



Toxicity testing on weathered groundwater diesel

Environmental Assessment Program (EAP)

Final Report

September 4, 2019
[Revised January 17, 2020]

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SIGNATURE PAGE



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This report has been prepared by Nautilus Environmental Company Inc. based on data and/or samples provided by our client and the results of this study are for their sole benefit. Any reliance on the data by a third party is at the sole and exclusive risk of that party. The results presented here relate only to the samples tested.

1.0 INTRODUCTION

The Washington State Department of Ecology (Ecology) Environmental Assessment Program (EAP) requested Nautilus Environmental to characterize the toxicity of weathered diesel to freshwater and marine organisms. Currently, there are few aquatic toxicity studies on weathered diesel in the literature. This project was conducted to obtain toxicity information on weathered diesel for developing water quality objectives. Specifically, the goal of the project was to estimate the no-observed effects levels (NOEL) and low-observed effects levels (LOEL) of weathered diesel in contaminated groundwater on freshwater and marine organisms. These effects levels will be directly applicable to WET testing that is carried out under the Model Toxics Control Act (MTCA) as it pertains to the cleanup and investigation of hydrocarbon-contaminated sites by Ecology's Toxics Cleanup Program (TCP). TCP will use the NOEL values to establish a screening level for hydrocarbon contamination in fresh and marine waters. Previous studies had been conducted on gasoline and diesel for this purpose.

Tests were conducted on diesel contaminated groundwater DW-3, that was collected from a site in Washington State. The contaminated groundwater was analysed to determine the concentration of hydrocarbons in the diesel range and was also tested for metals, TDS, and other constituents that might affect toxicity to aquatic organisms. Samples of the groundwater were shipped by EAP to Nautilus for toxicity testing. The following toxicity tests were performed on the groundwater sample:

- Topsmelt survival and growth (EPA/600/R-95/136)
- Echinoderm fertilization (EPA/600/R-95/136)
- Fathead minnow survival and growth (EPA-821-R-02_013)
- *Ceriodaphnia dubia* survival and reproduction (EPA-821-R-02-013)

The results of these toxicity tests are presented in this report. Copies of laboratory data sheets and printouts of statistical analyses are provided in Appendices A to D; analytical chemistry is provided in Appendix E; and the chain-of-custody form is provided in Appendix F.

2.0 METHODS

Methods for the toxicity tests are summarized in Tables 1 to 4. Testing was conducted according to procedures described by the US EPA (1995, 2002) and the Washington Department of Ecology (WDOE, 2016).

Preliminary toxicity tests using *C. dubia* were conducted in December 2018 with uncontaminated groundwater to ensure that it was non-toxic to the study organisms. This was done to ensure that any observable effects could be attributed to the weathered diesel contamination. Sample MW-11 was chosen as the site control for further testing based on the results of these tests; results of these tests are presented in a separate report.

The groundwater DW-3 used for the range finding tests was collected January 29, 2019 and received on January 30, 2019. Subsamples of 100% and 50% strength dilutions were sent to Ecology's Manchester Environmental Laboratory for initial analysis of groundwater diesel prior to testing to ensure the diesel concentrations were within detection limits.

To determine an appropriate concentration series that would capture NOEC and LOEC values, range-finding tests were performed for each test species prior to initiation of the definitive tests. Subsamples of the test solutions were taken at various points of the tests to determine the concentration and stability of weathered diesel in the toxicity tests. Samples were shipped to Manchester Environmental Laboratory for analysis.

Additional groundwater sample and site control was collected March 19, received on March 20, 2019 and was homogenized with the samples from January. Subsamples were collected and sent for re-analysis of diesel prior to definitive testing to ensure concentrations were similar to those used in the range-finding tests. Measured concentrations of the refresh sample were lower than the initial measurements though were comparable.

Definitive tests using topsmelt and fathead minnow were initiated on April 23, 2019; the echinoderm test was initiated May 1, 2019; and the definitive test using *C. dubia* was initiated on May 14, 2019. Subsamples of test solutions were once again taken at various time points and sent to Manchester Environmental Laboratory for analysis. The average measured concentration of DW-3 for the duration of the range-finding tests was 5.95 mg/L diesel; the average measured concentration for the definitive tests was 4.80 mg/L diesel. Statistical analyses were performed using the site control (MW-11) as the negative control with CETIS (Tidepool Scientific Software, 2013).

Table 1. Summary of test conditions: 7-d topsmelt (*Atherinops affinis*) survival and growth test.

Test species	<i>Atherinops affinis</i>
Organism source	Commercial supplier
Organism age	9-to 15-days post-hatch
Test type	Static-renewal
Test duration	7 days
Test vessel	500-mL glass container with lid
Test volume	500 mL
Test concentrations	Six concentrations, plus laboratory and methanol control
Test replicates	5 per treatment
Number of organisms	5 per replicate
Control/dilution water	Natural seawater
Test solution renewal	Daily (80% renewal)
Test temperature	20 ± 1°C
Test salinity	30 ± 2 ppt; sample salinity adjusted by addition of H ₂ Ocean Pro+ marine salts
Feeding	Twice a day with newly hatched brine shrimp nauplii (<i>Artemia</i> sp.)
Light intensity	Ambient laboratory lighting
Photoperiod	16 hours light / 8 hours dark
Aeration	None, unless dissolved oxygen falls below 4.0 mg/L
Test measurements	Temperature, dissolved oxygen, pH and salinity measured daily; survival checked daily
Test protocol	EPA/600/R-95/136
Statistical software	CETIS Version 1.9.4
Test endpoints	Survival and biomass
Test acceptability criteria for controls	≥80% survival; ≥0.85 mg mean dry weight
Reference toxicant	Copper (added as CuCl ₂)

Table 2. Summary of test conditions: 40-min echinoderm fertilization test.

Test species	<i>Strongylocentrotus purpuratus</i>
Organism source	Commercial supplier
Organism age	< 3 hours post gamete collection
Test type	Static
Test duration	10 minutes sperm exposure; 10 minutes egg fertilization
Test vessel	30-mL glass vials with snap cap
Test volume	30 mL
Test concentrations	Six concentrations, plus laboratory and methanol control
Test replicates	4 per treatment
Number of organisms	2000 eggs per replicate
Control/dilution water	Natural seawater
Test solution renewal	None
Test temperature	12 ± 1°C
Test salinity	30 ± 2 ppt; sample salinity adjusted by addition of H ₂ Ocean Pro+ marine salts
Feeding	None
Light intensity	Ambient laboratory lighting
Photoperiod	None
Aeration	None
Test measurements	Temperature, dissolved oxygen, pH and salinity measured at test initiation
Test protocol	EPA/600/R-95/136
Statistical software	CETIS Version 1.9.4
Test endpoint	Fertilization
Test acceptability criterion for controls	≥60% and <98% mean fertilization
Reference toxicant	Copper (added as CuCl ₂)

Table 3. Summary of test conditions: 7-d fathead minnow (*Pimephales promelas*) survival and growth test.

Test species	<i>Pimephales promelas</i>
Organism source	Commercial supplier
Organism age	<24 hours post-hatch
Test type	Static-renewal
Test duration	7 days
Test vessel	375-mL glass container with lid
Test volume	375 mL
Test concentrations	Six concentrations, plus laboratory and methanol control
Test replicates	4 per treatment
Number of organisms	10 per replicate
Control/dilution water	Moderately-hard reconstituted water
Test solution renewal	Daily (80% renewal)
Test temperature	25 ± 1°C
Feeding	Twice a day with approximately 1500-2250 newly hatched brine shrimp nauplii (<i>Artemia sp.</i>) in each test container
Light intensity	100 to 500 lux
Photoperiod	16 hours light / 8 hours dark
Aeration	None, unless dissolved oxygen falls to <40% saturation
Test measurements	Temperature, dissolved oxygen, pH and conductivity measured daily; hardness and alkalinity of undiluted sample measured at test initiation; survival checked daily
Test protocol	EPA-821-R-02_013
Statistical software	CETIS Version 1.9.4
Test endpoints	Survival and biomass
Test acceptability criteria for controls	≥80% survival; ≥250 µg mean dry weight
Reference toxicant	Sodium chloride (NaCl)

Table 4. Summary of test conditions: *Ceriodaphnia dubia* survival and reproduction test.

Test species	<i>Ceriodaphnia dubia</i>
Organism source	In-house culture
Organism age	<24 hour old neonates, produced within a 8 hour window
Test type	Static-renewal
Test duration	7 ± 1 day
Test vessel	20-mL glass test tube with snap cap
Test volume	20 mL
Test concentrations	Six concentrations, plus laboratory and methanol control
Test replicates	10 per treatment
Number of organisms	1 per replicate
Control/dilution water	20% Perrier water and 80% deionized water + 5 µg/L Se and 2 µg/L vitamin B12
Test solution renewal	Daily (100% renewal)
Test temperature	25 ± 1°C
Feeding	Daily with <i>Pseudokirchneriella subcapitata</i> and YCT (3:1 ratio)
Light intensity	100 to 600 lux at water surface
Photoperiod	16 hours light / 8 hours dark
Aeration	None
Test measurements	Temperature, dissolved oxygen, pH and conductivity measured daily; hardness and alkalinity of undiluted sample measured at test initiation; survival and reproduction checked daily
Test protocol	EPA-821-R-02-013
Statistical software	CETIS Version 1.9.4
Test endpoints	Survival and reproduction ≥80% survival; ≥15 young per surviving control producing three broods; ≥60% of controls producing three or more broods; no ephippia present
Test acceptability criteria for controls	
Reference toxicant	Sodium chloride (NaCl)

2.1 Subsampling Test Solutions

Subsamples of the fresh test solutions were taken during the filling of the test chambers. Subsamples of the stale test vessel solutions were collected as a composite of the test vessel replicates prior to renewal or at the end of the tests.

Test solutions were renewed daily for tests with topsmelt, fathead minnow and *C. dubia*; the 40 minute exposure echinoderm test did not require renewal of test solution. The site control MW-11 was used to calculate all test endpoints.

3.0 RESULTS

Samples of the fresh and stale test solutions were analyzed by the Manchester Environmental Laboratory. The nominal and measured concentrations for the range-finding tests are presented in Tables 5 to 7. The nominal and measured concentrations for the definitive tests are presented in Tables 8 to 10. Only the 100%, 6.3%, 0.39%, and lab (0%) and site control samples were analysed. Mean measured concentrations used for statistical analysis were calculated by plotting the measured concentrations as the dependent variable (y axis) vs. the nominal concentrations as the independent variable (x axis) for all time points in Microsoft Excel and then fitting a regression line to the data. Some data points were judged to be outliers and were not included in the regression analyses. The lab control was used in the regression lines as it was part of the test dilution series and the site control was not. The lab control concentrations were at or below detection limits. The reported analytical values were used for the lab control. The equation of the best fit regression line was then used to calculate the mean measured concentration at each time point and concentration. In all cases, an order 2 polynomial equation fit the plotted data best. The plots used to calculate the mean measured concentrations are shown in Figures 1 to 4. The calculated or mean measured concentrations are presented in Table 8.

Table 5. Results: Topsmelt and echinoderm nominal and measured test concentrations (mg diesel/L) (Range-finder).

Nominal Conc. (% v/v)	Measured Concentration (mg diesel/L)								
	t=0	t=24		t=48	t=72	t=96	t=120	t=144	t=168
	fresh	stale	fresh	fresh	fresh	fresh	fresh	fresh	stale
100	6.1	5.62	5.26	5.88	3.96	5.26	4.15	3.86	4.4
100 dupl.	6.08	--	--	--	--	--	--	--	--
25	--	--	--	--	--	--	--	--	--
6.25	0.38	0.2	0.3	0.31	0.22	0.34	0.36	0.16	0.25
6.25 dupl.	0.26	--	0.25	-	0.21	0.35	--	0.15	--
1.56	--	--	--	--	--	--	--	--	--
0.39	0.19	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.14
0.10	--	--	--	--	--	--	--	--	--
Site Control	0.31	--	--	--	--	--	--	0.26	--
Lab Control	0.15	--	--	--	--	--	--	0.15	--

t = time, Detection Limit = 0.15

Figure 1. Polynomial Regression of Test Solution Concentrations vs. Measured Concentrations – Topsmelt Range-finding Test

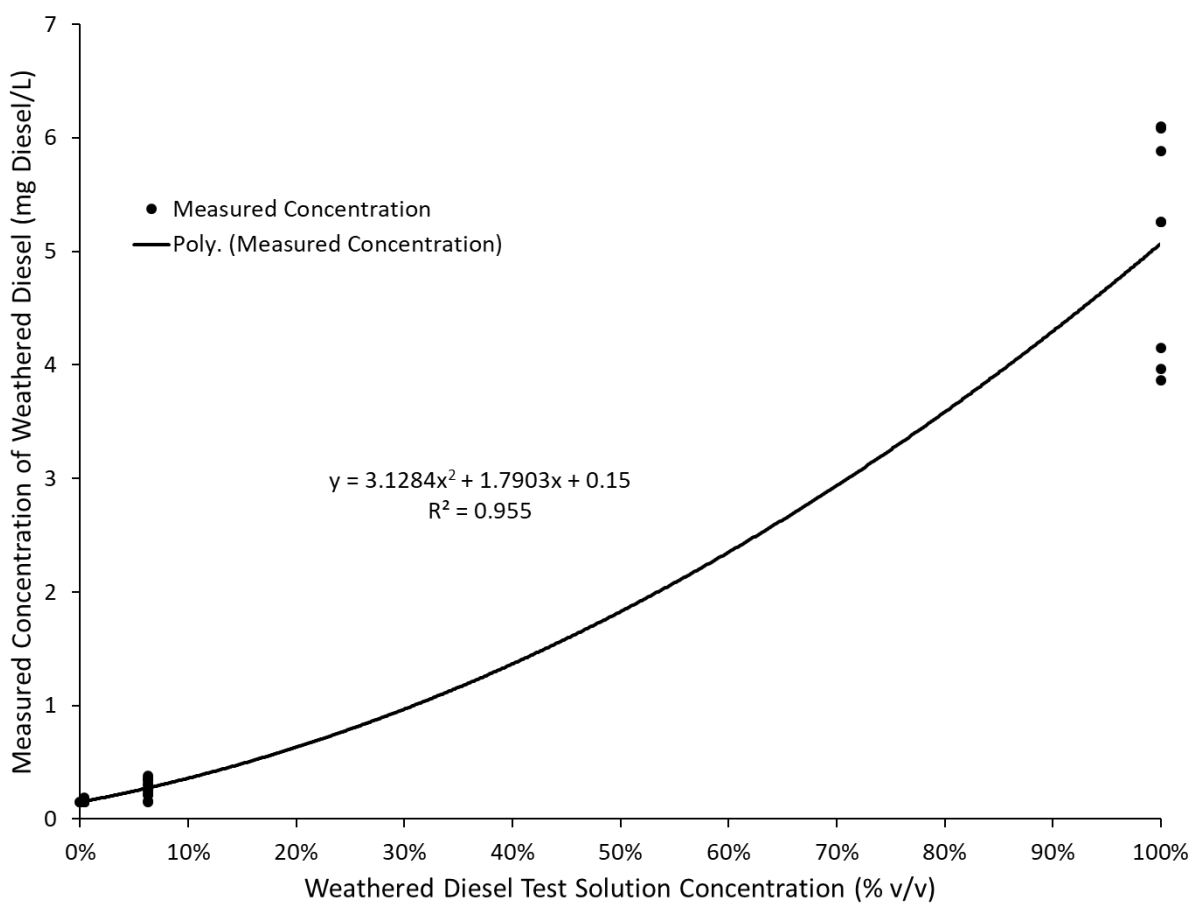


Table 6. Results: Fathead minnow nominal and measured test concentrations (mg diesel/L) (Range-finder).

Nominal Conc. (% v/v)	Measured Concentration (mg diesel/L)									
	t=0	t=24		t=48	t=72	t=96	t=120	t=144	t=168	
	fresh	stale	fresh	fresh	fresh	fresh	fresh	fresh	stale	fresh
100	6.16	5.39	6.33	6.35	7.04	0.32	4.8	6.63	5.7	6.69
100 dupl.	6.27	--	--	--	--	--	--	--	--	6.85
25	--	--	--	--	--	--	--	--	--	--
6.25	0.39	0.35	0.29	0.39	0.26	0.15	0.33	0.28	0.3	--
6.25 dupl.	0.29	--	0.3	-	0.4	0.15	--	0.28	--	--
1.56	--	--	--	--	--	--	--	--	--	--
0.39	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	--
0.10	--	--	--	--	--	--	--	--	--	--
Site Control	0.15	--	--	--	--	--	--	0.25	--	--
Lab Control	0.15	--	--	--	--	--	--	0.16	--	--

t = time, Detection Limit = 0.15

Figure 2. Polynomial Regression of Test Solution Concentrations vs. Measured Concentrations – Fathead Minnow Range-finding Test

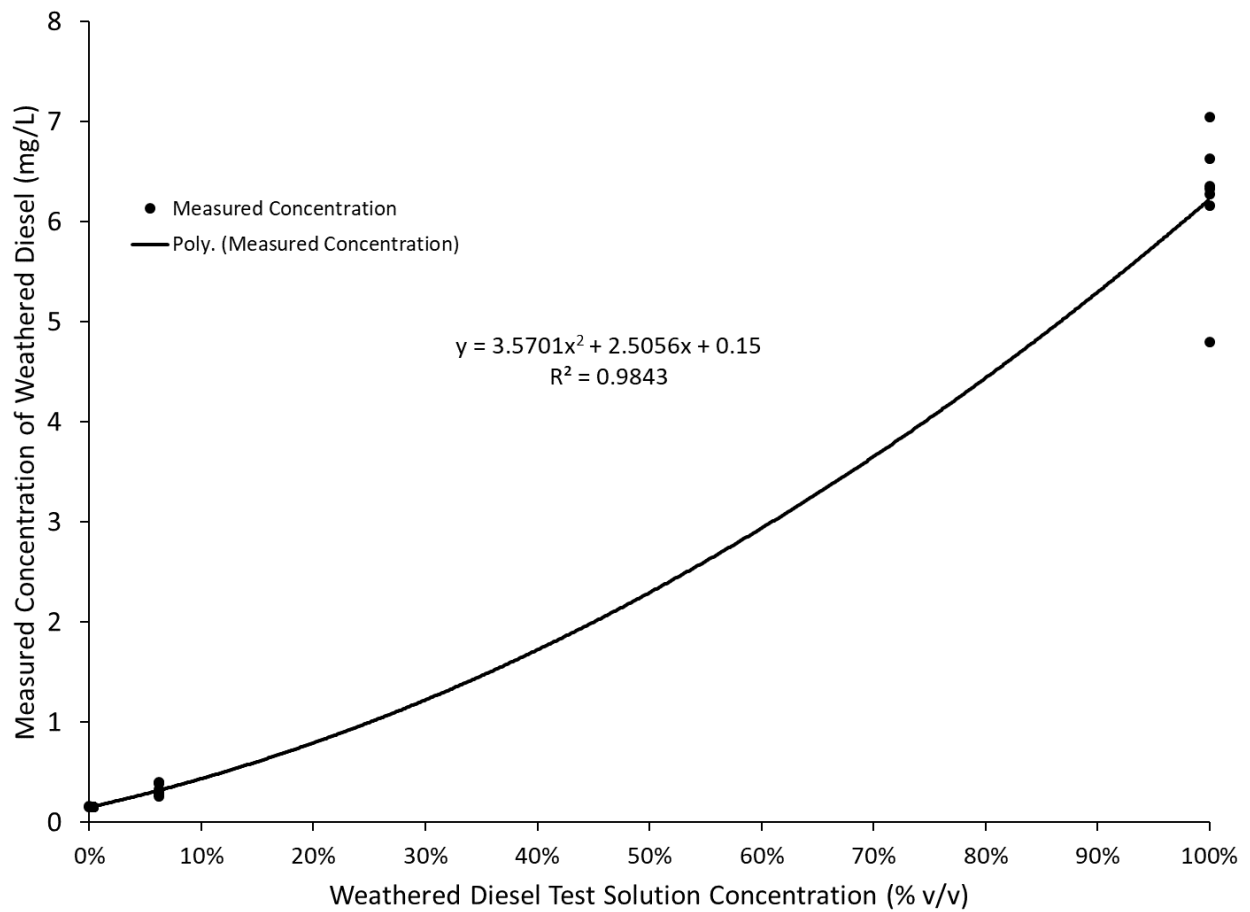


Table 7. Results: *Ceriodaphnia dubia* nominal and measured test concentrations (mg diesel/L) (Range-finder).

Nominal Conc. (% v/v)	Measured Concentration (mg diesel/L)						
	t = 0	t=24	t=48	t=72	t = 96	t = 120	t=144
	fresh	fresh	fresh	fresh	fresh	fresh	fresh
100	6.16	6.29	5.53	5.48	3.75	5.24	--
100 dupl.	6.26	--	--	--	--	--	--
25	--	--	--	--	--	--	--
6.25	0.32	0.34	0.27	0.28	0.25	0.30	--
6.25 dupl.	0.42	0.28	--	0.27	0.24	--	--
1.56	-	-	-	-	-	-	-
0.39	0.15	0.16	0.15	0.17	0.17	0.17	-
0.10	--	--	--	--	--	--	--
Site Control	0.28	--	--	--	--	--	0.23
Lab Control	0.22	--	--	--	--	--	0.16

t = time, Detection Limit = 0.16

Figure 3. Polynomial Regression of Test Solution Concentrations vs. Measured Concentrations – *C. dubia* Range-finding Test

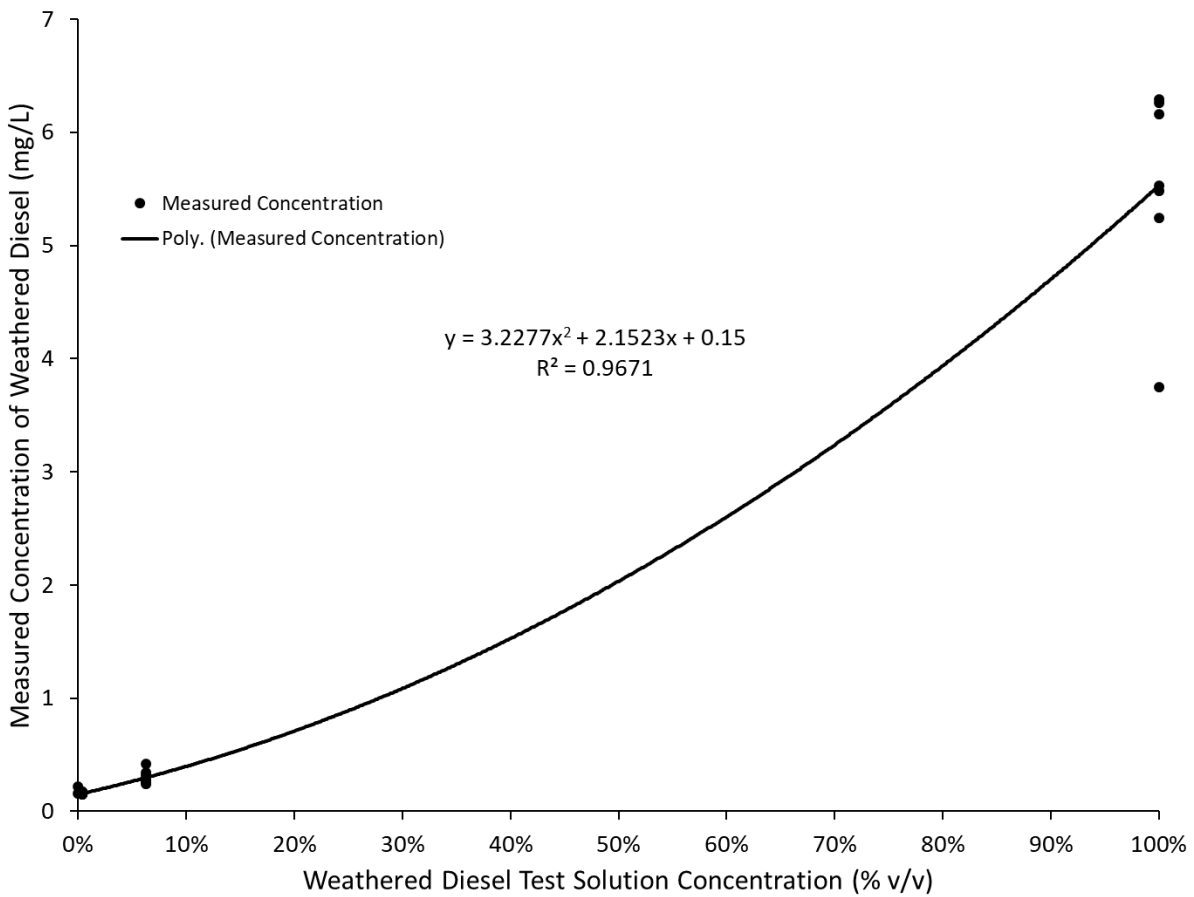


Table 8. Results: Topsmelt and echinoderm nominal and measured test concentrations (mg diesel/L) (Definitive).

Nominal Conc. (% v/v)	Measured Concentration (mg diesel/L)							
	t=0	t=24	t=48	t=72	t=96	t=120	t=144	t=168
	fresh	fresh	fresh	fresh	fresh	fresh	fresh	stale
100	3.94	2.74	3.06	3.19	4.43	2.97	2.66	2.86
100 dupl.	2.84	--	3.22	--	2.76	--	2.69	--
75	--	--	--	--	--	--	--	--
50	1.41	0.43	1.43	1.35	1.23	1.13	0.42	0.14
50 dupl.	--	--	--	--	0.27	--	1.2	--
25	0.67	0.62	0.65	0.62	0.55	0.58	0.64	0.51
12.5	--	--	--	--	--	--	--	--
Site Control	0.22	--	--	--	--	--	0.17	--
Lab Control	0.17	--	--	--	--	--	0.15	--

t = time

Table 9. Results: Echinoderm nominal and measured test concentrations (mg diesel/L) (Definitive).

Nominal Conc. (% v/v)	Measured Concentration (mg diesel/L)	Mean Value
	t=0	t=0
	fresh	fresh
100	2.83	2.71
100 dupl.	2.59	-
Site Control	0.28	0.22
Site Control dupl.	0.16	-
Lab Control dupl.	0.16	0.16
Lab Control	0.15	-

t = time

Figure 4. Polynomial Regression of Test Solution Concentrations vs. Measured Concentrations – Topsmelt Definitive Test

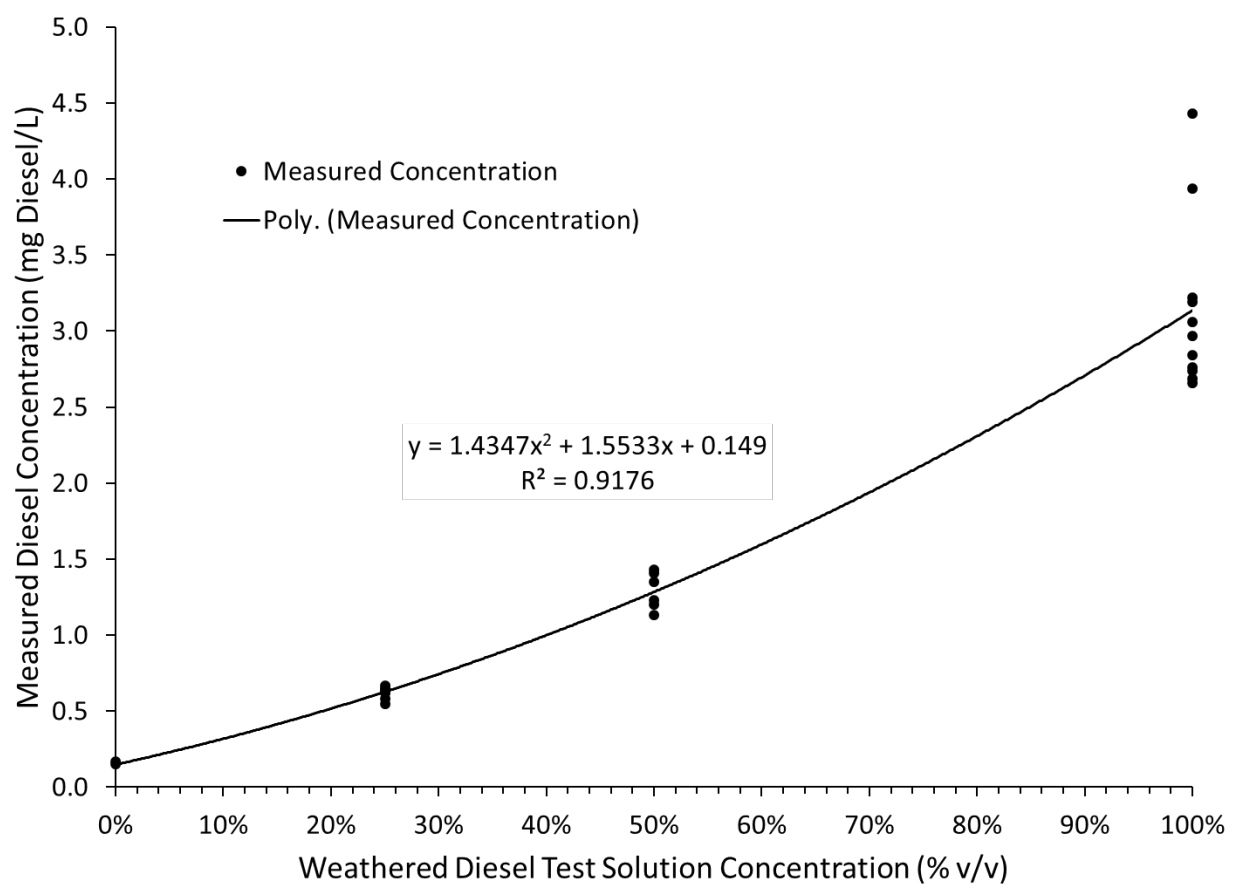


Table 10. Results: Fathead minnow nominal and measured test concentrations (mg diesel/L) (Definitive).

Nominal Conc. (% v/v)	Measured Concentration (mg diesel/L)							
	t=0	t=24	t=48	t=72	t=96	t=120	t=144	t=168
	fresh	fresh	fresh	fresh	fresh	fresh	fresh	stale
100	5.39	0.22	3.76	3.93	4	3.11	3.98	3.56
100 dupl.	5.83	--	3.98	--	4.12	--	4.02	--
75	--	--	--	--	--	--	--	--
50	2.28	0.19	1.8	1.97	1.82	2.42	1.89	1.72
50 dupl.	--	--	--	--	1.76	--	1.75	--
25	0.94	0.81	0.93	0.86	0.83	0.95	0.89	0.81
12.5	--	--	--	--	--	--	--	--
Site Control	0.2	--	--	--	--	--	0.21	--
Lab Control	0.17	--	--	--	--	--	0.17	--

t = time

Figure 5. Polynomial Regression of Test Solution Concentrations vs. Measured Concentrations – Fathead Minnow Definitive Test

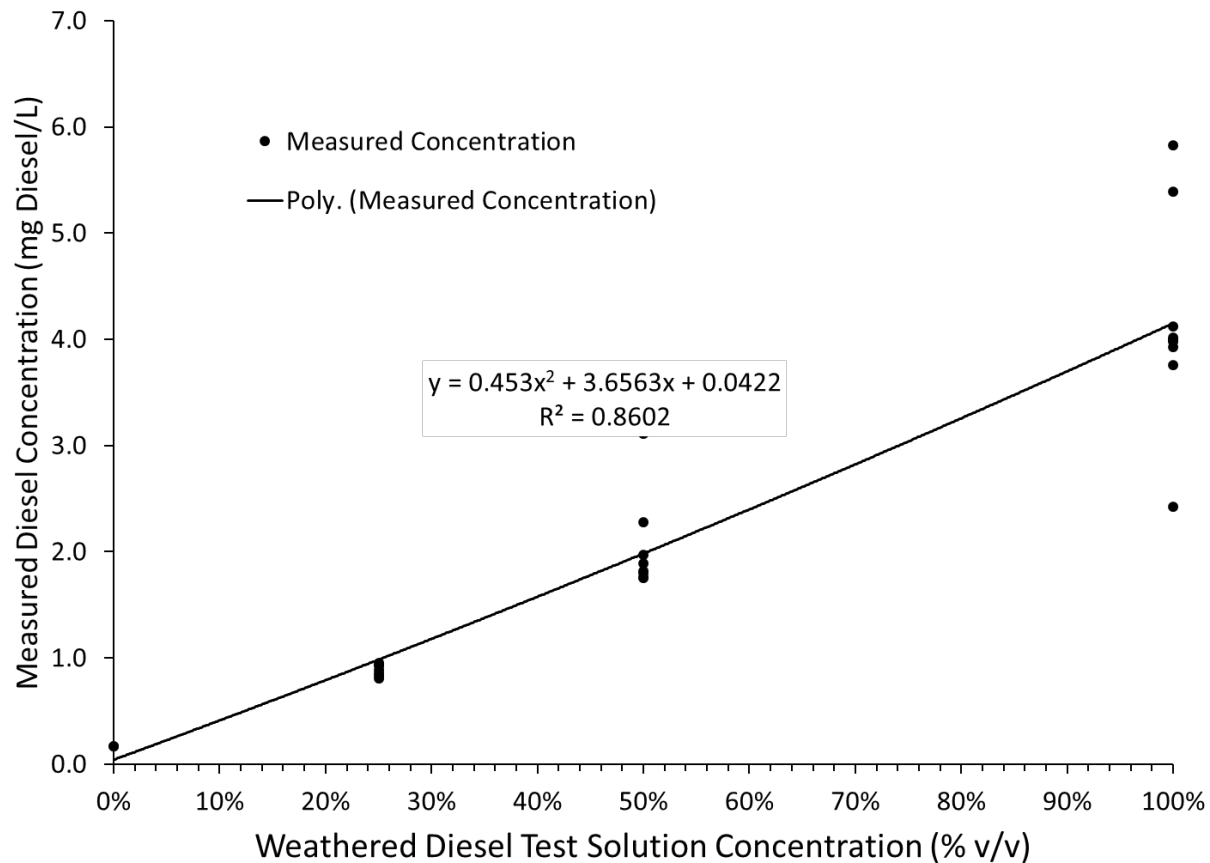


Table 11. Results: *Ceriodaphnia dubia* nominal and measured test concentrations (mg diesel/L) (Definitive).

Nominal Conc. (% v/v)	Measured Concentration (mg diesel/L)					
	t = 0	t=24	t=48	t=72	t = 96	t = 120
	fresh	fresh	fresh	fresh	fresh	fresh
100	3.76	4.02	4.13	4.24	4.02	--
100 dupl.	4.38	--	4.24	--	4.13	--
Site Control	0.22	--	--	--	--	0.28
Lab Control	0.17	--	--	--	--	0.16

t = time

3.1 Range-finding Toxicity Test Results

The results of the range-finding tests conducted in February 2019 are summarized in Tables 11 to 14. Results were compared against the laboratory control. Results of these tests were used to determine concentrations to use for the definitive tests.

Table 12. Results: Topsmelt 7-day survival and growth test (Range-finder).

Weathered Diesel Concentrations		Survival (%) (Mean ± SD)	Biomass (mg) (Mean ± SD)	Dry Weight (mg) (Mean ± SD)
Nominal (% v/v)	Mean Measured (mg diesel/L)			
Laboratory control	0.15 (DL)	100 ± 0.0	1.18 ± 0.02	1.18 ± 0.02
MW-11 (Site Control)	0.29	86.7 ± 11.6	1.04 ± 0.31	1.18 ± 0.19
0.10	<i>0.15</i>	93.3 ± 11.6	1.08 ± 0.18	1.16 ± 0.10
0.39	0.16	93.3 ± 11.6	1.16 ± 0.10	1.25 ± 0.05
1.6	<i>0.18</i>	93.3 ± 11.6	1.16 ± 0.13	1.25 ± 0.17
6.3	0.27	100 ± 0.0	1.25 ± 0.17	1.25 ± 0.17
25	<i>0.79</i>	93.3 ± 11.6	1.06 ± 0.14	1.16 ± 0.30
100	5.07	20.0 ± 20.0	0.13 ± 0.13	0.62 ± 0.07
Test endpoint (mg diesel/L) *				
LC50 (95% CL)		2.4 (1.7 – 3.3)	--	--
IC25 (95% CL)		--	1.3 (0.5 – 1.8)	2.1 (N/A – 2.9)
IC50 (95% CL)		--	2.3 (1.4 – 3.2)	>5.1 (N/A)_
NOEC		0.8	0.8	0.8
LOEC		5.1	5.1	5.1

SD = Standard Deviation, LC = Lethal Concentration, IC = Inhibition Concentration, CL = Confidence Limits, NOEC = No Observed Effect Concentration, LOEC = Lowest Observed Effect Concentration, DL = Detection Limit (0.15 mg Diesel/L), N/A = Not Available

* = result was calculated using the laboratory control as the negative control

Calculated concentrations are represented in italics.

Table 13. Results: Echinoderm (*Strongylocentrotus purpuratus*) fertilization test (Range-finder)

Weathered Diesel Concentrations		Fertilized eggs (%) (Mean ± SD)
Nominal (% v/v)	Mean Measured (mg diesel/L)	
Laboratory control	0.15 (DL)	86.0 ± 3.8
Salt-adjusted control	-	91.2 ± 3.0
MW-11 (Site Control)	0.31	90.8 ± 2.5
0.1	--	87.2 ± 7.3
0.39	0.19	94.0 ± 3.2
1.6	--	91.2 ± 1.9
6.3	0.32	89.0 ± 2.2
25	--	90.0 ± 3.6
100	6.1	86.0 ± 3.4
Test endpoint (mg diesel/L) *		
	IC25	>6.1
	IC50	>6.1
	NOEC	6.1
	LOEC	>6.1

SD = Standard Deviation, IC = Inhibition Concentration, NOEC = No Observed Effect Concentration, LOEC = Lowest Observed Effect Concentration, DL = Detection Limit (0.15 mg Diesel/L)

(*) = Result was calculated using nominal values and the laboratory control as the negative control

Table 14. Results: Fathead minnow 7-day survival and growth test (Range-finder).

Weathered Diesel Concentrations		Survival (%) (Mean \pm SD)	Biomass (mg) (Mean \pm SD)	Dry Weight (mg) (Mean \pm SD)
Nominal (% v/v)	Mean Measured (mg diesel/L)			
Laboratory control	0.16	100 \pm 0.0	0.69 \pm 0.14	0.69 \pm 0.14
MW-11 (Site Control)	0.20	96.7 \pm 5.8	0.80 \pm 0.12	0.82 \pm 0.09
0.10	<i>0.15</i>	93.3 \pm 5.8	0.63 \pm 0.06	0.67 \pm 0.06
0.39	0.15	100 \pm 0.0	0.75 \pm 0.03	0.75 \pm 0.03
1.6	<i>0.19</i>	100 \pm 0.0	0.75 \pm 0.04	0.75 \pm 0.04
6.3	0.32	93.3 \pm 11.6	0.66 \pm 0.19	0.70 \pm 0.13
25	<i>1.00</i>	96.7 \pm 5.8	0.69 \pm 0.03	0.71 \pm 0.01
100	6.23	86.7 \pm 15.3	0.35 \pm 0.13	0.40 \pm 0.08
Test endpoint (mg diesel /L) *				
LC50 (95% CL)		>6.28	--	--
IC25 (95% CL)		--	2.6 (N/A – 5.4)	3.1 (0.4– 4.9)
IC50 (95% CL)		--	>6.2	>6.2
NOEC		6.28	1.0	0.65
LOEC		>6.28	6.28	1.30

SD = Standard Deviation, LC = Lethal Concentration, IC = Inhibition Concentration, CL = Confidence Limits, N/A = Not Available, NOEC = No Observed Effect Concentration, LOEC = Lowest Observed Effect Concentration, DL = Detection Limit (0.15 mg Diesel/L)

* = result was calculated using the laboratory control as the negative control

Calculated concentrations are represented in italics.

Table 15. Results: *C. dubia* survival and reproduction test (Range-finder).

Weathered Diesel Concentrations		Survival (%)	Reproduction (Mean \pm SD)
Nominal (% v/v)	Mean Measured (mg diesel/L)		
Laboratory control	0.19	100	18.2 \pm 0.8
MW-11 (Site Control)	0.26	100	6.4 \pm 1.3
0.1	0.15	100	20.6 \pm 1.3
0.39	0.16	100	17.2 \pm 5.8
1.6	0.19	100	20.0 \pm 1.9
6.25	0.30	100	19.6 \pm 1.7
25	0.89	100	19.6 \pm 1.8
100	5.53	100	22.0 \pm 2.2
Test endpoint (mg diesel /L) *			
	LC50	>5.53	--
	IC25 (95% CL)	--	>5.53
	IC50 (95% CL)	--	>5.53
	NOEC	5.53	5.53
	LOEC	>5.53	>5.53

SD = Standard Deviation, DL = Detection Limit (0.16 mg Diesel/L)

(*) = Result was calculated using nominal values and the laboratory control as the negative control

3.2 Definitive Toxicity Test Results

The results of the definitive toxicity tests are summarized in Tables 15 to 18. All endpoints were calculated in CETIS using the site control as the negative control. Confidence limits (95%) are shown in brackets.

The results of the 7-day Topsmelt test are shown in 15. The LC50 estimate for survival was >3.14 mg weathered diesel/L and the IC25 estimate for biomass and dry weight was 2.0 (1.4 – 2.6) and 2.4 (1.7 – 3.0) mg weathered diesel/L, respectively. The resulting LOEC estimates for survival, biomass and dry weight were >3.14, 3.14 and 3.14 mg weathered diesel/L, respectively.

The results of the Echinoderm fertilization test are shown in Table 16. No significant difference was observed in the % fertilization when the highest concentration of the weathered diesel test sample, DW-3 (100% v/v) was compared to the laboratory control.

The results of the 7-day Fathead minnow test are shown in Table 17. The lethal concentration (LC50) estimate for survival was >4.33 mg weathered diesel/L, and the 25% inhibitive concentrations (IC25) estimate for biomass and dry weight were 4.3 (3.8 – 4.6) and 4.8 (N/A – 6.0) mg weathered diesel/L, respectively.

The results of the *C. dubia* survival and reproduction test are shown in Table 19. No significant effects were observed on either survival or reproduction when the highest concentration of the weathered diesel test sample, DW-3 (100% v/v) was compared to the laboratory control.

Table 16. Results: Topsmelt 7-day survival and growth test (Definitive).

Diesel Concentrations		Survival (%) (Mean \pm SD)	Biomass (mg) (Mean \pm SD)	Dry Weight (mg) (Mean \pm SD)
Nominal (% v/v)	Mean Measured (mg diesel/L)			
Laboratory control	0.16	100 \pm 0.0	1.83 \pm 0.41	1.83 \pm 0.41
MW-11 (Site Control)	0.20	100 \pm 0.0	2.04 \pm 0.26	2.04 \pm 0.26
12.5	0.37*	92.0 \pm 11.0	1.71 \pm 0.33	1.86 \pm 0.25
25	0.62	100 \pm 0.0	1.93 \pm 0.31	1.93 \pm 0.31
50	1.29	92.0 \pm 11.0	1.65 \pm 0.47	1.77 \pm 0.32
75	2.12*	92.0 \pm 11.0	1.30 \pm 0.26	1.42 \pm 0.33
100	3.14	72.0 \pm 17.9	0.90 \pm 0.40	1.21 \pm 0.28
Test endpoint (mg diesel/L)				
LC25 (95% CL)		3.0 (2.3 – N/A)	--	--
LC50 (95% CL)		>3.14	--	--
IC25 (95% CL)		--	2.0 (1.4 – 2.6)	2.4 (1.7 – 3.0)
IC50 (95% CL)		--	3.1 (2.4 – 4.0)	>3.14
NOEC		3.14	2.12	2.12
LOEC		>3.14	3.14	3.14

SD = Standard Deviation, LC = Lethal Concentration, IC = Inhibition Concentration, CL = Confidence Limits, NOEC = No Observed Effect Concentration, LOEC = Lowest Observed Effect Concentration, N/A = Not Available, DL = Detection Limit (0.15 mg Diesel/L)

(*) Value interpolated from nominal-measured relationship; results are calculated using the laboratory control as the negative control

Table 17. Results: Echinoderm (*Strongylocentrotus purpuratus*) fertilization test (Definitive).

Sample ID	Analytical (mg diesel/L)	Fertilized eggs (%) (Mean \pm SD)
Laboratory control	0.16	78.2 \pm 3.5
Salt-adjusted control	-	75.2 \pm 6.2
MW-11 (Site control)	0.22	78.2 \pm 5.2
DW-3 (100% v/v)	2.71	79.2 \pm 4.8

SD = Standard Deviation, DL = Detection Limit (0.15 mg Diesel/L)

Table 18. Results: Fathead minnow 7-day survival and growth test (Definitive).

Diesel Concentrations		Survival (%) (Mean \pm SD)	Biomass (mg) (Mean \pm SD)	Dry Weight (mg) (Mean \pm SD)
Nominal (% v/v)	Mean Measured (mg diesel/L)			
Laboratory control	0.17	100 \pm 0.0	0.63 \pm 0.03	0.63 \pm 0.03
MW-11 (Site Control)	0.21	95.0 \pm 5.8	0.62 \pm 0.05	0.65 \pm 0.03
12.5	0.51*	97.5 \pm 5.0	0.59 \pm 0.04	0.61 \pm 0.02
25	0.89	97.5 \pm 5.0	0.58 \pm 0.05	0.60 \pm 0.03
50	1.90	87.5 \pm 12.6	0.60 \pm 0.09	0.69 \pm 0.05
75	3.04*	95.0 \pm 5.8	0.58 \pm 0.05	0.61 \pm 0.05
100	4.33	82.5 \pm 12.6	0.44 \pm 0.10	0.54 \pm 0.07
Test endpoint (mg diesel/L)				
LC50 (95% CL)		>4.33	--	--
IC25 (95% CL)		--	4.3 (3.8 – 4.6)	>4.33
IC50 (95% CL)		--	>4.33	>4.33
NOEC		3.04	3.04	3.04
LOEC		4.33	4.33	4.33

SD = Standard Deviation, LC = Lethal Concentration, IC = Inhibition Concentration, CL = Confidence Limits, N/A = Not Available, NOEC = No Observed Effect Concentration, LOEC = Lowest Observed Effect Concentration, DL = Detection Limit (0.15 mg Diesel/L)

(*) Value interpolated from nominal-measured relationship; results are calculated using the laboratory control as the negative control

Table 19. Results: *C. dubia* survival and reproduction test (Definitive).

Diesel Concentrations		Survival (%)	Reproduction (Mean \pm SD)
Sample ID	Mean Measured (mg diesel/L)		
Laboratory Control	0.17	100	18.8 \pm 2.4
MW-11 (Site Control)	0.25	100	15.5 \pm 3.7
DW-3 (100% v/v)	4.12	100	17.4 \pm 4.2

SD = Standard Deviation, DL = Detection Limit (0.15 mg Diesel/L)

4.0 QA/QC

The health history of the test organisms used in the exposures was acceptable and met the requirements of the USEPA protocol. The tests met all control acceptability criteria and water quality parameters remained within ranges specified in the protocols throughout the tests. There were no deviations from the test methodologies, aside from decreased test replicates for the range-finding tests and the sample being outside of holding time for all tests. Subsamples were analyzed each testing period to ensure the diesel in the sample remained stable for the duration of the tests. Uncertainty associated with the test is best described by the standard deviation around the mean and/or the confidence intervals around the point estimates.

Results of the reference toxicant test conducted during the testing program are summarized in Table 19. Results for this test fell within the range for organism performance of the mean and two standard deviations, based on historical results obtained by the laboratory with this test. Thus, the sensitivity of the organisms used in this test was appropriate. The reference toxicant test was performed under the same conditions as those used for the sample.

Table 20. Reference toxicant test results.

Test Species	Endpoint	Historical Mean (2SD Range)	CV (%)	Test Date
<i>A. affinis</i>	Survival (LC50): 100.1 µg/L Cu	91.3 (60.4 – 138.0)	21	February 19, 2019
	Biomass (IC50): 101.1 µg/L Cu	96.0 (65.0 – 141.6)	20	
	Survival (LC50): 104.0 µg/L Cu	95.9 (68.6 – 133.9)	17	April 23, 2019
	Biomass (IC50): 95.1 µg/L Cu	91.3 (66.1 – 126.1)	16	
<i>S. purpuratus</i>	Fertilization (IC50): 16.4 µg/L Cu	18.4 (8.4 – 40.1)	41	February 19, 2019
	Fertilization (IC50): 10.21 µg/L Cu	18.0 (8.9 – 36.5)	36	May 1, 2019
<i>P. promelas</i>	Survival (LC50): 4.0 g/L NaCl	4.7 (3.7 – 6.1)	12	February 19, 2019
	Biomass (IC50): 4.0 g/L NaCl	4.5 (3.4 – 6.1)	15	
	Survival (LC50): 4.2 g/L NaCl	4.7 (3.7 – 6.1)	13	April 23, 2019
	Biomass (IC50): 3.5 g/L NaCl	4.5 (3.4 – 6.1)	15	
<i>C. dubia</i>	Survival (LC50): 2.0 g/L NaCl	2.0 (1.8 – 2.2)	5	March 5, 2019
	Biomass (IC50): 1.8 g/L NaCl	1.5 (1.0 – 2.1)	18	
	Survival (LC50): 2.0 g/L NaCl	2.0 (1.8 – 2.2)	4	May 14, 2019
	Biomass (IC50): 1.2 g/L NaCl	1.6 (1.1 – 2.3)	20	

SD = Standard Deviation, CV = Coefficient of Variation, LC = Lethal Concentration, IC = Inhibition Concentration, EC = Effective Concentration

5.0 REFERENCES

USEPA. 2002. Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Marine and Estuarine Organisms. EPA-821-R-02-014.

USEPA. 1995. Short-Term Method for Estimating the Chronic Toxicity of Effluents and Receiving Waters to the West Coast Marine and Estuarine Organisms. EPA-600-R-95-136.

Tidepool Scientific Software. 2013. CETIS comprehensive environmental toxicity information system, version 1.9.4 Tidepool Scientific Software, McKinleyville, CA. 275 pp.

WDOE. 2016. Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria. Washington State Department of Ecology. Water Quality Program. Publication number: WQ-R-95-80, Revised June 2016.

APPENDIX A – *Atherinops affinis* Toxicity Test Data

Topsmelt Test Summary Sheet

Client: WDOE Start Date & Time: Feb 19, 2019 @ 1510h

Work Order No.: 191279a Test Species: Atherinops affinis

Sample Information:

Sample ID: DW-3 (Range finder)
Sample Date: 29-Jan-19
Date Received: 30-Jan-19
Sample Volume: 5x 20L

Dilution Water:

Type: Natural Seawater
Source: Vancouver Aquarium
Vancouver, BC

Test Organism Information:

Batch No.: 021919
Source: Aquatic Biosystems, CO
Age: 9 days

Copper Reference Toxicant Results:

Reference Toxicant ID: AAC38
Stock Solution ID: 19Cu01
Date Initiated: 19-Feb-19
7-d EC50 (95% CL): 100.1 (89.9 - 111.4) µg/L Cu
7-d IC50 (95% CL): 101.1 (86.4 - 112.0) µg/L Cu

EC50 Reference Toxicant Mean (Acceptable Range): 91.3 (60.4 - 138.0) µg/L Cu CV (%): 21
IC50 Reference Toxicant Mean (Acceptable Range): 96.0 (65.0 - 141.6) µg/L Cu CV (%): 20

Test Results:

	Survival	Dry Biomass	Dry Weight
NOEC	0.8	0.8	0.8
LOEC	5.1	5.1	5.1
LC25 mg/L diesel (95% CL)	N/A		
LC50 mg/L diesel (95% CL)	2.4 (1.7 - 3.3)		
IC25 mg/L diesel (95% CL)		1.3 (0.5 - 1.8)	2.1 (n/a - 2.9)
IC50 mg/L diesel (95% CL)		2.3 (1.4 - 3.2)	>5.1

Reviewed by: 

Date reviewed: Dec-19, 2019

Chronic Marine Toxicity Test Initial and Final Water Quality Measurements

Client: WDOE
Sample ID: DW-3 (Pangea Gender)
Work Order #: 191279

Start Date & Time: Feb 19/19 @ 1510h
Stop Date & Time: Feb 26/19 @ 1430h
Test Species: Atherinops affinis

Concentration	Days													
	0	1		2		3		4		5		6		7
Control	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	20.0	19.0	19.0	19.0	19.0	19.5	19.0	19.5	19.0	24.5	19.0	19.0
DO (mg/L)	7.2	7.3	7.3	6.6	6.8	6.3	6.9	5.8	7.5	6.2	7.3	5.9	6.9	5.7
pH	7.9	7.8	7.9	7.7	7.8	7.6	7.8	7.5	7.6	7.6	7.7	7.5	7.7	7.5
Salinity (ppt)	29	29		29		29		30		30		29		29
Initials	ML	ML		ML		ML		A		A		ML		CL

Concentration	Days													
	0	1		2		3		4		5		6		7
Site Control	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	19.0	19.5	19.0	19.0	19.0	19.0
DO (mg/L)	6.8	7.3	7.4	6.5	7.1	6.4	6.9	5.9	7.3	6.1	7.2	6.0	7.2	6.6
pH	7.9	7.8	8.0	7.8	7.9	7.7	8.1	7.9	7.8	7.6	7.7	7.8	7.9	8.0
Salinity (ppt)	30	30		30		28		30		30		29		30
Initials	A	ML		ML		ML		A		A		ML		A

Concentration	Days													
	0	1		2		3		4		5		6		7
0.1	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	20.0	19.5	19.0	19.0	19.0	19.0
DO (mg/L)	5.8	7.3	7.2	6.7	6.9	6.3	6.9	5.9	7.2	6.0	7.2	5.8	6.9	5.9
pH	7.9	7.8	7.9	7.7	7.8	7.6	7.8	7.6	7.6	7.6	7.7	7.5	7.7	7.5
Salinity (ppt)	29	29		29		29		30		30		29		29
Initials	A	ML		ML		ML		A		A		ML		A

Concentration	Days													
	0	1		2		3		4		5		6		7
0.39	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	20.0	19.5	19.0	19.0	19.0	19.0
DO (mg/L)	6.8	7.1	7.2	6.7	6.9	6.3	6.9	6.0	7.2	6.1	7.2	5.9	6.9	6.2
pH	7.9	7.8	7.9	7.7	7.9	7.6	7.8	7.5	7.7	7.6	7.7	7.5	7.7	7.6
Salinity (ppt)	29	29		29		29		30		30		29		29
Initials	A	ML		ML		ML		A		A		ML		A

Analysts: ML, AWD, W, RL

Reviewed by: [Signature]
Date reviewed: July 17, 2019

Sample Description:

DW-3: clear, yellow, odourless, some particulates

Comments:

ML-11: clear, light yellow, odourless, no particulates

2/2

Chronic Marine Toxicity Test

Initial and Final Water Quality Measurements

Client:

WDOE

Start Date & Time:

Feb 19/19 @ 1510h

Sample ID:

DW-3 (Range finder)

Stop Date & Time:

Feb 26/19 @ 1430h

Work Order #:

191279

Test Species: *Atherinops affinis*

Concentration (% v/v) 1.6	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	20.0	19.5	19.0	19.0	19.0	19.0
DO (mg/L)	6.8	7.1	7.2	6.6	6.9	6.4	6.9	5.9	7.2	6.0	7.2	5.9	6.9	6.1
pH	7.9	7.8	7.9	7.7	7.9	7.7	7.8	7.5	7.7	7.6	7.7	7.6	7.7	7.6
Salinity (ppt)	29	29		29		29		30		30		29		29
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Concentration 6.3	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	20.0	19.5	19.0	19.0	19.0	19.0
DO (mg/L)	6.8	7.0	7.2	6.5	7.0	6.3	6.9	6.1	7.2	6.1	7.3	5.8	6.9	5.8
pH	7.9	7.8	7.9	7.7	7.9	7.7	7.8	7.6	7.7	7.9	7.7	7.6	7.8	7.6
Salinity (ppt)	29	29		29		29		30		30		29		29
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Concentration 25	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	20.0	19.5	19.0	19.0	19.0	19.0
DO (mg/L)	6.8	7.0	7.3	6.4	7.0	6.2	6.9	6.2	7.2	6.1	7.3	5.8	6.9	6.5
pH	7.9	7.9	7.9	7.8	7.9	7.8	7.9	7.6	7.7	7.9	7.7	7.8	7.9	7.7
Salinity (ppt)	29	30		30		29		30		30		29		30
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Concentration 100	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	19.0	19.5	19.0	19.0	19.0	19.0
DO (mg/L)	7.3	7.1	7.6	6.4	7.4	6.0	7.2	6.2	7.7	6.0	7.4	5.6	6.8	6.4
pH	7.9	7.9	7.9	7.9	7.9	8.0	7.9	7.8	7.8	7.8	7.8	8.0	8.0	8.0
Salinity (ppt)	28	31		31		30		30		30		30		30
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Analysts:

ML, AWD, CS, RC

Reviewed by:

[Signature]

Date reviewed:

July 17, 2019

Sample Description:

Comments:

Chronic Marine Toxicity Test

Initial and Final Water Quality Measurements

Client:

Sample ID:

Work Order #:

WDE
DW-3 (Range finder)
191279

Start Date & Time:

Stop Date & Time:

Test Species:

1910h
Feb 19/19 @ 1450h
Feb 26/19 @ 1400h
Atherinops affinis 1430h

(% v/v) Concentration Salt Adj. Ctrl	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	19.0	19.0	19.0	19.0	19.5	19.0	19.6	19.0	19.0	19.0	19.0
DO (mg/L)	7.1	7.2	7.2	6.6	7.0	6.4	7.2	6.0	7.2	6.3	7.3	6.4	7.1	6.5
pH	7.9	7.9	8.0	7.8	8.0	7.7	7.9	7.6	7.9	7.7	7.9	7.7	7.9	7.6
Salinity (ppt)	29	29		29		29		30		30		30		30
Initials	ML	ML		ML		ML		ML		ML		ML		ML

Concentration	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)														
DO (mg/L)														
pH														
Salinity (ppt)														
Initials														

Concentration	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)														
DO (mg/L)														
pH														
Salinity (ppt)														
Initials														

Concentration	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)														
DO (mg/L)														
pH														
Salinity (ppt)														
Initials														

Analysts:

Reviewed by:

Date reviewed:

ML, AWD, CJ, RC
[Signature]
July 17, 2019

Sample Description:

Comments:

7-d Topsmelt Toxicity Test **Daily Survival**

Client: WDOE
Sample ID: DW-3 (Range & Gender)
Work Order #: 191279

Start Date & Time: Feb 19/19 @ 1510h
Stop Date & Time: Feb 26/19 @ 1430h
Test Species: Atherinops affinis

Concentration <i>Normal</i> % (v/v)	Rep	Day of Test - No. of Survivors							Comments
		1	2	3	4	5	6	7	
control	A	5	5	5	5	5	5	5	
	B	5	5	5	5	5	5	5	
	C	5	5	5	5	5	5	5	
	D								
	E								
site c/v	A	5	5	5	5	4	4	4	
	B	5	5	5	5	5	4	4	
	C	5	5	5	5	5	5	5	
	D								
	E								
0.10	A	5	5	5	5	5	5	5	
	B	5	5	5	5	5	5	5	
	C	5	5	5	5	5	4	4	
	D								
	E								
0.39	A	5	5	5	5	5	4	4	
	B	5	5	5	5	5	5	5	
	C	5	5	5	5	5	5	5	
	D								
	E								
1.6	A	5	5	5	5	5	5	5	
	B	5	5	5	4	4	4	4	
	C	5	5	5	5	5	5	5	
	D								
	E								
6.3	A	5	5	5	5	5	5	5	
	B	5	5	5	5	5	5	5	
	C	5	5	5	5	5	5	5	
	D								
	E								
25	A	5	4	4	4	4	4	4	
	B	5	5	5	5	5	5	5	
	C	5	5	5	5	5	5	5	
	D								
	E								
100.	A	5	4	1	1	1	1	1	
	B	5	5	0					
	C	5	4	3	2	2	2	2	
	D								
	E								
Tech Initials		ML	ML	ML	mm	~	ML	ML	

Comments:

-remaining organisms appear normal

Reviewed by:

[Signature]

Date reviewed:

July 17, 2019

7-d Topsmelt Toxicity Test Daily Survival

Client: WDOE
Sample ID: DWI-3 (Range Finder)
Work Order #: 191279

Start Date & Time: Feb 19/190 1450h
Stop Date & Time: Feb 26/190 1450h
Test Species: Atherinops affinis

1510h
1430h

Concentration % (v/v)	Rep	Day of Test - No. of Survivors							Comments
		1	2	3	4	5	6	7	
Salt	A	5	5	5	5	5	5	5	
Adj.	B	↓	↓	↓	↓	↓	↓	↓	
chl	C	↓	↓	↓	↓	↓	↓	↓	
	D	↓	↓	↓	↓	↓	↓	↓	
	E	↓	↓	↓	↓	↓	↓	↓	
	A								
	B								
	C								
	D								
	E								
	A								
	B								
	C								
	D								
	E								
	A								
	B								
	C								
	D								
	E								
	A								
	B								
	C								
	D								
	E								
	A								
	B								
	C								
	D								
	E								
	A								
	B								
	C								
	D								
	E								
Tech Initials		mb	mb	mb	um	~	mb	mb	

Comments: _____

Reviewed by: [Signature]

Date reviewed: July 17, 2019

Topsmelt Toxicity Test Data Sheet

Dry Weight Data

Client: WDOEStart Date & Time: Feb 19/19 @ 1510hWork Order #: 191279Termination Date & Time: Feb 26/19 @ 1430hSample ID: (RangeFinder) DW-3

% (v/v) <i>Nominal</i>	Rep	RF Pan No. <i>red</i>	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
Control	A	1	5	MU	1005.44	1011.47	5	DG 1/1
	B	2	5		1022.51	1028.31	5	1/1
	C	3	5		1016.50	1022.38	5	1/1
	D	4			1023.78	—		
	E	5			1007.24	—		
Salt Adj. Control (site)	A	6	4		1026.12	1030.30	4	1/1
	B	7	4		1018.47	1022.87	4	1/1
	C	8	5		1003.51	1010.51	5	1/1
	D	9			1026.74	—		
	E	10			1018.23	—		
0.1	A	11	5		1036.19	1042.56	5	1/1
	B	12	5		1017.93	1023.26	5	1/1
	C	13	4		1028.24	1032.82	4	1/1
	D	14			1023.75	—		
	E	15			1021.54	—		
0.39	A	16	4		1028.71	1033.97	4	1/1
	B	17	5		1008.66	1014.75	5	1/1
	C	18	5		1019.13	1025.24	5	1/1
	D	19			1020.17	—		
	E	20			1016.51	—		
1.6	A	21	5		1029.50	1034.77	5	1/1
	B	22	4		1027.35	1032.89	4	1/1
	C	23	5		1029.90	1036.42	5	1/1
	D	24			1023.91	—		
	E	25			1017.25	—		
6.3	A	26	5		1024.96	1032.18	5	1/1
	B	27	5		1033.09	1039.09	5	1/1
	C	28	5		1031.22	1036.78	5	1/1
	D	29			1025.80	—		
	E	30			1012.64	—		✓

Comments: 10% reweigh: ① pan * 2 weight 1028.30 ② pan * 12 weight 1023.29
 ③ pan * 26 weight 1032.19

Reviewed by: Date Reviewed: July 17, 2019

Topsmelt Toxicity Test Data Sheet

Dry Weight Data

Client: WDOEStart Date & Time: Feb 19/19 @ 1510hWork Order #: 191275Termination Date & Time: Feb 26/19 @ 1430hSample ID: Range finder (Dw-3)

% (v/v) <i>Normal</i>	Rep	RF Pan No. <i>Rep</i>	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
<i>Control</i> <i>CS</i> 25	A	<i>CS</i> 131	4	<i>ML</i>	1021.23	1027.21	4	<i>DG</i> / <i>10</i>
	B	232	5		1024.52	1029.89	5	<i>10</i>
	C	333	5		1035.84	1040.42	5	<i>10</i>
	D	434			1028.55	—		
	E	535			1020.50	—		
<i>Salt-Adj.</i> <i>CS</i> <i>Control</i> 100	A	<i>CS</i> 636	1		1019.29	1019.86	1	<i>10</i>
	B	737	0		1005.70	—	—	
	C	838	2		1016.34	1017.68	2	<i>10</i>
	D	939			1010.38	—		
	E	1040			1013.95	—		<i>↓</i>
	A	11						
	B	12						
	C	13						
	D	14						
	E	15						
	A	16						
	B	17						
	C	18						
	D	19						
	E	20						
	A	21						
	B	22						
	C	23						
	D	24						
	E	25						
	A	26						
	B	27						
	C	28						
	D	29						
	E	30						

Comments:

Reviewed by:



Date Reviewed:

July 17, 2019

Topsmelt Toxicity Test Data Sheet

Dry Weight Data

Client: WDOE

Start Date & Time: Feb 19/19 14:50h ^{1510h}

Work Order #: 191279

Termination Date & Time: Feb 26/19 14:50h ^{1430h}

Sample ID: DW-3 (Range finder)

RT Black

% (v/v)	Rep	Pan No.	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
Salt Adj. Control	A	1 ^{ml} 26	5	ML	1034.74	1039.88	5	CA/CA
	B	2 27	5	↓	1026.92	1033.40	5	↓
	C	3 28	5	↓	1007.01	1013.83	5	↓
	D	4 29	5	↓	1021.46	1027.52	5	↓
	E	5 30	5	↓	1017.82	1024.45	5	↓
	A	6						
	B	7						
	C	8						
	D	9						
	E	10						
	A	11						
	B	12						
	C	13						
	D	14						
	E	15						
	A	16						
	B	17						
	C	18						
	D	19						
	E	20						
	A	21						
	B	22						
	C	23						
	D	24						
	E	25						
	A	26						
	B	27						
	C	28						
	D	29						
	E	30						

Comments:

Reviewed by: WU

Date Reviewed: July 17, 2019

CETIS Summary Report

Report Date: 19 Dec-19 14:29 (p 1 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Batch ID: 00-9075-6029	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 15:10	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 26 Feb-19 14:30	Species: Atherinops affinis	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 9d
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 15h	Client: WA State Dept of Ecology	

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓ Level	mg/L	95% LCL	95% UCL	TU	S
02-3344-4127	Mean Dry Biomass-mg	Linear Interpolation (ICPIN)	✓ IC5	0.4651	n/a	1.333		1
			✓ IC10	0.7864	n/a	1.258		
			✓ IC15	0.932	0.2029	1.423		
			✓ IC20	1.087	0.3168	1.591		
			✓ IC25	1.254	0.4612	1.801		
			IC40	1.841	0.9731	2.566		
			IC50	2.315	1.393	3.21		
08-7851-4664	Mean Dry Weight-mg	Linear Interpolation (ICPIN)	IC5	0.8057	n/a	1.302		1
			IC10	1.072	n/a	1.614		
			IC15	1.377	n/a	1.967		
			IC20	1.727	n/a	2.368		
			IC25	2.128	n/a	2.855		
			IC40	3.724	1.332	5.027		
			IC50	>5.07	n/a	n/a		

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits		Overlap	Decision
				Lower	Upper		
02-3344-4127	Mean Dry Biomass-mg	Control Resp	1.181	0.85	>>	Yes	Passes Criteria

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	SC	5	1.245	1.079	1.411	1.028	1.364	0.05979	0.1337	10.74%	0.00%
0.15	N	3	1.181	1.123	1.239	1.16	1.206	0.01348	0.02334	1.98%	5.18%
0.151		3	1.085	0.6387	1.532	0.916	1.274	0.1038	0.1798	16.57%	12.84%
0.16		3	1.164	0.923	1.405	1.052	1.222	0.05601	0.09701	8.33%	6.52%
0.18		3	1.155	0.8286	1.482	1.054	1.304	0.07595	0.1315	11.39%	7.22%
0.27		3	1.252	0.8247	1.679	1.112	1.444	0.09931	0.172	13.74%	-0.55%
0.29	XC	3	1.039	0.2594	1.818	0.836	1.4	0.1811	0.3137	30.20%	16.59%
0.79		3	1.062	0.7133	1.411	0.916	1.196	0.08105	0.1404	13.22%	14.71%
5.07		3	0.1273	-0.2068	0.4614	0	0.268	0.07765	0.1345	105.62%	89.77%

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	SC	5	1.245	1.079	1.411	1.028	1.364	0.05979	0.1337	10.74%	0.00%
0.15	N	3	1.181	1.123	1.239	1.16	1.206	0.01348	0.02334	1.98%	5.18%
0.151		3	1.162	0.9008	1.423	1.066	1.274	0.06063	0.105	9.04%	6.71%
0.16		3	1.252	1.115	1.388	1.218	1.315	0.03169	0.05489	4.38%	-0.52%
0.18		3	1.248	0.8191	1.676	1.054	1.385	0.09962	0.1725	13.83%	-0.20%
0.27		3	1.252	0.8247	1.679	1.112	1.444	0.09931	0.172	13.74%	-0.55%
0.29	XC	3	1.182	0.707	1.656	1.045	1.4	0.1103	0.1911	16.17%	5.10%
0.79		3	1.162	0.4182	1.905	0.916	1.495	0.1728	0.2993	25.76%	6.71%
5.07		2	0.62	-0.01516	1.255	0.57	0.67	0.04999	0.07069	11.40%	50.21%

SC = salt adjusted control

N = lab control

XC = site control

CETIS Summary Report

Report Date: 19 Dec-19 14:29 (p 2 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	1.028	1.296	1.364	1.212	1.326
0.15	N	1.206	1.16	1.176		
0.151		1.274	1.066	0.916		
0.16		1.052	1.218	1.222		
0.18		1.054	1.108	1.304		
0.27		1.444	1.2	1.112		
0.29	XC	0.836	0.88	1.4		
0.79		1.196	1.074	0.916		
5.07		0.114	0	0.268		

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	1.028	1.296	1.364	1.212	1.326
0.15	N	1.206	1.16	1.176		
0.151		1.274	1.066	1.145		
0.16		1.315	1.218	1.222		
0.18		1.054	1.385	1.304		
0.27		1.444	1.2	1.112		
0.29	XC	1.045	1.1	1.4		
0.79		1.495	1.074	0.916		
5.07		0.57		0.67		

SC = salt adjusted control

N = lab control

XC = site control

CETIS Analytical Report

Report Date: 16 Dec-19 13:42 (p 1 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 20-7028-7252	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 13:41	Analysis: Trimmed Spearman-Kärber	Status Level: 1
Batch ID: 00-9075-6029	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 15:10	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 26 Feb-19 14:30	Species: Atherinops affinis	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 9d
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 15h	Client: WA State Dept of Ecology	

Trimmed Spearman-Kärber Estimates

Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0	20.00%	0.3747	0.07319	2.37	1.692	3.32

7d Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	Code	Count	Mean	Min	Max	Std Dev	CV%	%Effect	A/B	Mean	%Effect
0.15	N	3	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	15/15	1	0.0%
0.151		3	0.9333	0.8000	1.0000	0.1155	12.37%	6.67%	14/15	0.95	5.0%
0.16		3	0.9333	0.8000	1.0000	0.1155	12.37%	6.67%	14/15	0.95	5.0%
0.18		3	0.9333	0.8000	1.0000	0.1155	12.37%	6.67%	14/15	0.95	5.0%
0.27		3	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	15/15	0.95	5.0%
0.79		3	0.9333	0.8000	1.0000	0.1155	12.37%	6.67%	14/15	0.9333	6.67%
5.07		3	0.2000	0.0000	0.4000	0.2000	100.00%	80.0%	3/15	0.2	80.0%

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.0000	1.0000	1.0000
0.151		1.0000	1.0000	0.8000
0.16		0.8000	1.0000	1.0000
0.18		1.0000	0.8000	1.0000
0.27		1.0000	1.0000	1.0000
0.79		0.8000	1.0000	1.0000
5.07		0.2000	0.0000	0.4000

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	5/5	5/5	5/5
0.151		5/5	5/5	4/5
0.16		4/5	5/5	5/5
0.18		5/5	4/5	5/5
0.27		5/5	5/5	5/5
0.79		4/5	5/5	5/5
5.07		1/5	0/5	2/5

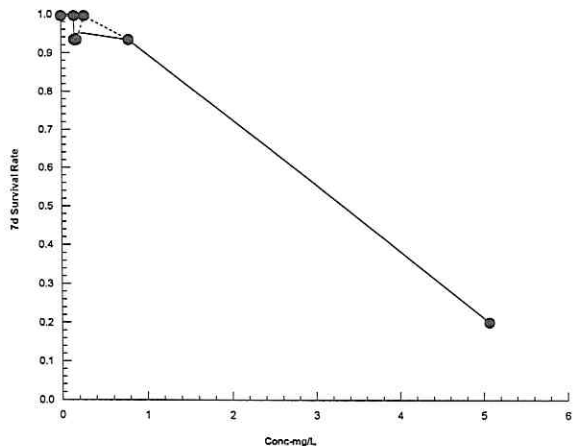
N = Lab control

CETIS Analytical Report

Report Date: 16 Dec-19 13:42 (p 2 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test			Nautilus Environmental
Analysis ID: 20-7028-7252	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4	
Analyzed: 16 Dec-19 13:41	Analysis: Trimmed Spearman-Kärber	Status Level: 1	

Graphics



CETIS Analytical Report

Report Date: 18 Dec-19 13:35 (p 1 of 2)
 Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 02-3344-4127	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 17 Dec-19 17:24	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 00-9075-6029	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 15:10	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 26 Feb-19 14:30	Species: Atherinops affinis	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 9d
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 15h	Client: WA State Dept of Ecology	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	2081880	200	Yes	Two-Point Interpolation

Test Acceptability Criteria

TAC Limits

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	1.181	0.85	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
IC5	0.4651	n/a	1.333
IC10	0.7864	n/a	1.258
IC15	0.932	0.2029	1.423
IC20	1.087	0.3168	1.591
IC25	1.254	0.4612	1.801
IC40	1.841	0.9731	2.566
IC50	2.315	1.393	3.21

Mean Dry Biomass-mg Summary

Calculated Variate

Isotonic Variate

Conc-mg/L	Code	Count	Mean	Min	Max	Std Dev	CV%	%Effect	Mean	%Effect
0.15	N	3	1.181	1.16	1.206	0.02334	1.98%	0.0%	1.181	0.0%
0.151		3	1.085	0.916	1.274	0.1798	16.57%	8.07%	1.164	1.4%
0.16		3	1.164	1.052	1.222	0.09701	8.34%	1.41%	1.164	1.4%
0.18		3	1.155	1.054	1.304	0.1315	11.39%	2.15%	1.164	1.4%
0.27		3	1.252	1.112	1.444	0.172	13.74%	-6.04%	1.164	1.4%
0.79		3	1.062	0.916	1.196	0.1404	13.22%	10.05%	1.062	10.05%
5.07		3	0.1273	0	0.268	0.1345	105.60%	89.22%	0.1273	89.22%

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.206	1.16	1.176
0.151		1.274	1.066	0.916
0.16		1.052	1.218	1.222
0.18		1.054	1.108	1.304
0.27		1.444	1.2	1.112
0.79		1.196	1.074	0.916
5.07		0.114	0	0.268

N= lab control

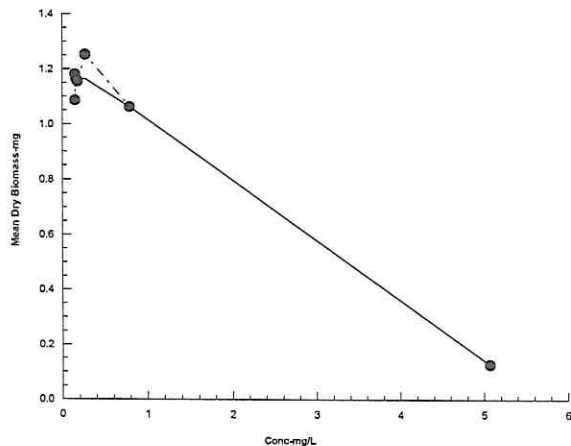
CETIS Analytical Report

Report Date: 18 Dec-19 13:35 (p 2 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test Nautilus Environmental

Analysis ID: 02-3344-4127	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 17 Dec-19 17:24	Analysis: Linear Interpolation (ICPIN)	Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 18 Dec-19 13:35 (p 1 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 08-7851-4664	Endpoint: Mean Dry Weight-mg	CETIS Version: CETISv1.9.4
Analyzed: 17 Dec-19 17:25	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 00-9075-6029	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 15:10	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 26 Feb-19 14:30	Species: Atherinops affinis	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 9d
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 15h	Client: WA State Dept of Ecology	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	2141035	200	Yes	Two-Point Interpolation

Point Estimates

Level	mg/L	95% LCL	95% UCL
IC5	0.8057	n/a	1.302
IC10	1.072	n/a	1.614
IC15	1.377	n/a	1.967
IC20	1.727	n/a	2.368
IC25	2.128	n/a	2.855
IC40	3.724	1.332	5.027
IC50	>5.07	n/a	n/a

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Calculated Variate						Isotonic Variate	
			Mean	Min	Max	Std Dev	CV%	%Effect	Mean	%Effect
0.15	N	3	1.181	1.16	1.206	0.02334	1.98%	0.0%	1.219	0.0%
0.151		3	1.162	1.066	1.274	0.105	9.04%	1.61%	1.219	0.0%
0.16		3	1.252	1.218	1.315	0.05489	4.39%	-6.01%	1.219	0.0%
0.18		3	1.248	1.054	1.385	0.1725	13.83%	-5.68%	1.219	0.0%
0.27		3	1.252	1.112	1.444	0.172	13.74%	-6.04%	1.219	0.0%
0.79		3	1.162	0.916	1.495	0.2993	25.76%	1.61%	1.162	4.68%
5.07		2	0.62	0.57	0.67	0.07069	11.40%	47.49%	0.62	49.13%

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.206	1.16	1.176
0.151		1.274	1.066	1.145
0.16		1.315	1.218	1.222
0.18		1.054	1.385	1.304
0.27		1.444	1.2	1.112
0.79		1.495	1.074	0.916
5.07		0.57	0.67	

N= Lab control

CETIS Analytical Report

Report Date: 18 Dec-19 13:35 (p 2 of 2)
Test Code/ID: 191279a / 13-1178-4533

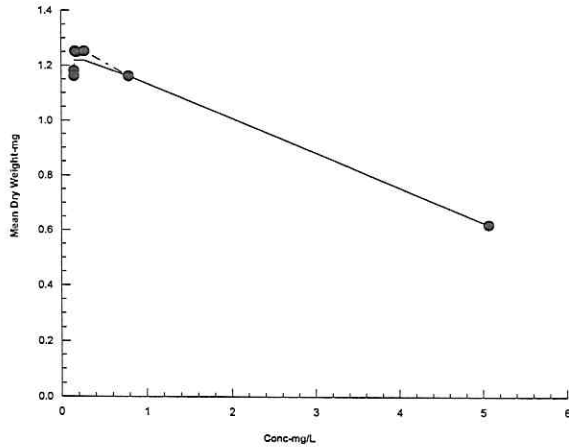
Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 08-7851-4664 Endpoint: Mean Dry Weight-mg
Analyzed: 17 Dec-19 17:25 Analysis: Linear Interpolation (ICPIN)

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 16 Dec-19 14:07 (p 1 of 2)
 Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 07-5682-5805	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 14:05	Analysis: Parametric-Control vs Treatments	Status Level: 1
Batch ID: 00-9075-6029	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 15:10	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 26 Feb-19 14:30	Species: Atherinops affinis	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 9d
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 15h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)	C > T	0.79	5.07	2.001		23.45%

Dunnett Multiple Comparison Test

Control	vs	Conc-mg/L	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.151	0.7179	2.532	0.28	4	CDF	0.5857	Non-Significant Effect
		0.16	0.7179	2.532	0.28	4	CDF	0.5857	Non-Significant Effect
		0.18	0.7179	2.532	0.28	4	CDF	0.5857	Non-Significant Effect
		0.27	0	2.532	0.28	4	CDF	0.8571	Non-Significant Effect
		0.79	0.7179	2.532	0.28	4	CDF	0.5857	Non-Significant Effect
		5.07*	8.026	2.532	0.28	4	CDF	4.4E-06	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	1.81551	0.302585	6	16.5	1.3E-05	Significant Effect
Error	0.256705	0.018336	14			
Total	2.07221		20			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:0.1%)
Variances	Levene Equality of Variance Test	2.932	7.436	0.0456	Equal Variances
Variances	Mod Levene Equality of Variance Test	0.7858	15.52	0.6076	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.8703	0.8118	0.0097	Normal Distribution

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15	N	3	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
0.151		3	0.9333	0.6465	1.0000	1.0000	0.8000	1.0000	0.0667	12.37%	6.67%
0.16		3	0.9333	0.6465	1.0000	1.0000	0.8000	1.0000	0.0667	12.37%	6.67%
0.18		3	0.9333	0.6465	1.0000	1.0000	0.8000	1.0000	0.0667	12.37%	6.67%
0.27		3	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
0.79		3	0.9333	0.6465	1.0000	1.0000	0.8000	1.0000	0.0667	12.37%	6.67%
5.07		3	0.2000	0.0000	0.6968	0.2000	0.0000	0.4000	0.1155	100.00%	80.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15	N	3	1.345	1.345	1.346	1.345	1.345	1.345	0	0.00%	0.00%
0.151		3	1.266	0.9244	1.607	1.345	1.107	1.345	0.07938	10.86%	5.90%
0.16		3	1.266	0.9244	1.607	1.345	1.107	1.345	0.07938	10.86%	5.90%
0.18		3	1.266	0.9244	1.607	1.345	1.107	1.345	0.07938	10.86%	5.90%
0.27		3	1.345	1.345	1.346	1.345	1.345	1.345	0	0.00%	0.00%
0.79		3	1.266	0.9244	1.607	1.345	1.107	1.345	0.07938	10.86%	5.90%
5.07		3	0.458	-0.1125	1.028	0.4636	0.2255	0.6847	0.1326	50.15%	65.96%

N = low control

Dec. 17/19

CETIS Analytical Report

Report Date: 16 Dec-19 14:07 (p 2 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 07-5682-5805 Endpoint: 7d Survival Rate
Analyzed: 16 Dec-19 14:05 Analysis: Parametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.0000	1.0000	1.0000
0.151		1.0000	1.0000	0.8000
0.16		0.8000	1.0000	1.0000
0.18		1.0000	0.8000	1.0000
0.27		1.0000	1.0000	1.0000
0.79		0.8000	1.0000	1.0000
5.07		0.2000	0.0000	0.4000

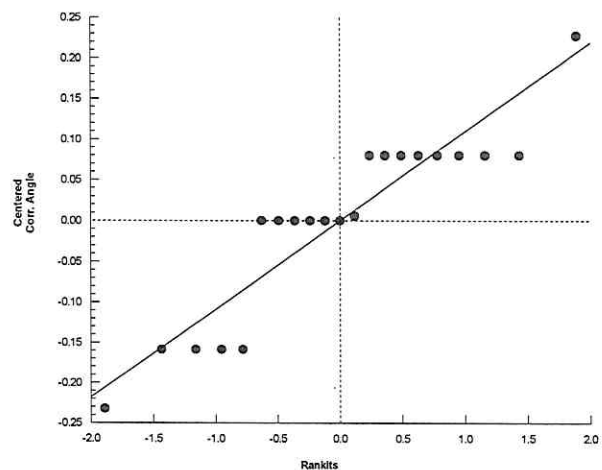
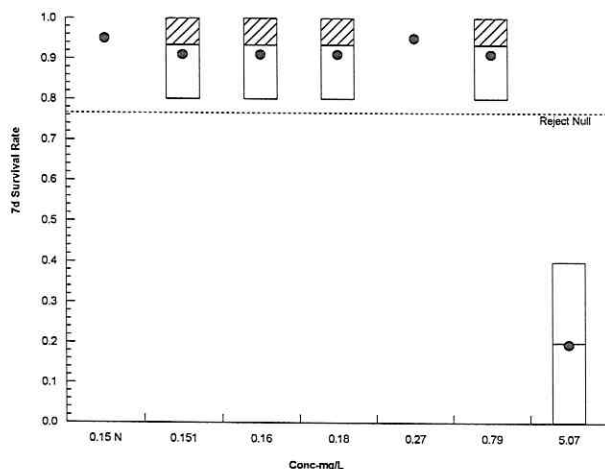
Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.345	1.345	1.345
0.151		1.345	1.345	1.107
0.16		1.107	1.345	1.345
0.18		1.345	1.107	1.345
0.27		1.345	1.345	1.345
0.79		1.107	1.345	1.345
5.07		0.4636	0.2255	0.6847

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	5/5	5/5	5/5
0.151		5/5	5/5	4/5
0.16		4/5	5/5	5/5
0.18		5/5	4/5	5/5
0.27		5/5	5/5	5/5
0.79		4/5	5/5	5/5
5.07		1/5	0/5	2/5

Graphics



N=(ab control)

CETIS Analytical Report

Report Date: 16 Dec-19 14:00 (p 1 of 2)
 Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 15-0029-9202	Endpoint: Mean Dry Weight-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 13:59	Analysis: Parametric-Multiple Comparison	Status Level: 1
Batch ID: 00-9075-6029	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 15:10	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 26 Feb-19 14:30	Species: Atherinops affinis	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 9d
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 15h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	C > T	0.79	5.07	2.001		33.93%

Bonferroni Adj t Test

Control	vs	Conc-mg/L	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.151	0.1456	2.746	0.358	4	CDF	1.0000	Non-Significant Effect
		0.16	-0.5441	2.746	0.358	4	CDF	1.0000	Non-Significant Effect
		0.18	-0.5135	2.746	0.358	4	CDF	1.0000	Non-Significant Effect
		0.27	-0.5467	2.746	0.358	4	CDF	1.0000	Non-Significant Effect
		0.79	0.1456	2.746	0.358	4	CDF	1.0000	Non-Significant Effect
		5.07*	3.843	2.746	0.401	3	CDF	0.0061	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.656295	0.109382	6	4.283	0.0134	Significant Effect
Error	0.332018	0.0255398	13			
Total	0.988313		19			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	9.993	16.81	0.1250	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.971	0.866	0.7761	Normal Distribution

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15	N	3	1.181	1.123	1.239	1.176	1.16	1.206	0.01348	1.98%	0.00%
0.151		3	1.162	0.9008	1.423	1.145	1.066	1.274	0.06063	9.04%	1.61%
0.16		3	1.252	1.115	1.388	1.222	1.218	1.315	0.03169	4.38%	-6.01%
0.18		3	1.248	0.8191	1.676	1.304	1.054	1.385	0.09962	13.83%	-5.68%
0.27		3	1.252	0.8247	1.679	1.2	1.112	1.444	0.09931	13.74%	-6.04%
0.79		3	1.162	0.4182	1.905	1.074	0.916	1.495	0.1728	25.76%	1.61%
5.07		2	0.62	-0.01516	1.255	0.62	0.57	0.67	0.04999	11.40%	47.49%

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.206	1.16	1.176
0.151		1.274	1.066	1.145
0.16		1.315	1.218	1.222
0.18		1.054	1.385	1.304
0.27		1.444	1.2	1.112
0.79		1.495	1.074	0.916
5.07		0.57	0.67	

N= (a) control

CETIS Analytical Report

Report Date: 16 Dec-19 14:00 (p 2 of 2)
Test Code/ID: 191279a / 13-1178-4533

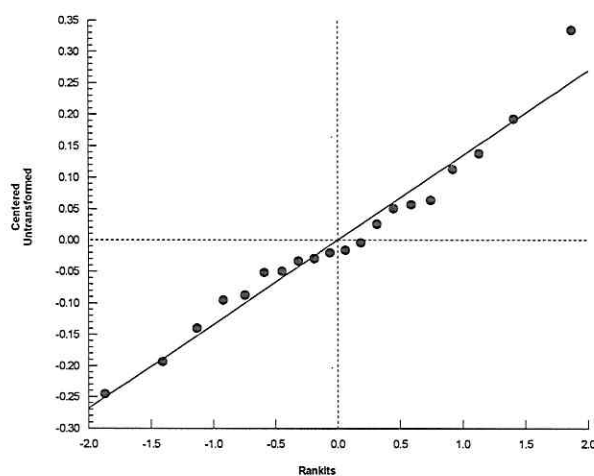
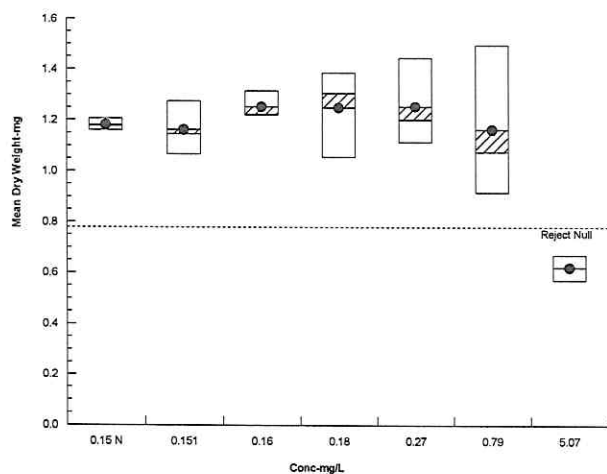
Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 15-0029-9202 Endpoint: Mean Dry Weight-mg
Analyzed: 16 Dec-19 13:59 Analysis: Parametric-Multiple Comparison

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 16 Dec-19 14:00 (p 1 of 2)
Test Code/ID: 191279a / 13-1178-4533

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 14-6247-1442	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 13:59	Analysis: Parametric-Control vs Treatments	Status Level: 1
Batch ID: 00-9075-6029	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 15:10	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 26 Feb-19 14:30	Species: Atherinops affinis	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 9d
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 15h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	C > T	0.79	5.07	2.001		23.58%

Dunnett Multiple Comparison Test

Control	vs	Conc-mg/L	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.151	0.8669	2.532	0.279	4	CDF	0.5175	Non-Significant Effect
		0.16	0.1516	2.532	0.279	4	CDF	0.8125	Non-Significant Effect
		0.18	0.2303	2.532	0.279	4	CDF	0.7861	Non-Significant Effect
		0.27	-0.6488	2.532	0.279	4	CDF	0.9659	Non-Significant Effect
		0.79	1.079	2.532	0.279	4	CDF	0.4218	Non-Significant Effect
		5.07*	9.579	2.532	0.279	4	CDF	1.3E-06	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	2.75926	0.459876	6	25.35	9.3E-07	Significant Effect
Error	0.253937	0.0181384	14			
Total	3.01319		20			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	5.142	16.81	0.5257	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9442	0.871	0.2641	Normal Distribution

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15	N	3	1.181	1.123	1.239	1.176	1.16	1.206	0.01348	1.98%	0.00%
0.151		3	1.085	0.6387	1.532	1.066	0.916	1.274	0.1038	16.57%	8.07%
0.16		3	1.164	0.923	1.405	1.218	1.052	1.222	0.05601	8.33%	1.41%
0.18		3	1.155	0.8286	1.482	1.108	1.054	1.304	0.07595	11.39%	2.15%
0.27		3	1.252	0.8247	1.679	1.2	1.112	1.444	0.09931	13.74%	-6.04%
0.79		3	1.062	0.7133	1.411	1.074	0.916	1.196	0.08105	13.22%	10.05%
5.07		3	0.1273	-0.2068	0.4614	0.114	0	0.268	0.07765	105.62%	89.22%

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.206	1.16	1.176
0.151		1.274	1.066	0.916
0.16		1.052	1.218	1.222
0.18		1.054	1.108	1.304
0.27		1.444	1.2	1.112
0.79		1.196	1.074	0.916
5.07		0.114	0	0.268

N=lab control

CETIS Analytical Report

Report Date: 16 Dec-19 14:00 (p 2 of 2)
Test Code/ID: 191279a / 13-1178-4533

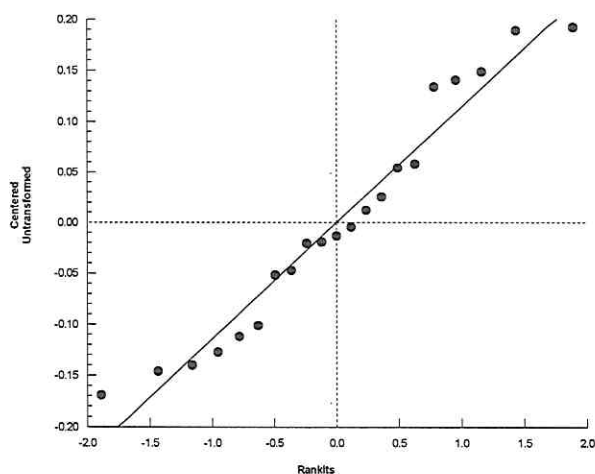
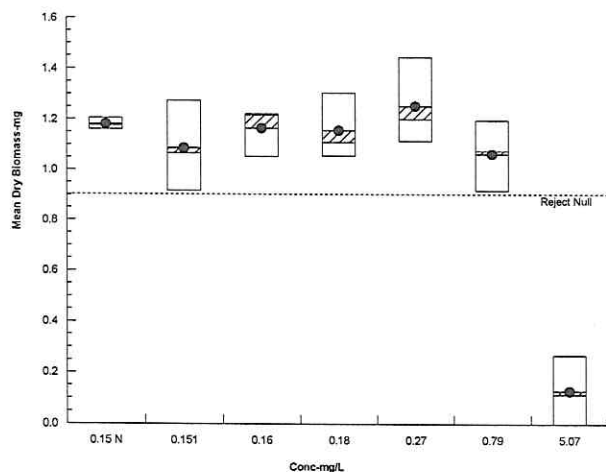
Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 14-6247-1442
Analyzed: 16 Dec-19 13:59
Endpoint: Mean Dry Biomass-mg
Analysis: Parametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



Topsmelt Test Summary Sheet

Client: WDOE Start Date & Time: April 23, 2019 @ 1230h

Work Order No.: 191279 Test Species: Atherinops affinis

Sample Information:

Sample ID: DW-3 (Definitive)
Sample Date: 29-Jan-19
Date Received: 30-Jan-19
Sample Volume: 5x 20L

Dilution Water:

Type: Natural Seawater
Source: Vancouver Aquarium
Vancouver, BC

Test Organism Information:

Batch No.: 042319
Source: Aquatic Biosystems, CO
Age: 12 days

Copper Reference Toxicant Results:

Reference Toxicant ID: AAC41
Stock Solution ID: 19Cu01
Date Initiated: 23-Apr-19
7-d EC50 (95% CL): 104.0 (88.3 - 122.5) µg/L Cu
7-d IC50 (95% CL): 95.1 (70.6 - 112.5) µg/L Cu

EC50 Reference Toxicant Mean (Acceptable Range) : 95.9 (68.6 - 133.9) µg/L Cu CV (%): 17
IC50 Reference Toxicant Mean (Acceptable Range) : 91.3 (66.1 - 126.1) µg/L Cu CV (%): 16

Test Results:

	Survival	Dry Biomass	Dry Weight
NOEC	3.14	2.12	2.12
LOEC	>3.14	3.14	3.14
LC25 mg/L diesel (95% CL)	3.0 (2.3 - n/a)		
LC50 mg/L diesel (95% CL)	>3.14		
IC25 mg/L diesel (95% CL)		2.0 (1.4 - 2.6)	2.4 (1.7 - 3.0)
IC50 mg/L diesel (95% CL)		3.1 (2.4 - 4.0)	>3.14

n/a = not available

Reviewed by: 

Date reviewed: Jan 16, 2020

Chronic Marine Toxicity Test Initial and Final Water Quality Measurements

Client: WDDF
 Sample ID: DW-3
 Work Order #: 191279

Start Date & Time: Apr 23/19 @ 1230h
 Stop Date & Time: Apr 30/19 @ 1315h
 Test Species: Atherinops affinis

Concentration (% v/v) Control	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	26.0	19.5	20.0	20.0	20.0	20.0	19.5	20.0	19.0	20.0	19.0	19.0	20.0	19.5
DO (mg/L)	7.4	6.5	7.7	6.6	7.4	5.87	7.3	5.2	7.5	5.6	7.4	5.6	7.4	5.9
pH	7.7	7.6	7.6	7.4	7.8	7.4	7.7	7.6	7.7	7.8	7.7	7.5	7.8	7.5
Salinity (ppt)	29	30		30		30		30		30		29		30
Initials	<i>AK</i>	<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>

Concentration Salt Adj. Control	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	19.0	19.0	19.0	20.0	19.0	20.0	19.5	20.0	19.0	20.0	19.0	19.0	19.0	19.5
DO (mg/L)	6.7	6.4	7.8	6.3	7.4	5.7	7.3	5.3	7.4	5.7	7.4	5.0	7.5	5.3
pH	7.9	7.8	7.7	7.6	7.8	7.6	7.9	7.8	7.9	7.9	7.9	7.7	7.9	7.7
Salinity (ppt)	31	31		31		31		30		30		31		31
Initials	<i>AK</i>	<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>

Concentration Site Control	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	20.0	19.0	20.0	20.0	20.5	20.0	19.5	20.0	19.0	20.0	19.0	19.0	19.0	20.0
DO (mg/L)	7.85	6.3	7.4	6.3	7.3	5.7	7.7	5.2	7.5	5.7	7.4	5.3	7.7	5.8
pH	8.0	8.0	8.0	7.9	7.9	7.9	7.9	7.7	7.6	7.8	7.7	8.0	8.1	8.1
Salinity (ppt)	29	30		29		30		30		30		31		31
Initials	<i>AK</i>	<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>

Concentration 12-5	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	20.0	19.5	19.0	20.0	20.0	20.0	19.5	20.0	19.0	20.0	19.0	20.0	20.0	20.5
DO (mg/L)	7.3	6.6	7.6	6.1	7.4	5.75	7.3	5.2	7.4	5.8	7.5	5.8	7.4	6.1
pH	7.7	7.6	7.6	7.5	7.7	7.5	7.7	7.7	7.7	7.8	7.8	7.6	7.8	7.8
Salinity (ppt)	29	30		30		30		30		30		30		30
Initials	<i>AK</i>	<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>		<i>AK</i>

Analysts: CS, AWD

Reviewed by: *CS*

Date reviewed: July 17, 2019

Sample Description: DW-3: clear, yellow, odorless, some particulates

Comments: MW-11 (Site Control): clear, light yellow, odorless, no particulates

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Chronic Marine Toxicity Test Initial and Final Water Quality Measurements

Client: WDOE
 Sample ID: DW-3
 Work Order #: 191279

Start Date & Time: Apr 23/19 @ 1230h
 Stop Date & Time: Apr 30/19 @ 1320h
 Test Species: Atherinops affinis

(% v/v) Concentration 25	Days														
	0	1		2		3		4		5		6		7	
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final	
Temperature (°C)	20.0	19.5	19.0	20.0	20.0	20.5	20.0	20.0	19.0	20.0	19.0	20.0	20.0	20.5	
DO (mg/L)	7.2	6.4	6.5	6.2	7.5	5.6	7.3	5.2	7.4	5.5	7.5	5.5	7.8	5.9	
pH	7.8	7.6	7.7	7.5	7.8	7.6	7.7	7.7	7.7	7.8	7.7	7.7	7.8	7.7	
Salinity (ppt)	29	30		30		30		30		30		30		30	
Initials	A	A		A		A		A		A		A		A	

Concentration 50	Days														
	0	1		2		3		4		5		6		7	
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final	
Temperature (°C)	20.0	20.0	19.0	20.0	20.0	20.5	20.0	20.0	19.0	20.0	19.0	20.0	20.0	20.5	
DO (mg/L)	7.4	6.8	6.2 7.1	6.2	7.5	5.7	7.4	5.1	7.4	5.7	7.5	5.6	7.6	6.0	
pH	7.8	7.7	7.8	7.6	7.9	7.7	7.7	7.7	7.6	7.8	7.7	7.8	7.9	7.9	
Salinity (ppt)	29	29		30		30		30		30		30		30	
Initials	A	A		A		A		A		A		A		A	

Concentration 75	Days														
	0	1		2		3		4		5		6		7	
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final	
Temperature (°C)	20.0	7.80	19.0	20.0	20.0	20.5	19.5	20.0	19.0	20.0	19.0	20.0	20.0	20.0	
DO (mg/L)	7.7	6.7	8.7	6.3	7.6	5.8	7.5	5.2	7.4	5.7	7.5	5.6	7.5	6.2	
pH	7.8	7.8	7.8	7.7	7.9	7.8	7.7	7.8	7.6	7.9	7.7	7.8	7.9	7.9	
Salinity (ppt)	29	29		30		30		30		30		30		31	
Initials	A	A		A		A		A		A		A		A	

Concentration 100	Days														
	0	1		2		3		4		5		6		7	
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final	
Temperature (°C)	19.5	19.5	19.5	20.0	20.0	20.5	19.0	20.0	19.0	20.0	19.0	20.0	20.0	20.5	
DO (mg/L)	8.4	6.7	8.0	6.3	7.6	5.8	7.8	5.2	7.4	5.6	7.5	6.0	7.6	6.3	
pH	7.8	7.8	7.8	7.8	7.9	7.8	7.7	7.8	7.8	7.9	7.7	7.9	7.9	7.9	
Salinity (ppt)	29	29		30		30		30		30		30		30	
Initials	A	A		A		A		A		A		A		A	

Analysts: CS, AWD

Reviewed by: [Signature]

Date reviewed: July 17, 2019

Sample Description: _____

Comments: _____

7-d Topsmelt Toxicity Test Daily Survival

Client: WD05
Sample ID: DW-3
Work Order #: 191279

Start Date & Time: Apr 23/19 @ 1230h
Stop Date & Time: Apr 30/19 @ 1320h
Test Species: Atherinops affinis

Concentration % (v/v)	Rep	Day of Test - No. of Survivors							Comments
		1	2	3	4	5	6	7	
Control	A	5	5	5	5	5	5	5	
	B	↓	↓	↓	↓	5	↓	↓	
	C	↓	↓	↓	↓	5	↓	↓	
	D	↓	↓	↓	↓	5	↓	↓	
	E	↓	↓	↓	↓	5	↓	↓	
Salt adj. Control	A	↓	↓	↓	↓	4	4	4	
	B	↓	↓	↓	↓	5	5	5	
	C	↓	↓	↓	↓	5	5	5	
	D	↓	4	4	↓	4	4	4	
	E	↓	5	5	5	5	5	5	
Site Control	A	↓	↓	↓	↓	5	5	5	
	B	↓	↓	↓	↓	5	↓	↓	
	C	↓	↓	↓	↓	5	↓	↓	
	D	↓	↓	↓	↓	5	↓	↓	
	E	↓	↓	↓	↓	5	↓	↓	
12.5	A	↓	↓	↓	↓	5	5	5	
	B	↓	↓	↓	↓	5	5	5	
	C	↓	↓	↓	4	4	4	4	
	D	↓	↓	↓	5	5	5	5	
	E	↓	↓	↓	5	4	4	4	
25	A	↓	↓	↓	↓	5	5	5	
	B	↓	↓	↓	↓	5	↓	↓	
	C	↓	↓	↓	↓	5	↓	↓	
	D	↓	↓	↓	↓	5	↓	↓	
	E	↓	↓	↓	↓	5	↓	↓	
50	A	↓	↓	↓	↓	4	4	4	
	B	↓	↓	↓	↓	5	5	5	
	C	↓	↓	↓	↓	5	5	5	
	D	↓	↓	↓	↓	5	5	5	
	E	↓	↓	↓	↓	4	4	4	
75	A	↓	↓	↓	↓	4	4	4	
	B	↓	↓	↓	↓	5	5	5	
	C	↓	↓	↓	↓	4	4	4	
	D	↓	↓	↓	↓	5	5	5	
	E	↓	↓	↓	↓	5	5	5	
100	A	↓	↓	↓	4	4	3	3	
	B	↓	↓	↓	4	3	3	3	
	C	↓	↓	↓	5	5	5	5	
	D	↓	↓	↓	3	3	3	3	
	E	↓	↓	↓	4	4	4	4	
Tech Initials		AL	AL	AL	AL	AL	AL	AL	

Comments: Chloragium 1 organism died due to technician error
- remaining organisms appear normal

Reviewed by: [Signature]

Date reviewed: July 17, 2019

Topsmelt Toxicity Test Data Sheet

Dry Weight Data

Client: WDOEStart Date & Time: Apr 23/19 @ 1230hWork Order #: 191279Termination Date & Time: Apr 30/19 @ 1320hSample ID: DW-3AA Red

% (v/v)	Rep	Pan No.	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
Control	A	1	5	α	1049.82	1060.36	5	α/α
	B	2	5	α	1072.62	1083.92	5	α/α
	C	3	5	α	984.87 ⁷⁶	990.72	5	α/α
	D	4	5	α	984.97	993.62	5	α/α
	E	5	5	α	1048.66	1058.01	5	α/α
Salt Adj. Control	A	6	4	α	1071.53	1079.54	4	α/α
	B	7	5	α	1052.93	1061.54	5	α/α
	C	8	5	α	1064.84	1073.45	5	α/α
	D	9	4	α	1043.07	1049.76	4	α/α
	E	10	5	α	1064.16	1072.62	5	α/α
^{12.5} Site Control	A	11	5	α	1091.91	1101.08	5	α/α
	B	12	5	α	1064.50	1076.80	5	α/α
	C	13	5	α	1049.34	1059.94	5	α/α
	D	14	5	α	1042.41	1051.94	5	α/α
	E	15	5	α	1044.92	1054.24	5	α/α
²⁵ 12.5	A	16	5	α	1062.63	1071.82	5	α/α
	B	17	5	α	1057.02	1067.87	5	α/α
	C	18	4	α	1053.96	1062.16	4	α/α
	D	19	5	α	1075.82	1084.00	5	α/α
	E	20	4	α	1048.40	1054.77	4	α/α
⁵⁰ 25	A	21	5	α	1060.26	1068.97	5	α/α
	B	22	5	α	1043.92	1052.75	5	α/α
	C	23	5	α	1033.55	1045.75	5	α/α
	D	24	5	α	1043.28	1051.76	5	α/α
	E	25	5	α	1045.72	1055.67	5	α/α
50	A	26	4	α	1062.50	1067.64	4	α/α
	B	27	5	α	1065.37	1075.11	5	α/α
	C	28	5	α	1071.88	1081.34	5	α/α
	D	29	5	α	1042.72	1053.26	5	α/α
	E	30	4	α	1033.05	1039.49	4	α/α

Comments: 10% re-weigh : pan 1) * 2 weight 1083.99 2) * 5 weight 1058.12
3) * 11 weight 1101.15 4) * 25 weight 1055.60

Reviewed by: Date Reviewed: July 17, 2019

Topsmelt Toxicity Test Data Sheet

Dry Weight Data

Client: WDOE

Start Date & Time: Apr 23/19

Work Order #: 191279

Termination Date & Time: Apr 30/19

Sample ID: DW-3

AA Red

%. (v/v)	Rep	Pan No.	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
75	A	31	4	GA	1059.19	1064.06	4	GA/CA
	B	32	5	GA	1053.39	1059.69	5	GA/CA
	C	33	4	GA	1057.78	1065.54	4	GA/CA
	D	34	5	GA	1059.62	1067.49	5	GA/CA
	E	35	5	GA	1031.62	1037.23	5	GA/CA
100	A	36	3	GA	1098.81	1101.94	3	GA/CA
	B	37	3	GA	1081.21	1084.18	3	GA/CA
	C	38	5	GA	1075.00	1082.80	5	GA/CA
	D	39	3	GA	1044.74	1047.80	3	GA/CA
	E	40	4	GA	1044.64	1051.21	4	GA/CA
	A							
	B							
	C							
	D							
	E							
	A							
	B							
	C							
	D							
	E							
	A							
	B							
	C							
	D							
	E							
	A							
	B							
	C							
	D							
	E							

Comments:

Reviewed by:

W

Date Reviewed:

July 17, 2019

CETIS Summary Report

Report Date: 15 Jan-20 14:26 (p 1 of 3)
Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Batch ID: 19-0277-6459 Test Type: Growth-Survival (7d) Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30 Protocol: EPA/600/R-95/136 (1995) Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20 Species: Atherinops affinis Brine:
Test Length: 7d 1h Taxon: Actinopterygii Source: Aquatic Biosystems, CO Age: 12d

Sample ID: 17-4881-3663 Code: 683CC75F Project:
Sample Date: 19 Mar-19 Material: Diesel Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07 CAS (PC): Station: DW-3
Sample Age: 35d 13h Client: WA State Dept of Ecology

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓ NOEL	LOEL	TOEL	TU	PMSD	S
12-3914-5165	7d Survival Rate	Steel Many-One Rank Sum Test	3.14	>3.14	n/a		16.0%	1
07-1613-2626	Mean Dry Biomass-mg	Dunnett Multiple Comparison Test	2.12	3.14	2.58		30.1%	1
13-4199-4736	Mean Dry Weight-mg	Dunnett Multiple Comparison Test	2.12	3.14	2.58		26.4%	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓ Level	mg/L	95% LCL	95% UCL	TU	S
01-5682-8765	7d Survival Rate	Linear Interpolation (ICPIN)	EC5	0.7664	0.04748	2.863		1
			EC10	2.21	0.3861	2.49		
			EC15	2.445	1.609	2.962		
			EC20	2.697	2.104	n/a		
			EC25	2.968	2.331	n/a		
			EC40	>3.14	n/a	n/a		
			EC50	>3.14	n/a	n/a		
16-1829-3962	Mean Dry Biomass-mg	NLR: 3P Log-Logistic	IC5	0.9821	n/a	1.567		1
			IC10	1.311	n/a	1.912		
			IC15	1.567	0.4562	2.179		
			IC20	1.793	1.018	2.393		
			IC25	2.004	1.364	2.578		
			IC40	2.619	2.085	3.209		
			IC50	3.063	2.359	3.978		
18-3332-0016	Mean Dry Weight-mg	NLR: 3P Log-Logistic	IC5	0.984	n/a	1.67		1
			IC10	1.405	n/a	2.138		
			IC15	1.751	0.7986	2.473		
			IC20	2.067	1.321	2.738		
			IC25	2.37	1.726	3.001		
			IC40	3.297	2.292	4.511		
			IC50	3.999	2.378	6.725		

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits		Overlap	Decision
				Lower	Upper		
01-5682-8765	7d Survival Rate	Control Resp	1	0.8	>>	Yes	Passes Criteria
16-1829-3962	Mean Dry Biomass-mg	Control Resp	1.832	0.85	>>	Yes	Passes Criteria
07-1613-2626	Mean Dry Biomass-mg	PMSD	0.3013	<<	0.5	No	Passes Criteria

CETIS Summary Report

 Report Date: 15 Jan-20 14:26 (p 2 of 3)
 Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	SC	5	0.9200	0.7840	1.0000	0.8000	1.0000	0.0490	0.1095	11.91%	0.00%
0.16	N	5	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	-8.70%
0.2	XC	5	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	-8.70%
0.37		5	0.9200	0.7840	1.0000	0.8000	1.0000	0.0490	0.1095	11.91%	0.00%
0.62		5	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	-8.70%
1.29		5	0.9200	0.7840	1.0000	0.8000	1.0000	0.0490	0.1095	11.91%	0.00%
2.12		5	0.9200	0.7840	1.0000	0.8000	1.0000	0.0490	0.1095	11.91%	0.00%
3.14		5	0.7200	0.4979	0.9421	0.6000	1.0000	0.0800	0.1789	24.85%	21.74%

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	SC	5	1.615	1.413	1.817	1.338	1.722	0.0727	0.1626	10.06%	0.00%
0.16	N	5	1.832	1.32	2.344	1.192	2.26	0.1845	0.4126	22.52%	-13.42%
0.2	XC	5	2.037	1.712	2.362	1.834	2.46	0.1171	0.2618	12.85%	-26.10%
0.37		5	1.712	1.305	2.118	1.274	2.17	0.1464	0.3273	19.12%	-5.97%
0.62		5	1.927	1.544	2.31	1.696	2.44	0.138	0.3085	16.01%	-19.29%
1.29		5	1.653	1.072	2.234	1.028	2.108	0.2092	0.4677	28.30%	-2.33%
2.12		5	1.296	0.969	1.624	0.974	1.574	0.1179	0.2637	20.34%	19.74%
3.14		5	0.9012	0.4066	1.396	0.594	1.36	0.1781	0.3983	44.20%	44.20%

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	SC	5	1.762	1.593	1.931	1.673	2.003	0.06081	0.136	7.72%	0.00%
0.16	N	5	1.832	1.32	2.344	1.192	2.26	0.1845	0.4126	22.52%	-3.96%
0.2	XC	5	2.037	1.712	2.362	1.834	2.46	0.1171	0.2618	12.85%	-15.58%
0.37		5	1.857	1.544	2.17	1.592	2.17	0.1128	0.2522	13.58%	-5.40%
0.62		5	1.927	1.544	2.31	1.696	2.44	0.138	0.3085	16.01%	-9.34%
1.29		5	1.769	1.365	2.172	1.285	2.108	0.1452	0.3247	18.36%	-0.36%
2.12		5	1.423	1.006	1.839	1.122	1.94	0.1499	0.3352	23.56%	19.27%
3.14		5	1.211	0.859	1.563	0.99	1.642	0.1268	0.2836	23.42%	31.27%

N = lab control

SC = salt adjusted control

XC = site control (MW-11)

CETIS Summary Report

Report Date: 15 Jan-20 14:26 (p 3 of 3)
 Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	0.8000	1.0000	1.0000	0.8000	1.0000
0.16	N	1.0000	1.0000	1.0000	1.0000	1.0000
0.2	XC	1.0000	1.0000	1.0000	1.0000	1.0000
0.37		1.0000	1.0000	0.8000	1.0000	0.8000
0.62		1.0000	1.0000	1.0000	1.0000	1.0000
1.29		0.8000	1.0000	1.0000	1.0000	0.8000
2.12		0.8000	1.0000	0.8000	1.0000	1.0000
3.14		0.6000	0.6000	1.0000	0.6000	0.8000

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	1.602	1.722	1.722	1.338	1.692
0.16	N	2.108	2.26	1.192	1.73	1.87
0.2	XC	1.834	2.46	2.12	1.906	1.864
0.37		1.838	2.17	1.64	1.636	1.274
0.62		1.742	1.766	2.44	1.696	1.99
1.29		1.028	1.948	1.892	2.108	1.288
2.12		0.974	1.26	1.552	1.574	1.122
3.14		0.626	0.594	1.36	0.612	1.314

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	2.003	1.722	1.722	1.673	1.692
0.16	N	2.108	2.26	1.192	1.73	1.87
0.2	XC	1.834	2.46	2.12	1.906	1.864
0.37		1.838	2.17	2.05	1.636	1.592
0.62		1.742	1.766	2.44	1.696	1.99
1.29		1.285	1.948	1.892	2.108	1.61
2.12		1.218	1.26	1.94	1.574	1.122
3.14		1.043	0.99	1.36	1.02	1.642

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	4/5	5/5	5/5	4/5	5/5
0.16	N	5/5	5/5	5/5	5/5	5/5
0.2	XC	5/5	5/5	5/5	5/5	5/5
0.37		5/5	5/5	4/5	5/5	4/5
0.62		5/5	5/5	5/5	5/5	5/5
1.29		4/5	5/5	5/5	5/5	4/5
2.12		4/5	5/5	4/5	5/5	5/5
3.14		3/5	3/5	5/5	3/5	4/5

N = lab control

SC = salt adjusted control

XC = site control (MW-11)

CETIS Analytical Report

Report Date: 14 Jan-20 16:50 (p 1 of 2)
 Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 01-5682-8765	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 14 Jan-20 16:48	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 19-0277-6459	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20	Species: Atherinops affinis	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 12d
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 13h	Client: WA State Dept of Ecology	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	180712	200	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits		Overlap	Decision
Attribute	Test Stat	Lower	Upper		
Control Resp	1	0.8	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC5	0.7664	0.04748	2.863
EC10	2.21	0.3861	2.49
EC15	2.445	1.609	2.962
EC20	2.697	2.104	n/a
EC25	2.968	2.331	n/a
EC40	>3.14	n/a	n/a
EC50	>3.14	n/a	n/a

7d Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	Code	Count	Mean	Min	Max	Std Dev	CV%	%Effect	A/B	Mean	%Effect
0.16	N	5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	25/25	1	0.0%
0.37		5	0.9200	0.8000	1.0000	0.1095	11.91%	8.0%	23/25	0.96	4.0%
0.62		5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	25/25	0.96	4.0%
1.29		5	0.9200	0.8000	1.0000	0.1095	11.91%	8.0%	23/25	0.92	8.0%
2.12		5	0.9200	0.8000	1.0000	0.1095	11.91%	8.0%	23/25	0.92	8.0%
3.14		5	0.7200	0.6000	1.0000	0.1789	24.85%	28.0%	18/25	0.72	28.0%

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	1.0000	1.0000	1.0000	1.0000	1.0000
0.37		1.0000	1.0000	0.8000	1.0000	0.8000
0.62		1.0000	1.0000	1.0000	1.0000	1.0000
1.29		0.8000	1.0000	1.0000	1.0000	0.8000
2.12		0.8000	1.0000	0.8000	1.0000	1.0000
3.14		0.6000	0.6000	1.0000	0.6000	0.8000

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	5/5	5/5	5/5	5/5	5/5
0.37		5/5	5/5	4/5	5/5	4/5
0.62		5/5	5/5	5/5	5/5	5/5
1.29		4/5	5/5	5/5	5/5	4/5
2.12		4/5	5/5	4/5	5/5	5/5
3.14		3/5	3/5	5/5	3/5	4/5

N = Lab Control

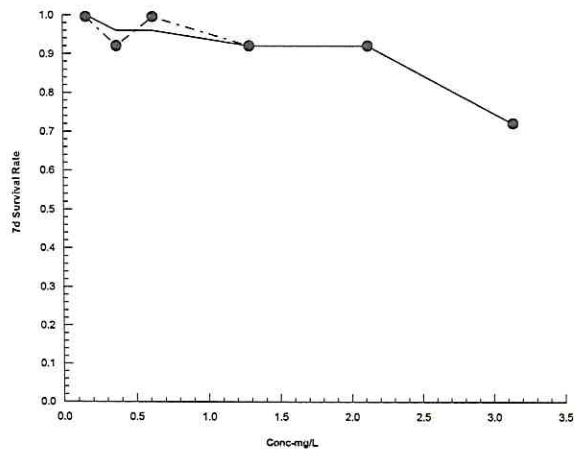
CETIS Analytical Report

Report Date: 14 Jan-20 16:50 (p 2 of 2)
Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test Nautilus Environmental

Analysis ID: 01-5682-8765	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 14 Jan-20 16:48	Analysis: Linear Interpolation (ICPIN)	Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 14 Jan-20 16:50 (p 1 of 2)
Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 16-1829-3962	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 14 Jan-20 16:49	Analysis: Nonlinear Regression (NLR)	Status Level: 1
Batch ID: 19-0277-6459	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20	Species: Atherinops affinis	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 12d
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 13h	Client: WA State Dept of Ecology	

Non-Linear Regression Options

Model Name and Function	Weighting Function	PTBS Function	X Trans	Y Trans
3P Log-Logistic: $\mu = \alpha / [1 + (x/\delta)^\gamma]$	Normal [$\omega=1$]	Off [$\mu^*=\mu$]	None	None

Regression Summary

Iters	Log LL	AICc	BIC	Adj R2	Optimize	F Stat	Critical	P-Value	Decision($\alpha:5\%$)
8	32.06	-57.19	-53.91	0.4745	Yes	0.3524	3.009	0.7878	Non-Significant Lack of Fit

Point Estimates

Level	mg/L	95% LCL	95% UCL
IC5	0.9821	n/a	1.567
IC10	1.311	n/a	1.912
IC15	1.567	0.4562	2.179
IC20	1.793	1.018	2.393
IC25	2.004	1.364	2.578
IC40	2.619	2.085	3.209
IC50	3.063	2.359	3.978

Test Acceptability Criteria

		TAC Limits			
Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	1.832	0.85	>>	Yes	Passes Criteria

Regression Parameters

Parameter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision($\alpha:5\%$)
α	1.831	0.1024	1.621	2.041	17.89	<1.0E-37	Significant Parameter
γ	2.589	1.13	0.2697	4.908	2.29	0.0300	Significant Parameter
δ	3.063	0.384	2.275	3.851	7.976	<1.0E-37	Significant Parameter

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision($\alpha:5\%$)
Model	75.97	25.32	3	199.8	<1.0E-37	Significant
Lack of Fit	0.1444	0.04812	3	0.3524	0.7878	Non-Significant
Pure Error	3.278	0.1366	24			
Residual	3.422	0.1267	27			

Residual Analysis

Attribute	Method	Test Stat	Critical	P-Value	Decision($\alpha:5\%$)
Variances	Bartlett Equality of Variance Test	1.598	11.07	0.9015	Equal Variances
	Mod Levene Equality of Variance	0.2746	2.773	0.9211	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9688	0.9303	0.5071	Normal Distribution
	Anderson-Darling A2 Normality Te	0.3541	2.492	0.4668	Normal Distribution

CETIS Analytical Report

Report Date: 14 Jan-20 16:50 (p 2 of 2)
Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 16-1829-3962
Analyzed: 14 Jan-20 16:49

Endpoint: Mean Dry Biomass-mg
Analysis: Nonlinear Regression (NLR)

CETIS Version: CETISv1.9.4
Status Level: 1

Mean Dry Biomass-mg Summary

Calculated Variate

Conc-mg/L	Code	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0.16	N	5	1.832	1.192	2.26	0.1845	0.4126	22.52%	0.0%
0.37		5	1.712	1.274	2.17	0.1464	0.3273	19.12%	6.57%
0.62		5	1.927	1.696	2.44	0.138	0.3085	16.01%	-5.17%
1.29		5	1.653	1.028	2.108	0.2092	0.4677	28.30%	9.78%
2.12		5	1.296	0.974	1.574	0.1179	0.2637	20.34%	29.24%
3.14		5	0.9012	0.594	1.36	0.1781	0.3983	44.20%	50.81%

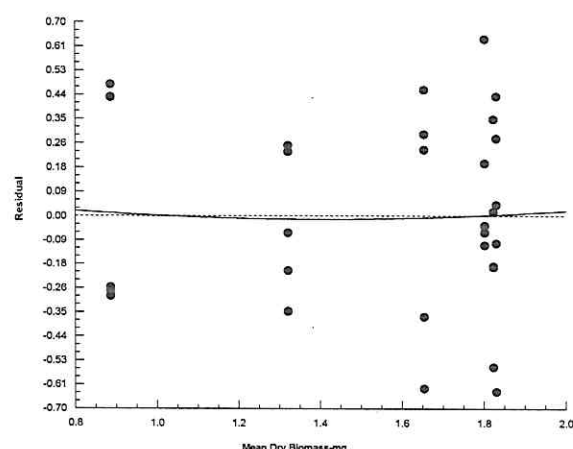
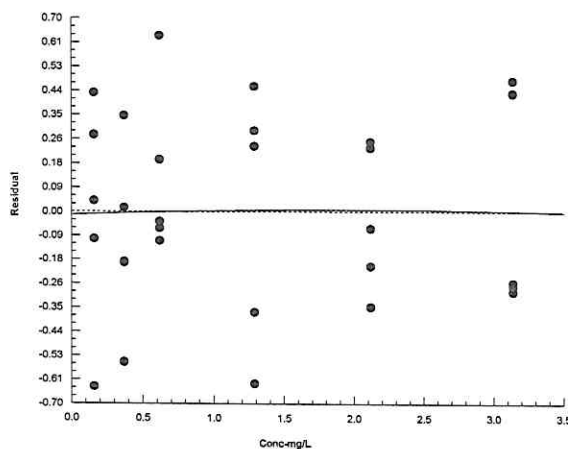
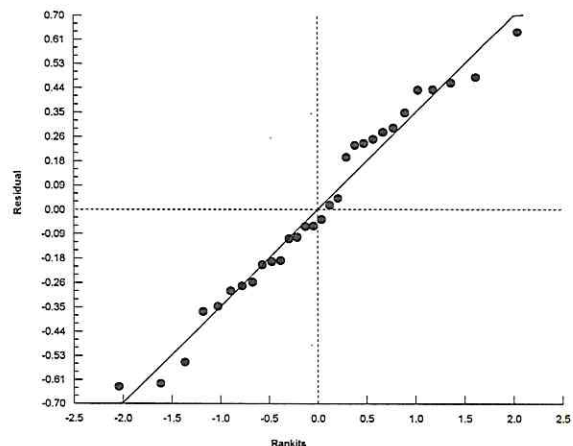
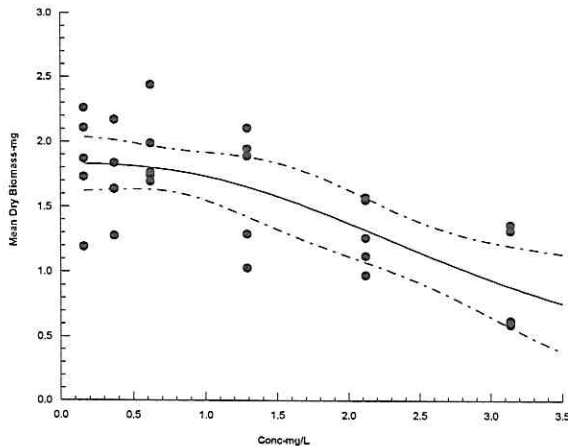
Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	2.108	2.26	1.192	1.73	1.87
0.37		1.838	2.17	1.64	1.636	1.274
0.62		1.742	1.766	2.44	1.696	1.99
1.29		1.028	1.948	1.892	2.108	1.288
2.12		0.974	1.26	1.552	1.574	1.122
3.14		0.626	0.594	1.36	0.612	1.314

N = lab control

Graphics

Model: 3P Log-Logistic: $\mu = \alpha / [1 + (x/\delta)^\gamma]$ Distribution: Normal [$\omega = 1$]



CETIS Analytical Report

Report Date: 15 Jan-20 12:46 (p 1 of 2)
 Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 18-3332-0016	Endpoint: Mean Dry Weight-mg	CETIS Version: CETISv1.9.4
Analyzed: 15 Jan-20 12:46	Analysis: Nonlinear Regression (NLR)	Status Level: 1
Batch ID: 19-0277-6459	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20	Species: Atherinops affinis	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 12d
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 13h	Client: WA State Dept of Ecology	

Non-Linear Regression Options

Model Name and Function	Weighting Function	PTBS Function	X Trans	Y Trans
3P Log-Logistic: $\mu = \alpha / [1 + (x/\delta)^\gamma]$	Normal [$\omega=1$]	Off [$\mu^* = \mu$]	None	None

Regression Summary

Iters	Log LL	AICc	BIC	Adj R2	Optimize	F Stat	Critical	P-Value	Decision($\alpha:5\%$)
14	36.22	-65.52	-62.23	0.3883	Yes	0.2675	3.009	0.8482	Non-Significant Lack of Fit

Point Estimates

Level	mg/L	95% LCL	95% UCL
IC5	0.984	n/a	1.67
IC10	1.405	n/a	2.138
IC15	1.751	0.7986	2.473
IC20	2.067	1.321	2.738
IC25	2.37	1.726	3.001
IC40	3.297 73.17	2.292	4.511
IC50	3.999 73.17	2.378	6.725

Regression Parameters

Parameter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision($\alpha:5\%$)
α	1.889	0.09771	1.688	2.089	19.33	<1.0E-37	Significant Parameter
γ	2.1	1.108	-0.1744	4.374	1.895	0.0689	Non-Significant Parameter
δ	3.999	0.8705	2.213	5.785	4.594	9.1E-05	Significant Parameter

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision($\alpha:5\%$)
Model	85.6	28.53	3	297.2	<1.0E-37	Significant
Lack of Fit	0.08388	0.02796	3	0.2675	0.8482	Non-Significant
Pure Error	2.509	0.1045	24			
Residual	2.593	0.09602	27			

Residual Analysis

Attribute	Method	Test Stat	Critical	P-Value	Decision($\alpha:5\%$)
Variances	Bartlett Equality of Variance Test	1.037	11.07	0.9595	Equal Variances
	Mod Levene Equality of Variance	0.1365	2.773	0.9816	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9787	0.9303	0.7888	Normal Distribution
	Anderson-Darling A2 Normality Te	0.3562	2.492	0.4618	Normal Distribution

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Calculated Variate						
			Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0.16	N	5	1.832	1.192	2.26	0.1845	0.4126	22.52%	0.0%
0.37		5	1.857	1.592	2.17	0.1128	0.2522	13.58%	-1.38%
0.62		5	1.927	1.696	2.44	0.138	0.3085	16.01%	-5.17%
1.29		5	1.769	1.285	2.108	0.1452	0.3247	18.36%	3.46%
2.12		5	1.423	1.122	1.94	0.1499	0.3352	23.56%	22.34%
3.14		5	1.211	0.99	1.642	0.1268	0.2836	23.42%	33.89%

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 18-3332-0016 Endpoint: Mean Dry Weight-mg
 Analyzed: 15 Jan-20 12:46 Analysis: Nonlinear Regression (NLR)

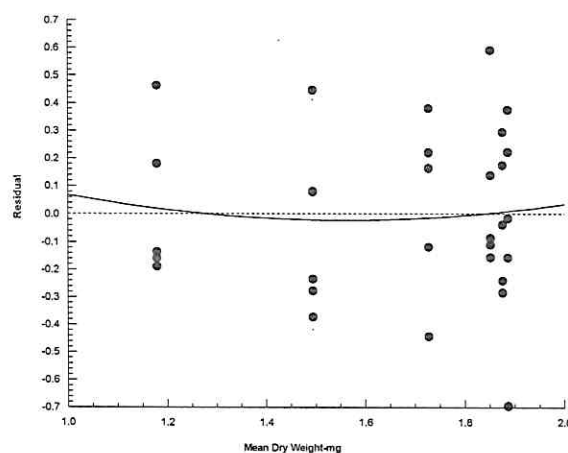
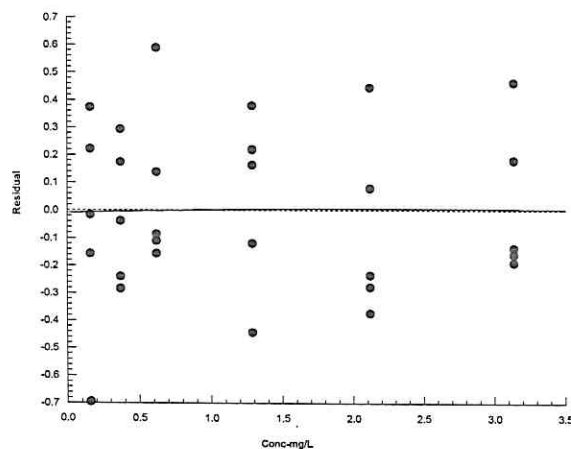
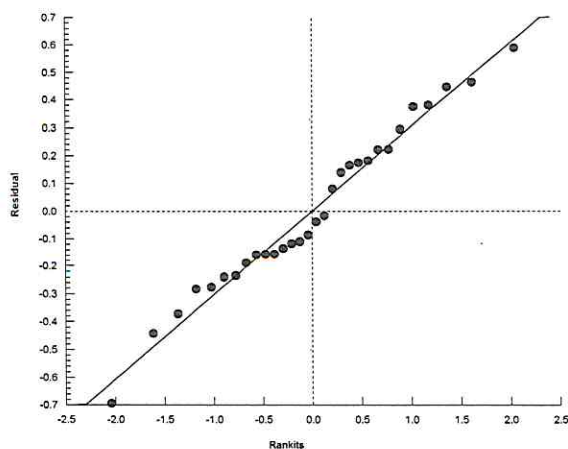
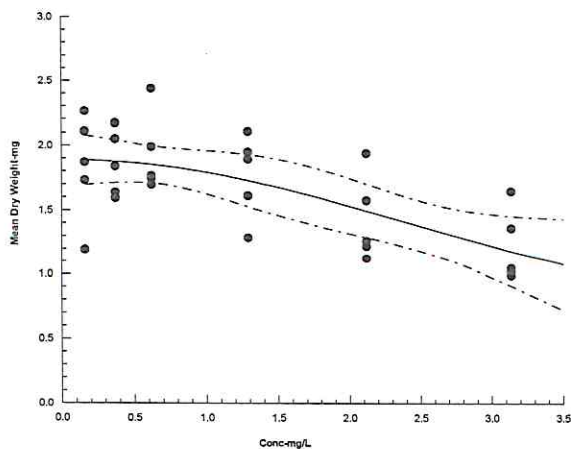
CETIS Version: CETISv1.9.4
 Status Level: 1

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	2.108	2.26	1.192	1.73	1.87
0.37		1.838	2.17	2.05	1.636	1.592
0.62		1.742	1.766	2.44	1.696	1.99
1.29		1.285	1.948	1.892	2.108	1.61
2.12		1.218	1.26	1.94	1.574	1.122
3.14		1.043	0.99	1.36	1.02	1.642

Graphics

Model: 3P Log-Logistic: $\mu = \alpha / [1 + (x/\delta)^\gamma]$ Distribution: Normal [$\omega = 1$]



CETIS Analytical Report

Report Date: 15 Jan-20 12:47 (p 1 of 2)
Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 01-7592-9747	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 15 Jan-20 12:47	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 19-0277-6459	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20	Species: Atherinops affinis	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 12d
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 13h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	Salt Control passed 7d survival rate	10.75%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Salt Control	1.633	1.86	0.109	8	CDF	0.0706	Non-Significant Effect

Test Acceptability Criteria

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.92	0.8	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0226832	0.0226832	1	2.667	0.1411	Non-Significant Effect
Error	0.0680495	0.0085062	8			
Total	0.0907326		9			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Levene Equality of Variance Test	96	11.26	9.9E-06	Unequal Variances
Variances	Mod Levene Equality of Variance Test	3	13.75	0.1340	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.8142	0.7411	0.0215	Normal Distribution

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	SC	5	0.9200	0.7840	1.0000	1.0000	0.8000	1.0000	0.0490	11.91%	0.00%
0.16	N	5	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-8.70%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	SC	5	1.25	1.088	1.412	1.345	1.107	1.345	0.05833	10.43%	0.00%
0.16	N	5	1.345	1.345	1.346	1.345	1.345	1.345	0	0.00%	-7.62%

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	0.8000	1.0000	1.0000	0.8000	1.0000
0.16	N	1.0000	1.0000	1.0000	1.0000	1.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	1.107	1.345	1.345	1.107	1.345
0.16	N	1.345	1.345	1.345	1.345	1.345

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	4/5	5/5	5/5	4/5	5/5
0.16	N	5/5	5/5	5/5	5/5	5/5

N=10ab Control, SC= salt adjusted control
001-406-995-6

CETIS™ v1.9.4.11

Analyst: *[Signature]*

QA: *[Signature]*

[Handwritten notes and signatures]

CETIS Analytical Report

Report Date: 15 Jan-20 12:47 (p 2 of 2)
Test Code/ID: 191279b / 16-4104-7007

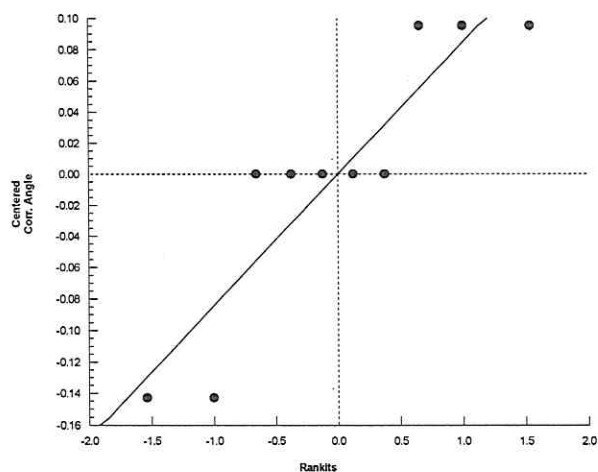
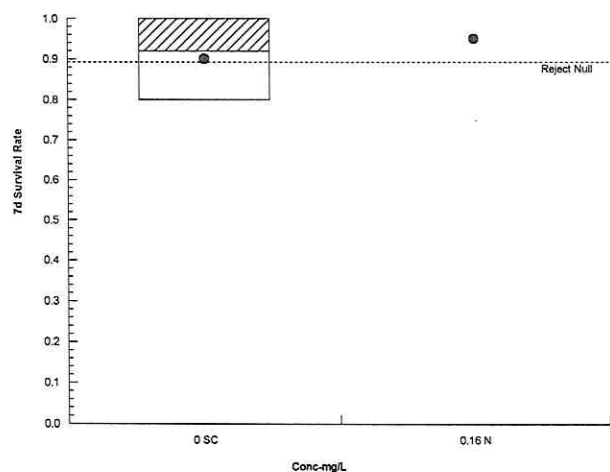
Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 01-7592-9747 Endpoint: 7d Survival Rate
Analyzed: 15 Jan-20 12:47 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 15 Jan-20 12:47 (p 1 of 2)
 Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 10-5445-1064	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 15 Jan-20 12:47	Analysis: Nonparametric-Two Sample	Status Level: 1
Batch ID: 19-0277-6459	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20	Species: Atherinops affinis	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 12d
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 13h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	Comparison Result
Angular (Corrected)	C > T	Site Control passed 7d survival rate

Wilcoxon Rank Sum Two-Sample Test

Control	vs	Control II	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Site Control	27.5	n/a	1	8	Exact	1.0000	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0	0	1	65540	<1.0E-37	Significant Effect
Error	0	0	8			
Total	0		9			

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.16	N	5	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
0.2	XC	5	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.16	N	5	1.345	1.345	1.346	1.345	1.345	1.345	0	0.00%	0.00%
0.2	XC	5	1.345	1.345	1.346	1.345	1.345	1.345	0	0.00%	0.00%

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	1.0000	1.0000	1.0000	1.0000	1.0000
0.2	XC	1.0000	1.0000	1.0000	1.0000	1.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	1.345	1.345	1.345	1.345	1.345
0.2	XC	1.345	1.345	1.345	1.345	1.345

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	5/5	5/5	5/5	5/5	5/5
0.2	XC	5/5	5/5	5/5	5/5	5/5

N = lab control
 XC = site control (MW-11)

CETIS Analytical Report

Report Date: 15 Jan-20 12:47 (p 2 of 2)
Test Code/ID: 191279b / 16-4104-7007

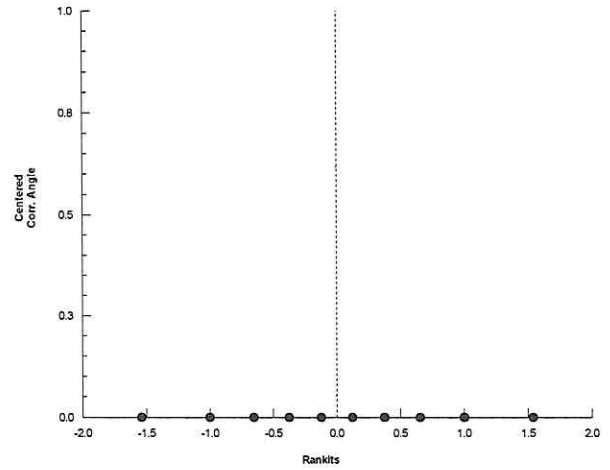
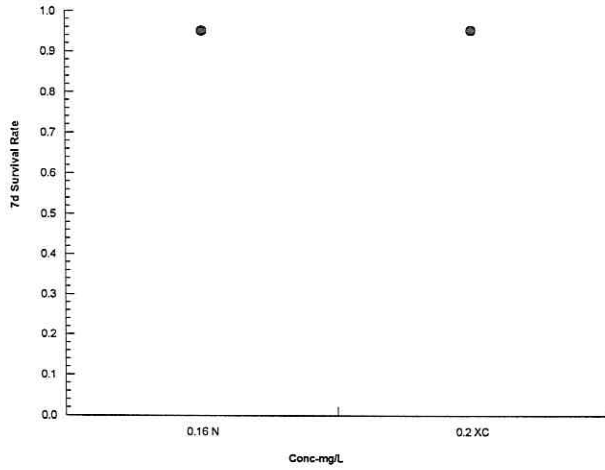
Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 10-5445-1064 Endpoint: 7d Survival Rate
Analyzed: 15 Jan-20 12:47 Analysis: Nonparametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 15 Jan-20 12:47 (p 1 of 1)
Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 17-9641-2332	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 15 Jan-20 12:47	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 19-0277-6459	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20	Species: Atherinops affinis	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 12d
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 13h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	Comparison Result	PMSD
Untransformed	C > T	Site Control passed mean dry biomass-mg	22.18%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Site Control	-0.9372	1.86	0.406	8	CDF	0.8120	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.104847	0.104847	1	0.8783	0.3761	Non-Significant Effect
Error	0.954969	0.119371	8			
Total	1.05982		9			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	2.484	23.15	0.3998	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9386	0.7411	0.5381	Normal Distribution

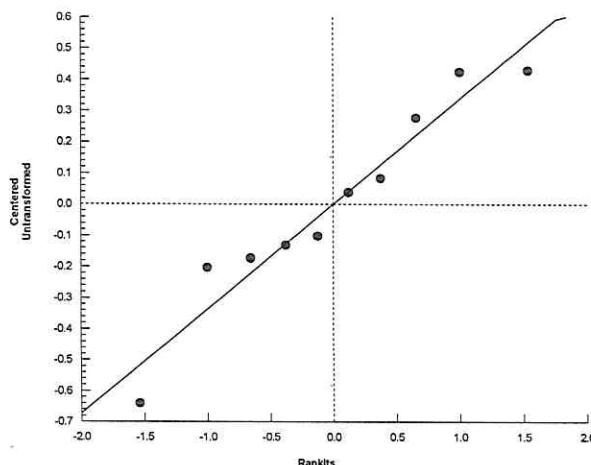
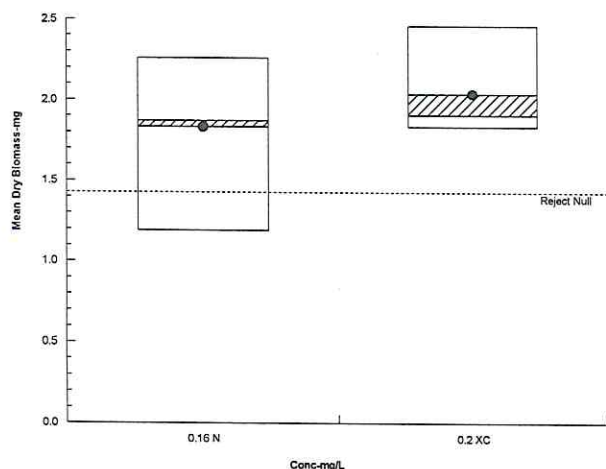
Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.16	N	5	1.832	1.32	2.344	1.87	1.192	2.26	0.1845	22.52%	0.00%
0.2	XC	5	2.037	1.712	2.362	1.906	1.834	2.46	0.1171	12.85%	-11.18%

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.16	N	2.108	2.26	1.192	1.73	1.87
0.2	XC	1.834	2.46	2.12	1.906	1.864

Graphics



N = lab control
XC = site control (MW-11)
001-406-995-6

CETIS™ v1.9.4.11

Analyst: *[Signature]*

QA: *[Signature]* 1/16/20

CETIS Analytical Report

Report Date: 15 Jan-20 12:47 (p 1 of 2)
 Test Code/ID: 191279b / 16-4104-7007

Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 01-9151-4622	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 15 Jan-20 12:47	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 19-0277-6459	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 12:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 30 Apr-19 13:20	Species: Atherinops affinis	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: 12d
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 13h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	Comparison Result	PMSD
Untransformed	C > T	Salt Control passed mean dry biomass-mg	20.13%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Salt Control	1.093	1.86	0.369	8	CDF	0.1531	Non-Significant Effect

Test Acceptability Criteria

TAC Limits

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	1.615	0.85	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.117508	0.117508	1	1.195	0.3061	Non-Significant Effect
Error	0.786545	0.0983181	8			
Total	0.904052		9			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	6.44	23.15	0.0987	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9311	0.7411	0.4586	Normal Distribution

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	SC	5	1.615	1.413	1.817	1.692	1.338	1.722	0.0727	10.06%	0.00%
0.16	N	5	1.832	1.32	2.344	1.87	1.192	2.26	0.1845	22.52%	-13.42%

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	SC	1.602	1.722	1.722	1.338	1.692
0.16	N	2.108	2.26	1.192	1.73	1.87

N = ^{K2L} ~~neg~~ lab control

SC = salt adjusted control

CETIS Analytical Report

Report Date: 15 Jan-20 12:47 (p 2 of 2)
Test Code/ID: 191279b / 16-4104-7007

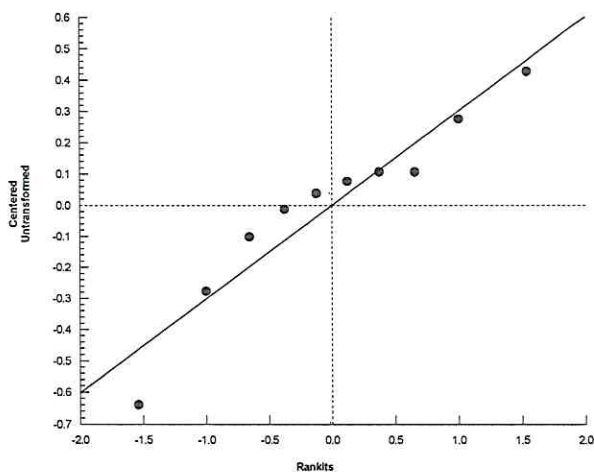
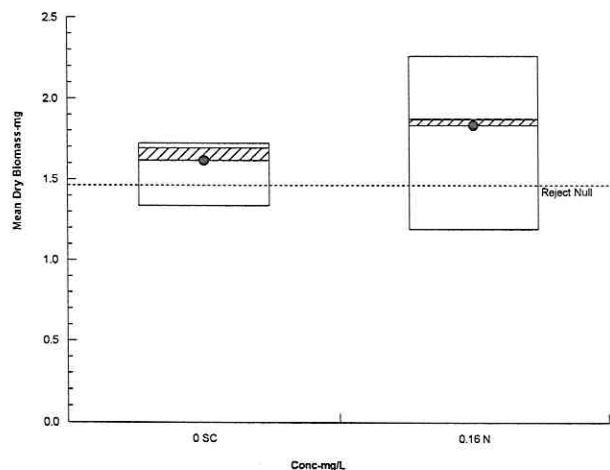
Pacific Topsmelt 7-d Survival and Growth Test

Nautilus Environmental

Analysis ID: 01-9151-4622 Endpoint: Mean Dry Biomass-mg
Analyzed: 15 Jan-20 12:47 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



APPENDIX B – *Strongylocentrotus purpuratus* Toxicity Test Data

Echinoderm Fertilization Test Summary Sheet

Client: WDOE

Start Date/Time: February 19, 2019 091745h

Work Order No.: 191280

Test Species: *S. purpuratus*

Sample Information:

Sample ID: DW-3 (magenta)

Sample Date: January 29, 2019

Date Received: January 30, 2019

Sample Volume: 5 x 20L

Dilution Water:

Type: Natural Seawater

Source: Vancouver Aquarium

Test Organism Information:

Batch No.: 021919

Source: Enthelby, San Diego, CA

Sperm:Egg Ratio: 200:1

Exposure Period: 20 min: 20 min

Reference Toxicant Results:

Reference Toxicant ID: 3PUS06

Stock Solution ID: 18C006

Date Initiated: February 19, 2019

IC50 (95% CL): 16.4 (15.2-18.6) ug/L Cu

Reference Toxicant Mean \pm 2 SD: 18.4 (8.4-40.1) ug/L Cu

Reference Toxicant CV (%): 41

Test Results: The fertilization rate ~~is~~ IC25 is estimated to be >100% (v/v)
The fertilization rate IC50 is estimated to be >100% (v/v)

Reviewed by: JCH

Date reviewed: July 8/19

Echinoderm Fertilization WQ Data Sheet

Client : WDOE

Analysts: YHL

Sample ID: DW-3 (orange-finder)

Test Date & Time: February 19, 2019 @ 1745h

WO No: 191280

Test Species: S. purpuratus

Initial Water Quality

	100% sample
Temp (°C)	5.5
DO (mg/L)	9.6
pH	7.6
Salinity	0.2

Brine/Salt Adjusted Water Quality

	100% sample
Temp (°C)	13.0
DO (mg/L)	8.3
pH	7.7
Salinity	29

Concentration % (v/v)	Water Quality			
	Temp. (°C)	DO (mg/L)	pH	Salinity (ppt)
Seawater Control	13.0	8.2	7.6	29
Salt Control	13.0	8.3	7.8	29
Site Control - ^{HW-1}	13.0	8.3	7.8	31
0.1	13.0	8.2	7.6	29
0.39	13.0	8.3	7.6	29
1.6	13.0	8.3	7.6	29
6.3	13.0	8.3	7.6	29
25	13.0	8.3	7.7	29
100	13.0	8.3	7.7	29
Analyst Initials	YHL	YHL	YHL	YHL

Thermometer: CER-3 DO meter: 313 pH meter: 313 Salinity: 313
probe probe probe

Sample Description: DW-3 - clear, yellow, some particulates, no odour.
HW-1 - clear, light yellow, no particulates, no odour.

Echinoderm Source: Enthalpy, San Diego, CA Date Received: February 19, 2019

Seawater Source: Vancouver Aquarium Date Collected: February 15, 2019

Comments: _____

Reviewed: YHL Date Reviewed: July 8/19

Echinoderm Fertilization Toxicity Test Data Sheet

Fertilized Egg Counts

Client: WDOE Start Date/Time: February 19, 2019 @ 1745h
 Sample ID: DW-3 (range finder) Test species: S. purpuratus
 Work Order #: 191280 Test set up by: YU
 Sample ID: 191280 Test Duration: 20 min: 20 min

Concentration (% v/v)	Rep	No. Fertilized Eggs	No. Unfertilized Eggs	Comments	Initials
Seawater Control	A	89	11		NW
	B	89	11		
	C	81	19		
	D	85	15		
Salt Control	A	95	5		
	B	92	8		
	C	90	10		
	D	88	12		
Site Control	A	94	6		
	B	90	10		
	C	88	12		
	D	91	9		
0.1	A	84	16		
	B	98	2		
	C	85	15		
	D	82	18		
0.39	A	98	2		
	B	95	5		
	C	91	9		
	D	92	8		
1.6	A	94	6		
	B	90	10		
	C	90	10		
	D	91	9		
6.3	A	89	11		
	B	90	10		
	C	91	9		
	D	86	14		
25	A	95	5		
	B	87	13		
	C	88	12		
	D	90	10		

Comments: _____

Reviewed by: Jul

Date Reviewed: July 8/19

Echinoderm Fertilization Toxicity Test Data Sheet

Fertilized Egg Counts

Client: WDOE Start Date/Time: February 19, 2019 @ 1745h
 Sample ID: OW-3 (range finder) Test species: S. purpuratus
 Work Order #: 191280 Test set up by: YMC
 Sample ID: W08 Test Duration: 20 min; 20 min

Concentration (% v/v)	Rep	No. Fertilized Eggs	No. Unfertilized Eggs	Comments	Initials
100	A	91	9		YMC
	B	84	16		↓
	C	85	15		
	D	84	16		
	A				
	B				
	C				
	D				
EGG Control	A	0	100		YMC
	B	0	100		↓
	C	0	100		
	D	0	100		
Effluent Control	A	0	100		
	B	0	100		
	C	0	100		
	D	0	100		↓
	A				
	B				
	C				
	D				
	A				
	B				
	C				
	D				
	A				
	B				
	C				
	D				

Comments: _____

Reviewed by: JCH Date Reviewed: July 8/19

CETIS Summary Report

Report Date: 12 Aug-19 12:03 (p 1 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Batch ID: 19-5334-0495	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 19 Feb-19 17:45	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 19 Feb-19 18:25	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m	Taxon: Echinoidea	Source: San Diego Lab Age:
Sample ID: 04-0118-8251	Code: 191282	Project:
Sample Date: 29 Jan-19	Material: Ground Water	Source: WDOE
Receipt Date: 30 Jan-19	CAS (PC):	Station: NWTPH-Dx
Sample Age: 21d 18h	Client: WDOE	

Single Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
09-1544-4894	Fertilization Rate	Equal Variance t Two-Sample Test	0.9640	Salt Control passed fertilization rate	1
19-1223-1912	Fertilization Rate	Equal Variance t Two-Sample Test	0.9605	Site Control passed fertilization rate	1

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓ NOEL	LOEL	TOEL	TU	PMSD	S
03-5399-0733	Fertilization Rate	Dunnett Multiple Comparison Test	100	>100	n/a	1	11.1%	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓ Level	%	95% LCL	95% UCL	TU	S
02-8166-1841	Fertilization Rate	Linear Interpolation (ICPIN)	IC1	34.06	n/a	n/a	2.936	1
			IC5	>100	n/a	n/a	<1	
			IC10	>100	n/a	n/a	<1	
			IC15	>100	n/a	n/a	<1	
			IC20	>100	n/a	n/a	<1	
			IC25	>100	n/a	n/a	<1	
			IC40	>100	n/a	n/a	<1	
			IC50	>100	n/a	n/a	<1	

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits		Overlap	Decision
				Lower	Upper		
02-8166-1841	Fertilization Rate	Control Resp	0.86	0.7	>>	Yes	Passes Criteria
03-5399-0733	Fertilization Rate	Control Resp	0.86	0.7	>>	Yes	Passes Criteria
09-1544-4894	Fertilization Rate	Control Resp	0.9125	0.7	>>	Yes	Passes Criteria
	Fertilization Rate	Control Resp	0.86	0.7	>>	Yes	Passes Criteria
19-1223-1912	Fertilization Rate	Control Resp	0.9075	0.7	>>	Yes	Passes Criteria
	Fertilization Rate	Control Resp	0.86	0.7	>>	Yes	Passes Criteria

Fertilization Rate Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	SC	4	0.9125	0.8650	0.9600	0.8800	0.9500	0.0149	0.0299	3.27%	0.00%
0	N	4	0.8600	0.7991	0.9209	0.8100	0.8900	0.0192	0.0383	4.45%	5.75%
0	XC	4	0.9075	0.8677	0.9473	0.8800	0.9400	0.0125	0.0250	2.75%	0.55%
0.1		4	0.8725	0.7567	0.9883	0.8200	0.9800	0.0364	0.0727	8.34%	4.38%
0.39		4	0.9400	0.8897	0.9903	0.9100	0.9800	0.0158	0.0316	3.36%	-3.01%
1.6		4	0.9125	0.8824	0.9426	0.9000	0.9400	0.0095	0.0189	2.07%	0.00%
6.3		4	0.8900	0.8556	0.9244	0.8600	0.9100	0.0108	0.0216	2.43%	2.47%
25		4	0.9000	0.8434	0.9566	0.8700	0.9500	0.0178	0.0356	3.95%	1.37%
100		4	0.8600	0.8064	0.9136	0.8400	0.9100	0.0168	0.0337	3.91%	5.75%

CETIS Summary Report

Report Date: 12 Aug-19 12:03 (p 2 of 2)
 Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Fertilization Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	SC	0.9500	0.9200	0.9000	0.8800
0	N	0.8900	0.8900	0.8100	0.8500
0	XC	0.9400	0.9000	0.8800	0.9100
0.1		0.8400	0.9800	0.8500	0.8200
0.39		0.9800	0.9500	0.9100	0.9200
1.6		0.9400	0.9000	0.9000	0.9100
6.3		0.8900	0.9000	0.9100	0.8600
25		0.9500	0.8700	0.8800	0.9000
100		0.9100	0.8400	0.8500	0.8400

Fertilization Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	SC	95/100	92/100	90/100	88/100
0	N	89/100	89/100	81/100	85/100
0	XC	94/100	90/100	88/100	91/100
0.1		84/100	98/100	85/100	82/100
0.39		98/100	95/100	91/100	92/100
1.6		94/100	90/100	90/100	91/100
6.3		89/100	90/100	91/100	86/100
25		95/100	87/100	88/100	90/100
100		91/100	84/100	85/100	84/100

CETIS Analytical Report

Report Date: 05 Jul-19 13:04 (p 1 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 02-8166-1841	Endpoint: Fertilization Rate	CETIS Version: CETISv1.9.4
Analyzed: 05 Jul-19 13:03	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 19-5334-0495	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 19 Feb-19 17:45	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 19 Feb-19 18:25	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m (20 min 20 min)	Taxon: Echinoidea	Source: San Diego Lab Age:
Sample ID: 04-0118-8251	Code: 191282	Project:
Sample Date: 29 Jan-19	Material: Ground Water	Source: WDOE
Receipt Date: 30 Jan-19	CAS (PC):	Station: NWTPH-Dx
Sample Age: 21d 18h	Client: WDOE	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	1549091	200	Yes	Two-Point Interpolation

Test Acceptability Criteria

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.86	0.7	>>	Yes	Passes Criteria

Point Estimates

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC1	34.06	n/a	n/a	2.936	n/a	n/a
IC5	>100	n/a	n/a	<1	n/a	n/a
IC10	>100	n/a	n/a	<1	n/a	n/a
IC15	>100	n/a	n/a	<1	n/a	n/a
IC20	>100	n/a	n/a	<1	n/a	n/a
IC25	>100	n/a	n/a	<1	n/a	n/a
IC40	>100	n/a	n/a	<1	n/a	n/a
IC50	>100	n/a	n/a	<1	n/a	n/a

Fertilization Rate Summary

Conc-%	Code	Count	Calculated Variate(A/B)							Isotonic Variate	
			Mean	Min	Max	Std Dev	CV%	%Effect	A/B	Mean	%Effect
0	N	4	0.8600	0.8100	0.8900	0.0383	4.45%	0.0%	344/400	0.8963	0.0%
0.1		4	0.8725	0.8200	0.9800	0.0727	8.34%	-1.45%	349/400	0.8963	0.0%
0.39		4	0.9400	0.9100	0.9800	0.0316	3.36%	-9.3%	376/400	0.8963	0.0%
1.6		4	0.9125	0.9000	0.9400	0.0189	2.07%	-6.11%	365/400	0.8963	0.0%
6.3		4	0.8900	0.8600	0.9100	0.0216	2.43%	-3.49%	356/400	0.895	0.14%
25		4	0.9000	0.8700	0.9500	0.0356	3.95%	-4.65%	360/400	0.895	0.14%
100		4	0.8600	0.8400	0.9100	0.0337	3.92%	0.0%	344/400	0.86	4.05%

Fertilization Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	0.8900	0.8900	0.8100	0.8500
0.1		0.8400	0.9800	0.8500	0.8200
0.39		0.9800	0.9500	0.9100	0.9200
1.6		0.9400	0.9000	0.9000	0.9100
6.3		0.8900	0.9000	0.9100	0.8600
25		0.9500	0.8700	0.8800	0.9000
100		0.9100	0.8400	0.8500	0.8400

CETIS Analytical Report

Report Date: 05 Jul-19 13:04 (p 2 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

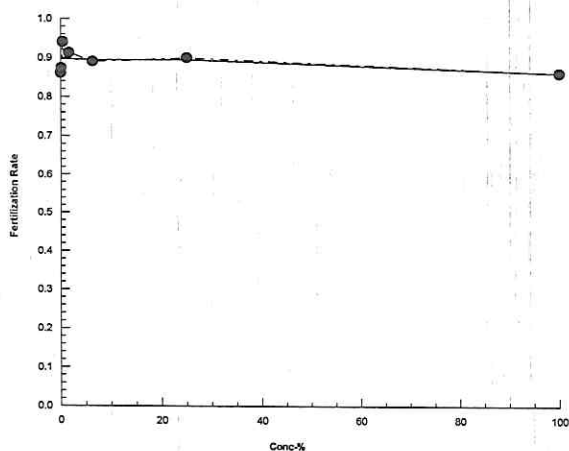
Analysis ID: 02-8166-1841 Endpoint: Fertilization Rate
Analyzed: 05 Jul-19 13:03 Analysis: Linear Interpolation (ICPIN)

CETIS Version: CETISv1.9.4
Status Level: 1

Fertilization Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	89/100	89/100	81/100	85/100
0.1		84/100	98/100	85/100	82/100
0.39		98/100	95/100	91/100	92/100
1.6		94/100	90/100	90/100	91/100
6.3		89/100	90/100	91/100	86/100
25		95/100	87/100	88/100	90/100
100		91/100	84/100	85/100	84/100

Graphics



CETIS Analytical Report

Report Date: 05 Jul-19 13:04 (p 1 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 09-1544-4894	Endpoint: Fertilization Rate	CETIS Version: CETISv1.9.4
Analyzed: 05 Jul-19 13:03	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 19-5334-0495	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 19 Feb-19 17:45	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 19 Feb-19 18:25	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m	Taxon: Echinoidea	Source: San Diego Lab Age:
Sample ID: 04-0118-8251	Code: 191282	Project:
Sample Date: 29 Jan-19	Material: Ground Water	Source: WDOE
Receipt Date: 30 Jan-19	CAS (PC):	Station: NWTPH-Dx
Sample Age: 21d 18h	Client: WDOE	

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	Salt Control passed fertilization rate	6.30%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Salt Control	-2.181	1.943	0.075	6	CDF	0.9640	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.9125	0.7	>>	Yes	Passes Criteria
Control Resp	0.86	0.7	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0142041	0.0142041	1	4.757	0.0720	Non-Significant Effect
Error	0.0179167	0.0029861	6			
Total	0.0321208		7			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	1.02	47.47	0.9873	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9502	0.6451	0.7136	Normal Distribution

Fertilization Rate Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	SC	4	0.9125	0.8650	0.9600	0.9100	0.8800	0.9500	0.0149	3.27%	0.00%
0	N	4	0.8600	0.7991	0.9209	0.8700	0.8100	0.8900	0.0192	4.45%	5.75%

Angular (Corrected) Transformed Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	SC	4	1.274	1.186	1.361	1.267	1.217	1.345	0.02746	4.31%	0.00%
0	N	4	1.19	1.103	1.276	1.203	1.12	1.233	0.02719	4.57%	6.62%

Fertilization Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	SC	0.9500	0.9200	0.9000	0.8800
0	N	0.8900	0.8900	0.8100	0.8500

Angular (Corrected) Transformed Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	SC	1.345	1.284	1.249	1.217
0	N	1.233	1.233	1.12	1.173

Fertilization Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	SC	95/100	92/100	90/100	88/100
0	N	89/100	89/100	81/100	85/100

live
July 8/19

CETIS Analytical Report

Report Date: 05 Jul-19 13:04 (p 2 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

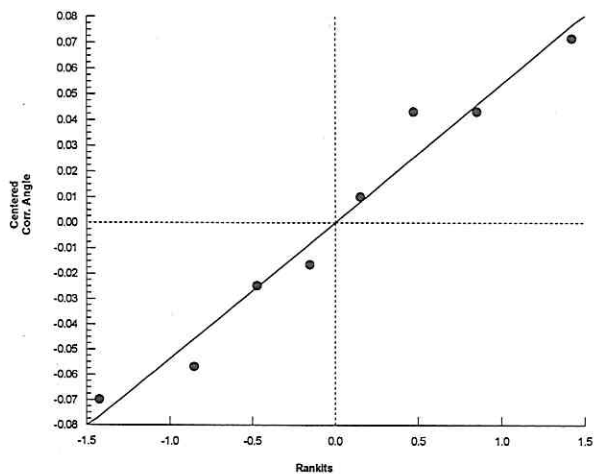
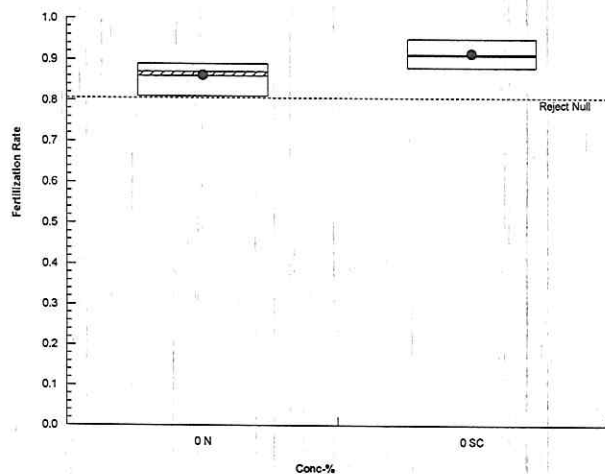
Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 09-1544-4894 Endpoint: Fertilization Rate
Analyzed: 05 Jul-19 13:03 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 05 Jul-19 13:04 (p 1 of 2)
 Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 19-1223-1912	Endpoint: Fertilization Rate	CETIS Version: CETISv1.9.4
Analyzed: 05 Jul-19 13:03	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 19-5334-0495	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 19 Feb-19 17:45	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 19 Feb-19 18:25	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m	Taxon: Echinoidea	Source: San Diego Lab Age:
Sample ID: 04-0118-8251	Code: 191282	Project:
Sample Date: 29 Jan-19	Material: Ground Water	Source: WDOE
Receipt Date: 30 Jan-19	CAS (PC):	Station: NWTPH-Dx
Sample Age: 21d 18h	Client: WDOE	

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	Site Control passed fertilization rate	5.68%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Site Control	-2.114	1.943	0.068	6	CDF	0.9605	Non-Significant Effect

Test Acceptability Criteria

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.9075	0.7	>>	Yes	Passes Criteria
Control Resp	0.86	0.7	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0110413	0.0110413	1	4.47	0.0789	Non-Significant Effect
Error	0.014821	0.0024702	6			
Total	0.0258624		7			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	1.49	47.47	0.7510	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9407	0.6451	0.6178	Normal Distribution

Fertilization Rate Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	0.8600	0.7991	0.9209	0.8700	0.8100	0.8900	0.0192	4.45%	0.00%
0	XC	4	0.9075	0.8677	0.9473	0.9050	0.8800	0.9400	0.0125	2.75%	-5.52%

Angular (Corrected) Transformed Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.19	1.103	1.276	1.203	1.12	1.233	0.02719	4.57%	0.00%
0	XC	4	1.264	1.193	1.335	1.258	1.217	1.323	0.02227	3.52%	-6.25%

Fertilization Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	0.8900	0.8900	0.8100	0.8500
0	XC	0.9400	0.9000	0.8800	0.9100

Angular (Corrected) Transformed Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.233	1.233	1.12	1.173
0	XC	1.323	1.249	1.217	1.266

Fertilization Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	89/100	89/100	81/100	85/100
0	XC	94/100	90/100	88/100	91/100

CETIS Analytical Report

Report Date: 05 Jul-19 13:04 (p 2 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

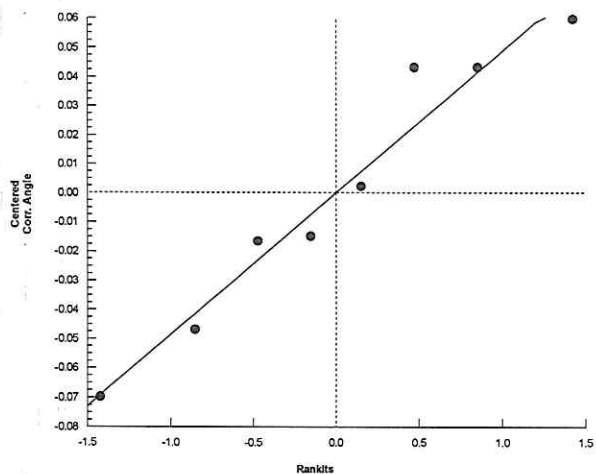
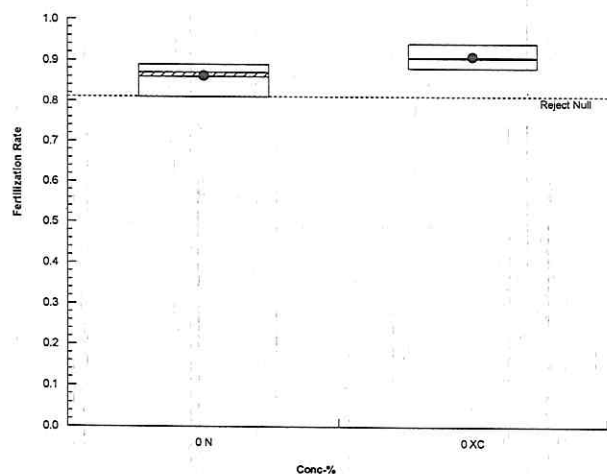
Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 19-1223-1912 Endpoint: Fertilization Rate
Analyzed: 05 Jul-19 13:03 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 12 Aug-19 12:01 (p 1 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 03-5399-0733	Endpoint: Fertilization Rate	CETIS Version: CETISv1.9.4
Analyzed: 12 Aug-19 12:01	Analysis: Parametric-Control vs Treatments	Status Level: 1
Batch ID: 19-5334-0495	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 19 Feb-19 17:45	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 19 Feb-19 18:25	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m (20 + 20)	Taxon: Echinoidea	Source: San Diego Lab Age:
Sample ID: 04-0118-8251	Code: 191282	Project:
Sample Date: 29 Jan-19	Material: Ground Water	Source: WDOE
Receipt Date: 30 Jan-19	CAS (PC):	Station: NWTPH-Dx
Sample Age: 21d 18h	Client: WDOE	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)	C > T	100	>100	n/a	1	11.07%

Dunnett Multiple Comparison Test

Control	vs	Conc-%	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.1	-0.6632	2.448	0.125	6	CDF	0.9679	Non-Significant Effect
		0.39	-2.768	2.448	0.125	6	CDF	1.0000	Non-Significant Effect
		1.6	-1.61	2.448	0.125	6	CDF	0.9981	Non-Significant Effect
		6.3	-0.865	2.448	0.125	6	CDF	0.9813	Non-Significant Effect
		25	-1.247	2.448	0.125	6	CDF	0.9939	Non-Significant Effect
		100	0.002783	2.448	0.125	6	CDF	0.8564	Non-Significant Effect

Test Acceptability Criteria

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.86	0.7	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0595077	0.009918	6	1.898	0.1286	Non-Significant Effect
Error	0.10974	0.0052257	21			
Total	0.169247		27			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	8.272	16.81	0.2188	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9027	0.8975	0.0132	Normal Distribution

Fertilization Rate Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	0.8600	0.7991	0.9209	0.8700	0.8100	0.8900	0.0192	4.45%	0.00%
0.1		4	0.8725	0.7567	0.9883	0.8450	0.8200	0.9800	0.0364	8.34%	-1.45%
0.39		4	0.9400	0.8897	0.9903	0.9350	0.9100	0.9800	0.0158	3.36%	-9.30%
1.6		4	0.9125	0.8824	0.9426	0.9050	0.9000	0.9400	0.0095	2.07%	-6.10%
6.3		4	0.8900	0.8556	0.9244	0.8950	0.8600	0.9100	0.0108	2.43%	-3.49%
25		4	0.9000	0.8434	0.9566	0.8900	0.8700	0.9500	0.0178	3.95%	-4.65%
100		4	0.8600	0.8064	0.9136	0.8450	0.8400	0.9100	0.0168	3.91%	0.00%

Angular (Corrected) Transformed Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.19	1.103	1.276	1.203	1.12	1.233	0.02719	4.57%	0.00%
0.1		4	1.223	1.004	1.443	1.166	1.133	1.429	0.06899	11.28%	-2.85%
0.39		4	1.331	1.214	1.448	1.315	1.266	1.429	0.03675	5.52%	-11.89%
1.6		4	1.272	1.216	1.328	1.258	1.249	1.323	0.01761	2.77%	-6.92%
6.3		4	1.234	1.18	1.288	1.241	1.187	1.266	0.01693	2.74%	-3.72%
25		4	1.253	1.151	1.356	1.233	1.202	1.345	0.03219	5.14%	-5.36%
100		4	1.189	1.107	1.271	1.166	1.159	1.266	0.02576	4.33%	0.01%

CETIS Analytical Report

Report Date: 12 Aug-19 12:01 (p 2 of 2)
Test Code/ID: 191280-RF / 17-6447-3179

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 03-5399-0733 Endpoint: Fertilization Rate
Analyzed: 12 Aug-19 12:01 Analysis: Parametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

Fertilization Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	0.8900	0.8900	0.8100	0.8500
0.1		0.8400	0.9800	0.8500	0.8200
0.39		0.9800	0.9500	0.9100	0.9200
1.6		0.9400	0.9000	0.9000	0.9100
6.3		0.8900	0.9000	0.9100	0.8600
25		0.9500	0.8700	0.8800	0.9000
100		0.9100	0.8400	0.8500	0.8400

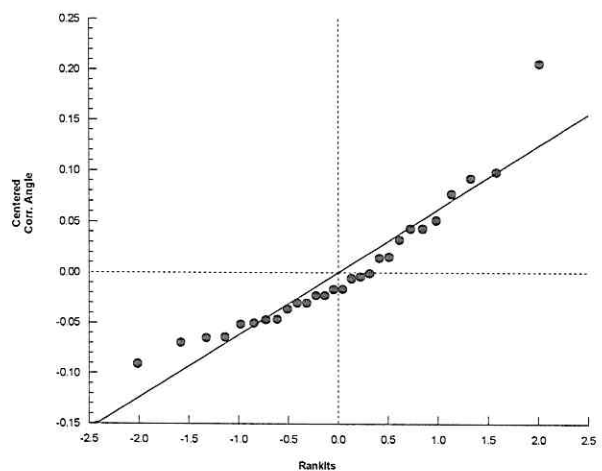
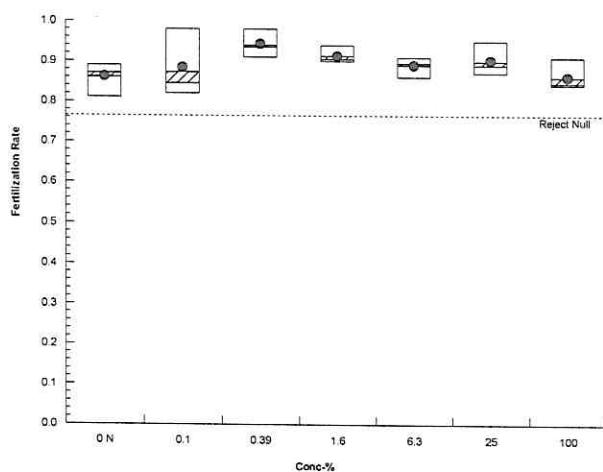
Angular (Corrected) Transformed Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.233	1.233	1.12	1.173
0.1		1.159	1.429	1.173	1.133
0.39		1.429	1.345	1.266	1.284
1.6		1.323	1.249	1.249	1.266
6.3		1.233	1.249	1.266	1.187
25		1.345	1.202	1.217	1.249
100		1.266	1.159	1.173	1.159

Fertilization Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	89/100	89/100	81/100	85/100
0.1		84/100	98/100	85/100	82/100
0.39		98/100	95/100	91/100	92/100
1.6		94/100	90/100	90/100	91/100
6.3		89/100	90/100	91/100	86/100
25		95/100	87/100	88/100	90/100
100		91/100	84/100	85/100	84/100

Graphics



Echinoderm Fertilization Test Summary Sheet

Client: WDOE

Start Date/Time: May 1, 2019 @ 1630h

Work Order No.: 1918280

Test Species: S. purpuratus

Sample Information:

Sample ID: DW-3

Sample Date: January 29, 2019

Date Received: January 30, 2019

Sample Volume: 5 x 20L

Dilution Water:

Type: Natural Seawater

Source: Vancouver Aquarium

Test Organism Information:

Batch No.: 050119

Source: Eschschol, San Diego, CA

Sperm:Egg Ratio: 200:1

Exposure Period: 20 min: 20 min

Reference Toxicant Results:

Reference Toxicant ID: SPUS 07

Stock Solution ID: 19Cu01 (100 ng/L Cu)

Date Initiated: May 1, 2019

IC50 (95% CL): 10.42 (8.1 - 14.2) ug/L Cu

Reference Toxicant Mean \pm 2 SD: 18.0 (8.9 - 36.5) ug/L Cu

Reference Toxicant CV (%): 36

Sample ID:	Fertilization Rate \pm SD (%)
Seawater Control	78.2 \pm 3.5
Salt Control	76.2 \pm 6.2
MW-11 (Site control)	78.2 \pm 5.2
DW-3	79.2 \pm 4.8

**** sample was not significantly different relative to the Seawater Control or Site controls*

Reviewed by: JCU

Date reviewed: July 8/19

Echinoderm Fertilization WQ Data Sheet

Client: WDOE

Analysts: NLC

Sample ID: DW-3

Test Date & Time: May 1, 2019 @ 1630h

WO No: 191280

Test Species: S. purpuratus

Initial Water Quality

	100% sample
Temp (°C)	12.5
DO (mg/L)	8.9
pH	7.6
Salinity	0.2

Brine/Salt Adjusted Water Quality

	100% sample
Temp (°C)	13.0
DO (mg/L)	8.3
pH	7.8
Salinity	30

Concentration ~ % (v/v)	Water Quality			
	Temp. (°C)	DO (mg/L)	pH	Salinity (ppt)
Seawater control	13.0	8.2	7.8	29
Salt control	13.0	8.3	8.0	30
Site Control - MW-1	13.0	8.3	8.0	30
DW-3	13.0	8.3	7.8	30
(100% v/v)				
Analyst Initials	NLC	NLC	NLC	NLC

Thermometer: CER#3 DO meter: 313 pH meter: 313 Salinity: 313

Sample Description: DW-3 - clear, yellow, some particulates, no odor.
MW-1 (Site control) - clear, light yellow, no odor, no particulates

Echinoderm Source: Enthelmy, San Diego, CA Date Received: May 1, 2019

Seawater Source: Vancouver Aquarium Date Collected: April 26, 2019

Comments: _____

Reviewed: NLC Date Reviewed: July 8/19

Echinoderm Fertilization Toxicity Test Data Sheet

Fertilized Egg Counts

Client: WDOE Start Date/Time: May 1, 2018 @ 1630h
 Work Order #: 191280 Test species: *S. purpuratus*
 Sample ID: DW-3 Test set up by: YML
 Test Duration: 20 min; 70 min

Sample ID	Rep	No. Fertilized Eggs	No. Unfertilized Eggs	Comments	Initials
Seawater Control	A	77	23		YML
	B	74	26		
	C	82	18		
	D	80	20		
Salt Control	A	75	25		
	B	82	18		
	C	80	20		
	D	68	32		
Site Control - MW-11	A	71	29		
	B	83	17		
	C	81	19		
	D	78	22		
100% (v/v)	A	81	19		✓
	B	77	23		
	C	85	15		
	D	74	26		
	A				
	B				
	C				
	D				
	A				
	B				
	C				
	D				
	A				
	B				
	C				
	D				

Comments: _____

Reviewed by: YML Date Reviewed: July 8/19

CETIS Summary Report

Report Date: 05 Jul-19 13:18 (p 1 of 1)
Test Code/ID: 191280 / 19-5959-5356

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Batch ID: 02-3159-5361 Test Type: Fertilization Analyst: Yvonne Lam
Start Date: 01 May-19 16:30 Protocol: EPA/600/R-95/136 (1995) Diluent: Natural seawater
Ending Date: 01 May-19 17:10 Species: Strongylocentrotus purpuratus Brine:
Test Length: 40m (20 min, 20 min) Taxon: Echinoidea Source: San Diego Lab Age:

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
Control SW	09-4780-6032	01 May-19	01 May-19	16h	WDOE	
Salt Control	18-7858-9023	01 May-19	01 May-19	16h		
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	92d 16h	WA State Dept of Ecolog	
DW-3	16-5145-0355	29 Jan-19	30 Jan-19 08:46	92d 16h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
Control SW	Control SW	WDOE	Control	
Salt Control	Control water	WDOE	Salt Control	
MW-11	Diesel	WDOE	MW-11	
DW-3	Diesel	WDOE	DW-3	

Single Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.5347	Control SW passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.5347	Salt Control passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.5347	MW-11 passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.5347	DW-3 passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.7586	Control SW passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.7586	Salt Control passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.7586	MW-11 passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.7586	DW-3 passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.8465	Control SW passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.8465	Salt Control passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.8465	MW-11 passed fertilization rate	1
14-4363-1114	Fertilization Rate	Dunnett Multiple Comparison Test	0.8465	DW-3 passed fertilization rate	1

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits		Overlap	Decision
14-4363-1114	Fertilization Rate	Control Resp	0.7825	Lower	Upper	Yes	Passes Criteria

Fertilization Rate Summary

Sample	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
Control SW	N	4	0.7825	0.7268	0.8382	0.7400	0.8200	0.0175	0.0350	4.47%	0.00%
Salt Control	SC	4	0.7625	0.6632	0.8618	0.6800	0.8200	0.0312	0.0624	8.18%	2.56%
MW-11	XC	4	0.7825	0.6989	0.8661	0.7100	0.8300	0.0263	0.0525	6.71%	0.00%
DW-3		4	0.7925	0.7163	0.8687	0.7400	0.8500	0.0239	0.0479	6.04%	-1.28%

Fertilization Rate Detail

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	0.7700	0.7400	0.8200	0.8000
Salt Control	SC	0.7500	0.8200	0.8000	0.6800
MW-11	XC	0.7100	0.8300	0.8100	0.7800
DW-3		0.8100	0.7700	0.8500	0.7400

Fertilization Rate Binomials

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	77/100	74/100	82/100	80/100
Salt Control	SC	75/100	82/100	80/100	68/100
MW-11	XC	71/100	83/100	81/100	78/100
DW-3		81/100	77/100	85/100	74/100

CETIS Analytical Report

Report Date: 05 Jul-19 13:18 (p 1 of 2)
Test Code/ID: 191280 / 19-5959-5356

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 14-4363-1114	Endpoint: Fertilization Rate	CETIS Version: CETISv1.9.4
Analyzed: 05 Jul-19 13:15	Analysis: Parametric-Control vs Treatments	Status Level: 1
Batch ID: 02-3159-5361	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 01 May-19 16:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 01 May-19 17:10	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m (20 min, 20 min)	Taxon: Echinoidea	Source: San Diego Lab Age:

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
Control SW	09-4780-6032	01 May-19	01 May-19	16h	WDOE	
Salt Control	18-7858-9023	01 May-19	01 May-19	16h		
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	92d 16h	WA State Dept of Ecolog	
DW-3	16-5145-0355	29 Jan-19	30 Jan-19 08:46	92d 16h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
Control SW	Control SW	WDOE	Control	
Salt Control	Control water	WDOE	Salt Control	
MW-11	Diesel	WDOE	MW-11	
DW-3	Diesel	WDOE	DW-3	

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	Salt Control passed fertilization rate	10.80%
		MW-11 passed fertilization rate	10.80%
		DW-3 passed fertilization rate	10.80%

Dunnett Multiple Comparison Test

Sample I	vs	Sample II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Salt Control	0.5253	2.287	0.098	6	CDF	0.5347	Non-Significant Effect
		MW-11	-0.02433	2.287	0.098	6	CDF	0.7586	Non-Significant Effect
		DW-3	-0.3111	2.287	0.098	6	CDF	0.8465	Non-Significant Effect

Test Acceptability Criteria

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.7825	0.7	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0026505	0.0008835	3	0.2429	0.8648	Non-Significant Effect
Error	0.0436502	0.0036375	12			
Total	0.0463006		15			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	0.7372	11.34	0.8644	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9461	0.8408	0.4309	Normal Distribution

Fertilization Rate Summary

Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
Control SW	N	4	0.7825	0.7268	0.8382	0.7850	0.7400	0.8200	0.0175	4.47%	0.00%
Salt Control		4	0.7625	0.6632	0.8618	0.7750	0.6800	0.8200	0.0312	8.18%	2.56%
MW-11		4	0.7825	0.6989	0.8661	0.7950	0.7100	0.8300	0.0263	6.71%	0.00%
DW-3		4	0.7925	0.7163	0.8687	0.7900	0.7400	0.8500	0.0239	6.04%	-1.28%

Angular (Corrected) Transformed Summary

Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
Control SW	N	4	1.087	1.019	1.154	1.089	1.036	1.133	0.02119	3.90%	0.00%
Salt Control		4	1.064	0.9487	1.18	1.077	0.9695	1.133	0.03626	6.82%	2.06%
MW-11		4	1.088	0.988	1.187	1.101	1.002	1.146	0.0313	5.76%	-0.10%
DW-3		4	1.1	1.005	1.195	1.095	1.036	1.173	0.0299	5.44%	-1.22%

CETIS Analytical Report

Report Date: 05 Jul-19 13:18 (p 2 of 2)
Test Code/ID: 191280 / 19-5959-5356

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 14-4363-1114
Analyzed: 05 Jul-19 13:15
Endpoint: Fertilization Rate
Analysis: Parametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

Fertilization Rate Detail

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	0.7700	0.7400	0.8200	0.8000
Salt Control		0.7500	0.8200	0.8000	0.6800
MW-11		0.7100	0.8300	0.8100	0.7800
DW-3		0.8100	0.7700	0.8500	0.7400

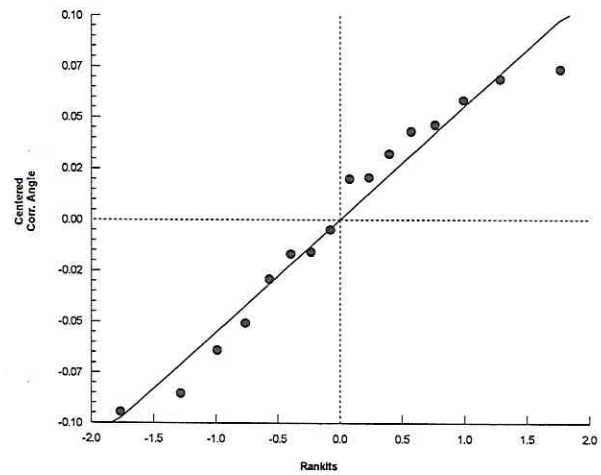
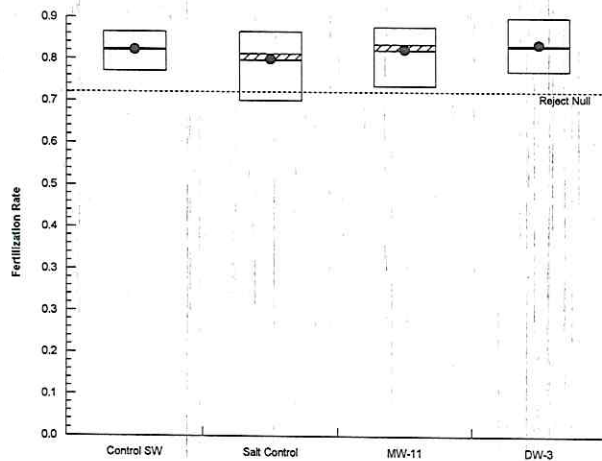
Angular (Corrected) Transformed Detail

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	1.071	1.036	1.133	1.107
Salt Control		1.047	1.133	1.107	0.9695
MW-11		1.002	1.146	1.12	1.083
DW-3		1.12	1.071	1.173	1.036

Fertilization Rate Binomials

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	77/100	74/100	82/100	80/100
DW-3		81/100	77/100	85/100	74/100

Graphics



CETIS Analytical Report

Report Date: 05 Jul-19 13:18 (p 1 of 2)
Test Code/ID: 191280 / 19-5959-5356

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 11-4262-9726	Endpoint: Fertilization Rate	CETIS Version: CETISv1.9.4
Analyzed: 05 Jul-19 13:15	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 02-3159-5361	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 01 May-19 16:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 01 May-19 17:10	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m (20 min/20 min)	Taxon: Echinoidea	Source: San Diego Lab Age:

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
Control SW	09-4780-6032	01 May-19	01 May-19	16h	WDOE	
Salt Control	18-7858-9023	01 May-19	01 May-19	16h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
Control SW	Control SW	WDOE	Control	
Salt Control	Control water	WDOE	Salt Control	

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	Salt Control passed fertilization rate	8.94%

Equal Variance t Two-Sample Test

Sample I	vs	Sample II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Salt Control	0.5334	1.943	0.082	6	CDF	0.3065	Non-Significant Effect

Test Acceptability Criteria

Test Acceptability Criteria		TAC Limits			
Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.7825	0.7	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0010038	0.0010038	1	0.2845	0.6129	Non-Significant Effect
Error	0.021167	0.0035278	6			
Total	0.0221708		7			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	2.93	47.47	0.4009	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9514	0.6451	0.7258	Normal Distribution

Fertilization Rate Summary

Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
Control SW	N	4	0.7825	0.7268	0.8382	0.7850	0.7400	0.8200	0.0175	4.47%	0.00%
Salt Control	SC	4	0.7625	0.6632	0.8618	0.7750	0.6800	0.8200	0.0312	8.18%	2.56%

Angular (Corrected) Transformed Summary

Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
Control SW	N	4	1.087	1.019	1.154	1.089	1.036	1.133	0.02119	3.90%	0.00%
Salt Control	SC	4	1.064	0.9487	1.18	1.077	0.9695	1.133	0.03626	6.82%	2.06%

Fertilization Rate Detail

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	0.7700	0.7400	0.8200	0.8000
Salt Control	SC	0.7500	0.8200	0.8000	0.6800

Angular (Corrected) Transformed Detail

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	1.071	1.036	1.133	1.107
Salt Control	SC	1.047	1.133	1.107	0.9695

CETIS Analytical Report

Report Date: 05 Jul-19 13:18 (p 2 of 2)
Test Code/ID: 191280 / 19-5959-5356

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

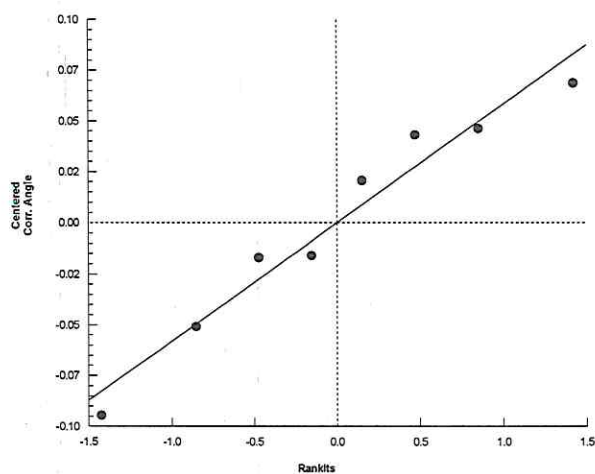
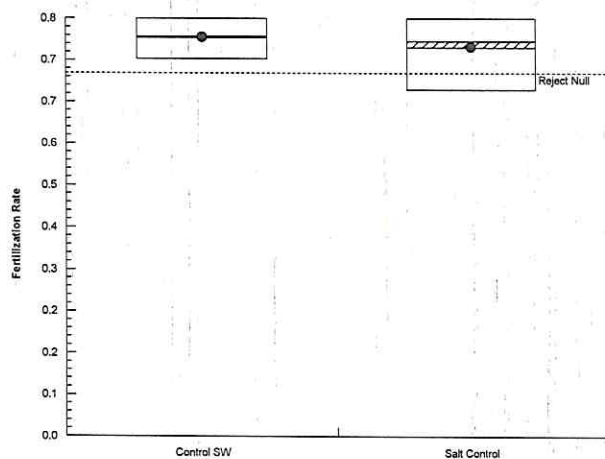
Analysis ID: 11-4262-9726 Endpoint: Fertilization Rate
Analyzed: 05 Jul-19 13:15 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Fertilization Rate Binomials

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
Control SW	N	77/100	74/100	82/100	80/100
Salt Control	SC	75/100	82/100	80/100	68/100

Graphics



CETIS Analytical Report

Report Date: 05 Jul-19 13:18 (p 1 of 2)
Test Code/ID: 191280 / 19-5959-5356

Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 05-0589-8278	Endpoint: Fertilization Rate	CETIS Version: CETISv1.9.4
Analyzed: 05 Jul-19 13:17	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 02-3159-5361	Test Type: Fertilization	Analyst: Yvonne Lam
Start Date: 01 May-19 16:30	Protocol: EPA/600/R-95/136 (1995)	Diluent: Natural seawater
Ending Date: 01 May-19 17:10	Species: Strongylocentrotus purpuratus	Brine:
Test Length: 40m (20 min, 20 min)	Taxon: Echinoidea	Source: San Diego Lab Age:

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	92d 16h	WA State Dept of Ecolog	
DW-3	16-5145-0355	29 Jan-19	30 Jan-19 08:46	92d 16h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
MW-11	Diesel	WDOE	MW-11	
DW-3	Diesel	WDOE	DW-3	

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	DW-3 passed fertilization rate	9.11%

Equal Variance t Two-Sample Test

Sample I	vs	Sample II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Site Control		DW-3	-0.2825	1.943	0.084	6	CDF	0.6065	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0002991	0.0002991	1	0.07983	0.7870	Non-Significant Effect
Error	0.0224832	0.0037472	6			
Total	0.0227823		7			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	1.096	47.47	0.9419	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9598	0.6451	0.8079	Normal Distribution

Fertilization Rate Summary

Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
MW-11	XC	4	0.7825	0.6989	0.8661	0.7950	0.7100	0.8300	0.0263	6.71%	0.00%
DW-3		4	0.7925	0.7163	0.8687	0.7900	0.7400	0.8500	0.0239	6.04%	-1.28%

Angular (Corrected) Transformed Summary

Sample	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
MW-11	XC	4	1.088	0.988	1.187	1.101	1.002	1.146	0.0313	5.76%	0.00%
DW-3		4	1.1	1.005	1.195	1.095	1.036	1.173	0.0299	5.44%	-1.12%

Fertilization Rate Detail

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
MW-11	XC	0.7100	0.8300	0.8100	0.7800
DW-3		0.8100	0.7700	0.8500	0.7400

Angular (Corrected) Transformed Detail

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
MW-11	XC	1.002	1.146	1.12	1.083
DW-3		1.12	1.071	1.173	1.036

Fertilization Rate Binomials

Sample	Code	Rep 1	Rep 2	Rep 3	Rep 4
MW-11	XC	71/100	83/100	81/100	78/100
DW-3		81/100	77/100	85/100	74/100

CETIS Analytical Report

Report Date: 05 Jul-19 13:18 (p 2 of 2)
Test Code/ID: 191280 / 19-5959-5356

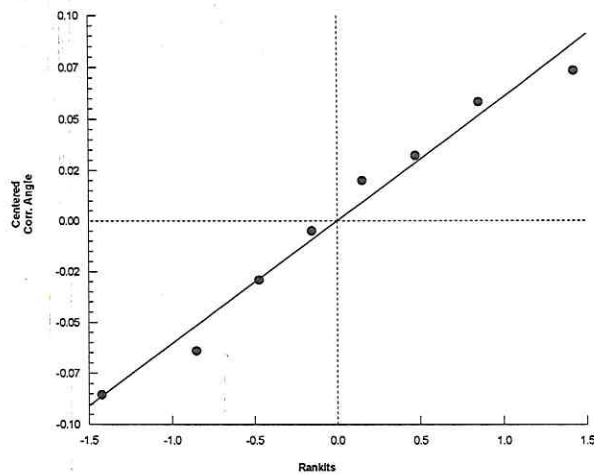
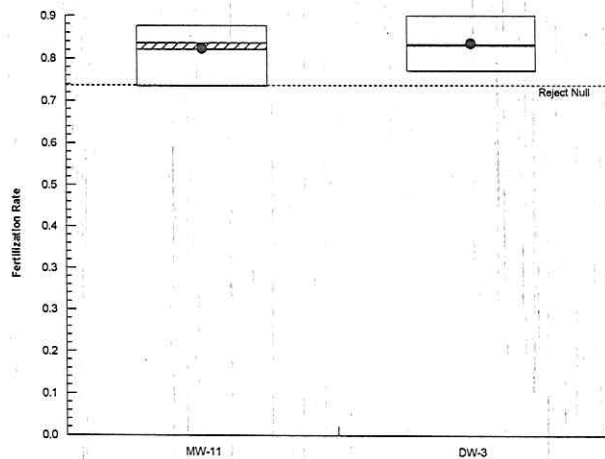
Echinoid Sperm Cell Fertilization Test

Nautilus Environmental

Analysis ID: 05-0589-8278 Endpoint: Fertilization Rate
Analyzed: 05 Jul-19 13:17 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



APPENDIX C – *Pimephales promelas* Toxicity Test Data

Fathead Minnow Test Summary Sheet

Client: WDOE

Start Date & Time: Feb 19/19 @ 1615h

Work Order No.: 191281a

Test Species: Pimephales promelas

Sample Information:

Sample ID: DW-3 (range finder)

Sample Date: Jan 29/19

Date Received: Jan 30/19

Sample Volume: 5 x 20L

Dilution Water:

Type: Moderately - hard water

Hardness (mg/L CaCO₃): 100

Alkalinity (mg/L CaCO₃): 74

Test Organism Information:

Batch No.: 021919

Source: Aquatic BioSystems, CO

Loading Density: 10 fish/250mL

NaCl Reference Toxicant Results:

Reference Toxicant ID: PP 138

Stock Solution ID: NaCl

Date Initiated: Feb 19/19

7-d EC50 (95% CL): 4.0 (3.4-4.7) g/L NaCl

7-d IC50 (95% CL): 4.0 (3.5-4.7) g/L NaCl

Survival:

Reference Toxicant Mean \pm 2 SD: 4.7 (3.7-6.1) g/L NaCl CV (%): 12

Biomass

Reference Toxicant Mean \pm 2 SD: 4.5 (3.4-6.1) g/L NaCl CV (%): 15

Test Results:

	Survival	Biomass
NOEC % (v/v)	6.2	1.0
LOEC % (v/v)	>6.2	6.2
EC25 % (v/v) (95% CL)	>6.2	
EC50 % (v/v) (95% CL)	>6.2	2.6 (1.9-5.4)
IC25 % (v/v) (95% CL)		3.9 (2.6-7.8)
IC50 % (v/v) (95% CL)		>6.2

Reviewed by: [Signature]

Date reviewed: Dec-19, 2019

7-d Chronic Freshwater Toxicity Test **Initial and Final Water Quality Measurements**

1/2

Client: WDOE
 Sample ID: DW-3 (Range-finder)
 Work Order #: 191281

Start Date & Time: Feb 19/19 @ 1615h
 Stop Date & Time: Feb 26/19 @ 1545h
 CER #: 11
 Test Species: Pimephales promelas

Concentration control	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0
DO (mg/L)	7.7	6.2	6.8	7.8	4.8	8.2	4.4	8.7	5.1	8.0	6.0	8.0	5.2	7.6
pH	8.0	7.6	8.0	7.5	8.0	7.5	7.9	7.2	7.7	7.4	7.6	7.3	7.9	7.1
Cond. (µS/cm)	323	321		322		322		332		328		325		337
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Concentration site control	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0
DO (mg/L)	7.8	7.0	7.9	5.4	8.0	4.5	7.9	5.4	8.7	5.9	8.0	5.5	7.8	5.8
pH	7.7	7.8	7.5	7.9	7.7	7.8	7.9	7.7	7.6	7.5	7.6	7.4	7.9	7.6
Cond. (µS/cm)	324	320		323		325		324		328		327		331
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Concentration 0.10	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0
DO (mg/L)	7.7	6.4	6.7	7.8	4.9	8.2	4.5	7.7	4.2	8.1	5.8	8.0	5.0	7.6
pH	8.0	7.6	8.0	7.5	8.0	7.5	7.9	7.3	7.7	7.4	7.6	7.2	7.9	7.4
Cond. (µS/cm)	320	321		322		323		329		330		327		338
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Concentration 0.39	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0
DO (mg/L)	7.7	6.3	6.7	7.7	5.0	8.3	4.4	7.7	4.3	8.1	5.9	8.1	5.1	7.6
pH	8.0	7.6	8.0	7.5	8.0	7.5	7.9	7.3	7.7	7.4	7.6	7.3	7.9	7.4
Cond. (µS/cm)	320	321		321		322		331		330		326		333
Initials	AK	ML		ML		ML		ML		ML		ML		AK

Thermometer: 11 DO meter/probe: 4, 4 pH meter/probe: 4, 1, 4 Conductivity meter/probe: 4, 4

	Control			
Hardness*	100			
Alkalinity*	34			

Analysts: ML, CS, ASD

Reviewed by: [Signature]

Date reviewed: July 17, 2019

* mg/L as CaCO₃

Sample Description:

Comments:

DW-3 : clear, yellow, some particulates, odourless
 MW-11 : clear, light yellow, no particulates, odourless

$\frac{2}{2}$

Start Date & Time: Feb 19/19 @ 1615h
Stop Date & Time: Feb 26/19 @ 1545h
CER #:
Test Species: *Pimephales promelas*

Concentration 1.6	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.5	24.0	24.0	24.0
DO (mg/L) 7.8	6.4	6.6	7.8	5.0	8.2	4.6	7.7	4.1	8.1	5.8	5.0	5.5	7.6	5.0
pH	8.0 ^m	7.6	8.0	7.5	8.0	7.5	7.9	7.2	7.7	7.3	7.6	7.3	7.9	7.4
Cond. (µS/cm)	320	320		321		321		330		330		325		334
Initials	A	ML		ML		ML		RL		A		ML		A

Concentration 6.3	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0
DO (mg/L) 7.8	6.5	6.5	7.7	5.0	8.2	4.5	7.7	5.2	8.1	5.9	8.0	5.4	7.7	5.0
pH	7.9	7.5	8.0	7.5	8.0	7.6	7.9	7.2	7.7	7.2	7.7	7.4	7.9	7.3
Cond. (µS/cm)	320	319		320		320		329		330		323		331
Initials	AT	ML7		ML9		ML7		M		a		ML9		AT

Concentration	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0
DO (mg/L) F.8	6.6	5.3	7.8	5.2	7.9	4.7	7.9	4.4	8.0	5.8	8.0	5.5	7.7	5.0
pH	7.7	7.5	7.8	7.6	7.8	7.7	7.9	7.2	7.7	7.3	7.7	7.4	7.8	7.3
Cond. (µS/cm)	315	313		312		313		321		321		316		324
Initials	GK	MN		MN		MN		m		A		MN		AK

Concentration	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
100.														
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.5		24.0	24.0
DO (mg/L) 8.2	7.9	6.9	7.8	5.6	7.8	5.0	8.0	5.5	8.5	5.7	8.0	5.4	7.8	4.3
pH	7.2	7.4	7.4	7.4	7.3	7.7	7.6	7.3	7.3	7.7	7.5	7.5	7.6	7.3
Cond. (µS/cm)	300	291		290		288		290		292		288		297
Initials	AK	ML		ML		ML		L		M		ML		AK

Thermometer: 1 DO meter/probe: 4 / 4 pH meter/probe: 4 / 4 Conductivity meter/probe: 4 / 4

	Control			
Hardness*	100			
Alkalinity*	74			

Analysts: MO, CS, AWD

Reviewed by: _____

Date reviewed: July 17, 2019

* mg/L as CaCO3

Sample Description: same as page 1

Comments:

7-d Fathead Minnow Toxicity Test **Daily Survival**

Client: WDOE
 Sample ID: DW-3 (Range finder)
 Work Order #: 191281

Start Date & Time: Feb 19/19 @ 1615h
 Stop Date & Time: Feb 26/19 @ 1545h
 Test Species: Pimephales promelas

Concentration % (v/v) <i>Normal</i>	Rep	Day of Test - No. of Survivors							Comments
		1	2	3	4	5	6	7	
Control	A	10	10	10	10	10	10	10	
	B	10	10	10	10	10	10	10	
	C	10	10	10	10	10	10	10	
	D								
Site Control	A	10	10	10	9	9	9	9	
	B	10	10	10	10	10	10	10	
	C	10	10	10	10	10	10	10	
	D								
0.10	A	10	10	10	10	10	10	9	
	B	10	10	10	10	10	10	10	
	C	10	10	10	10	9	9	9	
	D								
0.39	A	10	10	10	10	10	10	10	
	B	10	10	10	10	10	10	10	
	C	10	10	10	10	10	10	10	
	D								
1.6	A	10	10	10	10	10	10	10	
	B	10	10	10	10	10	10	10	
	C	10	10	10	10	10	10	10	
	D								
6.3	A	10	10	10	10	10	10	10	
	B	10	10	10	10	10	10	10 ^{mm} 8	
	C	10	10	10	10	10	10	10 ^{mm} 8 10	
	D								
25	A	10 ^{mm} 10	10	10	10	10	9	9	
	B	10	10	10	10	10	10	10	
	C	10	10	10	10	10	10	10	
	D								
100	A	10	10	10	10	10	10	10	
	B	10	10	10	10	10	7	7	
	C	10	10	10	9	9	9	9	
	D								
Tech Initials		ML	ML	MM	mm	A	MT	MO	

Comments: nb. 5cm solution depth
- remaining organisms appear normal

Reviewed by:  Date reviewed: July 17, 2019

Fathead Minnow Toxicity Test Data Sheet

Dry Weight Data

Client: WDOE
 Sample ID: DW-3 (Range Finder)

Start Date & Time: Feb 19/19 @ 1615h
 Termination Date & Time: Feb 26/19 @ 1545h

Work Order No.: 191281
PP Green

Balance ID: Bal- 1

nominal Concentration 0/0/0/0	Rep	Pan No.	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
control	A	1	10	MLG	1021.62	1030.04	10	SSK/ A
	B	2	10		1048.59	1055.51	10	1/A
	C	3	10		1011.22	1016.81	10	1/A
site ctrl	A	4	9		1032.18	1038.93	9	1/A
	B	5	10		1043.95	1051.96	10	1/A
	C	6	10		1030.81	1039.00 1040.01	10	1/A
0.10	A	7	9		1033.57	1039.15	9	1/A
	B	8	10		1034.12	1040.76	10	1/A
	C	9	9		1042.54	1049.13	9	1/A
0.39	A	10	10		1019.33	1027.07	10	1/A
	B	11	10		1078.68	1085.86	10	1/A
	C	12	10		1051.74	1059.42	10	1/A
1.6	A	13	10		1025.54	1033.45	10	1/A
	B	14	10		1035.30	1042.83	10	1/A
	C	15	10		1019.33	1026.38	10	1/A
6.3	A	16	10		1058.68	1066.36	10	1/A
	B	17	8		1019.68	1024.11	8	1/A
	C	18	10		1062.92	1070.73	10	1/A
25	A	19	9		998.43	1004.96	9	1/A
	B	20	10		1050.32	1057.47	10	1/A
	C	21	10		1038.03	1045.06	10	1/A
100	A	22	10		1032.73	1037.54	10	1/A
	B	23	7		1046.46	1048.80 75	7	1/A
	C	24	9		1037.55	1041.06	9	1/A

Comments: 10% reweigh (mg): # 4 = 1038.80 # 20 = 1057.31
12 = 1059.35

Reviewed by: [Signature]
 Version 1.1; Issued June 18, 2015

Date Reviewed: July 17, 2019
 Nautilus Environmental Company Inc.

CETIS Summary Report

Report Date: 19 Dec-19 15:34 (p 1 of 3)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Batch ID: 03-4091-2684	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 16:15	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 26 Feb-19 15:45	Species: Pimephales promelas	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 16h	Client: WA State Dept of Ecology	

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓ Level	mg/L	95% LCL	95% UCL	TU	S
03-6347-4466	7d Survival Rate	Linear Interpolation (ICPIN)	EC5	1.885	n/a	n/a		1
			EC10	5.118	n/a	n/a		
			EC15	>6.23	n/a	n/a		
			EC20	>6.23	n/a	n/a		
			EC25	>6.23	n/a	n/a		
			EC40	>6.23	n/a	n/a		
			EC50	>6.23	n/a	n/a		
20-6823-9408	Mean Dry Biomass-mg	Linear Interpolation (ICPIN)	IC5	1.051	n/a	1.659		1
			IC10	1.36	n/a	2.157		
			IC15	1.716	n/a	2.823		
			IC20	2.124	n/a	3.914		
			IC25	2.595	n/a	5.414		
			IC40	4.475	0.8971	n/a		
			IC50	>6.23	n/a	n/a		
09-0897-9947	Mean Dry Weight-mg	Linear Interpolation (ICPIN)	IC5	1.231	n/a	1.506		1
			IC10	1.591	n/a	2.036		
			IC15	2.009	n/a	2.813		
			IC20	2.494	0.1173	3.794		
			IC25	3.057	0.3557	4.9		
			IC40	5.353	1.787	n/a		
			IC50	>6.23	n/a	n/a		

CETIS Summary Report

Report Date: 19 Dec-19 15:34 (p 2 of 3)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0.15		3	0.9333	0.7899	1.0000	0.9000	1.0000	0.0333	0.0577	6.19%	0.00%
0.151		3	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	-7.14%
0.16	N	3	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	-7.14%
0.19		3	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	-7.14%
0.2	XC	3	0.9667	0.8232	1.0000	0.9000	1.0000	0.0333	0.0577	5.97%	-3.57%
0.32		3	0.9333	0.6465	1.0000	0.8000	1.0000	0.0667	0.1155	12.37%	0.00%
1		3	0.9667	0.8232	1.0000	0.9000	1.0000	0.0333	0.0577	5.97%	-3.57%
6.23		3	0.8667	0.4872	1.0000	0.7000	1.0000	0.0882	0.1528	17.63%	7.14%

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0.15		3	0.627	0.4784	0.7756	0.558	0.664	0.03453	0.0598	9.54%	0.00%
0.151		3	0.7533	0.6769	0.8297	0.718	0.774	0.01775	0.03075	4.08%	-20.15%
0.16	N	3	0.6943	0.3418	1.047	0.559	0.842	0.08193	0.1419	20.44%	-10.74%
0.19		3	0.7497	0.6426	0.8567	0.705	0.791	0.02488	0.04309	5.75%	-19.56%
0.2	XC	3	0.7987	0.4943	1.103	0.675	0.92	0.07073	0.1225	15.34%	-27.38%
0.32		3	0.664	0.1883	1.14	0.443	0.781	0.1106	0.1915	28.84%	-5.90%
1		3	0.6903	0.6086	0.772	0.653	0.715	0.01899	0.03288	4.76%	-10.10%
6.23		3	0.3537	0.04061	0.6667	0.229	0.481	0.07276	0.126	35.63%	43.59%

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0.15		3	0.6721	0.5316	0.8125	0.62	0.7322	0.03264	0.05654	8.41%	0.00%
0.151		3	0.7533	0.6769	0.8297	0.718	0.774	0.01775	0.03075	4.08%	-12.09%
0.16	N	3	0.6943	0.3418	1.047	0.559	0.842	0.08193	0.1419	20.44%	-3.31%
0.19		3	0.7497	0.6426	0.8567	0.705	0.791	0.02488	0.04309	5.75%	-11.54%
0.2	XC	3	0.8237	0.607	1.04	0.75	0.92	0.05036	0.08723	10.59%	-22.56%
0.32		3	0.7009	0.3839	1.018	0.5537	0.781	0.07368	0.1276	18.21%	-4.29%
1		3	0.7145	0.6865	0.7426	0.703	0.7256	0.006516	0.01129	1.58%	-6.32%
6.23		3	0.3994	0.2072	0.5916	0.3271	0.481	0.04466	0.07736	19.37%	40.57%

N = lab control
XC = site control

CETIS Summary Report

 Report Date: 19 Dec-19 15:34 (p 3 of 3)
 Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15		0.9000	1.0000	0.9000
0.151		1.0000	1.0000	1.0000
0.16	N	1.0000	1.0000	1.0000
0.19		1.0000	1.0000	1.0000
0.2	XC	0.9000	1.0000	1.0000
0.32		1.0000	0.8000	1.0000
1		0.9000	1.0000	1.0000
6.23		1.0000	0.7000	0.9000

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15		0.558	0.664	0.659
0.151		0.774	0.718	0.768
0.16	N	0.842	0.682	0.559
0.19		0.791	0.753	0.705
0.2	XC	0.675	0.801	0.92
0.32		0.768	0.443	0.781
1		0.653	0.715	0.703
6.23		0.481	0.229	0.351

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15		0.62	0.664	0.7322
0.151		0.774	0.718	0.768
0.16	N	0.842	0.682	0.559
0.19		0.791	0.753	0.705
0.2	XC	0.75	0.801	0.92
0.32		0.768	0.5537	0.781
1		0.7256	0.715	0.703
6.23		0.481	0.3271	0.39

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15		9/10	10/10	9/10
0.151		10/10	10/10	10/10
0.16	N	10/10	10/10	10/10
0.19		10/10	10/10	10/10
0.2	XC	9/10	10/10	10/10
0.32		10/10	8/10	10/10
1		9/10	10/10	10/10
6.23		10/10	7/10	9/10

N = (ab Control)

XC = site control

CETIS Analytical Report

 Report Date: 16 Dec-19 14:27 (p 1 of 2)
 Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 11-0367-2582	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 14:27	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 03-4091-2684	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 16:15	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 26 Feb-19 15:45	Species: Pimephales promelas	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 16h	Client: WA State Dept of Ecology	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	515536	200	Yes	Two-Point Interpolation

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC5	1	n/a	n/a
EC10	3.324	n/a	n/a
EC15	>6.23	n/a	n/a
EC20	>6.23	n/a	n/a
EC25	>6.23	n/a	n/a
EC40	>6.23	n/a	n/a
EC50	>6.23	n/a	n/a

7d Survival Rate Summary

Conc-mg/L	Code	Count	Calculated Variate(A/B)							Isotonic Variate	
			Mean	Min	Max	Std Dev	CV%	%Effect	A/B	Mean	%Effect
0.15	N	3	0.9333	0.9000	1.0000	0.0577	6.19%	0.0%	28/30	0.9833	0.0%
0.151		3	1.0000	1.0000	1.0000	0.0000	0.00%	-7.14%	30/30	0.9833	0.0%
0.16		3	1.0000	1.0000	1.0000	0.0000	0.00%	-7.14%	30/30	0.9833	0.0%
0.19		3	1.0000	1.0000	1.0000	0.0000	0.00%	-7.14%	30/30	0.9833	0.0%
0.32		3	0.9333	0.8000	1.0000	0.1155	12.37%	0.0%	28/30	0.95	3.39%
1		3	0.9667	0.9000	1.0000	0.0577	5.97%	-3.57%	29/30	0.95	3.39%
6.23		3	0.8667	0.7000	1.0000	0.1528	17.63%	7.14%	26/30	0.8667	11.86%

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	0.9000	1.0000	0.9000
0.151		1.0000	1.0000	1.0000
0.16		1.0000	1.0000	1.0000
0.19		1.0000	1.0000	1.0000
0.32		1.0000	0.8000	1.0000
1		0.9000	1.0000	1.0000
6.23		1.0000	0.7000	0.9000

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	9/10	10/10	9/10
0.151		10/10	10/10	10/10
0.16		10/10	10/10	10/10
0.19		10/10	10/10	10/10
0.32		10/10	8/10	10/10
1		9/10	10/10	10/10
6.23		10/10	7/10	9/10

N= lab control

CETIS Analytical Report

Report Date: 16 Dec-19 14:27 (p 2 of 2)
Test Code/ID: 191281a / 01-3018-0898

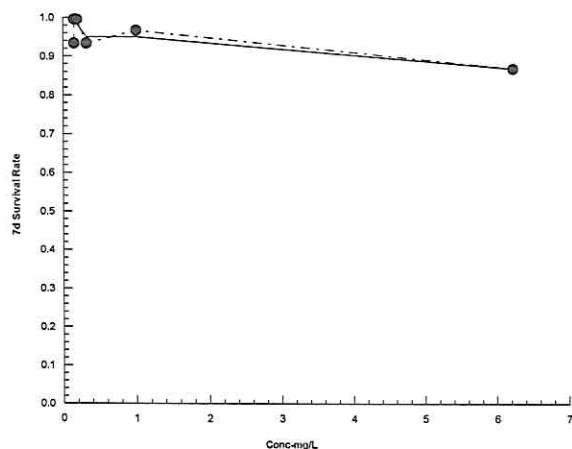
Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 11-0367-2582 Endpoint: 7d Survival Rate
Analyzed: 16 Dec-19 14:27 Analysis: Linear Interpolation (ICPIN)

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 18 Dec-19 13:36 (p 1 of 2)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 20-6823-9408	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 17 Dec-19 17:26	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 03-4091-2684	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 16:15	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 26 Feb-19 15:45	Species: Pimephales promelas	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 16h	Client: WA State Dept of Ecology	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	1068237	200	Yes	Two-Point Interpolation

Point Estimates

Level	mg/L	95% LCL	95% UCL
IC5	1.051	n/a	1.659
IC10	1.36	n/a	2.157
IC15	1.716	n/a	2.823
IC20	2.124	n/a	3.914
IC25	2.595	n/a	5.414
IC40	4.475	0.8971	n/a
IC50	>6.23	n/a	n/a

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Calculated Variate						Isotonic Variate	
			Mean	Min	Max	Std Dev	CV%	%Effect	Mean	%Effect
0.15	N	3	0.627	0.558	0.664	0.0598	9.54%	0.0%	0.7061	0.0%
0.151		3	0.7533	0.718	0.774	0.03075	4.08%	-20.15%	0.7061	0.0%
0.16		3	0.6943	0.559	0.842	0.1419	20.44%	-10.74%	0.7061	0.0%
0.19		3	0.7497	0.705	0.791	0.04309	5.75%	-19.56%	0.7061	0.0%
0.32		3	0.664	0.443	0.781	0.1915	28.84%	-5.9%	0.6772	4.1%
1		3	0.6903	0.653	0.715	0.03288	4.76%	-10.1%	0.6772	4.1%
6.23		3	0.3537	0.229	0.481	0.126	35.63%	43.59%	0.3537	49.91%

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	0.558	0.664	0.659
0.151		0.774	0.718	0.768
0.16		0.842	0.682	0.559
0.19		0.791	0.753	0.705
0.32		0.768	0.443	0.781
1		0.653	0.715	0.703
6.23		0.481	0.229	0.351

CETIS Analytical Report

Report Date: 18 Dec-19 13:36 (p 2 of 2)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

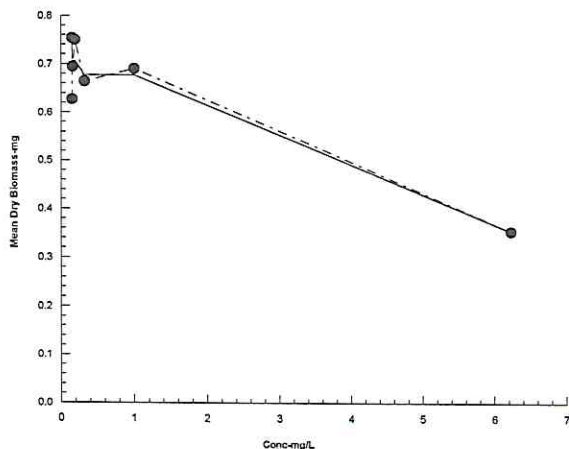
Nautilus Environmental

Analysis ID: 20-6823-9408
Analyzed: 17 Dec-19 17:26

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

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 16 Dec-19 14:34 (p 1 of 2)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test					Nautilus Environmental	
Analysis ID:	06-6817-8280	Endpoint:	Mean Dry Weight-mg	CETIS Version:	CETISv1.9.4	
Analyzed:	16 Dec-19 14:34	Analysis:	Linear Interpolation (ICPIN)	Status Level:	1	
Batch ID:	03-4091-2684	Test Type:	Growth-Survival (7d)	Analyst:	Mimi Tran	
Start Date:	19 Feb-19 16:15	Protocol:	EC/EPS 1/RM/22	Diluent:	Mod-Hard Synthetic Water	
Ending Date:	26 Feb-19 15:45	Species:	Pimephales promelas	Brine:		
Test Length:	6d 23h	Taxon:	Actinopterygii	Source:	Aquatic Biosystems, CO	Age: <24
Sample ID:	14-5122-1569	Code:	567FE241	Project:		
Sample Date:	29 Jan-19	Material:	Diesel	Source:	WA Dept of Ecology	
Receipt Date:	30 Jan-19 08:46	CAS (PC):		Station:	DW-3	
Sample Age:	21d 16h	Client:	WA State Dept of Ecology			

Linear Interpolation Options					
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	626733	200	Yes	Two-Point Interpolation

Point Estimates			
Level	mg/L	95% LCL	95% UCL
IC5	1.231	n/a	1.588
IC10	1.591	n/a	2.203
IC15	2.009	n/a	2.962
IC20	2.494	0.0176	3.889
IC25	3.057	0.2682	5.022
IC40	5.353	1.411	n/a
IC50	>6.23	n/a	n/a

Mean Dry Weight-mg Summary			Calculated Variate						Isotonic Variate	
Conc-mg/L	Code	Count	Mean	Min	Max	Std Dev	CV%	%Effect	Mean	%Effect
0.15	N	3	0.6721	0.62	0.7322	0.05654	8.41%	0.0%	0.7174	0.0%
0.151		3	0.7533	0.718	0.774	0.03075	4.08%	-12.09%	0.7174	0.0%
0.16		3	0.6943	0.559	0.842	0.1419	20.44%	-3.31%	0.7174	0.0%
0.19		3	0.7497	0.705	0.791	0.04309	5.75%	-11.54%	0.7174	0.0%
0.32		3	0.7009	0.5537	0.781	0.1276	18.21%	-4.29%	0.7077	1.34%
1		3	0.7145	0.703	0.7256	0.01129	1.58%	-6.32%	0.7077	1.34%
6.23		3	0.3994	0.3271	0.481	0.07736	19.37%	40.57%	0.3994	44.33%

Mean Dry Weight-mg Detail				
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	0.62	0.664	0.7322
0.151		0.774	0.718	0.768
0.16		0.842	0.682	0.559
0.19		0.791	0.753	0.705
0.32		0.768	0.5537	0.781
1		0.7256	0.715	0.703
6.23		0.481	0.3271	0.39

N= lab control

CETIS Analytical Report

Report Date: 16 Dec-19 14:34 (p 2 of 2)
Test Code/ID: 191281a / 01-3018-0898

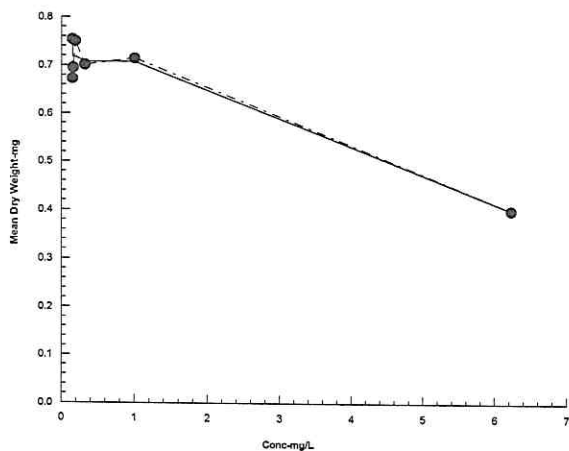
Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 06-6817-8280 Endpoint: Mean Dry Weight-mg
Analyzed: 16 Dec-19 14:34 Analysis: Linear Interpolation (ICPIN)

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 16 Dec-19 14:28 (p 1 of 2)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 20-6335-2881	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 14:28	Analysis: Parametric-Control vs Treatments	Status Level: 1
Batch ID: 03-4091-2684	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 16:15	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 26 Feb-19 15:45	Species: Pimephales promelas	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 16h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)	C > T	6.23	>6.23	n/a		15.02%

Dunnett Multiple Comparison Test

Control	vs	Conc-mg/L	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.15	1.15	2.532	0.239	4	CDF	0.3911	Non-Significant Effect
		0.151	0	2.532	0.239	4	CDF	0.8571	Non-Significant Effect
		0.19	0	2.532	0.239	4	CDF	0.8571	Non-Significant Effect
		0.32	1.076	2.532	0.239	4	CDF	0.4234	Non-Significant Effect
		1	0.575	2.532	0.239	4	CDF	0.6495	Non-Significant Effect
		6.23	2.06	2.532	0.239	4	CDF	0.1118	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0984944	0.0164157	6	1.226	0.3503	Non-Significant Effect
Error	0.187438	0.0133884	14			
Total	0.285933		20			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Levene Equality of Variance Test	5.372	4.456	0.0046	Unequal Variances
Variances	Mod Levene Equality of Variance Test	1.243	7.191	0.3871	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9068	0.871	0.0475	Normal Distribution

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15		3	0.9333	0.7899	1.0000	0.9000	0.9000	1.0000	0.0333	6.19%	0.00%
0.151		3	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-7.14%
0.16	N	3	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-7.14%
0.19		3	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-7.14%
0.32		3	0.9333	0.6465	1.0000	1.0000	0.8000	1.0000	0.0667	12.37%	0.00%
1		3	0.9667	0.8232	1.0000	1.0000	0.9000	1.0000	0.0333	5.97%	-3.57%
6.23		3	0.8667	0.4872	1.0000	0.9000	0.7000	1.0000	0.0882	17.63%	7.14%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15		3	1.303	1.07	1.537	1.249	1.249	1.412	0.05432	7.22%	0.00%
0.151		3	1.412	1.411	1.413	1.412	1.412	1.412	0	0.00%	-8.34%
0.16	N	3	1.412	1.411	1.413	1.412	1.412	1.412	0	0.00%	-8.34%
0.19		3	1.412	1.411	1.413	1.412	1.412	1.412	0	0.00%	-8.34%
0.32		3	1.31	0.8731	1.748	1.412	1.107	1.412	0.1016	13.43%	-0.54%
1		3	1.358	1.124	1.591	1.412	1.249	1.412	0.05432	6.93%	-4.17%
6.23		3	1.217	0.6903	1.745	1.249	0.9912	1.412	0.1225	17.43%	6.60%

N = 8 low control

CETIS Analytical Report

Report Date: 16 Dec-19 14:28 (p 2 of 2)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 20-6335-2881 Endpoint: 7d Survival Rate
Analyzed: 16 Dec-19 14:28 Analysis: Parametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	0.9000	1.0000	0.9000
0.151		1.0000	1.0000	1.0000
0.16		1.0000	1.0000	1.0000
0.19		1.0000	1.0000	1.0000
0.32		1.0000	0.8000	1.0000
1		0.9000	1.0000	1.0000
6.23		1.0000	0.7000	0.9000

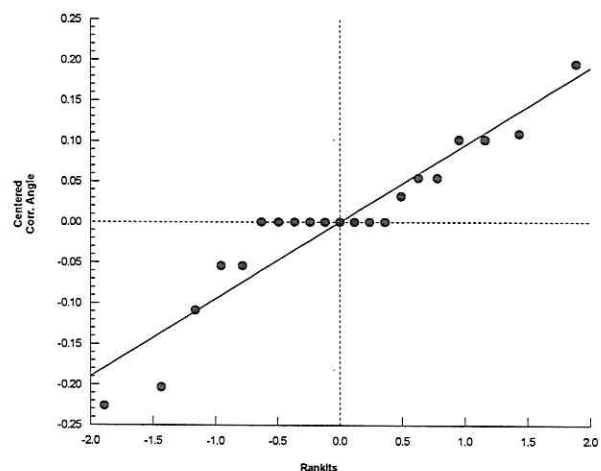
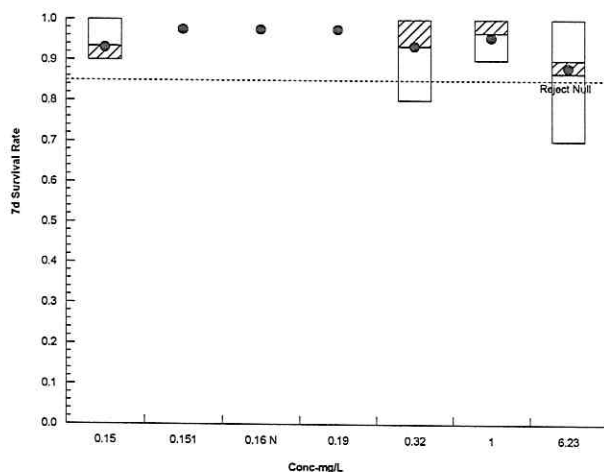
Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	1.249	1.412	1.249
0.151		1.412	1.412	1.412
0.16		1.412	1.412	1.412
0.19		1.412	1.412	1.412
0.32		1.412	1.107	1.412
1		1.249	1.412	1.412
6.23		1.412	0.9912	1.249

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15	N	9/10	10/10	9/10
0.151		10/10	10/10	10/10
0.16		10/10	10/10	10/10
0.19		10/10	10/10	10/10
0.32		10/10	8/10	10/10
1		9/10	10/10	10/10
6.23		10/10	7/10	9/10

Graphics



N = lab control

CETIS Analytical Report

Report Date: 16 Dec-19 14:29 (p 1 of 2)
Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 06-3059-3958	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 14:28	Analysis: Parametric-Control vs Treatments	Status Level: 1
Batch ID: 03-4091-2684	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 16:15	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 26 Feb-19 15:45	Species: Pimephales promelas	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 16h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	C > T	1	6.23	2.496		31.86%

Dunnett Multiple Comparison Test

Control	vs	Conc-mg/L	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.15	0.7707	2.532	0.221	4	CDF	0.5616	Non-Significant Effect
		0.151	-0.6752	2.532	0.221	4	CDF	0.9681	Non-Significant Effect
		0.19	-0.6332	2.532	0.221	4	CDF	0.9645	Non-Significant Effect
		0.32	0.3473	2.532	0.221	4	CDF	0.7434	Non-Significant Effect
		1	0.0458	2.532	0.221	4	CDF	0.8445	Non-Significant Effect
		6.23*	3.899	2.532	0.221	4	CDF	0.0039	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.338081	0.0563469	6	4.921	0.0066	Significant Effect
Error	0.1603	0.01145	14			
Total	0.498381		20			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	9.712	16.81	0.1373	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9539	0.871	0.4030	Normal Distribution

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15		3	0.627	0.4784	0.7756	0.659	0.558	0.664	0.03453	9.54%	0.00%
0.151		3	0.7533	0.6769	0.8297	0.768	0.718	0.774	0.01775	4.08%	-20.15%
0.16	N	3	0.6943	0.3418	1.047	0.682	0.559	0.842	0.08193	20.44%	-10.74%
0.19		3	0.7497	0.6426	0.8567	0.753	0.705	0.791	0.02488	5.75%	-19.56%
0.32		3	0.664	0.1883	1.14	0.768	0.443	0.781	0.1106	28.84%	-5.90%
1		3	0.6903	0.6086	0.772	0.703	0.653	0.715	0.01899	4.76%	-10.10%
6.23		3	0.3537	0.04061	0.6667	0.351	0.229	0.481	0.07276	35.63%	43.59%

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15		0.558	0.664	0.659
0.151		0.774	0.718	0.768
0.16	N	0.842	0.682	0.559
0.19		0.791	0.753	0.705
0.32		0.768	0.443	0.781
1		0.653	0.715	0.703
6.23		0.481	0.229	0.351

N = lab control

CETIS Analytical Report

Report Date: 16 Dec-19 14:29 (p 2 of 2)
Test Code/ID: 191281a / 01-3018-0898

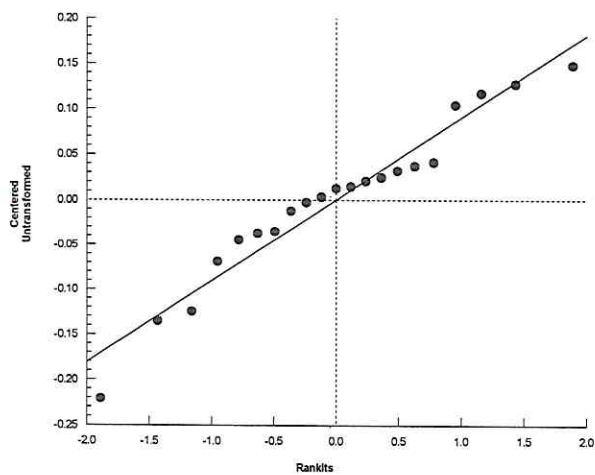
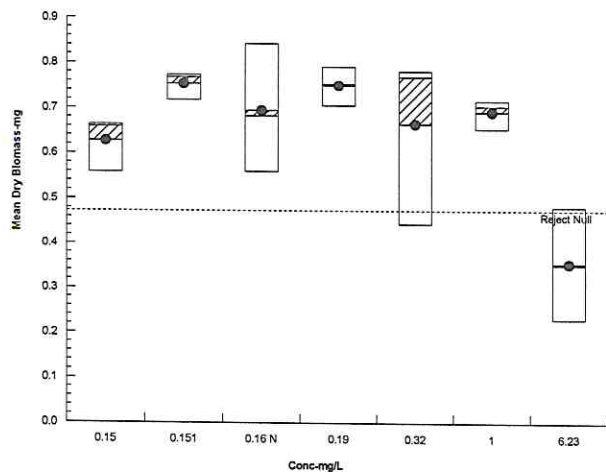
Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 06-3059-3958
Analyzed: 16 Dec-19 14:28
Endpoint: Mean Dry Biomass-mg
Analysis: Parametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 16 Dec-19 14:29 (p 1 of 2)
 Test Code/ID: 191281a / 01-3018-0898

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 06-5739-3261	Endpoint: Mean Dry Weight-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Dec-19 14:28	Analysis: Parametric-Control vs Treatments	Status Level: 1
Batch ID: 03-4091-2684	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 19 Feb-19 16:15	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 26 Feb-19 15:45	Species: Pimephales promelas	Brine:
Test Length: 6d 23h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 14-5122-1569	Code: 567FE241	Project:
Sample Date: 29 Jan-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 30 Jan-19 08:46	CAS (PC):	Station: DW-3
Sample Age: 21d 16h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	C > T	1	6.23	2.496		24.79%

Dunnett Multiple Comparison Test

Control	vs	Conc-mg/L	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.15	0.3274	2.532	0.172	4	CDF	0.7509	Non-Significant Effect
		0.151	-0.8677	2.532	0.172	4	CDF	0.9806	Non-Significant Effect
		0.19	-0.8137	2.532	0.172	4	CDF	0.9776	Non-Significant Effect
		0.32	-0.0967	2.532	0.172	4	CDF	0.8815	Non-Significant Effect
		1	-0.2969	2.532	0.172	4	CDF	0.9219	Non-Significant Effect
		6.23*	4.338	2.532	0.172	4	CDF	0.0017	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.270155	0.0450259	6	6.494	0.0019	Significant Effect
Error	0.0970641	0.0069332	14			
Total	0.367219		20			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	10.6	16.81	0.1016	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9687	0.871	0.7050	Normal Distribution

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15		3	0.6721	0.5316	0.8125	0.664	0.62	0.7322	0.03264	8.41%	0.00%
0.151		3	0.7533	0.6769	0.8297	0.768	0.718	0.774	0.01775	4.08%	-12.09%
0.16	N	3	0.6943	0.3418	1.047	0.682	0.559	0.842	0.08193	20.44%	-3.31%
0.19		3	0.7497	0.6426	0.8567	0.753	0.705	0.791	0.02488	5.75%	-11.54%
0.32		3	0.7009	0.3839	1.018	0.768	0.5537	0.781	0.07368	18.21%	-4.29%
1		3	0.7145	0.6865	0.7426	0.715	0.703	0.7256	0.006516	1.58%	-6.32%
6.23		3	0.3994	0.2072	0.5915	0.39	0.3271	0.481	0.04466	19.37%	40.57%

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3
0.15		0.62	0.664	0.7322
0.151		0.774	0.718	0.768
0.16	N	0.842	0.682	0.559
0.19		0.791	0.753	0.705
0.32		0.768	0.5537	0.781
1		0.7256	0.715	0.703
6.23		0.481	0.3271	0.39

N = last control

CETIS Analytical Report

Report Date: 16 Dec-19 14:29 (p 2 of 2)
Test Code/ID: 191281a / 01-3018-0898

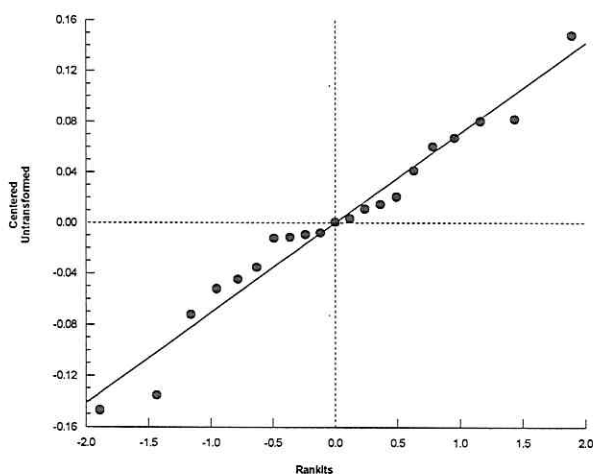
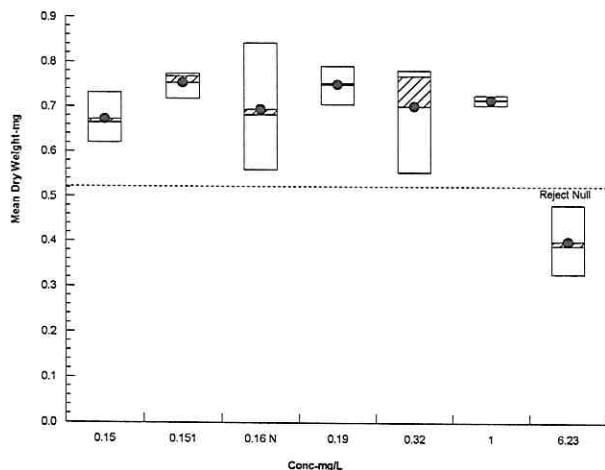
Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 06-5739-3261 Endpoint: Mean Dry Weight-mg
Analyzed: 16 Dec-19 14:28 Analysis: Parametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



Fathead Minnow Test Summary Sheet

Client: WDOE

Start Date & Time: Apr 23/19 @ 1340h

Work Order No.: 191281

Test Species: Pimephales promelas

Sample Information:

Sample ID: DW-3 (definitive)

Sample Date: Jan 29/19

Date Received: Jan 30/19

Sample Volume: 5x20L

Dilution Water:

Type: Moderately - hard water

Hardness (mg/L CaCO₃): 100

Alkalinity (mg/L CaCO₃): 70

Test Organism Information:

Batch No.: 042319

Source: Aquatic BioSystems, CO

Loading Density: 10 fish / 250 mL

NaCl Reference Toxicant Results:

Reference Toxicant ID: PP139

Stock Solution ID: NaCl

Date Initiated: Apr 23/19

7-d EC50 (95% CL): 4.2(3.6-4.9) g/L NaCl

7-d IC50 (95% CL): 3.5(3.1-4.2) g/L NaCl

Survival:

Reference Toxicant Mean \pm 2 SD: 4.7(3.7-6.1) g/L NaCl CV (%): 13

Biomass

Reference Toxicant Mean \pm 2 SD: 4.5(3.4-6.1) g/L NaCl CV (%): 15

Test Results:

	Survival	Biomass
NOEC % (v/v)	3.04	3.04
LOEC % (v/v)	4.33	4.33
EC25 % (v/v) (95% CL)	>4.33	
EC50 % (v/v) (95% CL)	>4.33	
IC25 % (v/v) (95% CL)		4.3(3.8-4.6)
IC50 % (v/v) (95% CL)		>4.33

Reviewed by: [Signature]

Date reviewed: Jan 16, 2020

7-d Chronic Freshwater Toxicity Test Initial and Final Water Quality Measurements

1/2

Client: WDOE
Sample ID: DW-3
Work Order #: 191281

Start Date & Time: April 23/19 @ 1240h
Stop Date & Time: Feb 19/19 MW
CER #: 11
Test Species: Pimephales promelas

Concentration 0% (v/v) control	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	25.0	24.0	24.0	24.0	25.0	24.0	25.0	24.0	25.0	24.0
DO (mg/L)	8.0	6.6	8.1	6.4	8.0	6.2	7.8	6.0	7.9	6.2	7.8	5.5	7.7	5.3
pH	8.0	7.6	8.1	7.6	8.1	7.6	8.0	7.4	7.9	7.6	7.9	7.6	8.0	7.4
Cond. (µS/cm)	330	321		323		326		321		322		329		320
Initials	MW	MW		MW		MW		A		A		MW		MW

Concentration Site ctrl	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0	24.0	24.0	24.0
DO (mg/L)	8.2	6.8	7.9	6.3	8.1	6.3	8.0	5.9	7.9	6.3	7.9	5.0	8.1	5.4
pH	7.7	7.9	8.0	7.9	8.0	7.7	7.7	7.8	7.7	7.7	7.7	7.7	8.0	7.4 MW
Cond. (µS/cm)	332	334		342		345		329		331		342		357
Initials	MW	MW		MW		MW		A		A		MW		MW

Concentration 12.5	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0	24.0	25.0	24.0
DO (mg/L)	8.1	6.7	7.9	6.3	8.0	6.2	7.8	6.0	7.7	6.2	7.8	5.0	7.7	5.3
pH	7.8	7.6	7.8	7.6	8.0	7.6	8.0	7.5	7.9	7.6	7.9	7.6	7.8	7.4
Cond. (µS/cm)	474	483		489		486		471		465		496		496
Initials	MW	MW		MW		MW		A		A		MW		MW

Concentration 25	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0	24.0	25.0	24.0
DO (mg/L)	8.1	6.7	7.9	6.3	8.0	6.3	7.9	5.9	7.8	6.2	7.8	4.6	7.8	5.3
pH	7.7	7.7	7.9	7.7	7.8	7.6	7.9	7.6	7.9	7.6	7.9	7.5	7.6	7.4
Cond. (µS/cm)	617	611		612		602		600		602		611		620
Initials	MW	MW		MW		MW		A		A		MW		MW

Thermometer: 4 DO meter/probe: 4, 4 pH meter/probe: 4, 4 Conductivity meter/probe: 4, 4

	Control			
Hardness*	100			
Alkalinity*	70			

Analysts: MW, AWD, CS

Reviewed by: [Signature]

Date reviewed: July 17/19

* mg/L as CaCO₃

Sample Description: DW-3: clear yellow, odorless, some particulates
Comments: MW-11: clear, light yellow, odorless, no particulates

7-d Chronic Freshwater Toxicity Test Initial and Final Water Quality Measurements

2/2

Client: WDOE
Sample ID: DW-3
Work Order #: 191281

Start Date & Time: Apr 23/19 @ 1340h
Stop Date & Time: Apr 30/19 @ 1425h
CER #: 11
Test Species: Pimephales promelas

Concentration 50	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0	24.0	25.0	24.0
DO (mg/L)	8.1	6.7	7.8	6.4	8.0	6.4	8.0	5.9	7.7	6.3	7.8	5.2	7.8	5.4
pH	7.4	7.7	7.5	7.7	7.8	7.7	7.7	7.6	7.9	7.6	7.8	7.6	7.4	7.4
Cond. (µS/cm)	897	889		896		894		886		879		903		917
Initials	ML	ML		ML		ML		ML		ML		ML		ML

Concentration 75 150	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.5	24.0	24.5	24.0	25.0	24.0
DO (mg/L)	8.2	6.7	7.8	6.4	8.1	6.4	8.1	6.0	7.9	6.2	7.8	5.7	7.9	5.9
pH	7.3	7.7	7.4	7.7	7.5	7.7	7.7	7.7	7.8	7.7	7.8	7.7	7.3	7.5
Cond. (µS/cm)	1185	1164		1201		1194		1186		1190		1188		1148
Initials	ML	ML		ML		ML		ML		ML		ML		ML

Concentration 150	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0
DO (mg/L)	8.3	6.8	7.9	6.3	8.0	6.5	8.3	6.0	8.0	6.3	7.9	5.9	8.1	5.7
pH	7.1	7.7	7.3	7.6	7.5	7.7	7.6	7.7	7.7	7.7	7.6	7.7	7.0	7.5
Cond. (µS/cm)	1469	1404		1490		1455		1442		1445		1482		1494
Initials	ML	ML		ML		ML		ML		ML		ML		ML

Concentration	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)														
DO (mg/L)														
pH														
Cond. (µS/cm)														
Initials														

Thermometer: 11 DO meter/probe: 4, 4 pH meter/probe: 4, 4 Conductivity meter/probe: 4, 4

	Control			
Hardness*	150			
Alkalinity*	70			

* mg/L as CaCO₃

Analysts: ML, ALW, CS

Reviewed by: ML
Date reviewed: July 17, 2019

Sample Description: _____

Comments: _____

7-d Fathead Minnow Toxicity Test **Daily Survival**

Client: WDOE
Sample ID: DW-3
Work Order #: 191281

Start Date & Time: Apr 23/19 08:40h
Stop Date & Time: Apr 30/19 14:25h
Test Species: Pimephales promelas

Concentration 0/0(v/v)	Rep	Day of Test - No. of Survivors							Comments
		1	2	3	4	5	6	7	
control	A	10 0	10	10	10	10	10	10	
	B	1			1	1	10 0	10	
	C						10	10	
	D						10	10	
site ctrl	A						10	10	
	B						9	9	
	C						9	9	
	D						10	10	
12.5	A						10	10	
	B						10	10	
	C						9	9	
	D						10	10	
25	A				↓	10	10	10	
	B				9	9	9	9	
	C				10	10	10	10	
	D						10	10	
50	A						9 0	7	
	B						9	9	
	C						9	9	
	D						10	10	
75	A						10	9	
	B						10	10	
	C				↓	1	10	10	
	D		↓		9	9	9	9	
100	A		10		10	10	9	7	
	B		10	↓	10	10	10	10	
	C		9 0	9	9	9	8	8	
	D	↓	10	10	10	10	9	8	
	A								
	B								
	C								
	D								
Tech Initials		ML	ML	ML	ML	8	ML	JW	

Comments: n.b. 5cm
-remaining organism appear normal

Reviewed by: [Signature]

Date reviewed: July 17, 2019


Fathead Minnow Toxicity Test Data Sheet

Dry Weight Data

Client: WDOEStart Date & Time: April 23/19 1345hSample ID: DM-3Termination Date & Time: April 30/19 1425hWork Order No.: PP B11x

Sample ID % (v/v)	Rep	Pan No.	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
Control	A	1	10	JW	1042.14	1048.93	10	GH/AT
	B	2	10 9		1047.36	1053.38	10	GH/AT
	C	3	10		1060.55	1066.81	10	GH/AT
	D	4	10		1009.57	1015.73	10	GH/AT
site control	A	5	10		1009.05	1015.95	10	GH/AT
	B	6	9		1055.09	1060.77	9	GH/AT
	C	7	9		1062.72	1068.70	9	GH/AT
	D	8	10		1046.48	1052.62	10	GH/AT
12.5	A	9	10		1073.01	1078.84	10	GH/AT
	B	10	10		1068.54	1074.58 ⁵ 7	10	GH/AT
	C	11	9		1079.60	1085.13	9	GH/AT
	D	12	10		1061.30	1067.69	10	GH/AT
25	A	13	10		1059.75	1066.04	10	GH/AT
	B	14	9		1046.75	1051.89	9	GH/AT
	C	15	10		1044.72	1055 ⁵ 0.40	10	GH/AT
	D	16	10		1051.24	1057.50	10	GH/AT
50	A	17	7		1055.97	1060.88 ⁵ 2	7	GH/AT
	B	18	9		1023.35	1028.97 ⁵ 8	9	GH/AT
	C	19	9		1063.16	1069.94	9	GH/AT
	D	20	10		1080.25	1086.97	10	GH/AT
75	A	21	9		1070.87	1075.91	9	GH/AT
	B	22	10		983.59	989.47	10	GH/AT
	C	23	10		993.21	999.35	10	GH/AT
	D	24	9		1052.49	1058.68 ⁵ 5	9	GH/AT

Comments: 10% re-weigh: (1) pan: 2 weight: 1053.40 (2) pan: 17 weight: 1060.77
(3) pan: 26 weight: 1059.43

Reviewed by: Date Reviewed: July 17, 2019


Fathead Minnow Toxicity Test Data Sheet

Dry Weight Data

Client: INDOEStart Date & Time: Apr 23/19 @ 1340hSample ID: DW-3Termination Date & Time: Apr 30/19 @ 1425hWork Order No.: PP Blue

Sample ID o/o (v/v)	Rep	Pan No.	No. alive	Initials	Pan weight (mg)	Pan + organism (mg)	No. weighed	Initials
100	A	25	7	JW	1052.99	1057.18	7	GH/AT
	B	26	10	↓	1053.61	1059.57	10	GH/AT
	C	27	8	↓	1055.99	1059.71	8	GH/AT
	D	28	8	↓	1056.39	1060.31	8	GH/AT
	A							
	B							
	C							
	D							
	A							
	B							
	C							
	D							
	A							
	B							
	C							
	D							
	A							
	B							
	C							
	D							
	A							
	B							
	C							
	D							

Comments: _____

Reviewed by: Date Reviewed: July 17, 2019

W.O.#: 191281

[illegible]

Reviewed by:

Date Reviewed:

CETIS Summary Report

Report Date: 16 Jan-20 11:09 (p 1 of 3)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Batch ID: 05-6678-6169	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 13:40	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 30 Apr-19 14:25	Species: Pimephales promelas	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 14h	Client: WA State Dept of Ecology	

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓ NOEL	LOEL	TOEL	TU	PMSD	S
09-7701-8224	7d Survival Rate	Dunnett Multiple Comparison Test	3.04	4.33	3.628		12.5%	1
03-5664-9281	Mean Dry Biomass-mg	Dunnett Multiple Comparison Test	3.04	4.33	3.628		18.2%	1
19-2257-6075	Mean Dry Weight-mg	Dunnett Multiple Comparison Test	3.04	4.33	3.628		12.7%	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓ Level	mg/L	95% LCL	95% UCL	TU	S
15-1142-7854	7d Survival Rate	Linear Interpolation (ICPIN)	✓ EC5	1.243	0.3571	4.283		1
			EC10	3.203	0.3878	5.006		
			EC15	3.924	2.509	n/a		
			EC20	>4.33	n/a	n/a		
			EC25	>4.33	n/a	n/a		
			EC40	>4.33	n/a	n/a		
			EC50	>4.33	n/a	n/a		
18-8963-1070	Mean Dry Biomass-mg	NLR: 3P Log-Logistic	IC5	3.194	n/a	3.997		1
			IC10	3.598	n/a	4.294		
			IC15	3.873	2.495	4.425		
			IC20	4.095	3.512	4.506		
			IC25	4.287	3.844	4.647		
			IC40	4.788	3.677	5.791		
			IC50	5.108	3.273	7.972		
08-3875-3809	Mean Dry Weight-mg	NLR: 3P Log-Logistic	IC5	3.589	n/a	4.489		1
			IC10	4.037	n/a	4.61		
			IC15	4.343	3.707	4.757		
			IC20	4.588	2.813	5.316		
			IC25	4.801	n/a	6.025		
			IC40	5.355	n/a	n/a		
			IC50	5.708	n/a	n/a		

CETIS Summary Report

Report Date: 16 Jan-20 11:09 (p 2 of 3)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0.17	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%
0.21	XC	4	0.9500	0.8581	1.0000	0.9000	1.0000	0.0289	0.0577	6.08%	5.00%
0.51		4	0.9750	0.8954	1.0000	0.9000	1.0000	0.0250	0.0500	5.13%	2.50%
0.89		4	0.9750	0.8954	1.0000	0.9000	1.0000	0.0250	0.0500	5.13%	2.50%
1.9		4	0.8750	0.6748	1.0000	0.7000	1.0000	0.0629	0.1258	14.38%	12.50%
3.04		4	0.9500	0.8581	1.0000	0.9000	1.0000	0.0289	0.0577	6.08%	5.00%
4.33		4	0.8250	0.6248	1.0000	0.7000	1.0000	0.0629	0.1258	15.25%	17.50%

Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0.17	N	4	0.6308	0.5772	0.6843	0.602	0.679	0.01682	0.03364	5.33%	0.00%
0.21	XC	4	0.6175	0.5348	0.7002	0.568	0.69	0.02598	0.05196	8.41%	2.10%
0.51		4	0.5945	0.5371	0.6519	0.553	0.639	0.01804	0.03608	6.07%	5.75%
0.89		4	0.5843	0.4974	0.6711	0.514	0.629	0.0273	0.0546	9.35%	7.37%
1.9		4	0.5995	0.4518	0.7472	0.485	0.678	0.04642	0.09284	15.49%	4.96%
3.04		4	0.5805	0.4969	0.6641	0.504	0.616	0.02628	0.05257	9.06%	7.97%
4.33		4	0.4448	0.2814	0.6081	0.372	0.596	0.05133	0.1027	23.08%	29.49%

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0.17	N	4	0.6308	0.5772	0.6843	0.602	0.679	0.01682	0.03364	5.33%	0.00%
0.21	XC	4	0.6499	0.5958	0.7039	0.614	0.69	0.01698	0.03397	5.23%	-3.03%
0.51		4	0.6099	0.5727	0.647	0.583	0.639	0.01168	0.02337	3.83%	3.31%
0.89		4	0.5985	0.5452	0.6518	0.568	0.629	0.01675	0.0335	5.60%	5.11%
1.9		4	0.6859	0.6016	0.7703	0.6256	0.7533	0.0265	0.05301	7.73%	-8.75%
3.04		4	0.6116	0.5268	0.6965	0.56	0.6844	0.02666	0.05333	8.72%	3.03%
4.33		4	0.5374	0.4261	0.6486	0.465	0.5986	0.03496	0.06992	13.01%	14.80%

N = (ab) control
XC = site control (MW-11)

CETIS Summary Report

Report Date: 16 Jan-20 11:09 (p 3 of 3)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	1.0000	1.0000	1.0000	1.0000
0.21	XC	1.0000	0.9000	0.9000	1.0000
0.51		1.0000	1.0000	0.9000	1.0000
0.89		1.0000	0.9000	1.0000	1.0000
1.9		0.7000	0.9000	0.9000	1.0000
3.04		0.9000	1.0000	1.0000	0.9000
4.33		0.7000	1.0000	0.8000	0.8000

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	0.679	0.602	0.626	0.616
0.21	XC	0.69	0.568	0.598	0.614
0.51		0.583	0.603	0.553	0.639
0.89		0.629	0.514	0.568	0.626
1.9		0.485	0.563	0.678	0.672
3.04		0.504	0.588	0.614	0.616
4.33		0.419	0.596	0.372	0.392

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	0.679	0.602	0.626	0.616
0.21	XC	0.69	0.6311	0.6644	0.614
0.51		0.583	0.603	0.6144	0.639
0.89		0.629	0.5711	0.568	0.626
1.9		0.6929	0.6256	0.7533	0.672
3.04		0.56	0.588	0.614	0.6844
4.33		0.5986	0.596	0.465	0.49

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	10/10	10/10	10/10	10/10
0.21	XC	10/10	9/10	9/10	10/10
0.51		10/10	10/10	9/10	10/10
0.89		10/10	9/10	10/10	10/10
1.9		7/10	9/10	9/10	10/10
3.04		9/10	10/10	10/10	9/10
4.33		7/10	10/10	8/10	8/10

N = lab control

XC = site control (MW-11)

CETIS Analytical Report

Report Date: 16 Jan-20 11:13 (p 1 of 2)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 15-1142-7854	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 16 Jan-20 11:05	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 05-6678-6169	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 13:40	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 30 Apr-19 14:25	Species: Pimephales promelas	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 14h	Client: WA State Dept of Ecology	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	1567867	200	Yes	Two-Point Interpolation

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC5	1.243	0.3571	4.283
EC10	3.203	0.3878	5.006
EC15	3.924	2.509	n/a
EC20	>4.33	n/a	n/a
EC25	>4.33	n/a	n/a
EC40	>4.33	n/a	n/a
EC50	>4.33	n/a	n/a

7d Survival Rate Summary

Conc-mg/L	Code	Count	Calculated Variate(A/B)							Isotonic Variate	
			Mean	Min	Max	Std Dev	CV%	%Effect	A/B	Mean	%Effect
0.17	N	4	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	40/40	1	0.0%
0.51		4	0.9750	0.9000	1.0000	0.0500	5.13%	2.5%	39/40	0.975	2.5%
0.89		4	0.9750	0.9000	1.0000	0.0500	5.13%	2.5%	39/40	0.975	2.5%
1.9		4	0.8750	0.7000	1.0000	0.1258	14.38%	12.5%	35/40	0.9125	8.75%
3.04		4	0.9500	0.9000	1.0000	0.0577	6.08%	5.0%	38/40	0.9125	8.75%
4.33		4	0.8250	0.7000	1.0000	0.1258	15.25%	17.5%	33/40	0.825	17.5%

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	1.0000	1.0000	1.0000	1.0000
0.51		1.0000	1.0000	0.9000	1.0000
0.89		1.0000	0.9000	1.0000	1.0000
1.9		0.7000	0.9000	0.9000	1.0000
3.04		0.9000	1.0000	1.0000	0.9000
4.33		0.7000	1.0000	0.8000	0.8000

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	10/10	10/10	10/10	10/10
0.51		10/10	10/10	9/10	10/10
0.89		10/10	9/10	10/10	10/10
1.9		7/10	9/10	9/10	10/10
3.04		9/10	10/10	10/10	9/10
4.33		7/10	10/10	8/10	8/10

CETIS Analytical Report

Report Date: 16 Jan-20 11:13 (p 2 of 2)
Test Code/ID: 191281b / 12-5973-4283

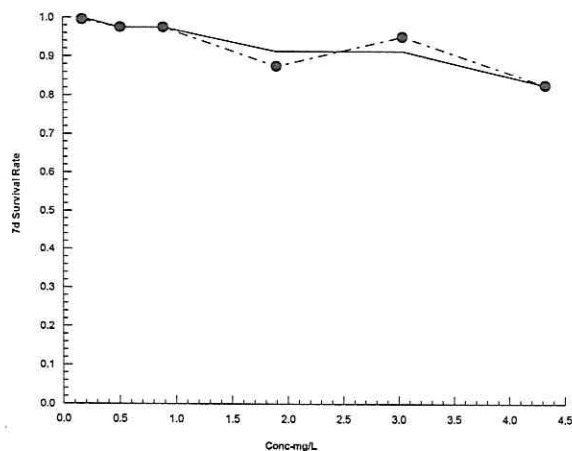
Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 15-1142-7854 Endpoint: 7d Survival Rate
Analyzed: 16 Jan-20 11:05 Analysis: Linear Interpolation (ICPIN)

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 16 Jan-20 11:08 (p 1 of 2)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 18-8963-1070	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Jan-20 11:05	Analysis: Nonlinear Regression (NLR)	Status Level: 1
Batch ID: 05-6678-6169	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 13:40	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 30 Apr-19 14:25	Species: Pimephales promelas	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 14h	Client: WA State Dept of Ecology	

Non-Linear Regression Options

Model Name and Function	Weighting Function	PTBS Function	X Trans	Y Trans
3P Log-Logistic: $\mu=\alpha/[1+(x/\delta)^y]$	Normal [$w=1$]	Off [$\mu^*=\mu$]	None	None

Regression Summary

Iters	Log LL	AICc	BIC	Adj R2	Optimize	F Stat	Critical	P-Value	Decision($\alpha:5\%$)
16	66.96	-126.7	-124.4	0.4291	Yes	0.3506	3.16	0.7892	Non-Significant Lack of Fit

Point Estimates

Level	mg/L	95% LCL	95% UCL
IC5	3.194	n/a	3.997
IC10	3.598	n/a	4.294
IC15	3.873	2.495	4.425
IC20	4.095	3.512	4.506
IC25	4.287	3.844	4.647
IC40	4.788	3.677	5.791
IC50	5.108	3.273	7.972

Regression Parameters

Parameter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision($\alpha:5\%$)
α	0.6026	0.01658	0.5681	0.6371	36.35	<1.0E-37	Significant Parameter
y	6.27	4.716	-3.537	16.08	1.33	0.1979	Non-Significant Parameter
δ	5.108	0.6789	3.696	6.52	7.524	2.2E-07	Significant Parameter

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision($\alpha:5\%$)
Model	7.942	2.647	3	640.5	<1.0E-37	Significant
Lack of Fit	0.004792	0.001597	3	0.3506	0.7892	Non-Significant
Pure Error	0.08201	0.004556	18			
Residual	0.0868	0.004133	21			

Residual Analysis

Attribute	Method	Test Stat	Critical	P-Value	Decision($\alpha:5\%$)
Variances	Bartlett Equality of Variance Test	5.559	11.07	0.3515	Equal Variances
	Mod Levene Equality of Variance	0.8775	2.773	0.5157	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9809	0.9169	0.9121	Normal Distribution
	Anderson-Darling A2 Normality Te	0.2193	2.492	0.8715	Normal Distribution

Mean Dry Biomass-mg Summary

			Calculated Variate						
Conc-mg/L	Code	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0.17	N	4	0.6308	0.602	0.679	0.01682	0.03364	5.33%	0.0%
0.51		4	0.5945	0.553	0.639	0.01804	0.03608	6.07%	5.75%
0.89		4	0.5843	0.514	0.629	0.0273	0.0546	9.35%	7.37%
1.9		4	0.5995	0.485	0.678	0.04642	0.09284	15.49%	4.96%
3.04		4	0.5805	0.504	0.616	0.02628	0.05257	9.06%	7.97%
4.33		4	0.4448	0.372	0.596	0.05133	0.1027	23.08%	29.49%

CETIS Analytical Report

Report Date: 16 Jan-20 11:08 (p 2 of 2)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 18-8963-1070
Analyzed: 16 Jan-20 11:05

Endpoint: Mean Dry Biomass-mg
Analysis: Nonlinear Regression (NLR)

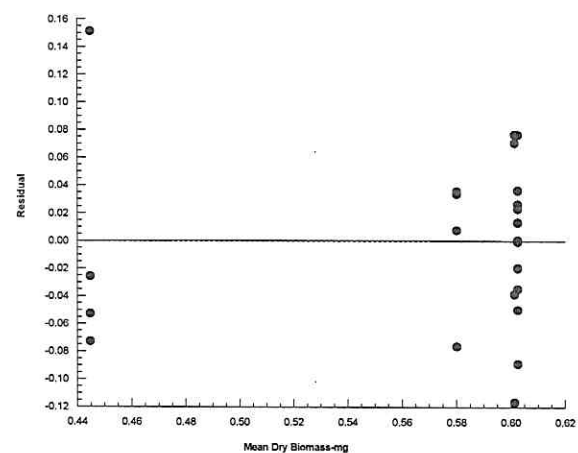
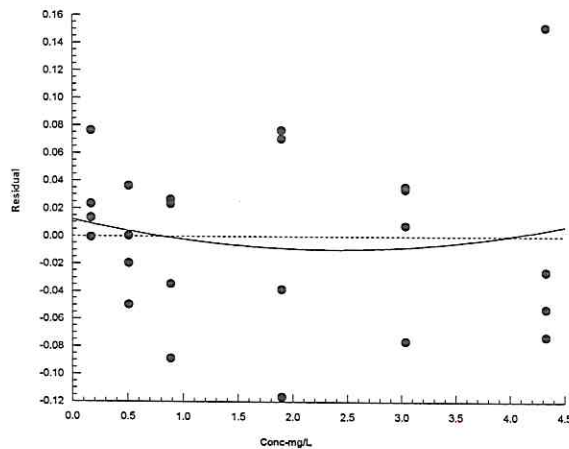
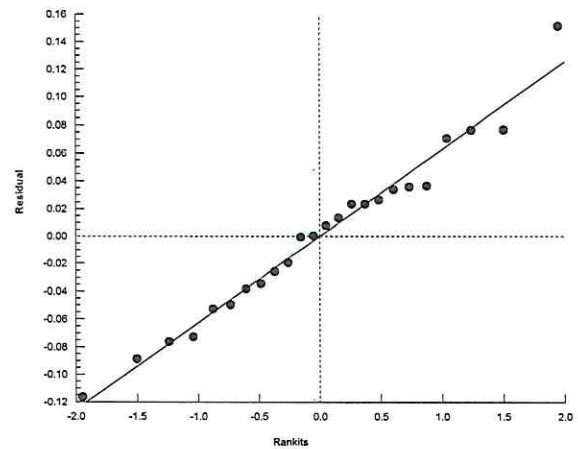
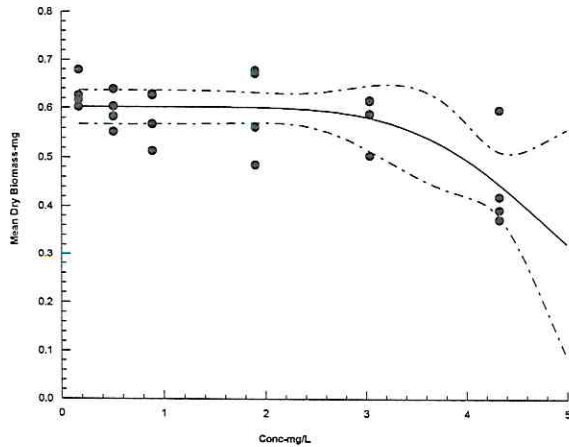
CETIS Version: CETISv1.9.4
Status Level: 1

Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	0.679	0.602	0.626	0.616
0.51		0.583	0.603	0.553	0.639
0.89		0.629	0.514	0.568	0.626
1.9		0.485	0.563	0.678	0.672
3.04		0.504	0.588	0.614	0.616
4.33		0.419	0.596	0.372	0.392

Graphics

Model: 3P Log-Logistic: $\mu = \alpha / [1 + (x/\delta)^\gamma]$ Distribution: Normal [$\omega=1$]



CETIS Analytical Report

Report Date: 16 Jan-20 11:08 (p 1 of 2)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 08-3875-3809	Endpoint: Mean Dry Weight-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Jan-20 11:05	Analysis: Nonlinear Regression (NLR)	Status Level: 1
Batch ID: 05-6678-6169	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 13:40	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 30 Apr-19 14:25	Species: Pimephales promelas	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 14h	Client: WA State Dept of Ecology	

Non-Linear Regression Options

Model Name and Function	Weighting Function	PTBS Function	X Trans	Y Trans
3P Log-Logistic: $\mu=\alpha/[1+(x/\delta)^y]$	Normal [$w=1$]	Off [$\mu^*=\mu$]	None	None

Regression Summary

Iters	Log LL	AICc	BIC	Adj R2	Optimize	F Stat	Critical	P-Value	Decision($\alpha:5\%$)
34	71.67	-136.1	-133.8	0.2565	Yes	2.782	3.16	0.0707	Non-Significant Lack of Fit

Point Estimates

Level	mg/L	95% LCL	95% UCL
IC5	3.589	n/a	4.489
IC10	4.037	n/a	4.61
IC15	4.343	3.707	4.757
IC20	4.588	2.813	5.316
IC25	4.801	n/a	6.025
IC40	5.355	n/a	n/a
IC50	5.708	n/a	n/a

Regression Parameters

Parameter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision($\alpha:5\%$)
α	0.6298	0.0136	0.6016	0.6581	46.32	<1.0E-37	Significant Parameter
y	6.345	7.501	-9.254	21.94	0.8459	0.4072	Non-Significant Parameter
δ	5.708	1.873	1.814	9.602	3.048	0.0061	Significant Parameter

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision($\alpha:5\%$)
Model	9.027	3.009	3	1079	<1.0E-37	Significant
Lack of Fit	0.01856	0.006187	3	2.782	0.0707	Non-Significant
Pure Error	0.04003	0.002224	18			
Residual	0.05859	0.00279	21			

Residual Analysis

Attribute	Method	Test Stat	Critical	P-Value	Decision($\alpha:5\%$)
Variances	Bartlett Equality of Variance Test	3.972	11.07	0.5534	Equal Variances
	Mod Levene Equality of Variance	1.835	2.773	0.1567	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9349	0.9169	0.1257	Normal Distribution
	Anderson-Darling A2 Normality Te	0.6582	2.492	0.0861	Normal Distribution

Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Calculated Variate						
			Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0.17	N	4	0.6308	0.602	0.679	0.01682	0.03364	5.33%	0.0%
0.51		4	0.6099	0.583	0.639	0.01168	0.02337	3.83%	3.31%
0.89		4	0.5985	0.568	0.629	0.01675	0.0335	5.60%	5.11%
1.9		4	0.6859	0.6256	0.7533	0.0265	0.05301	7.73%	-8.75%
3.04		4	0.6116	0.56	0.6844	0.02666	0.05333	8.72%	3.04%
4.33		4	0.5374	0.465	0.5986	0.03496	0.06992	13.01%	14.8%

CETIS Analytical Report

Report Date: 16 Jan-20 11:08 (p 2 of 2)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 08-3875-3809
Analyzed: 16 Jan-20 11:05
Endpoint: Mean Dry Weight-mg
Analysis: Nonlinear Regression (NLR)

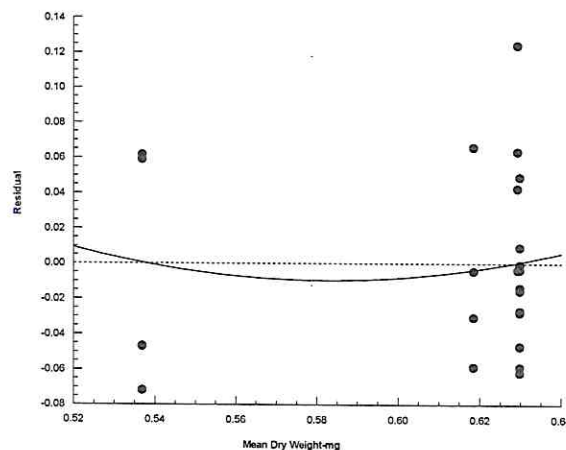
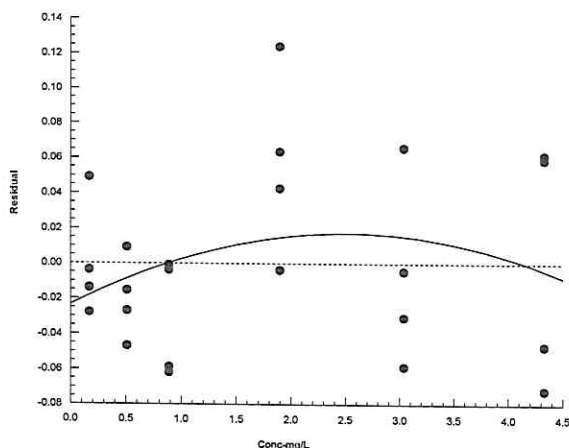
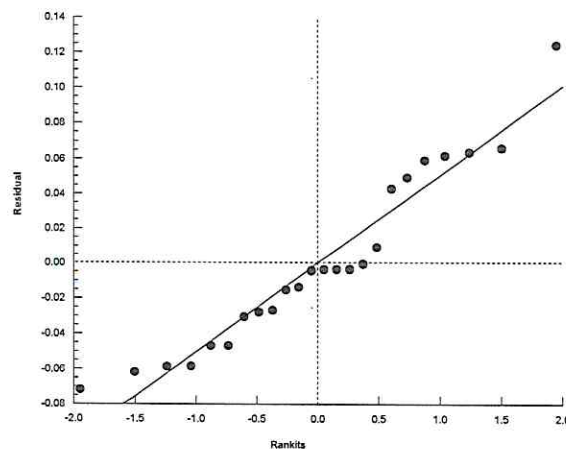
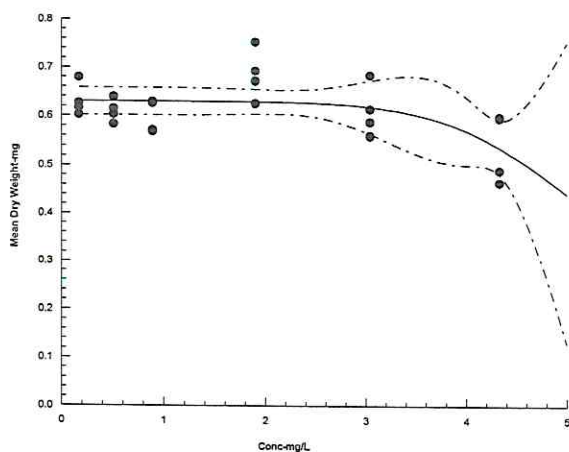
CETIS Version: CETISv1.9.4
Status Level: 1

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	0.679	0.602	0.626	0.616
0.51		0.583	0.603	0.6144	0.639
0.89		0.629	0.5711	0.568	0.626
1.9		0.6929	0.6256	0.7533	0.672
3.04		0.56	0.588	0.614	0.6844
4.33		0.5986	0.596	0.465	0.49

Graphics

Model: 3P Log-Logistic: $\mu = a/[1+(x/\delta)^\gamma]$ Distribution: Normal [$\omega=1$]



CETIS Analytical Report

Report Date: 16 Jan-20 11:09 (p 1 of 2)
 Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 18-2774-3121	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 16 Jan-20 11:09	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 05-6678-6169	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 13:40	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 30 Apr-19 14:25	Species: Pimephales promelas	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 14h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	Site Control passed 7d survival rate	6.13%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Site Control	1.732	1.943	0.091	6	CDF	0.0670	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0132797	0.0132797	1	3	0.1340	Non-Significant Effect
Error	0.0265593	0.0044266	6			
Total	0.039839		7			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Distribution	Shapiro-Wilk W Normality Test	0.8489	0.6451	0.0929	Normal Distribution

7d Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.17	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
0.21	XC	4	0.9500	0.8581	1.0000	0.9500	0.9000	1.0000	0.0289	6.08%	5.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.17	N	4	1.412	1.412	1.412	1.412	1.412	1.412	0	0.00%	0.00%
0.21	XC	4	1.331	1.181	1.48	1.331	1.249	1.412	0.04705	7.07%	5.77%

7d Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	1.0000	1.0000	1.0000	1.0000
0.21	XC	1.0000	0.9000	0.9000	1.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	1.412	1.412	1.412	1.412
0.21	XC	1.412	1.249	1.249	1.412

7d Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	10/10	10/10	10/10	10/10
0.21	XC	10/10	9/10	9/10	10/10

N = lab control
 XC = site control (MW-11)

CETIS Analytical Report

Report Date: 16 Jan-20 11:09 (p 2 of 2)
Test Code/ID: 191281b / 12-5973-4283

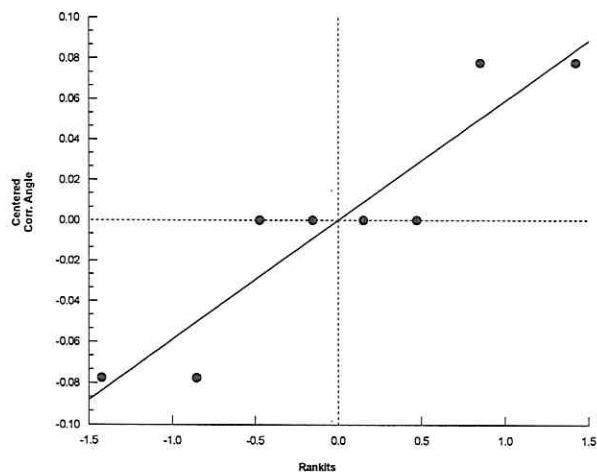
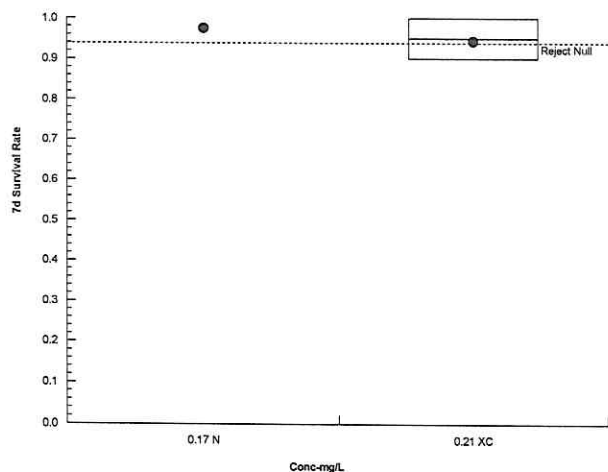
Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 18-2774-3121 Endpoint: 7d Survival Rate
Analyzed: 16 Jan-20 11:09 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 16 Jan-20 11:09 (p 1 of 1)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 11-7099-3661	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Jan-20 11:09	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 05-6678-6169	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 13:40	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 30 Apr-19 14:25	Species: Pimephales promelas	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 14h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	Comparison Result	PMSD
Untransformed	C > T	Site Control passed mean dry biomass-mg	9.53%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Site Control	0.4281	1.943	0.060	6	CDF	0.3418	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0003511	0.0003511	1	0.1833	0.6835	Non-Significant Effect
Error	0.011494	0.0019157	6			
Total	0.0118451		7			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	2.385	47.47	0.4939	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.897	0.6451	0.2715	Normal Distribution

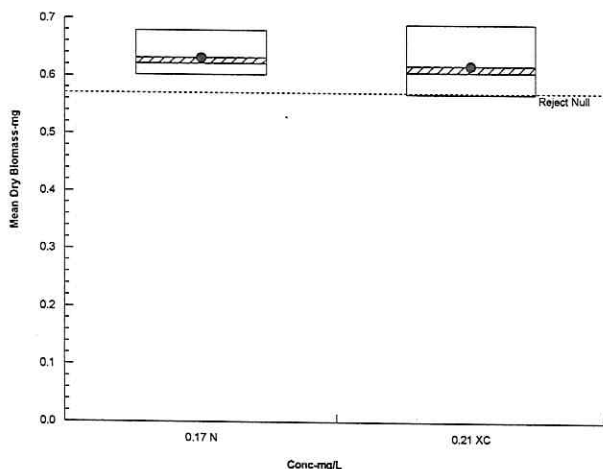
Mean Dry Biomass-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.17	N	4	0.6308	0.5772	0.6843	0.621	0.602	0.679	0.01682	5.33%	0.00%
0.21	XC	4	0.6175	0.5348	0.7002	0.606	0.568	0.69	0.02598	8.41%	2.10%

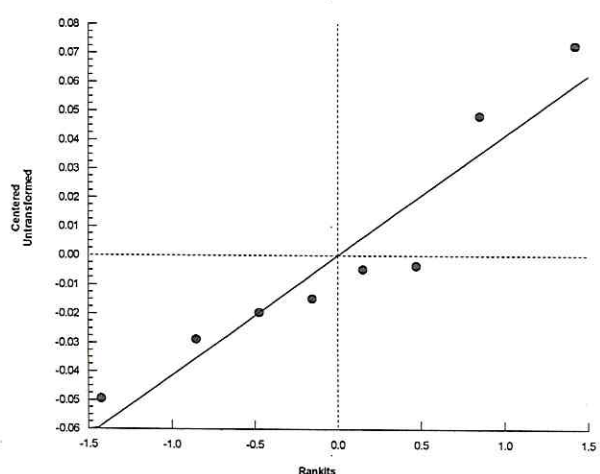
Mean Dry Biomass-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	0.679	0.602	0.626	0.616
0.21	XC	0.69	0.568	0.598	0.614

Graphics



N = lab control
XC = site control



CETIS Analytical Report

Report Date: 16 Jan-20 11:10 (p 1 of 1)
Test Code/ID: 191281b / 12-5973-4283

Fathead Minnow 7-d Larval Survival and Growth Test

Nautilus Environmental

Analysis ID: 19-0918-7801	Endpoint: Mean Dry Weight-mg	CETIS Version: CETISv1.9.4
Analyzed: 16 Jan-20 11:09	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 05-6678-6169	Test Type: Growth-Survival (7d)	Analyst: Mimi Tran
Start Date: 23 Apr-19 13:40	Protocol: EC/EPS 1/RM/22	Diluent: Mod-Hard Synthetic Water
Ending Date: 30 Apr-19 14:25	Species: Pimephales promelas	Brine:
Test Length: 7d 1h	Taxon: Actinopterygii	Source: Aquatic Biosystems, CO Age: <24
Sample ID: 17-4881-3663	Code: 683CC75F	Project:
Sample Date: 19 Mar-19	Material: Diesel	Source: WA Dept of Ecology
Receipt Date: 20 Mar-19 11:07	CAS (PC):	Station: DW-3
Sample Age: 35d 14h	Client: WA State Dept of Ecology	

Data Transform	Alt Hyp	Comparison Result	PMSD
Untransformed	C > T	Site Control passed mean dry weight-mg	7.36%

Equal Variance t Two-Sample Test

Control	vs	Control II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		Site Control	-0.8007	1.943	0.046	6	CDF	0.7731	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0007327	0.0007327	1	0.6412	0.4538	Non-Significant Effect
Error	0.0068563	0.0011427	6			
Total	0.0075889		7			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	1.019	47.47	0.9877	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9124	0.6451	0.3713	Normal Distribution

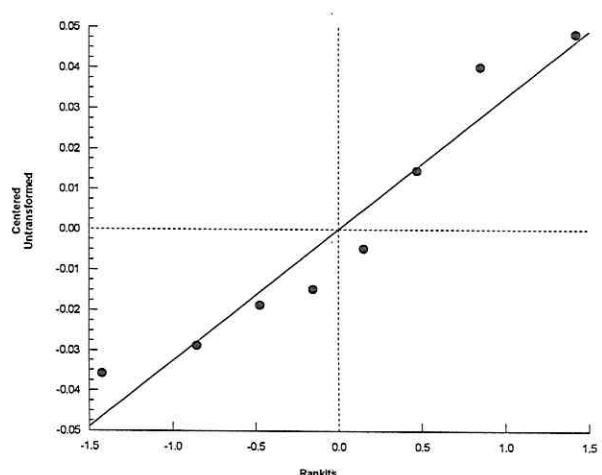
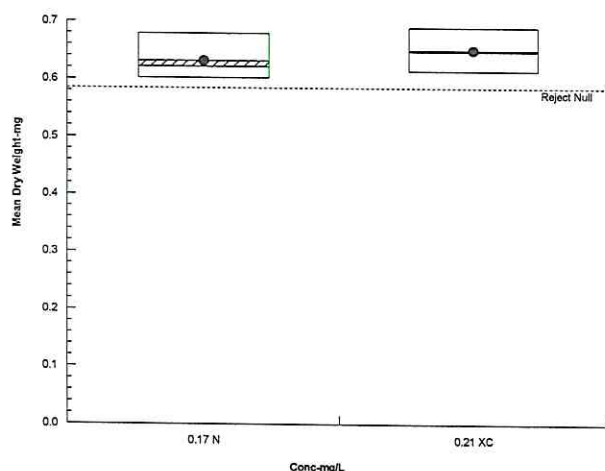
Mean Dry Weight-mg Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.17	N	4	0.6308	0.5772	0.6843	0.621	0.602	0.679	0.01682	5.33%	0.00%
0.21	XC	4	0.6499	0.5958	0.7039	0.6478	0.614	0.69	0.01698	5.23%	-3.03%

Mean Dry Weight-mg Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0.17	N	0.679	0.602	0.626	0.616
0.21	XC	0.69	0.6311	0.6644	0.614

Graphics



N=lab control
XC=site control

APPENDIX D – *Ceriodaphnia dubia* Toxicity Test Data

Ceriodaphnia dubia Summary Sheet

Client: WDOE
Work Order No.: 191282

Start Date/Time: Mar 11/19 @ 800h
Set up by: KL/SSIC

Sample Information:

Sample ID: DW-3 (range finder)
Sample Date: Jan 29/19
Date Received: Jan 30/19
Sample Volume: 5x20L

Test Validity Criteria:

- 1) Mean survival of first generation controls is $\geq 80\%$
- 2) At least 60% of controls have produced three broods within 8 days
- 3) An average of ≥ 15 live young produced per surviving female in the control solutions during the first three broods.
- 4) Invalid if ephippia observed in any control solution at any time.

WQ Ranges:

T ($^{\circ}\text{C}$) = 25 ± 1 ; DO (mg/L) = 4.0 to 8.4; pH = 6.0 to 9.0

Test Organism Information:

Broodstock No.: B5022719A
Age of young (Day 0): <24-h (within 8-h)
Avg No. young in first 3 broods of previous 7 d: 42
Mortality (%) in previous 7 d: 0
Individual female # used ≥ 8 young on test day: 4, 6-8, 10-12, 17, 20

NaCl Reference Toxicant Results:

Reference Toxicant ID: Cd215
Stock Solution ID: 18Na05 (100g/L)
Date Initiated: Mar 5/19

7-d LC50 (95% CL): 2.0 (1.7-2.3) g/L NaCl
7-d IC50 (95% CL): 1.8 (1.4-2.0) g/L NaCl

7-d LC50 Reference Toxicant Mean and Historical Range: 2.0 (1.8-2.2) g/L NaCl CV (%): 5
7-d IC50 Reference Toxicant Mean and Historical Range: 1.5 (1.0-2.1) g/L NaCl CV (%): 18

Test Results:

	Survival	Reproduction
LC50 %v/v (95% CL)	<u>>100 5.53</u>	
IC25 %v/v (95% CL)		<u>>100 5.53</u>
IC50 %v/v (95% CL)		<u>>100 5.53</u>
NOEC %v/v	<u>100 5.53</u>	<u>>100 5.53</u>
LOEC %v/v	<u>>100 >5.53</u>	<u>>100 >5.53</u>

Reviewed by: [Signature]

Date reviewed: Aug 14/19

Chronic Freshwater Toxicity Test Initial and Final Water Quality Measurements

Client: WDOE
Sample ID: DW-3
Work Order #: WDOE cerro range binder 191282

Start Date & Time: Mar 11/19 @ 1500h
Stop Date & Time: Mar 17/20 @ 1500h
CER #: 4
Test Species: Ceriodaphnia dubia

7-(V10) Concentration CONTROL	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.1	8.3	8.0	8.1	8.1	8.1	8.0	8.1	8.1	7.3	5.1	7.2		
pH	8.2	8.2	8.2	8.2	7.9	7.9	8.2	8.1	8.2	7.9	5.2	7.9		
Cond. (µS/cm)	219	223		223		221		220		220		229		
Initials	JB	SSK		SSK		SSK		SSK		A		A		

Concentration SITE CONTROL	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.4	8.3	8.2	8.3	8.4	8.3	8.4	8.7	8.1	7.4	5.0	7.4		
pH	7.9	8.2	8.2	8.1	8.1	8.2	8.3	8.2	8.2	8.1	5.1	8.0		
Cond. (µS/cm)	212	321		325		320		318		312		322		
Initials	JB	SSK		SSK		SSK		SSK		A		A		

①8.0 ②3.5

Concentration 0.10	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.3	8.3	8.3	8.3	8.3	8.3	8.3	7.3	8.1	7.3	5.0	7.2		
pH	7.9	8.1	8.1	8.1	8.1	8.2	8.3	8.0	8.2	8.1	5.1	5.0		
Cond. (µS/cm)	212	221		222		223		224		226		235		
Initials	JB	SSK		SSK		SSK		SSK		A		A		

Concentration 0.39	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.4	8.3	8.3	8.3	8.3	8.3	8.3	7.3	8.1	7.4	5.1	7.3		
pH	7.9	8.1	8.1	8.1	8.1	8.2	8.3	8.0	8.2	8.0	5.1	5.0		
Cond. (µS/cm)	219	216		219		220		225		226		236		
Initials	JB	SSK		SSK		SSK		SSK		A		A		

Thermometer: 4 DO meter/probe: 1 / 1 pH meter/probe: 1 / 1 Conductivity meter/probe: 1 / 1

	Control	MW11 (SRL) DW-3 (W-2)
Hardness*	102	180
Alkalinity*	100	164

* mg/L as CaCO₃

Analysts: JB, AWD, SSK

Reviewed by: [Signature]

Date reviewed: Aug. 14, 2019

Sample Description: Brown, some particulates, organic (hydrocarbon odour),

Comments: Broodboard Used: 000271A (4, 6-8, 10-12, 17, 20)

2/2

Chronic Freshwater Toxicity Test Initial and Final Water Quality Measurements

Client: WDOE
Sample ID: DW-3
Work Order #: WDOE Cero Range Finder
191282

Start Date & Time: MARCH 9 @ 1500h
Stop Date & Time: MARCH 12 @ 1500h
CER #: 4
Test Species: Ceriodaphnia dubia

Concentration <u>106</u>	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.4	8.3	8.3	8.3	8.3	8.3	8.3	7.3	8.0	7.3	8.1	7.3		
pH	7.9	8.2	8.1	8.2	8.0	8.2	8.3	8.2	8.3	8.1	8.2	8.0		
Cond. (µS/cm)	218	222		224		221		226		227		236		
Initials	JB	SSK		SSK		SSK		SSK		A		r		

Concentration <u>6.3</u>	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.4	8.3	8.4	8.3	8.4	8.3	8.3	7.3	7.9	7.3	8.1	7.4		
pH	8.0	8.2	8.1	8.2	8.0	8.2	8.3	8.1	8.3	8.1	8.1	8.0		
Cond. (µS/cm)	219	221		222		224		226		230		238		
Initials	JB	SSK		SSK		SSK		SSK		r		r		

Concentration <u>25</u>	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.4	8.3	8.4	8.3	8.4	8.3	8.4	7.1	7.6	7.4	8.1	7.5		
pH	7.9	8.2	8.1	8.2	8.0	8.2	8.3	8.1	8.2	8.1	8.1	8.0		
Cond. (µS/cm)	232	233		231		231		235		233		239		
Initials	JB	SSK		SSK		SSK		SSK		r		A		

Concentration <u>100</u>	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	24.0	25.0	24.0	25.0		
DO (mg/L)	8.4	8.3	8.4	8.3	8.4	8.3	8.4	7.1	8.0	7.3	8.1	7.4		
pH	7.8	8.1	7.9	8.2	7.9	8.2	8.1	8.0	8.1	8.1	8.2	8.1		
Cond. (µS/cm)	285	286		287		283		282		288		285		
Initials	JB	SSK		SSK		SSK		SSK		r		r		

Thermometer: 4 DO meter/probe: 1, 1 pH meter/probe: 1, 1 Conductivity meter/probe: 1, 1

	Control	MWIL (SK CONT)	DW-3 (100%)
Hardness*	102	150	400
Alkalinity*	100	164	30

* mg/L as CaCO3

Analysts: JB, AWD, SSK

Reviewed by: SSK

Date reviewed: AUG. 14, 2014

Sample Description: see pg 1

Comments: Broodboard Used: see pg 1

**Chronic Freshwater Toxicity Test
C. dubia Reproduction Data**

Client: WDOE
Sample ID: DW-3
Work Order: WDOE (endo rangefinder) 191282

Start Date & Time: Mar 11/19 @ 1500h
Stop Date & Time: Mar 17, 2019 1500h
Set up by: JB/KL

Days	Concentration: <u>CONTROL</u>											Concentration: <u>SIDE CNTRL 7-(V/V)</u>											Concentration: <u>0.10</u>										
	A	B	C	D	E	F	G	H	I	J	Init	A	B	C	D	E	F	G	H	I	J	Init	A	B	C	D	E	F	G	H	I	J	Init
1	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK
2	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK
3	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK
4	3	4	3	8	4						SSK	2	✓	2	✓	2						SSK	3	6	4	6	4						SSK
5	6	5	6	8	6						A	2	3	3	3	3						A	8	9	8	7	8						A
6	9	9	8	8	9						m	3	2	3	2	2						A	8	7	8	9	8						m
7																																	
8																																	
Total	18	18	18	19	19						JB	7	5	8	5	7						JB	19	22	20	20	20						JB
	17 30											0-SSK 1.6											6.3										
Days	Concentration: <u>0.30</u>											Concentration: <u>0-SSK 1.6</u>											Concentration: <u>6.3</u>										
	A	B	C	D	E	F	G	H	I	J	Init	A	B	C	D	E	F	G	H	I	J	Init	A	B	C	D	E	F	G	H	I	J	Init
1	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK
2	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK
3	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK
4	4	✓	5	4	5						SSK	4	5	3	4	4						SSK	4	4	4	3	5						SSK
5	8	✓	7	8	8						A	7	8	7	9	7						A	7	7	8	6	7						A
6	7	7	8	7	8						m	9	8	9	9	7						m	8	9	9	8	9						m
7																																	
8																																	
Total	19	7	20	19	21						JB	20	21	19	22	18						JB	19	20	21	17	21						JB
Days	Concentration: <u>25</u>											Concentration: <u>100</u>											Concentration:										
	A	B	C	D	E	F	G	H	I	J	Init	A	B	C	D	E	F	G	H	I	J	Init	A	B	C	D	E	F	G	H	I	J	Init
1	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK											
2	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK											
3	✓	✓	✓	✓	✓						SSK	✓	✓	✓	✓	✓						SSK											
4	3	5	4	4	5						SSK	5	4	3	4	6						SSK											
5	7	6	7	8	9						A	9	8	7	8	9						A											
6	8	8	7	9	8						m	9	10	9	9	10						m											
7																																	
8																																	
Total	18	19	18	21	22						JB	23	22	19	21	25						JB											

Notes: X = mortality.

Comments: 1. Total # Young only based on the first 3 Broods. Fourth and subsequent broods not included in total count.

2. Ehippia present in Controls (Y) (N) ?

Reviewed by: [Signature]

Date reviewed: Aug 14, 2019

CETIS Analytical Report

Report Date: 18 Dec-19 13:56 (p 1 of 2)
 Test Code/ID: 191282-RF / 05-3511-2209

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 10-2017-7832	Endpoint: 6d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 18 Dec-19 13:56	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 00-6510-9183	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 11 Mar-19 15:00	Protocol: Washington DOE (2016)	Diluent: 20% Perrier Water
Ending Date: 17 Mar-19 15:00	Species: Ceriodaphnia dubia	Brine:
Test Length: 6d 0h	Taxon: Branchiopoda	Source: In-House Culture Age: <24
Sample ID: 10-2477-5369	Code: DW-3	Project:
Sample Date: 10 Mar-19 Jan 29/19	Material: Water Sample	Source: WDOE
Receipt Date: 11 Mar-19 Jan 30/19	CAS (PC):	Station: wdoe
Sample Age: 39h 4846	Client: WDOE	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	1038395	200	Yes	Two-Point Interpolation

Point Estimates

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
EC5	>5.53	n/a	n/a	<18.08	n/a	n/a
EC10	>5.53	n/a	n/a	<18.08	n/a	n/a
EC15	>5.53	n/a	n/a	<18.08	n/a	n/a
EC20	>5.53	n/a	n/a	<18.08	n/a	n/a
EC25	>5.53	n/a	n/a	<18.08	n/a	n/a
EC40	>5.53	n/a	n/a	<18.08	n/a	n/a
EC50	>5.53	n/a	n/a	<18.08	n/a	n/a

6d Survival Rate Summary

Conc-%	Code	Count	Calculated Variate(A/B)							Isotonic Variate	
			Mean	Min	Max	Std Dev	CV%	%Effect	A/B	Mean	%Effect
0.15	N	5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	5/5	1	0.0%
0.16		5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	5/5	1	0.0%
0.19		5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	5/5	1	0.0%
0.191		5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	5/5	1	0.0%
0.3		5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	5/5	1	0.0%
0.89		5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	5/5	1	0.0%
5.53		5	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	5/5	1	0.0%

6d Survival Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.15	N	1.0000	1.0000	1.0000	1.0000	1.0000
0.16		1.0000	1.0000	1.0000	1.0000	1.0000
0.19		1.0000	1.0000	1.0000	1.0000	1.0000
0.191		1.0000	1.0000	1.0000	1.0000	1.0000
0.3		1.0000	1.0000	1.0000	1.0000	1.0000
0.89		1.0000	1.0000	1.0000	1.0000	1.0000
5.53		1.0000	1.0000	1.0000	1.0000	1.0000

6d Survival Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.15	N	1/1	1/1	1/1	1/1	1/1
0.16		1/1	1/1	1/1	1/1	1/1
0.19		1/1	1/1	1/1	1/1	1/1
0.191		1/1	1/1	1/1	1/1	1/1
0.3		1/1	1/1	1/1	1/1	1/1
0.89		1/1	1/1	1/1	1/1	1/1
5.53		1/1	1/1	1/1	1/1	1/1

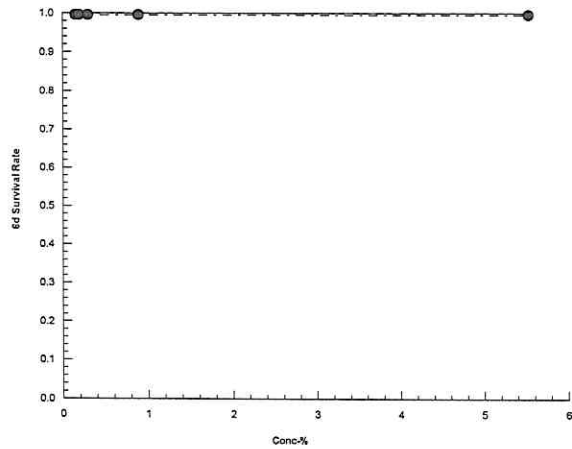
N= lab control

CETIS Analytical Report

Report Date: 18 Dec-19 13:56 (p 2 of 2)
Test Code/ID: 191282-RF / 05-3511-2209

Ceriodaphnia 7-d Survival and Reproduction Test			Nautilus Environmental
Analysis ID: 10-2017-7832	Endpoint: 6d Survival Rate	CETIS Version: CETISv1.9.4	
Analyzed: 18 Dec-19 13:56	Analysis: Linear Interpolation (ICPIN)	Status Level: 1	

Graphics



CETIS Analytical Report

Report Date: 18 Dec-19 13:57 (p 1 of 2)
 Test Code/ID: 191282-RF / 05-3511-2209

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 06-3368-0096	Endpoint: Reproduction	CETIS Version: CETISv1.9.4
Analyzed: 18 Dec-19 13:56	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Batch ID: 00-6510-9183	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 11 Mar-19 15:00	Protocol: Washington DOE (2016)	Diluent: 20% Perrier Water
Ending Date: 17 Mar-19 15:00	Species: Ceriodaphnia dubia	Brine:
Test Length: 6d 0h	Taxon: Branchiopoda	Source: In-House Culture Age: <24
Sample ID: 10-2477-5369	Code: DW-3	Project:
Sample Date: 10 Mar-19 29-Jan-19	Material: Water Sample	Source: WDOE
Receipt Date: 11 Mar-19 30-Jan-19	CAS (PC):	Station: wdoe
Sample Age: 39h 08:46	Client: WDOE	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	1865638	200	Yes	Two-Point Interpolation

Point Estimates

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC5	>5.53	n/a	n/a	<18.08	n/a	n/a
IC10	>5.53	n/a	n/a	<18.08	n/a	n/a
IC15	>5.53	n/a	n/a	<18.08	n/a	n/a
IC20	>5.53	n/a	n/a	<18.08	n/a	n/a
IC25	>5.53	n/a	n/a	<18.08	n/a	n/a
IC40	>5.53	n/a	n/a	<18.08	n/a	n/a
IC50	>5.53	n/a	n/a	<18.08	n/a	n/a

Reproduction Summary

Conc-%	Code	Count	Calculated Variate						Isotonic Variate	
			Mean	Min	Max	Std Dev	CV%	%Effect	Mean	%Effect
0.15	N	5	20.6	19	22	1.342	6.51%	0.0%	20.6	0.0%
0.16		5	17.2	7	21	5.762	33.50%	16.5%	19.43	5.66%
0.19		5	18.2	17	19	0.8367	4.60%	11.65%	19.43	5.66%
0.191		5	20	18	22	1.581	7.91%	2.91%	19.43	5.66%
0.3		5	19.6	17	21	1.673	8.54%	4.85%	19.43	5.66%
0.89		5	19.6	18	22	1.817	9.27%	4.85%	19.43	5.66%
5.53		5	22	19	25	2.236	10.16%	-6.8%	19.43	5.66%

Reproduction Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.15	N	19	22	20	22	20
0.16		19	7	20	19	21
0.19		18	18	17	19	19
0.191		20	21	19	22	18
0.3		19	20	21	17	21
0.89		18	19	18	21	22
5.53		23	22	19	21	25

N = lab control

CETIS Analytical Report

Report Date: 18 Dec-19 13:57 (p 2 of 2)
Test Code/ID: 191282-RF / 05-3511-2209

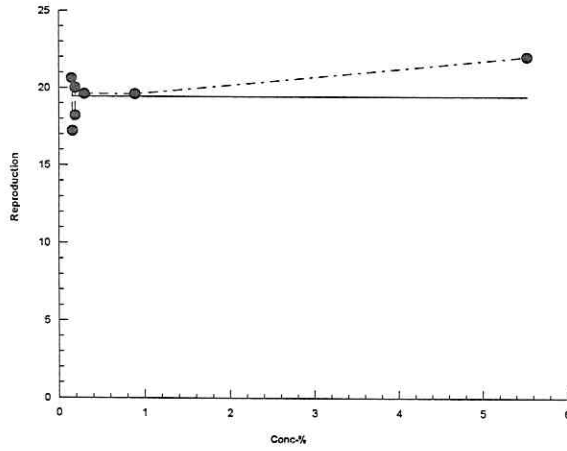
Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 06-3368-0096 Endpoint: Reproduction
Analyzed: 18 Dec-19 13:56 Analysis: Linear Interpolation (ICPIN)

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 18 Dec-19 14:07 (p 1 of 2)
 Test Code/ID: 191282-RF / 05-3511-2209

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 12-2294-4036	Endpoint: 6d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 18 Dec-19 14:07	Analysis: STP 2xK Contingency Tables	Status Level: 1
Batch ID: 00-6510-9183	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 11 Mar-19 15:00	Protocol: Washington DOE (2016)	Diluent: 20% Perrier Water
Ending Date: 17 Mar-19 15:00	Species: Ceriodaphnia dubia	Brine: -
Test Length: 6d 0h	Taxon: Branchiopoda	Source: In-House Culture Age: <24
Sample ID: 10-2477-5369	Code: DW-3	Project: -
Sample Date: 10 Mar-19 29-Jan-19	Material: Water Sample	Source: WDOE
Receipt Date: 11 Mar-19 30-Jan-19	CAS (PC): -	Station: wdoe
Sample Age: 39h 084b	Client: WDOE	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU
Untransformed	C > T	5.53	>5.53	n/a	18.08

Fisher Exact/Bonferroni-Holm Test

Control	vs	Group	Test Stat	P-Type	P-Value	Decision(α:5%)
Negative Control		0.15	1.0000	Exact	1.0000	Non-Significant Effect
		0.16	1.0000	Exact	1.0000	Non-Significant Effect
		0.191	1.0000	Exact	1.0000	Non-Significant Effect
		0.3	1.0000	Exact	1.0000	Non-Significant Effect
		0.89	1.0000	Exact	1.0000	Non-Significant Effect
		5.53	1.0000	Exact	1.0000	Non-Significant Effect

Data Summary

Conc-%	Code	NR	R	NR + R	Prop NR	Prop R	%Effect
0.15		5	0	5	1	0	0.0%
0.16		5	0	5	1	0	0.0%
0.19	N	5	0	5	1	0	0.0%
0.191		5	0	5	1	0	0.0%
0.3		5	0	5	1	0	0.0%
0.89		5	0	5	1	0	0.0%
5.53		5	0	5	1	0	0.0%

6d Survival Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.15		1.0000	1.0000	1.0000	1.0000	1.0000
0.16		1.0000	1.0000	1.0000	1.0000	1.0000
0.19	N	1.0000	1.0000	1.0000	1.0000	1.0000
0.191		1.0000	1.0000	1.0000	1.0000	1.0000
0.3		1.0000	1.0000	1.0000	1.0000	1.0000
0.89		1.0000	1.0000	1.0000	1.0000	1.0000
5.53		1.0000	1.0000	1.0000	1.0000	1.0000

6d Survival Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.15		1/1	1/1	1/1	1/1	1/1
0.16		1/1	1/1	1/1	1/1	1/1
0.19	N	1/1	1/1	1/1	1/1	1/1
0.191		1/1	1/1	1/1	1/1	1/1
0.3		1/1	1/1	1/1	1/1	1/1
0.89		1/1	1/1	1/1	1/1	1/1
5.53		1/1	1/1	1/1	1/1	1/1

N = lab control

CETIS Analytical Report

Report Date: 18 Dec-19 14:07 (p 2 of 2)
Test Code/ID: 191282-RF / 05-3511-2209

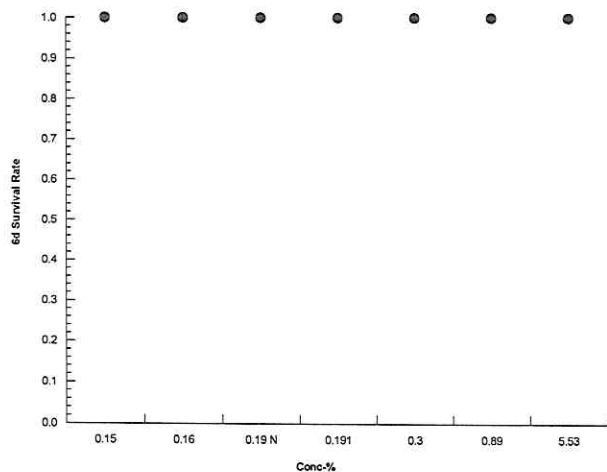
Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 12-2294-4036 Endpoint: 6d Survival Rate
Analyzed: 18 Dec-19 14:07 Analysis: STP 2xK Contingency Tables

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Analytical Report

Report Date: 18 Dec-19 14:07 (p 1 of 2)
 Test Code/ID: 191282-RF / 05-3511-2209

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 01-4092-6723	Endpoint: Reproduction	CETIS Version: CETISv1.9.4
Analyzed: 18 Dec-19 14:07	Analysis: Nonparametric-Control vs Treatments	Status Level: 1
Batch ID: 00-6510-9183	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 11 Mar-19 15:00	Protocol: Washington DOE (2016)	Diluent: 20% Perrier Water
Ending Date: 17 Mar-19 15:00	Species: Ceriodaphnia dubia	Brine:
Test Length: 6d 0h	Taxon: Branchiopoda	Source: In-House Culture Age: <24
Sample ID: 10-2477-5369	Code: DW-3	Project:
Sample Date: 10 Mar-19 29-Jan-19	Material: Water Sample	Source: WDOE
Receipt Date: 11 Mar-19 30-Jan-19	CAS (PC):	Station: wdoe
Sample Age: 39h ~ 0846h	Client: WDOE	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	C > T	5.53	>5.53	n/a	18.08	22.20%

Steel Many-One Rank Sum Test

Control	vs	Conc-%	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		0.15	39	16	2	8	CDF	1.0000	Non-Significant Effect
		0.16	33	16	2	8	CDF	0.9932	Non-Significant Effect
		0.191	36	16	3	8	CDF	0.9994	Non-Significant Effect
		0.3	34.5	16	3	8	CDF	0.9979	Non-Significant Effect
		0.89	33	16	3	8	CDF	0.9932	Non-Significant Effect
		5.53	39	16	2	8	CDF	1.0000	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	73.2	12.2	6	1.732	0.1503	Non-Significant Effect
Error	197.2	7.04286	28			
Total	270.4		34			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	18.92	16.81	0.0043	Unequal Variances
Distribution	Shapiro-Wilk W Normality Test	0.829	0.9146	8.0E-05	Non-Normal Distribution

Reproduction Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0.15		5	20.6	18.93	22.27	20	19	22	0.6	6.51%	0.00%
0.16		5	17.2	10.05	24.35	19	7	21	2.577	33.50%	16.50%
0.19	N	5	18.2	17.16	19.24	18	17	19	0.3742	4.60%	11.65%
0.191		5	20	18.04	21.96	20	18	22	0.7071	7.91%	2.91%
0.3		5	19.6	17.52	21.68	20	17	21	0.7483	8.54%	4.85%
0.89		5	19.6	17.34	21.86	19	18	22	0.8124	9.27%	4.85%
5.53		5	22	19.22	24.78	22	19	25	1	10.16%	-6.80%

Reproduction Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0.15		19	22	20	22	20
0.16		19	7	20	19	21
0.19	N	18	18	17	19	19
0.191		20	21	19	22	18
0.3		19	20	21	17	21
0.89		18	19	18	21	22
5.53		23	22	19	21	25

N= lab control

CETIS Analytical Report

Report Date: 18 Dec-19 14:07 (p 2 of 2)
Test Code/ID: 191282-RF / 05-3511-2209

Ceriodaphnia 7-d Survival and Reproduction Test

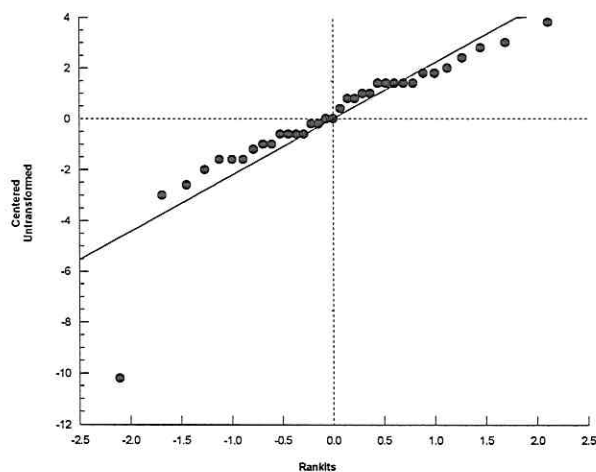
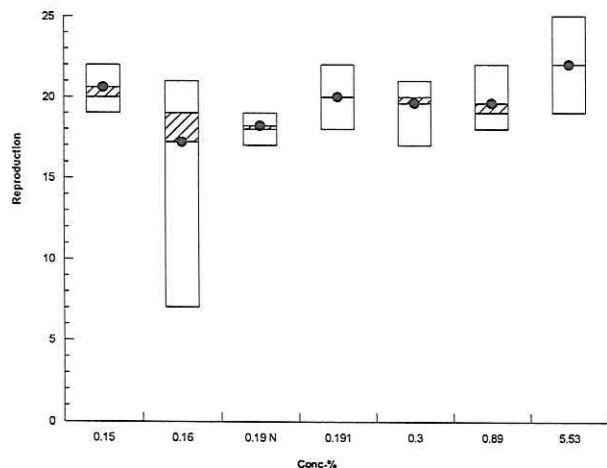
Nautilus Environmental

Analysis ID: 01-4092-6723
Analyzed: 18 Dec-19 14:07

Endpoint: Reproduction
Analysis: Nonparametric-Control vs Treatments

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



W.O.#: 191282

[illegible]

Notes: (i) sample diluted to 100ml w/ DI water

Reviewed by:

Date Reviewed:

Aug 14, 2019

Ceriodaphnia dubia Summary Sheet

Client: WDOE
Work Order No.: 191202

Start Date/Time: May 14/19 @ 1200h
Set up by: KLJSSIC

Sample Information:

Sample ID: MW11, DW-3
Sample Date: Jan 29/19, Mar 19/19
Date Received: Jan 30/19, Mar 20/19
Sample Volume: (2X20L, 5X20L) 3
(3X4L, 5X20L)

Test Validity Criteria:

- 1) Mean survival of first generation controls is $\geq 80\%$
- 2) At least 60% of controls have produced three broods within 8 days
- 3) An average of ≥ 15 live young produced per surviving female in the control solutions during the first three broods.
- 4) Invalid if ephippia observed in any control solution at any time.

WQ Ranges:

T ($^{\circ}\text{C}$) = 25 ± 1 ; DO (mg/L) = 3.3 to 8.4; pH = 6.0 to 8.5

Test Organism Information:

Broodstock No.:
Age of young (Day 0):
Avg No. young in first 3 broods of previous 7 d:
Mortality (%) in previous 7 d:
Individual female # used ≥ 8 young on test day

BB050619A+B & BB050819A KSK
<24-h (within 12-h)~
20
3
10,19,27 (BB050619A+B)
1,2,4,6-9 (BB050819A)

NaCl Reference Toxicant Results:

Reference Toxicant ID: Cd 220
Stock Solution ID: 18Na06 (100g/L NaCl)
Date Initiated: May 01/19

7-d LC50 (95% CL): 2.0 (1.7-2.3) g/L NaCl
7-d IC50 (95% CL): 1.2 (1.1-1.3) g/L NaCl

7-d LC50 Reference Toxicant Mean and Historical Range: 2.0 (1.8-2.2) g/L NaCl CV (%): 5
7-d IC50 Reference Toxicant Mean and Historical Range: 1.5 (1.1-2.3) g/L NaCl CV (%): 20
1-6

Test Results:

	Survival (%)	Reproduction (Mean \pm SD)
Negative Control	100	18.8 \pm 2.4
MW-11 (size control)	100	15.5 \pm 3.7
DW-3	100	17.4 \pm 4.2
		\pm
		\pm
		\pm
		\pm
		\pm
		\pm

Reviewed by: Jen

Date reviewed: July 9/19

Chronic Freshwater Toxicity Test Initial and Final Water Quality Measurements

Client: WDOE
Sample ID: DW-3 (weathered diesel)
Work Order #: 191282

Start Date & Time: May 14/19 @ noon
Stop Date & Time: May 20/19 @ 1400h
CER #: 4
Test Species: Ceriodaphnia dubia

Concentration lab Control	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	25.0	24.0	24.0	24.0	25.0	25.0	25.0	25.0	24.0	25.0	24.0	24.0	24.0	
DO (mg/L)	8.1	7.3	8.0	7.2	8.0	7.2	8.1	7.4	8.2	7.4	8.2	7.5	7.5	
pH	8.3	8.0	8.2	8.0	8.3	8.0	8.3	7.9	8.2	7.9	8.2	7.8	7.8	
Cond. (µS/cm)	216	216		216		216		214		215		223		
Initials	SSK	SSK		SSK		SSK						W		

MW-11

Site Ctrl. Concentration +50 SSK	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	25.0	24.0	24.5	25.0	25.0	25.0	24.0	25.0	24.0	24.0	24.0	
DO (mg/L)	8.2	7.4	8.2	7.2	8.2	7.5	8.2	7.3	8.1	7.1	8.0	7.0	7.0	
pH	7.6	7.8	7.6	7.6	7.7	7.7	7.6	7.7	7.6	7.8	7.5	7.6	7.6	
Cond. (µS/cm)	326	315		326		320		329		328		340		
Initials	SSK	SSK		SSK		SSK						W		

8.2

Concentration 12.5 SSK	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)	24.0	24.0	25.0	24.0	24.5	25.0	25.0	25.0	24.0	25.0	24.0	24.0	24.0	
DO (mg/L)	8.2	7.4	8.2	7.2	8.2	7.5	8.2	7.3	8.1	6.9	8.0	7.0	7.0	
pH	6.7	6.8	6.5	7.0	6.7	7.0	6.6	6.8	6.7	6.9	6.7	7.0	7.0	
Cond. (µS/cm)	1414	1400		1432		1408		1439		1452		1446		
Initials	SSK	SSK		SSK		SSK						W		

8.2 W

Concentration 100 SSK	Days													
	0	1		2		3		4		5		6		7
	init.	old	new	old	new	old	new	old	new	old	new	old	new	final
Temperature (°C)														
DO (mg/L)														
pH														
Cond. (µS/cm)														
Initials														

Thermometer: 4 DO meter/probe: 1 / 1 pH meter/probe: 1 / 1 Conductivity meter/probe: 1 / 1

	Control	100%	sik control	
Hardness*	100	400	180	
Alkalinity*	106	30	164	

* mg/L as CaCO3

Analysts: KL, AND, SSK, JB

Reviewed by: JKW

Date reviewed: July 9/19

Sample Description:

MW-11 clear, yellow, no particulates, no odor
DW-3 clear, yellow, ~~no~~ particulates, no odor

Comments:

Broodboard Used: 00050619AAB (K 10, 19, 27) & 00050819A (H1, 2, 4, 6-9)

**Chronic Freshwater Toxicity Test
C. dubia Reproduction Data**

Client: WDOE
Sample ID: DW-3 (weathered diesel)
Work Order: 191282

(% v/v)

Start Date & Time: May 14/19 @ 1200h
Stop Date & Time: May 20/19 @ 1400h
Set up by: KL/AWD/JB/SSK

Days	Concentration: CONTROL (lab)											Init	Concentration: SITE (H ₂ O) (100) MW-1											Init	Concentration: MW-2 (100) DW-3											Init
	A	B	C	D	E	F	G	H	I	J	A		B	C	D	E	F	G	H	I	J	A	B		C	D	E	F	G	H	I	J				
1	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	SSK	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	SSK	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	SSK			
2	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	SSK	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	SSK	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	SSK			
3	4	3	4	3	3	4	4	4	3	✓	SSK	✓	4	3	2	4	✓	2	3	4	3	SSK	4	3	3	3	3	3	✓	4	3	3	SSK			
4	✓	✓	5	5	✓	6	6	6	6	3	N	3	✓	6	✓	4	✓	5	6	6	2	✓	✓	6	✓	✓	7	6	6	✓	✓	✓	✓			
5	5	5	✓	✓	5	✓	✓	✓	✓	6	2	6	7	✓	8	8	7	7	✓	✓	2	7	5	✓	6	7	7	✓	✓	✓	✓	✓	✓			
6	10	7	12	8	10	11	12	11	9	8	Wk	✓	7	10	✓	9	✓	5	9	8	9	Wk	10	8	5	7	10	9	12	9	8	10	Wk			
7																																				
8																																				
Total	19	15	21	16	18	21	22	21	18	17	JW	9	18	13	16	21	11	14	17	18	18	Wk	21	16	14	16	20	19	25	19	11	13	Wk			

Days	Concentration:											Init	Concentration:											Init	Concentration:											Init
	A	B	C	D	E	F	G	H	I	J	A		B	C	D	E	F	G	H	I	J	A	B		C	D	E	F	G	H	I	J				
1																																				
2																																				
3																																				
4																																				
5																																				
6																																				
7																																				
8																																				
Total																																				

Days	Concentration:											Init	Concentration:											Init	Concentration:											Init
	A	B	C	D	E	F	G	H	I	J	A		B	C	D	E	F	G	H	I	J	A	B		C	D	E	F	G	H	I	J				
1																																				
2																																				
3																																				
4																																				
5																																				
6																																				
7																																				
8																																				
Total																																				

Notes: X = mortality.

Comments: 1. Total # Young only based on the first 3 Broods. Fourth and subsequent broods not included in total count.

2. Ehippia present in Controls (Y) (N) ?

Reviewed by: JG

Date reviewed: July 9/19

CETIS Analytical Report

Report Date: 26 Jun-19 14:00 (p 1 of 1)
Test Code/ID: 191282a / 10-2817-1520

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 07-8517-0948	Endpoint: 6d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 26 Jun-19 14:00	Analysis: Single 2x2 Contingency Table	Status Level: 1
Batch ID: 06-1681-4698	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 14 May-19 12:00	Protocol: EPA/821/R-02-013 (2002)	Diluent:
Ending Date: 20 May-19 14:00	Species: Ceriodaphnia dubia	Brine:
Test Length: 6d 2h	Taxon: Branchiopoda	Source: In-House Culture Age: <24

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	105d 12h	WA State Dept of Ecolog	
DW-3	16-5145-0355	29 Jan-19	30 Jan-19 08:46	105d 12h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
MW-11	Diesel	WDOE	MW-11	
DW-3	Diesel	WDOE	DW-3	

Fisher Exact Test

Sample I	vs	Sample II	Test Stat	P-Type	P-Value	Decision(α:5%)
Site Control		DW-3	1.0000	Exact	1.0000	Non-Significant Effect

Data Summary

Conc-%	Code	NR	R	NR + R	Prop NR	Prop R	%Effect
MW-11	XC	10	0	10	1	0	0.0%
DW-3		10	0	10	1	0	0.0%

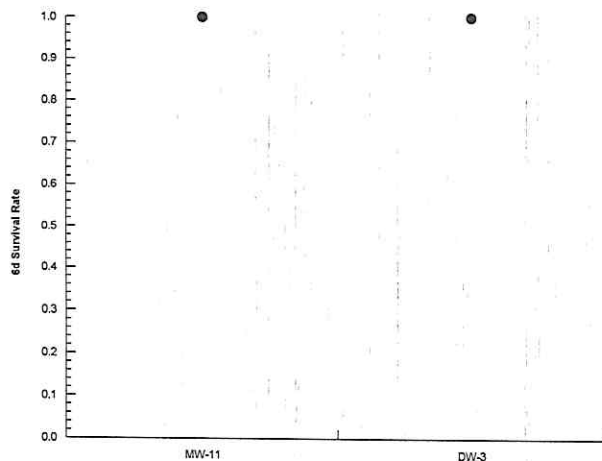
6d Survival Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
MW-11	XC	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
DW-3		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000

6d Survival Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
MW-11	XC	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
DW-3		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1

Graphics



CETIS Analytical Report

Report Date: 26 Jun-19 14:05 (p 1 of 2)
Test Code/ID: 191282a / 10-2817-1520

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 10-2638-7894	Endpoint: Reproduction	CETIS Version: CETISv1.9.4
Analyzed: 26 Jun-19 14:05	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 06-1681-4698	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 14 May-19 12:00	Protocol: EPA/821/R-02-013 (2002)	Diluent:
Ending Date: 20 May-19 14:00	Species: Ceriodaphnia dubia	Brine:
Test Length: 6d 2h	Taxon: Branchiopoda	Source: In-House Culture Age: <24

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	105d 12h	WA State Dept of Ecolog	
DW-3	16-5145-0355	29 Jan-19	30 Jan-19 08:46	105d 12h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
MW-11	Diesel	WDOE	MW-11	
DW-3	Diesel	WDOE	DW-3	

Data Transform	Alt Hyp	Comparison Result	PMSD
Untransformed	C > T	DW-3 passed reproduction	19.76%

Equal Variance t Two-Sample Test

Sample I	vs	Sample II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Site Control		DW-3	-1.075	1.734	3.064	18	CDF	0.8518	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	18.05	18.05	1	1.157	0.2964	Non-Significant Effect
Error	280.9	15.6056	18			
Total	298.95		19			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	1.293	6.541	0.7080	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.964	0.866	0.6270	Normal Distribution

Reproduction Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
MW-11	XC	10	15.5	12.86	18.14	16.5	9	21	1.167	23.80%	0.00%
DW-3		10	17.4	14.4	20.4	17.5	11	25	1.327	24.11%	-12.26%

Reproduction Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
MW-11	XC	9	18	13	16	21	11	14	17	18	18
DW-3		21	16	14	16	20	19	25	19	11	13

CETIS Analytical Report

Report Date: 26 Jun-19 14:05 (p 2 of 2)
Test Code/ID: 191282a / 10-2817-1520

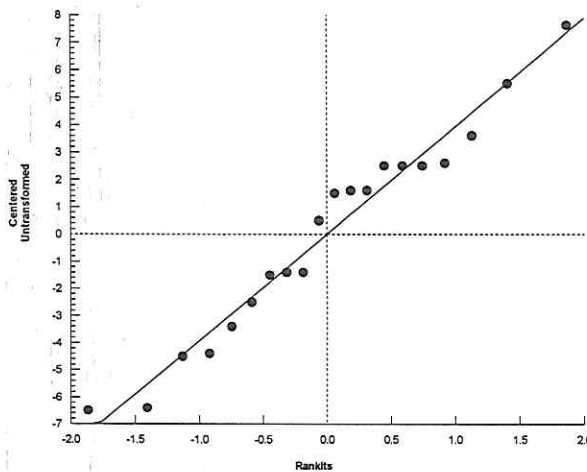
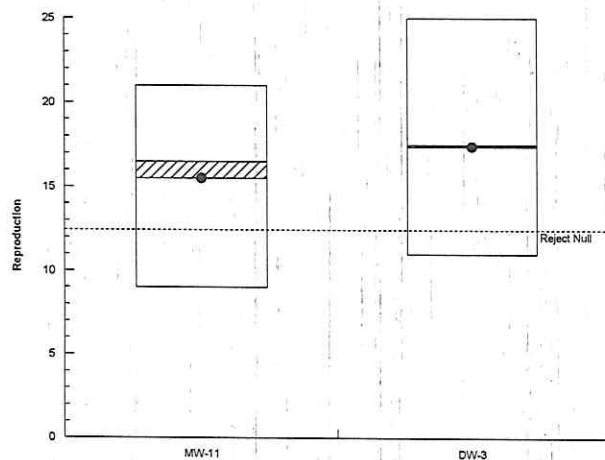
Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 10-2638-7894
Analyzed: 26 Jun-19 14:05
Endpoint: Reproduction
Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



CETIS Summary Report

Report Date: 26 Jun-19 14:01 (p 1 of 1)
Test Code/ID: 191282a / 10-2817-1520

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Batch ID: 06-1681-4698 Test Type: Reproduction-Survival (7d) Analyst: Kania Lywe
Start Date: 14 May-19 12:00 Protocol: EPA/821/R-02-013 (2002) Diluent:
Ending Date: 20 May-19 14:00 Species: Ceriodaphnia dubia Brine:
Test Length: 6d 2h Taxon: Branchiopoda Source: In-House Culture Age: <24

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
Lab Control	13-4083-0602	14 May-19	14 May-19	12h	WA State Dept of Ecolog	
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	105d 12h		
DW-3	16-5145-0355	29 Jan-19	30 Jan-19 08:46	105d 12h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
Lab Control	Water Sample	WDOE	Lab Control	
MW-11	Diesel	WDOE	MW-11	
DW-3	Diesel	WDOE	DW-3	

Single Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
07-8517-0948	6d Survival Rate	Fisher Exact Test	1.0000	Lab Control passed 6d survival rate	1
07-8517-0948	6d Survival Rate	Fisher Exact Test	1.0000	MW-11 passed 6d survival rate	1
07-8517-0948	6d Survival Rate	Fisher Exact Test	1.0000	DW-3 passed 6d survival rate	1
15-6851-5108	Reproduction	Equal Variance t Two-Sample Test	0.1858	Lab Control passed reproduction	1
15-6851-5108	Reproduction	Equal Variance t Two-Sample Test	0.1858	MW-11 passed reproduction	1
15-6851-5108	Reproduction	Equal Variance t Two-Sample Test	0.1858	DW-3 passed reproduction	1

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits		Overlap	Decision
15-6851-5108	Reproduction	Control Resp	18.8	Lower 15	Upper >>	Yes	Passes Criteria

6d Survival Rate Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
Lab Control	L	10	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%
MW-11	XC	10	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%
DW-3		10	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%

Reproduction Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
Lab Control	L	10	18.8	17.09	20.51	15	22	0.7572	2.394	12.74%	0.00%
MW-11	XC	10	15.5	12.86	18.14	9	21	1.167	3.689	23.80%	17.55%
DW-3		10	17.4	14.4	20.4	11	25	1.327	4.195	24.11%	7.45%

6d Survival Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
Lab Control	L	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
MW-11	XC	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
DW-3		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000

Reproduction Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
Lab Control	L	19	15	21	16	18	21	22	21	18	17
MW-11	XC	9	18	13	16	21	11	14	17	18	18
DW-3		21	16	14	16	20	19	25	19	11	13

6d Survival Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
Lab Control	L	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
MW-11	XC	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
DW-3		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1

CETIS Analytical Report

Report Date: 26 Jun-19 14:01 (p 1 of 1)
Test Code/ID: 191282a / 10-2817-1520

Ceriodaphnia 7-d Survival and Reproduction Test Nautilus Environmental

Analysis ID: 20-2246-1753	Endpoint: 6d Survival Rate	CETIS Version: CETISv1.9.4
Analyzed: 26 Jun-19 14:01	Analysis: Single 2x2 Contingency Table	Status Level: 1
Batch ID: 06-1681-4698	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 14 May-19 12:00	Protocol: EPA/821/R-02-013 (2002)	Diluent:
Ending Date: 20 May-19 14:00	Species: Ceriodaphnia dubia	Brine:
Test Length: 6d 2h	Taxon: Branchiopoda	Source: In-House Culture Age: <24

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
Lab Control	13-4083-0602	14 May-19	14 May-19	12h	WA State Dept of Ecolog	
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	105d 12h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
Lab Control	Water Sample	WDOE	Lab Control	
MW-11	Diesel	WDOE	MW-11	

Fisher Exact Test

Sample I	vs	Sample II	Test Stat	P-Type	P-Value	Decision(α:5%)
Lab Water		Site Control	1.0000	Exact	1.0000	Non-Significant Effect

Data Summary

Conc-%	Code	NR	R	NR + R	Prop NR	Prop R	%Effect
Lab Control	L	10	0	10	1	0	0.0%
MW-11	XC	10	0	10	1	0	0.0%

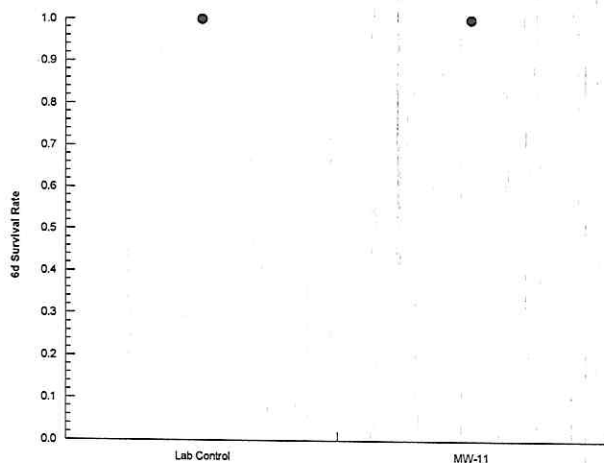
6d Survival Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
Lab Control	L	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
MW-11	XC	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000

6d Survival Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
Lab Control	L	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
MW-11	XC	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1

Graphics



CETIS Analytical Report

Report Date: 26 Jun-19 14:01 (p 1 of 2)
Test Code/ID: 191282a / 10-2817-1520

Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 10-1694-2397	Endpoint: Reproduction	CETIS Version: CETISv1.9.4
Analyzed: 26 Jun-19 14:01	Analysis: Parametric-Two Sample	Status Level: 1
Batch ID: 06-1681-4698	Test Type: Reproduction-Survival (7d)	Analyst: Kania Lywe
Start Date: 14 May-19 12:00	Protocol: EPA/821/R-02-013 (2002)	Diluent:
Ending Date: 20 May-19 14:00	Species: Ceriodaphnia dubia	Brine:
Test Length: 6d 2h	Taxon: Branchiopoda	Source: In-House Culture Age: <24

Sample Code	Sample ID	Sample Date	Receipt Date	Sample Age	Client Name	Project
Lab Control	13-4083-0602	14 May-19	14 May-19	12h	WA State Dept of Ecolog	
MW-11	19-3462-2452	29 Jan-19	30 Jan-19 08:46	105d 12h		

Sample Code	Material Type	Sample Source	Station Location	Lat/Long
Lab Control	Water Sample	WDOE	Lab Control	
MW-11	Diesel	WDOE	MW-11	

Data Transform	Alt Hyp	Comparison Result	PMSD
Untransformed	C > T	MW-11 failed reproduction	12.83%

Equal Variance t Two-Sample Test

Sample I	vs	Sample II	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)
Lab Water		Site Control*	2.373	1.734	2.412	18	CDF	0.0145	Significant Effect

Test Acceptability Criteria

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	18.8	15	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	54.45	54.45	1	5.63	0.0290	Significant Effect
Error	174.1	9.67222	18			
Total	228.55		19			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Variance Ratio F Test	2.374	6.541	0.2138	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9619	0.866	0.5825	Normal Distribution

Reproduction Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
Lab Control	L	10	18.8	17.09	20.51	18.5	15	22	0.7572	12.74%	0.00%
MW-11	XC	10	15.5	12.86	18.14	16.5	9	21	1.167	23.80%	17.55%

Reproduction Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
Lab Control	L	19	15	21	16	18	21	22	21	18	17
MW-11	XC	9	18	13	16	21	11	14	17	18	18

CETIS Analytical Report

Report Date: 26 Jun-19 14:01 (p 2 of 2)
Test Code/ID: 191282a / 10-2817-1520

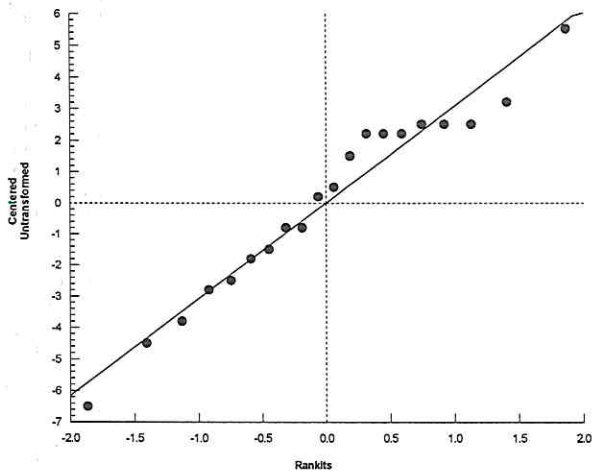
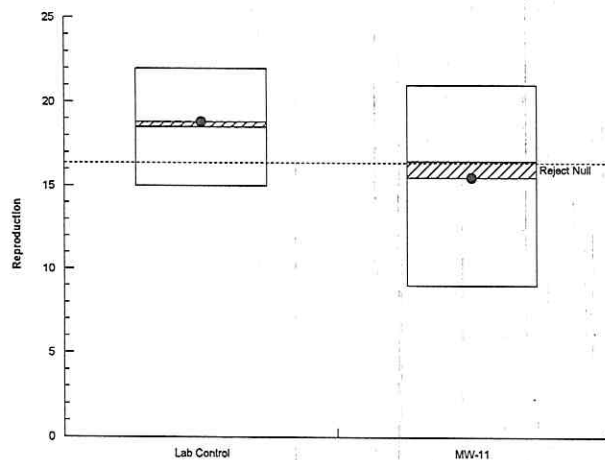
Ceriodaphnia 7-d Survival and Reproduction Test

Nautilus Environmental

Analysis ID: 10-1694-2397 Endpoint: Reproduction
Analyzed: 26 Jun-19 14:01 Analysis: Parametric-Two Sample

CETIS Version: CETISv1.9.4
Status Level: 1

Graphics



W.O.#: 191282

Hardness and Alkalinity Datasheet

[illegible]

Notes: ① sample diluted to 100ml w/D.I water.

Reviewed by:

Jon

Date Reviewed:

July 9/10

APPENDIX E – Analytical Chemistry

DEPARTMENT OF ECOLOGY
Manchester Environmental Laboratory
7411 Beach Drive East • Port Orchard, Washington 98366-8204

Case Narrative

February 14, 2019

To: Hobbs, William

Project: NWTPH-Dx Tox

Work Order: 1902025

Subject: Semivolatile Petroleum Products

From: Karin Bailey

LB

Sample Receipt

Enclosed are the TPHD results for the samples received by MEL on February 7, 2019. All samples were received in acceptable condition unless noted in Analyst Comments. All samples were prepared and analyzed within holding times unless noted in Analyst Comments.

Analytical Methods

These samples were prepared, analyzed, and verified by MEL according to the submitted chain-of-custody and MEL's procedures. A Sample Correlation Table with batch summary is located in Appendix A. The samples were:

- extracted following a modification of method SW3535A.
- analyzed following a modification of method NWTPH-DX.

Analyst Comments

TPHD by GCFID. The samples were received at the lab out of temperature. As a result, the sample final concentrations are qualified as estimates.

Sample Qualification

The samples were qualified according to MEL's procedures. The table in Appendix B summarizes the manual qualifiers added by MEL. All results reported below the method reporting limit (RL) were automatically qualified as estimates, but not included in Appendix B.

The qualifiers are defined in Appendix C.

Sample Verification

All analyses met QC acceptance criteria except as noted in Appendix D. All analytes met linearity requirements unless noted in Appendix E.

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-stk-100-0-1

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: 1902025-01
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	6.29	J	0.15	
NULL	Residual Range Organics	4.75	J	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.272	0.220	124	50-150

Authorized by: LB

Release Date: 2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-stk-100-0-2

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: 1902025-02
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	5.67	J	0.15	
NULL	Residual Range Organics	4.29	J	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.260	0.220	118	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-stk-100-0-3

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 995 mL
Final Vol: 3 mL

Lab ID #: 1902025-03
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	5.90	J	0.15	
NULL	Residual Range Organics	4.50	J	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.241	0.221	109	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-stk-0.5-0-1

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 985 mL
Final Vol: 3 mL

Lab ID #: 1902025-04
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	2.77	J	0.15	
NULL	Residual Range Organics	2.05	J	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.253	0.223	113	50-150

Authorized by: LR

Release Date: 2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-stk-0.5-0-2

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 995 mL
Final Vol: 3 mL

Lab ID #: 1902025-05
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	2.38	J	0.15	
NULL	Residual Range Organics	1.71	J	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.223	0.221	101	50-150

Authorized by: LB

Release Date: 2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-stk-0.5-0-3

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 995 mL
Final Vol: 3 mL

Lab ID #: 1902025-06
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	2.95	J	0.15	
NULL	Residual Range Organics	2.13	J	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.258	0.221	117	50-150

Authorized by:

Release Date:

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-control-0-1

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: 1902025-07
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/14/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.30	J	0.15	
NULL	Residual Range Organics	0.38	UJ	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.227	0.220	103	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-control-0-2

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 995 mL
Final Vol: 3 mL

Lab ID #: 1902025-08
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/14/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.29	J	0.15	
NULL	Residual Range Organics	0.38	UJ	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.241	0.221	109	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-control-0-3

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 990 mL
Final Vol: 3 mL

Lab ID #: 1902025-09
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/14/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.27	J	0.15	
NULL	Residual Range Organics	0.38	UJ	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.214	0.222	96	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-Ppro-BLNK-0-1

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 995 mL
Final Vol: 3 mL

Lab ID #: 1902025-10
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/14/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.15	UJ	0.15	
NULL	Residual Range Organics	0.38	UJ	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.242	0.221	109	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-Cdub-BLNK-0-1

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 990 mL
Final Vol: 3 mL

Lab ID #: 1902025-11
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/14/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.15	UJ	0.15	
NULL	Residual Range Organics	0.38	UJ	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.228	0.222	103	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: mix-MARINE-BLNK-0-1

Work Order: 1902025
Project Officer: Hobbs, William
Initial Vol: 1010 mL
Final Vol: 3 mL

Lab ID #: 1902025-12
Collected: 2/5/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/14/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.17	J	0.15	
NULL	Residual Range Organics	0.37	UJ	0.37	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.238	0.218	109	50-150

Authorized by: LB

Release Date: 2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : Method Blank

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19B033-BLK1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19B033-BLK1

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.15	UJ	0.15	
NULL	Residual Range Organics	0.38	U	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.167	0.220	76	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : LCS

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19B033-BS1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19B033-BS1

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: %

Analyte	Result	Spike Level	RL	%Rec	%Rec Limits
Diesel Range Organics	2.31	3.00	0.15	77	70-130

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.233	0.220	106	50-150

Authorized by: _____

Release Date: _____

2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : LCS Dup

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19B033-BSD1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19B033-BSD1

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: %

Analyte	Sample Result	Spike Level	%Rec	RPD	%Rec Limits	RPD Limit
Diesel Range Organics	1.03	3.00	34	77	70-130	40

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.123	0.220	56	50-150

Authorized by: *LS*

Release Date: 2/14/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : Duplicate

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 995 mL
Final Vol: 3 mL

Lab ID #: B19B033-DUP1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19B033-DUP1
Source Lab ID #: 1902025-01

Batch ID: B19B033
Prepared: 2/8/2019
Analyzed: 2/13/2019
Matrix: Water
Units: mg/L

Analyte	Sample Result	Sample Qual	Source Result	RPD	RPD Limit
Diesel Range Organics	5.67		6.29	10	40
Residual Range Organics	3.94		4.75	19	40

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.251	0.221	113	50-150

Authorized by: _____

Release Date: _____

2/14/19

Appendix A

Sample Correlation Table

Batch ID: B19B033		Prep Method: SW3535A	
Prepared: 2/8/2019		Analysis Method: NWTPH-DX	
<u>Field ID</u>		<u>MEL ID</u>	
mix-stk-100-0-1		1902025-01	
mix-stk-100-0-2		1902025-02	
mix-stk-100-0-3		1902025-03	
mix-stk-0.5-0-1		1902025-04	
mix-stk-0.5-0-2		1902025-05	
mix-stk-0.5-0-3		1902025-06	
mix-control-0-1		1902025-07	
mix-control-0-2		1902025-08	
mix-control-0-3		1902025-09	
mix-Ppro-BLNK-0-1		1902025-10	
mix-Cdub-BLNK-0-1		1902025-11	
mix-MARINE-BLNK-0-1		1902025-12	
Blank		B19B033-BLK1	
LCS		B19B033-BS1	
LCS Dup		B19B033-BSD1	
Duplicate (mix-stk-100-0-1)		B19B033-DUP1	

Appendix B

Manual Qualification Table

WO: 1902025

Analysis: TPHD

Reported result is estimated; sample was not properly preserved.

Diesel Range Organics J: 1902025-01, 1902025-02, 1902025-03, 1902025-04, 1902025-05, 1902025-06, 1902025-07, 1902025-08, 1902025-09, 1902025-12,

Residual Range Organics J: 1902025-01, 1902025-02, 1902025-03, 1902025-04, 1902025-05, 1902025-06,

Analyte was not detected at or above the estimated MRL; LCS recovery exceeded QC limits.

Diesel Range Organics UJ: B19B033-BLK1,

Analyte was not detected at or above the estimated MRL; sample was not properly preserved

Diesel Range Organics UJ: 1902025-10, 1902025-11,

Residual Range Organics UJ: 1902025-07, 1902025-08, 1902025-09, 1902025-10, 1902025-11, 1902025-12,

Appendix C

Data Qualifier Definitions

Code	Definition
E	Reported result is an estimate because it exceeds the calibration range.
J	The analyte was positively identified; the associated numerical value is the approximate concentration of the analyte in the sample.
N	The analysis indicates the present of an analyte for which there is presumptive evidence to make a “tentative identification”.
NJ	The analysis indicates the presence of an analyte that has been “tentatively identified” and the associated numerical value represents its approximate concentration.
NAF	Not analyzed for.
NC	Not calculated.
REJ	The sample results are rejected due to serious deficiencies in the ability to analyze the sample and meet quality control criteria. The presence or absence of the analyte cannot be verified.
U	The analyte was not detected at or above the reported sample quantitation limit.
UJ	The analyte was not detected at or above the reported sample quantitation limit. However, the reported quantitation limit is approximate and may or may not represent the actual limit of quantitation necessary to accurately measure the analyte in the sample.
bold	The analyte was present in the sample. (Visual aid to locate detected compounds on the analytical report.)

Appendix D

QC Exceptions Report

Lab ID	Analyte	Exception
B19B033-BSD1	Diesel Range Organics	Exceeds lower control limit
B19B033-BSD1	Diesel Range Organics	Exceeds RPD control limit

Appendix E
Initial Calibration Exceptions Report

Calibration ID: NA

Analysis: TPHD

Lab Number

Analyte

QC Exception

No initial calibration exceptions noted.

DEPARTMENT OF ECOLOGY
Manchester Environmental Laboratory
7411 Beach Drive East • Port Orchard, Washington 98366-8204

Case Narrative

April 8, 2019

To: Hobbs, William

Project: NWTPH-Dx Tox

Work Order: 1903027

Subject: Semivolatile Petroleum Products

From: Karin Bailey

LB

Sample Receipt

Enclosed are the TPHD results for the samples received by MEL on March 20, 2019. All samples were received in acceptable condition unless noted in Analyst Comments. All samples were prepared and analyzed within holding times unless noted in Analyst Comments.

Analytical Methods

These samples were prepared, analyzed, and verified by MEL according to the submitted chain-of-custody and MEL's procedures. A Sample Correlation Table with batch summary is located in Appendix A. The samples were:

- extracted following a modification of method SW3535A.
- analyzed following a modification of method NWTPH-DX.

Analyst Comments

TPHD by GCFID. The samples were received at the laboratory out of temperature. As a result, the sample final concentrations are qualified as estimates. The MRL was raised due to background in samples 1903027-02, -03, -06, -07, -10, -11, and -12.

Sample Qualification

The samples were qualified according to MEL's procedures. The table in Appendix B

summarizes the manual qualifiers added by MEL. All results reported below the method reporting limit (RL) were automatically qualified as estimates, but not included in Appendix B. The qualifiers are defined in Appendix C.

Sample Verification

All analyses met QC acceptance criteria except as noted in Appendix D. All analytes met linearity requirements unless noted in Appendix E.

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-100-72-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 940 mL
Final Vol: 3 mL

Lab ID #: 1903027-01
Collected: 3/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	5.48	J	0.16	
NULL	Residual Range Organics	5.71	J	0.40	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.190	0.234	81	50-150

Authorized by:

Release Date:

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-6.3-72-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 920 mL
Final Vol: 3 mL

Lab ID #: 1903027-02
Collected: 3/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.28	UJ	0.28	
NULL	Residual Range Organics	0.41	UJ	0.41	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.245	0.239	102	50-150

Authorized by:

Release Date:

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-6.3-72-2

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 890 mL
Final Vol: 3 mL

Lab ID #: 1903027-03
Collected: 3/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.27	UJ	0.27	
NULL	Residual Range Organics	0.42	UJ	0.42	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.240	0.247	97	50-150

Authorized by:

Release Date: 4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-0.39-72-1

Work Order: 1903027
Project Officer: . Hobbs, William
Initial Vol: 880 mL
Final Vol: 3 mL

Lab ID #: 1903027-04
Collected: 3/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.17	UJ	0.17	
NULL	Residual Range Organics	0.43	UJ	0.43	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.258	0.250	103	50-150

Authorized by: _____

Release Date: _____

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-100-96-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 870 mL
Final Vol: 3 mL

Lab ID #: 1903027-05
Collected: 3/15/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	3.75	J	0.17	
NULL	Residual Range Organics	3.71	J	0.43	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.216	0.253	85	50-150

Authorized by: _____

Release Date: _____

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-6.3-96-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 820 mL
Final Vol: 3 mL

Lab ID #: 1903027-06
Collected: 3/15/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.25	UJ	0.25	
NULL	Residual Range Organics	0.46	UJ	0.46	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.279	0.268	104	50-150

Authorized by:

Release Date: 4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-6.3-96-2

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 870 mL
Final Vol: 3 mL

Lab ID #: 1903027-07
Collected: 3/15/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.24	UJ	0.24	
NULL	Residual Range Organics	0.43	UJ	0.43	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.252	0.253	100	50-150

Authorized by: _____

Release Date: _____

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-0.39-96-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 890 mL
Final Vol: 3 mL

Lab ID #: 1903027-08
Collected: 3/15/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.17	UJ	0.17	
NULL	Residual Range Organics	0.42	UJ	0.42	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.243	0.247	98	50-150

Authorized by:

Release Date: 4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-100-120-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 940 mL
Final Vol: 3 mL

Lab ID #: 1903027-09
Collected: 3/16/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	5.24	J	0.16	
NULL	Residual Range Organics	5.38	J	0.40	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.210	0.234	90	50-150

Authorized by:

Release Date: 4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-6.3-120-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 940 mL
Final Vol: 3 mL

Lab ID #: 1903027-10
Collected: 3/16/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.30	UJ	0.30	
NULL	Residual Range Organics	0.40	UJ	0.40	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.239	0.234	102	50-150

Authorized by: _____

Release Date: _____

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-0.39-120-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 890 mL
Final Vol: 3 mL

Lab ID #: 1903027-11
Collected: 3/16/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.17	UJ	0.17	
NULL	Residual Range Organics	0.42	UJ	0.42	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.234	0.247	95	50-150

Authorized by: _____

LB

Release Date: _____

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-0-144-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 890 mL
Final Vol: 3 mL

Lab ID #: 1903027-12
Collected: 3/17/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.23	UJ	0.23	
NULL	Residual Range Organics	0.42	UJ	0.42	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.241	0.247	97	50-150

Authorized by: KB

Release Date: 4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for -
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: RF-Cdub-LabCont-144-1

Work Order: 1903027
Project Officer: Hobbs, William
Initial Vol: 940 mL
Final Vol: 3 mL

Lab ID #: 1903027-13
Collected: 3/17/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.16	UJ	0.16	
NULL	Residual Range Organics	0.40	UJ	0.40	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.235	0.234	101	50-150

Authorized by:

Release Date:

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : Method Blank

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19C132-BLK1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19C132-BLK1

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.15	U	0.15	
NULL	Residual Range Organics	0.38	U	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.205	0.220	93	50-150

Authorized by: _____

LB

Release Date: _____

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : LCS

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19C132-BS1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19C132-BS1

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: %

Analyte	Result	Spike Level	RL	%Rec	%Rec Limits
Diesel Range Organics	2.46	3.00	0.15	82	70-130

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.214	0.220	97	50-150

Authorized by:

Release Date:

4/8/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : LCS Dup

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19C132-BSD1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19C132-BSD1

Batch ID: B19C132
Prepared: 3/21/2019
Analyzed: 4/5/2019
Matrix: Water
Units: %

Analyte	Sample Result	Spike Level	%Rec	RPD	%Rec Limits	RPD Limit
Diesel Range Organics	2.46	3.00	82	0.2	70-130	40

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.216	0.220	.98	50-150

Authorized by: LB

Release Date: 4/8/19

Appendix A Sample Correlation Table

Batch ID: B19C132

Prep Method: SW3535A

Prepared: 3/21/2019

Analysis Method: NWTPH-DX

<u>Field ID</u>	<u>MEL ID</u>
RF-Cdub-100-72-1	1903027-01
RF-Cdub-6.3-72-1	1903027-02
RF-Cdub-6.3-72-2	1903027-03
RF-Cdub-0.39-72-1	1903027-04
RF-Cdub-100-96-1	1903027-05
RF-Cdub-6.3-96-1	1903027-06
RF-Cdub-6.3-96-2	1903027-07
RF-Cdub-0.39-96-1	1903027-08
RF-Cdub-100-120-1	1903027-09
RF-Cdub-6.3-120-1	1903027-10
RF-Cdub-0.39-120-1	1903027-11
RF-Cdub-0-144-1	1903027-12
RF-Cdub-LabCont-144-1	1903027-13
Blank	B19C132-BLK1
LCS	B19C132-BS1
LCS Dup	B19C132-BSD1

Appendix B

Manual Qualification Table

WO: 1903027

Analysis: TPHD

Reported result is estimated; sample was not properly preserved.

Diesel Range Organics J: 1903027-01, 1903027-05, 1903027-09,

Residual Range Organics J: 1903027-01, 1903027-05, 1903027-09,

Analyte was not detected at or above the estimated MRL; sample was not properly preserved

Diesel Range Organics UJ: 1903027-02, 1903027-03, 1903027-04, 1903027-06, 1903027-07, 1903027-08,
1903027-10, 1903027-11, 1903027-12, 1903027-13,

Residual Range Organics UJ: 1903027-02, 1903027-03, 1903027-04, 1903027-06, 1903027-07, 1903027-08,
1903027-10, 1903027-11, 1903027-12, 1903027-13,

Appendix C

Data Qualifier Definitions

Code	Definition
E	Reported result is an estimate because it exceeds the calibration range.
J	The analyte was positively identified; the associated numerical value is the approximate concentration of the analyte in the sample.
N	The analysis indicates the present of an analyte for which there is presumptive evidence to make a “tentative identification”.
NJ	The analysis indicates the presence of an analyte that has been “tentatively identified” and the associated numerical value represents its approximate concentration.
NAF	Not analyzed for.
NC	Not calculated.
REJ	The sample results are rejected due to serious deficiencies in the ability to analyze the sample and meet quality control criteria. The presence or absence of the analyte cannot be verified.
U	The analyte was not detected at or above the reported sample quantitation limit.
UJ	The analyte was not detected at or above the reported sample quantitation limit. However, the reported quantitation limit is approximate and may or may not represent the actual limit of quantitation necessary to accurately measure the analyte in the sample.
bold	The analyte was present in the sample. (Visual aid to locate detected compounds on the analytical report.)

Appendix D

QC Exceptions Report

Lab ID	Analyte	Exception
No QC exceptions reported.		

Appendix E
Initial Calibration Exceptions Report

Calibration ID: B9D0401

Analysis: TPHD

LabNumber Analyte

QC Exception

No ICAL exceptions.

DEPARTMENT OF ECOLOGY
Manchester Environmental Laboratory
7411 Beach Drive East • Port Orchard, Washington 98366-8204

Case Narrative

June 17, 2019

To: Hobbs, William

Project: NWTPH-Dx Tox

Work Order: 1905056

Subject: Semivolatile Petroleum Products

From: Karin Bailey

LB

Sample Receipt

Enclosed are the TPHD results for the samples received by MEL on May 29, 2019. All samples were received in acceptable condition unless noted in Analyst Comments. All samples were prepared and analyzed within holding times unless noted in Analyst Comments.

Analytical Methods

These samples were prepared, analyzed, and verified by MEL according to the submitted chain-of-custody and MEL's procedures. A Sample Correlation Table with batch summary is located in Appendix A. The samples were:

- extracted following a modification of method SW3535A.
- analyzed following a modification of method NWTPH-DX.

Analyst Comments

TPHD by GCFID. Sample 1905056-02 arrived at the laboratory over temperature. Sample 1905056-09 arrived at the laboratory unpreserved and past the preparation holding time. Samples 1905056-01, -10, -11, -12, -13, and B19F001-DUP1 arrived at the laboratory past the preparation holding time.

Sample 1905056-06 was accidentally concentrated to a low volume of approximately 0.25mL during the final volume step of the extraction process. Enough methylene chloride was added to adjust the final volume to the standard 3.0mL and the extract was analyzed. The surrogate

recovery was within QC limits indicating that the extract was not significantly affected by the low concentration volume.

Sample Qualification

The samples were qualified according to MEL's procedures. The table in Appendix B summarizes the manual qualifiers added by MEL. All results reported below the method reporting limit (RL) were automatically qualified as estimates, but not included in Appendix B. The qualifiers are defined in Appendix C.

Sample Verification

All analyses met QC acceptance criteria except as noted in Appendix D. All analytes met linearity requirements unless noted in Appendix E.

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-24-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 920 mL
Final Vol: 3 mL

Lab ID #: 1905056-01
Collected: 5/15/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	4.02	J	0.16	
NULL	Residual Range Organics	4.48	J	0.41	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.221	0.217	102	50-150

Authorized by: _____

LB

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-48-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 970 mL
Final Vol: 3 mL

Lab ID #: 1905056-02
Collected: 5/16/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	4.13	J	0.15	
NULL	Residual Range Organics	4.60	J	0.39	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.213	0.206	103	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-48-2

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 900 mL
Final Vol: 3 mL

Lab ID #: 1905056-03
Collected: 5/16/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	4.24		0.17	
NULL	Residual Range Organics	4.56		0.42	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.235	0.222	106	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-72-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 820 mL
Final Vol: 3 mL

Lab ID #: 1905056-04
Collected: 5/17/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	4.24		0.18	
NULL	Residual Range Organics	4.49		0.46	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.253	0.244	104	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-96-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 900 mL
Final Vol: 3 mL

Lab ID #: 1905056-05
Collected: 5/18/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	4.02		0.17	
NULL	Residual Range Organics	4.29		0.42	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.244	0.222	110	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-96-2

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 940 mL
Final Vol: 3 mL

Lab ID #: 1905056-06
Collected: 5/18/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	4.13		0.16	
NULL	Residual Range Organics	4.56		0.40	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.238	0.213	112	50-150

Authorized by: LB

Release Date: 6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
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Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-0-120-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 825 mL
Final Vol: 3 mL

Lab ID #: 1905056-08
Collected: 5/19/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.28		0.18	
NULL	Residual Range Organics	0.45	U	0.45	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.279	0.242	115	50-150

Authorized by:

Release Date:

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
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Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-LabCont-120-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 970 mL
Final Vol: 3 mL

Lab ID #: 1905056-09
Collected: 5/19/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.16	J	0.15	
NULL	Residual Range Organics	0.39	UJ	0.39	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.246	0.206	119	50-150

Authorized by:

Release Date:

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-0-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 895 mL
Final Vol: 3 mL

Lab ID #: 1905056-10
Collected: 5/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	3.76	J	0.17	
NULL	Residual Range Organics	3.97	J	0.42	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.251	0.223	112	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-100-0-2

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 990 mL
Final Vol: 3 mL

Lab ID #: 1905056-11
Collected: 5/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	4.38	J	0.15	
NULL	Residual Range Organics	4.77	J	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.241	0.202	120	50-150

Authorized by: _____

Release Date: _____

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-0-0-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 1010 mL
Final Vol: 3 mL

Lab ID #: 1905056-12
Collected: 5/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.22	J	0.15	
NULL	Residual Range Organics	0.37	UJ	0.37	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.215	0.198	108	50-150

Authorized by: _____

LB

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

Field ID: Ctox-Cdub-LabCont-0-1

Work Order: 1905056
Project Officer: Hobbs, William
Initial Vol: 930 mL
Final Vol: 3 mL

Lab ID #: 1905056-13
Collected: 5/14/2019
Prep Method: SW3535A
Analysis Method: NWTPH-DX

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.17	J	0.16	
NULL	Residual Range Organics	0.40	UJ	0.40	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.235	0.215	109	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
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Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : Method Blank

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19F001-BLK1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19F001-BLK1

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

CAS#	Analyte	Result	Qualifier	RL	MDL
68476-34-6	Diesel Range Organics	0.15	U	0.15	
NULL	Residual Range Organics	0.38	U	0.38	

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.223	0.200	111	50-150

Authorized by: _____

Release Date: _____

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**Washington State Department of Ecology
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Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : LCS

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19F001-BS1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19F001-BS1

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: %

Analyte	Result	Spike Level	RL	%Rec	%Rec Limits
Diesel Range Organics	2.52	3.00	0.15	84	70-130

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.228	0.200	114	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
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Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : LCS Dup

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1000 mL
Final Vol: 3 mL

Lab ID #: B19F001-BSD1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19F001-BSD1

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: %

Analyte	Sample Result	Spike Level	%Rec	RPD	%Rec Limits	RPD Limit
Diesel Range Organics	2.42	3.00	81	4	70-130	40

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.199	0.200	99	50-150

Authorized by: _____

Release Date: _____

6/17/19

**Washington State Department of Ecology
Manchester Environmental Laboratory
Final Report for
Semivolatile Petroleum Products**

Project: NWTPH-Dx Tox

QC Type : Duplicate

Work Order: Batch QC
Project Officer: Hobbs, William
Initial Vol: 1010 mL
Final Vol: 3 mL

Lab ID #: B19F001-DUP1
Prep Method: SW3535A
Analysis Method: NWTPH-DX
Source Field ID: B19F001-DUP1
Source Lab ID #: 1905056-11

Batch ID: B19F001
Prepared: 5/30/2019
Analyzed: 6/13/2019
Matrix: Water
Units: mg/L

Analyte	Sample Result	Sample Qual	Source Result	RPD	RPD Limit
Diesel Range Organics	3.76	J	4.38	15	40
Residual Range Organics	4.33	J	4.77	10	40

Surrogate Recovery:

CAS#	Analyte	Result	Spike Level	% Rec.	% Rec. Limits
629-99-2	Pentacosane	0.223	0.198	113	50-150

Authorized by: _____

Release Date: _____

6/17/19

Appendix A Sample Correlation Table

Batch ID: B19F001

Prep Method: SW3535A

Prepared: 5/30/2019

Analysis Method: NWTPH-DX

<u>Field ID</u>	<u>MEL ID</u>
Ctox-Cdub-100-24-1	1905056-01
Ctox-Cdub-100-48-1	1905056-02
Ctox-Cdub-100-48-2	1905056-03
Ctox-Cdub-100-72-1	1905056-04
Ctox-Cdub-100-96-1	1905056-05
Ctox-Cdub-100-96-2	1905056-06
Ctox-Cdub-0-120-1	1905056-08
Ctox-Cdub-LabCont-120-1	1905056-09
Ctox-Cdub-100-0-1	1905056-10
Ctox-Cdub-100-0-2	1905056-11
Ctox-Cdub-0-0-1	1905056-12
Ctox-Cdub-LabCont-0-1	1905056-13
Blank	B19F001-BLK1
LCS	B19F001-BS1
LCS Dup	B19F001-BSD1
Duplicate (Ctox-Cdub-100-0-2)	B19F001-DUP1

Appendix B

Manual Qualification Table

WO: 1905056

Analysis: TPHD

Reported result is estimated; Prep and/or analytical holdtime expired.

Diesel Range Organics J: 1905056-01, 1905056-10, 1905056-11, 1905056-12, 1905056-13, B19F001-DUP1,

Residual Range Organics J: 1905056-01, 1905056-10, 1905056-11, B19F001-DUP1,

Reported result is estimated; sample was not properly preserved.

Diesel Range Organics J: 1905056-02, 1905056-09,

Residual Range Organics J: 1905056-02,

Analyte was not detected at or above the estimated MRL; prep and/or analytical holdtime expired.

Residual Range Organics UJ: 1905056-12, 1905056-13,

Analyte was not detected at or above the estimated MRL; sample was not properly preserved

Residual Range Organics UJ: 1905056-09,

Appendix C Data Qualifier Definitions

Code	Definition
E	Reported result is an estimate because it exceeds the calibration range.
J	The analyte was positively identified; the associated numerical value is the approximate concentration of the analyte in the sample.
N	The analysis indicates the present of an analyte for which there is presumptive evidence to make a “tentative identification”.
NJ	The analysis indicates the presence of an analyte that has been “tentatively identified” and the associated numerical value represents its approximate concentration.
NAF	Not analyzed for.
NC	Not calculated.
REJ	The sample results are rejected due to serious deficiencies in the ability to analyze the sample and meet quality control criteria. The presence or absence of the analyte cannot be verified.
U	The analyte was not detected at or above the reported sample quantitation limit.
UJ	The analyte was not detected at or above the reported sample quantitation limit. However, the reported quantitation limit is approximate and may or may not represent the actual limit of quantitation necessary to accurately measure the analyte in the sample.
bold	The analyte was present in the sample. (Visual aid to locate detected compounds on the analytical report.)

Appendix D

QC Exceptions Report

Lab ID	Analyte	Exception
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No QC exceptions reported.

Appendix E

Initial Calibration Exceptions Report

Calibration ID: B9F1201

Analysis: TPHD

LabNumber **Analyte**

QC Exception

No ICAL exceptions.

APPENDIX F – Chain-of-Custody Forms



TESTING LOCATION (Please Circle)

Burnaby
8664 Commerce Court
Burnaby, British Columbia, Canada
V5A 4N7
Phone 604.420.8773

Calgary
#4, 6125 12 Street SE
Calgary, Alberta, Canada
T2H 2K1
Phone 403.253.7121

Chain of Custody

Date _____ Page ____ of ____

Report to: Company WA Dept of Ecology Address PO Box 47600 City/State/Zip Olympia, WA 98504 Contact Will Hobbs Phone 360-407-7512 Email whob461@ecy.wa.gov						Invoice To: Company WA Dept of Ecology Address PO Box 47612 City/State/Zip Olympia, WA 98504-7612 Contact Accounts Payable or Nicki Christoffer Phone 360-407-7021 Email nchr461@ecy.wa.gov PO No. 19-23608						ANALYSES REQUIRED <div style="display: flex; justify-content: space-between;"> <div style="writing-mode: vertical-rl; transform: rotate(180deg);">as per project QAPP WDOE</div> <div style="writing-mode: vertical-rl; transform: rotate(180deg);">4-d topsmelt</div> <div style="writing-mode: vertical-rl; transform: rotate(180deg);">Echinoderm larvae dev. ecino</div> <div style="writing-mode: vertical-rl; transform: rotate(180deg);">7-d fatheads</div> <div style="writing-mode: vertical-rl; transform: rotate(180deg);">7-d Ceriodaphnia</div> </div>										Receipt Temperature (°C)
Sample Collection By: Hobbs Sample Type: Grab <input type="radio"/> OR Composite <input type="radio"/>																						
SAMPLE ID	DATE	TIME	MATRIX	# OF CONTAINERS AND VOLUME (e.g. 1 x 20 L)	COMMENTS																	
DW-3	1/29/2019		water	5 x 20L	contaminated well	x	x	x	x	x							4.5					
MW-11	1/29/2019		water	2 x 20L	on-site control well	x	x	x	x	x							4.5					
SPECIAL INSTRUCTIONS/COMMENTS (CLIENT)						SAMPLE RECEIPT DETAILS (LABORATORY)				SAMPLE DESCRIPTION AND COMMENTS (LABORATORY)												
as per project plan						1. Total No. of Containers			4. Ice Present in Cooler?		Y / N											
						2. Courier			5. Seal Present?		Y / N											
						3. Good Condition?		Y / N	6. Initials Present on Seal?		Y / N											
RELINQUISHED BY (CLIENT)						RECEIVED BY (LABORATORY)																
(Printed Name) _____ (Signature) _____						(Signature) _____ (Date and Time) Jan 30/19 0846																
(Company) _____ (Date and Time) _____						(Company) _____ (Date and Time) _____																
Additional costs may be required for sample disposal or storage. Payment net 30 unless otherwise contracted.																Form 020; Version 1.0; Revised by CC 2016/20/09						

END OF REPORT
