.7	-	
400	3	5	CV3	10	<u> </u>	1/	30 <u>p</u>	30,2				24.	26,1	25.2		10004	67	6.1	6.7	ada sa			21.10	8.03		9 - C
	19	5	1	Ø	<u> </u>	400			305					24.8					38					1.83	>	
	16	5	1	0		ter						-														
	9	5	3	0	<u> </u>	p	1		129.5		-	N 11 1		1			1,7	6.2	16.7	-	-	701	1	8.00	- -	
800	5	5	2	6		1		\$ 30.1	f 31	¥/IS	5 A4654	J.H.	126,5	25.4			6.7	0.1	1.			1.	/ (f		
	24	5	2	0	\rightarrow	fl.	14 218		30.4	. 1973) 1. AAAA	n staffs National			24.5	1				5.8					7.86		
	18	5	0	0	17	100	1313			n neder 1 1 de de d								8 (1993) 1 (1993)								
Rand # QC	17 : AL	5	3			1		a aag	1.997							1		4 - 43 <u>5</u> 5	1.4245			1	1			
Initial Count QC			_																							
				A	651	2	3/3/1	6			Δле	at Init	iation	:	41								Fe	eding 1	limes	
Animal Source/Da	ate Rec	erve	u:		<u> </u>		<u>) / </u>	/		-	Age	at mit	1411011		12	·						0	24	48	72	96
0 a		i i.	nitiol -	raadi	na in f	roch f	est sol	ution	f = fin	al rea	dina in	test cl	hambe	er prior	to rer	newal					AM	: New York	6820	083	083	D MOC
Comments:							iation,			A\\		1001 0								_	DM	. 1101	510	TIPE	155	
		(A)	P CH	is red	1 prior		6	unule		<u>y</u> '	<u>)</u>											<u>``</u>		1	·	
DC Charles	V	'B		7 [13]	-	5.0	2							•					F	inal R	eview	:th	pЗ	24	15	
QC Check:	×	-4-1		1	· · · ·		-	2120																4		
Nautilus Environmer	ntal, 434	iU Van	dever	Aven	ue. Sa	n Dieg	O, CA 9	z120.																		

Topsmelt Acute Reference Toxicant Test March 2015

CETIS Summary Report

13 Mar-15 11:19 (p 1 of 1) 150305aara | 19-3155-0811

elt 96-h Acute Su	urvival	Test								Nautilus	Environme	ental (CA)
15-8595-8712 05 Mar-15 16:00 09 Mar-15 14:35 95h	P S	rotocol: pecies:	EPA/821/ Atherinop	R-02-01 s affinis	5			-	:: Di No	ot Applicable	Seawater	
11-0049-8953 05 Mar-15 05 Mar-15 16h	N	laterial: ource:	Copper cl Reference	iloride Toxica	ant			Client: Project		ternal		
Summary Endpoint		NOEL	LOE	L	TOEL	PMSD	τU	r	Method	I		
	e	50	100		70.71	15.3%		ę	Steel M	any-One Rank	Sum Test	
e Summary Endpoint 96h Survival Rat	e				95% LCL 105.1	95% UCL 154.6	TU					
Rate Summary												
Control Type	Count	Mear	n 95%	LCL	95% UCL	Min	Max				CV%	%Effect
Lab Control	4 4 4 4 4 4	1 1 0.65 0.2 0 0	1 1 0.49 0 0 0	09	1 1 0.8091 0.4598 0 0	1 1. 0.6 0 0 0	1 0.8 0.4 0		0 0.05 0.0816 0	0 0.1	0.0% 0.0% 15.38% 81.65%	0.0% 0.0% 35.0% 80.0% 100.0% 100.0%
Rate Detail												
Control Type	Rep 1	Rep	2 Rep	3	Rep 4							
Lab Control	1 1 0.6 0 0	1 1 0.8 0.2 0	1 1 0.6 0.4 0		1 1 0.6 0.2 0							
	15-8595-8712 05 Mar-15 16:00 09 Mar-15 14:35 95h 11-0049-8953 05 Mar-15 05 Mar-15 16h 300 Mar-15 300 Mar-15 16h 300 Mar-15 300 Mar-15 3000	15-8595-8712 T 05 Mar-15 16:00 P 09 Mar-15 14:35 S 95h S 11-0049-8953 C 05 Mar-15 S 16h S 5ummary Endpoint 96h Survival Rate S Rate Summary Count 4 4 4 4 4 4 4 4 4 4 4 4 4 1 Control Type Rep 1 Lab Control 1 1ab Control 1 0.6 0	05 Mar-15 16:00 Protocol: 09 Mar-15 14:35 Species: 95h Source: 11-0049-8953 Code: 05 Mar-15 Material: 05 Mar-15 Source: 16h Station: 35mmary Endpoint NOEL 96h Survival Rate 50 96h Survival Rate EC50 Rate Summary Ecc50 Control Type Count Mear Lab Control 4 1 4 0,6 0,4 0 Rate Detail Control Type Rep 1 Rep Lab Control 1 1 1 0 4 0,6 0,8 0 0 0,2 0	15-8595-8712 Test Type: Survival (9 05 Mar-15 16:00 Protocol: EPA/821/6 09 Mar-15 14:35 Species: Atherinops 95h Source: Aquatic Bi 11-0049-8953 Code: 150305aa 05 Mar-15 Material: Copper ch 05 Mar-15 Source: Reference 16h Station: Copper ch 30 S Mar-15 Source: Reference 16h Station: Copper ch 96h Survival Rate 50 100 96h Survival Rate 50 127. Rate Summary Ec50 127. Rate Summary Ec50 127. Rate Summary Countrol Type Count Mean 95% Lab Control 4 1 1 1 4 0.65 0.49 4 0 0 4 0.2 0 4 0 0 0 Rate Detail Control Type Rep 1 Rep 2 Rep Lab Control 1 1 1 <td< td=""><td>15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-07 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosyste 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxica 16h Station: Copper Chloride 3ummary Endpoint NOEL LOEL 96h Survival Rate 50 100 96h Survival Rate EC50 127.5 Rate Summary Ecc50 127.5 Rate Summary Ecc50 127.5 Rate Summary Ecc50 127.5 Rate Summary Ecc50 127.5 Rate Summary 4 1 1 4 0.65 0.4909 4 4 0.2 0 4 4 0 0 0 4 0 0 0 4 0 0 0 4 0 <</td><td>15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosystems, CO 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 96h Survival Rate 50 100 70.71 96h Survival Rate EC50 127.5 105.1 8ate Summary Ec50 127.5 105.1 96h Survival Rate EC50 127.5 105.1 8ate Summary Ec50 127.5 105.1 Control Type Count Mean 95% LCL 95% UCL 96h Survival Rate EC50 127.5 105.1 Rate Detail 0 0 0.4598 0.4598 4 0.2 0 0.4598 0.459</td><td>15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosystems, CO 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 95h NOEL LOEL TOEL PMSD 96h Survival Rate 50 100 70.71 15.3% 96h Survival Rate EC50 127.5 105.1 154.6 Rate Summary Endpoint Level µg/L 95% LCL 95% UCL 96h Survival Rate EC50 127.5 105.1 154.6 Rate Summary Control Type Count Mean 95% LCL 95% UCL Min Lab Control 4 1 1 1 1 1 4 0</td></td<> <td>15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosystems, CO 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 95h V Code: 100 96h Survival Rate 50 100 70.71 15.3% 96h Survival Rate EC50 127.5 105.1 154.6 96h Survival Rate EC50 127.5 105.1 154.6 Rate Summary EC50 127.5 105.1 154.6 84 0 0 0.44 0.4 1 1 96h Survival Rate EC50 127.5 105.1 154.6 1 1ab Control 4 1 1 1</td> <td>15-8595-8712 Test Type: Survival (96h) Analysi 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) Diluend 09 Mar-15 14:35 Species: Atherinops affinis Brine: 95h Source: Aquatic Biosystems, CO Age: 11-0049-8953 Code: 150305aara Client: 05 Mar-15 Material: Copper chloride Project 05 Mar-15 Source: Reference Toxicant Froject 05 Mar-15 Source: Reference Toxicant TU 16h Station: Copper Chloride Project 96h Survival Rate 50 100 70.71 15.3% 10 96h Survival Rate 50 127.5 105.1 154.6 10 96h Survival Rate EC50 127.5 105.1 154.6 1 96h Survival Rate EC50 127.5 105.1 154.6 1 96h Survival Rate EC50 127.5 105.1 144.6 1 1 124 1 1 1 1 1 1 1</td> <td>15-8595-8712 Test Type: Survival (96h) Analyst: 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) Diluent: Dil 99 Mar-15 14:35 Species: Atherinops affinis Brine: Na 95h Source: Aquatic Biosystems, CO Age: 12 11-0049-8953 Code: 150305aara Client: In 05 Mar-15 Material: Copper chloride Project: In 05 Mar-15 Source: Reference Toxicant Project: In 05 Mar-15 Source: Reference Toxicant Project: In 16h Station: Copper Chloride V Method 96h Survival Rate 50 100 70.71 15.3% Steel M e Summary Endpoint Level µg/L 95% LCL 95% UCL TU Method 96h Survival Rate EC50 127.5 105.1 154.6 Spearm 8 Summary Ec50 127.5 105.1 154.6 Spearm 1ab Control 4 1 1 1</td> <td>Analyst: 15-8595-8712 Test Type: Survival (96h) Analyst: Diluent: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent</td> <td>15-8595-8712 Test Type: Survival (96h) Analyst: 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) Diluent: Diluent:</td>	15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-07 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosyste 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxica 16h Station: Copper Chloride 3ummary Endpoint NOEL LOEL 96h Survival Rate 50 100 96h Survival Rate EC50 127.5 Rate Summary Ecc50 127.5 Rate Summary Ecc50 127.5 Rate Summary Ecc50 127.5 Rate Summary Ecc50 127.5 Rate Summary 4 1 1 4 0.65 0.4909 4 4 0.2 0 4 4 0 0 0 4 0 0 0 4 0 0 0 4 0 <	15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosystems, CO 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 96h Survival Rate 50 100 70.71 96h Survival Rate EC50 127.5 105.1 8ate Summary Ec50 127.5 105.1 96h Survival Rate EC50 127.5 105.1 8ate Summary Ec50 127.5 105.1 Control Type Count Mean 95% LCL 95% UCL 96h Survival Rate EC50 127.5 105.1 Rate Detail 0 0 0.4598 0.4598 4 0.2 0 0.4598 0.459	15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosystems, CO 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 95h NOEL LOEL TOEL PMSD 96h Survival Rate 50 100 70.71 15.3% 96h Survival Rate EC50 127.5 105.1 154.6 Rate Summary Endpoint Level µg/L 95% LCL 95% UCL 96h Survival Rate EC50 127.5 105.1 154.6 Rate Summary Control Type Count Mean 95% LCL 95% UCL Min Lab Control 4 1 1 1 1 1 4 0	15-8595-8712 Test Type: Survival (96h) 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) 09 Mar-15 14:35 Species: Atherinops affinis 95h Source: Aquatic Biosystems, CO 11-0049-8953 Code: 150305aara 05 Mar-15 Material: Copper chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 05 Mar-15 Source: Reference Toxicant 16h Station: Copper Chloride 95h V Code: 100 96h Survival Rate 50 100 70.71 15.3% 96h Survival Rate EC50 127.5 105.1 154.6 96h Survival Rate EC50 127.5 105.1 154.6 Rate Summary EC50 127.5 105.1 154.6 84 0 0 0.44 0.4 1 1 96h Survival Rate EC50 127.5 105.1 154.6 1 1ab Control 4 1 1 1	15-8595-8712 Test Type: Survival (96h) Analysi 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) Diluend 09 Mar-15 14:35 Species: Atherinops affinis Brine: 95h Source: Aquatic Biosystems, CO Age: 11-0049-8953 Code: 150305aara Client: 05 Mar-15 Material: Copper chloride Project 05 Mar-15 Source: Reference Toxicant Froject 05 Mar-15 Source: Reference Toxicant TU 16h Station: Copper Chloride Project 96h Survival Rate 50 100 70.71 15.3% 10 96h Survival Rate 50 127.5 105.1 154.6 10 96h Survival Rate EC50 127.5 105.1 154.6 1 96h Survival Rate EC50 127.5 105.1 154.6 1 96h Survival Rate EC50 127.5 105.1 144.6 1 1 124 1 1 1 1 1 1 1	15-8595-8712 Test Type: Survival (96h) Analyst: 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) Diluent: Dil 99 Mar-15 14:35 Species: Atherinops affinis Brine: Na 95h Source: Aquatic Biosystems, CO Age: 12 11-0049-8953 Code: 150305aara Client: In 05 Mar-15 Material: Copper chloride Project: In 05 Mar-15 Source: Reference Toxicant Project: In 05 Mar-15 Source: Reference Toxicant Project: In 16h Station: Copper Chloride V Method 96h Survival Rate 50 100 70.71 15.3% Steel M e Summary Endpoint Level µg/L 95% LCL 95% UCL TU Method 96h Survival Rate EC50 127.5 105.1 154.6 Spearm 8 Summary Ec50 127.5 105.1 154.6 Spearm 1ab Control 4 1 1 1	Analyst: 15-8595-8712 Test Type: Survival (96h) Analyst: Diluent: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent:: Diluent	15-8595-8712 Test Type: Survival (96h) Analyst: 05 Mar-15 16:00 Protocol: EPA/821/R-02-012 (2002) Diluent: Diluent:

Analyst: AC QA: PP3/26/15

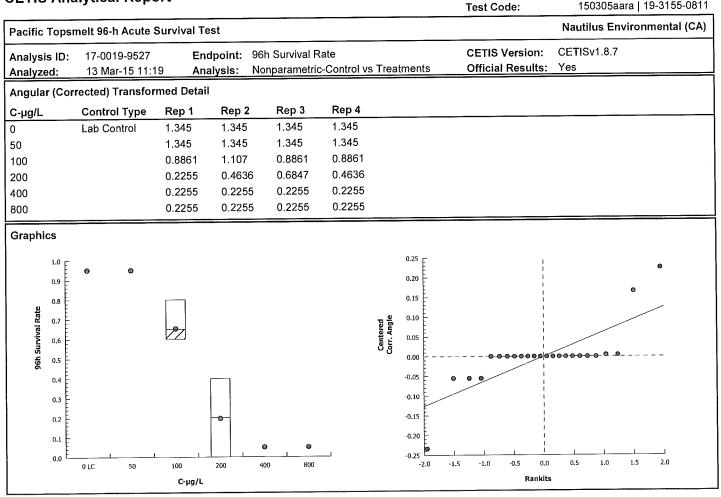
CETIS Ana	lytical Repo	ort			•	ort Date: Code:		ar-15 11:19)5aara 19-			
Pacific Topsn	nelt 96-h Acute S	Survival Te	st						Nautilus	Environm	ental (CA)
Analysis ID: Analyzed:	17-0019-9527 13 Mar-15 11:1		lpoint: 96h Ilysis: Non	Survival Ra parametric-		Freatments		IS Version: ial Results:	CETISv1. Yes	8.7	
Data Transfor	rm	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU
Angular (Corre	ected)	NA	C > T	NA	NA		15.3%	50	100	70.71	
Steel Many-O	ne Rank Sum Te	est									
Control	vs C-µg/L		Test Stat	Critical	Ties Dł	P-Value	Р-Туре	Decision(
Lab Control	50		18	10	16	0.7500	Asymp	-	icant Effect		
	100*		10	10	0 6	0.0276	Asymp	Significant			
	200*		10	10	0 6	0.0276	Asymp	Significant	Effect		
ANOVA Table	9										
Source	Sum Squ	ares	Mean Squ	are	DF	F Stat	P-Value	Decision(
Between	2.128261		0.7094204		3	59.88	<0.0001	Significant	t Effect		
Error	0.142162	2	0.0118468	5	12						
Total	2.270423				15						
Distributiona	I Tests										
Attribute	Test			Test Stat	Critical	P-Value	Decision				
Variances			y of Variance		5.953	0.2391	Equal Va				
Variances		Equality of V		2.818	5.953	0.0841	Equal Va				
Distribution	Shapiro-	Wilk W No	mality	0.7731	0.8408	0.0012	Non-norr	nal Distributio			
96h Survival	Rate Summary									• • •	0/ 555 4
C-µg/L	Control Type	Count	Mean	95% LCL	95% UCL	. Median	Min	Max	Std Err	CV%	%Effect
0	Lab Control	4	1	1	1	1	1	1	0	0.0% 0.0%	0.0% 0.0%
50		4	1	1	1	1	1	1 0.8	0 0.05	0.0% 15.38%	35.0%
100		4	0.65	0.4909	0.8091	0.6	0.6 0	0.8 0.4	0.03	81.65%	80.0%
200		4	0.2	0	0.4598	0.2 0	0	0.4	0.00100	01.0070	100.0%
400		4	0	0 0	0 0	0	0	0	0		100.0%
800		4	0	0	0	v					
Angular (Coi	rrected) Transfo	rmed Sum	mary					M		CV%	%Effect
C-μg/L	Control Type	Count	Mean		95% UCI	N74-	Min	Max	Std Err	0.0%	0.0%
0	Lab Control	4	1.345	1.345	1.346	1.345	1.345	1.345	0 0	0.0%	0.0%
50		4	1.345	1.345	1.346	1.345	1.345	1.345 1.107	0.05527	0.0 <i>%</i> 11.74%	30.03%
100		4	0.9413	0.7655	1.117	0.8861	0.8861	0.6847	0.09377	40.82%	65.85%
200		4	0.4594	0.161	0.7578	0.4636 0.2255	0.2255 0.2255	0.8847	0.05377	0.0%	83.24%
400		4	0.2255 0.2255	0.2255 0.2255	0.2256 0.2256	0.2255	0.2255	0.2255	0	0.0%	83.24%
800		4	0.2255	0.2200	0.2200						
96h Surviva		_ ·	- -	D -	D 4						
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4						·····
0	Lab Control	1	1	1	1						
50		1	1	1	1						
100		0.6	0.8	0.6	0.6						
200		0	0.2	0.4	0.2						
400		0	0	0	0						
800		0	0	0	0						

000-089-170-2

CETIS™ v1.8.7.20

CETIS Analytical	Report
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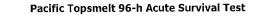
Report Date:

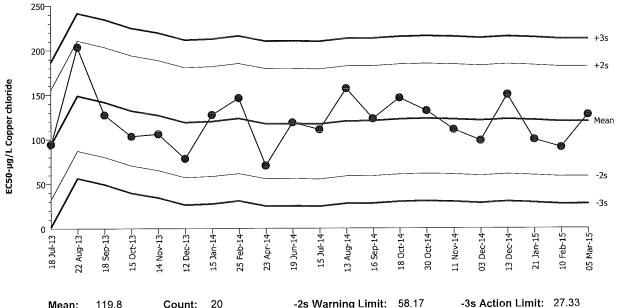


CETIS A	nalytical Re	port					-	ort Date: Code:			1:19 (p 1 of 1 19-3155-081	
Pacific Top	osmelt 96-h Acu	te Survival Te	st						Nautilus	s Enviro	nmental (CA	
Analysis II Analyzed:	D: 19-8561-473 13 Mar-15 1			h Survival F htrimmed Sp	Rate bearm a n-Kä	rber		S Version: ial Results:	CETISv1.8.7 ; Yes			
	-Kärber Estimat	es										
Threshold	Option	Threshold	Trim	Mu	Sigma		EC50	95% LCL	95% UCL			
Control Thr		0	0.00%	2,105	0.0419		127.5	105.1	154.6			
96h Surviv	al Rate Summa	ry			Calc	ulated Varia	te(A/B)					
C-µg/L	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	Α	В	
0 - pg/2	Lab Control	4	1	1	1	0	0	0.0%	0.0%	20	20	
50	202 201101	4	1	1	1	0	0	0.0%	0.0%	20	20	
100		4	0.65	0.6	0.8	0.05	0.1	15.38%	35.0%	13	20	
200		4	0.2	0	0.4	0.08165	0.1633	81.65%	80.0%	4	20	
400		4	0	0	0	0	0		100.0%	0	20	
800		4	0	0	0	0	0		100.0%	0	20	
96h Surviv	/al Rate Detail											
C-µg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4							
0	Lab Control	1	1	1	1							
50		1	1	1	1							
100		0.6	0.8	0.6	0.6							
200		0	0.2	0.4	0.2							
400		0	0	0	0							
800		0	0	0	0							
96h Survival Rate		200 400		<u>- +</u>	0							
	£	300 400 C-µg/L	, , , , , , , , , , , , , , , , , , , 	<u>, + ⊨,</u> 700 80) 00							

CETIS QC Plot

Pacific Topsmelt 96-h Acute Survival Te	st	Nautilus Enviro	onmental (CA)
Test Type: Survival (96h)	Organism: Atherinops affinis (Topsmelt)	Material: Copper chloride	
Protocol: EPA/821/R-02-012 (2002)	Endpoint: 96h Survival Rate	Source: Reference Toxicant-R	.EF





Mean:	119.8	Count:	20	-2s Warning Limit:	58.17	-3s Action Limit:	27.33
Sigma:	30.84	CV:	25.70%	+2s Warning Limit:	181.5	+3s Action Limit:	212.4

Qualit	ty Con	trol Data	a								
Point	Year	Month	Day	Time	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2013	Jul	18	12:50	94.59	-25.21	-0.8175			19-2632-6339	04-8526-8990
2		Aug	22	14:40	203.7	83.88	2.72	(+)		16-8357-2725	11-7110-5550
3		Sep	18	13:50	127.5	7.656	0.2483			09-9085-4812	11-5673-1751
4		Oct	15	15:20	103.5	-16.27	-0.5277			00-5901-5898	17-6384-6991
5		Nov	14	9:40	106.1	-13.68	-0.4437			06-5418-8921	10-2371-6330
6		Dec	12	13:20	78.46	-41.34	-1.341			12-4998-2305	03-2148-1441
7	2014	Jan	15	15:25	127.5	7.656	0.2483			13-3854-5258	05-1070-1044
8		Feb	25	13:55	146.4	26.61	0.8628			20-0325-5939	07-6658-0335
9		Apr	23	16:00	70.71	-49.09	-1.592			11-8272-9093	14-5541-7971
10		Jun	19	15:35	118.9	-0.8793	-0.02851			11-2944-5183	19-5384-3170
11		Jul	15	14:15	111	-8.843	-0.2867			00-8730-8108	10-9428-5566
12		Aug	13	14:10	156.9	37.12	1.204			12-9208-7415	02-1974-5349
13		Sep	16	13:10	123.1	3.314	0.1075			05-7478-8365	11-8140-9628
14		Oct	18	10:45	146.4	26.61	0.8628			18-6908-7115	01-0100-8379
15			30	11:50	132	12.15	0.394			17-2734-9303	12-4790-6162
16		Nov	11	10:50	111	-8.843	-0.2867			19-6246-9477	14-2586-1124
17		Dec	3	16:45	98.53	-21.27	-0.6897			07-3639-8754	16-6449-6521
18			13	15:30	150.1	30.28	0.9818			03-6652-6590	12-5127-9321
19	2015	Jan	21	15:35	99.71	-20.09	-0.6514			16-8270-8063	13-6137-3732
20		Feb	10	14:45	91.04	-28.76	-0.9325			05-4867-3928	21-3305-5409
21		Mar	5	16:00	127.5	7.656	0.2483			19-3155-0811	19-8561-4737

Marine Acute Bioassay Static-Renewal Conditions

Water Quality Measurements & Test Organism Survival

Client: Internal						Test Species: A. affinis													Tech Initials							
Sample ID: CuCl ₂						-	Start Date/Time: 3/5/2015 \600									-			0	24	48	72	96			
Test No.: 150305aara						-	End Date/Time: <u>3/9/2015</u> \435								 Counts:			CH	15	AD	AB	Eh				
restrio <u>roosoodaru</u>							-						·····	1.0/			Readings:				4	Polas		AG		
																			Dilutio					AB	-	-
																			conc.				1	200		
																	١		stock				1	4.6	•	••
									Cu st	ock co	oncenti	ration	(µg/L):	86	,90	0			Final V				1	2000		
																	•									
0					of Live	•		;	Salinity Temperature							Dissolved Oxygen						рН				
Concentration (µg/L)	Rand #	Organisms						(ppt)	I	1		T . Wildowski	<u>୍ଟ</u> ୍ରେ	Powersterner 1884	Personala		1	(mg/L) 	La companya da la		(units	 		
		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
Lab Control	22	5	5	5	5	5	30,0	30,3	20.9	30:7	30:7	19.8	юя	9.2	19,7	19.7	7.2	6.5	12	10	69	7,45	7,85	803	7.89	7,88
	2	5	5	5	5	5	0.000		30.7					20.9					6.8					r 7.83		
	10	5	5	5	5	5						ļ														
	15	5	2	5	5	115	Eb 310																			
50	24	5	5	5	5	5	24,9	10-10-10-10-10-10-10-10-10-10-10-10-10-1	1.	30.4	30.4	19.9	21.0	1.	201	19.8	7,0	6.4	72	69	6.8	7.97	7.85	804	7.93	7,40
	8	5	5	2	5	5			30.6					みる					6.7					7.86		
	17	5	5	5	5	5						ļ														
	11	5		5	5	5	240		1.					110.					11.0					1		
100	12	5	3	3	3	3	244		F	30.5	30.7	<u>19.5</u>	20.9		19.9	19.4	1.0	6.4	F	-71	7.0	7.97			7.95	7.94
	4	5	4	4	4	4			30.7					20.9					6.8					7.92		
	1	5	H II	3	3	3	-											-								
	19 23	5	4	2	ļ	0	049		600			10.4		in a					100			700		1.00		-
200	23 13	5 5	み 5	2	3		24.1	130,5	f	30.5	30.8	11.6		f	1951	195	1,0	1 %.4	7.2 1		7.((,५ (7,85	-	7.96	1.95
	14	5 5	14	4	4	2			30.9					20.9					6.3					793		
	18	5	1		1	1																				
400	9	5	0	<u> `</u>		÷	296	n 9 9	1			19/	20.6	i			7.0	6.6	i			7.96	7 86	I		
400	16	5	ň			\nearrow		61.0	f		\leftarrow	110	2018	f	/				f		/	.,		f	A	
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APPENDIX C

Glossary of Qualifier Codes



Glossary of Qualifier Codes:

Laboratory Procedures

- Q1 Temperatures out of recommended range; corrective action taken and recorded in Test Temperature Correction Log
- Q2 Temperatures out of recommended range; no action taken, test terminated same day
- Q3 Sample aerated prior to initiation or renewal due to dissolved oxygen (D.O.) levels below 6.0 mg/L
- Q4 Test aerated; D.O. levels dropped below 4.0 mg/L
- Q5 Test initiated with aeration due to an anticipated drop in D.O.
- Q6 Airline obstructed or fell out of replicate and replaced; drop in D.O. occurred
- Q7 Salinity out of recommended range
- Q8 Spilled test chamber/ Unable to recover test organism(s)
- Q9 Inadequate sample volume remaining, 50% renewal performed
- Q10 Inadequate sample volume remaining, no renewal performed
- Q11 Sample out of holding time; refer to QA section of report
- Q12 Replicate(s) not initiated; excluded from data analysis
- Q13 Survival counts not recorded due to poor visibility or heavy debris
- Q14 D.O. percent saturation was checked and was $\leq 110\%$

Data Analysis/Reporting

- 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.

Error Correction

- Q18 Incorrect Entry
- Q19 Illegible Entry
- Q20 Miscalculation
- Q21 Other (provide reason in comments section)