

**ACUTE TOXICITY TESTING  
ON FLOC LOG 708X**

**APPLIED POLYMER SYSTEMS, INC.**  
Woodstock, GA

Test Period: August 3-7, 2010

**Prepared for:**  
**Applied Polymer Systems, Inc.**  
Woodstock, Georgia

**Prepared by:**  
**MACTEC Engineering and Consulting, Inc.**  
Biology-Toxicology Laboratory  
404 SW 140<sup>th</sup> Terrace  
Newberry, FL 32669  
Telephone (352) 332-3318  
NELAC# E82998

MACTEC Project Number 6067100022.01

**August 31, 2010**



engineering and constructing a better tomorrow

August 31, 2010

Mr. Steven R. Iwinski  
Applied Polymer Systems, Inc.  
519 Industrial Drive  
Woodstock, GA 30189

Subject: **Applied Polymer Systems, Inc. Acute Toxicity Testing on Floc Log 708x  
August 3-7, 2010  
MACTEC Project No. 6067100022.01**

Dear Mr. Iwinski:

MACTEC Engineering and Consulting, Inc. (MACTEC), Biology-Toxicology Laboratory has completed 96-hour acute toxicity testing on samples of Floc Log 708x using the rainbow trout, *Oncorhynchus mykiss* and a 48-hour acute toxicity testing on samples of Floc Log 708x using the Water Flea, *Daphnia magna*. Tests were conducted in accordance with *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms*, EPA-821-R-02-012 (USEPA, 2002). The test endpoints for the acute toxicity tests are LC50 (lethal concentration of effluent tested that exhibit toxicity by mortality in 50% of the organisms tested), and the NOEC (the No Observed Effect Concentration, or the highest concentration tested that did not exhibit acute toxicity).

Test samples for the water flea were prepared by adding 840 milligrams (mg) of polymer to 1.0 liters (L) of dilute mineral water and allowing mixing on a stir plate for several hours (i.e., overnight). This stock test sample was then diluted with dilute mineral water to the following test concentrations: 52.5 ppm, 105 ppm, 210 ppm, 420 ppm, and 840 ppm. Test samples for the rainbow trout were prepared by adding 60 grams (g) of polymer to 17.86 liters (L) of mod hard water and allowing mixing on a stir plate for several hours (i.e., overnight). This stock test sample was then diluted with mod hard water to the following test concentrations: 210 ppm, 420 ppm, 840 ppm, 1680 ppm and 3360 ppm. Rainbow trout and water flea test organisms were exposed to the individual test concentrations. The rainbow trout exposures were renewed once with fresh polymer after 48 hours, and the water flea was not renewed. The survival endpoints were determined at the end of the test period for the water flea and rainbow trout.

The results at the conclusion of the acute toxicity test conducted with the water flea during the test period August 3 through 5, 2010 are: survival NOEC was 840 ppm and the LC50 was >840 ppm of Floc Log 708X for water flea survival.

The results at the conclusion of the acute toxicity test conducted with the rainbow trout during the test period August 3 through 7, 2010 are: survival NOEC was 3360 ppm and the LC50 was >3360 ppm of Floc Log 708X for water flea survival.

Results are summarized in the accompanying report. Detailed testing documentation, including chemical

and physical measurements (Appendix A), rainbow trout documentation (Appendix B), and polymer sample test data/reference toxicant data (Appendix C).

We appreciate the opportunity to provide these services to Applied Polymer Systems, Inc. Should you have any questions or need further assistance, please contact Joy Ryan or William Tucker at (352) 332-3318.

Sincerely,

**MACTEC Engineering and Consulting, Inc.**



R. Dave Dickens  
Biology-Toxicology Laboratory Manager  
Technical Specialist



William A. Tucker, PhD  
Laboratory Quality Assurance Officer  
Principal Scientist

Attachment:        Data Report

## TEST SUMMARY

### I. Client

Client: Applied Polymer Systems, Inc.  
519 Industrial Drive  
Woodstock, GA 30189

Product Tested: Floc Log 708x

### II. Laboratory Accreditation

Laboratory: Biology Toxicology Laboratory  
MACTEC Engineering and Consulting, Inc.  
404 SW 140 Terrace  
Newberry, Florida 32669

Accreditor: State of Florida, Department of Health  
Bureau of Laboratories

Accreditation ID: Lab ID. E82998

Category: Non-Potable Water Whole Effluent Toxicity

Effective: July 1, 2010 through June 30, 2011

### III. Tests Conducted

Test: **Water Flea, *Daphina magna***  
Methods for Estimating the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms,  
EPA-821-R-02-012, Test Method 2021.0 (EPA, 2002).  
Acute Definitive Survival Test

**Rainbow Trout, *Oncorhynchus mykiss***  
Methods for Estimating the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms,  
EPA-821-R-02-012, Test Method 2019.0 (EPA, 2002).  
Acute Definitive Survival Test

Test conditions and test acceptability criteria can be found in the Additional Tables (Tables 3 & 4) section at the end of this report.

Test Dates/Times: **Water Flea Acute:** August 3(1430)-5(1300), 2010  
**Rainbow Trout Acute:** August 3 (1600) - 7 (1400), 2010

Source/Age of Organisms: **Water Flea:** Marinco, ID# 2010-43/< 24 hours  
**Rainbow Trout:** Aquatic Research Organisms, ID# 2010-42 / 15 days

Source organism information can be found in Appendix B of this report.

Test Concentrations: **Water Flea:** Control, 52.5 ppm, 105ppm, 210ppm, 420 ppm, and 840 ppm  
**Rainbow Trout:** Control, 210 ppm, 420 ppm, 840 ppm, 1,680 ppm, and 3,360 ppm

Control Water: **Water Flea:** DMW, Dilute Mineral Water, 20% Perrier.  
**Rainbow Trout:** MHSFW, Moderately Hard Synthetic Freshwater

Deviation from Test Protocol: None

Statistical Package: NOEC determined using Comprehensive Environmental Toxicity Information System (CETIS) software (Tidepool, 2008)

#### IV. Results

**Table 1. Survival and Reproduction Data for Water Fleas Exposed for Two Days to Floc Log 708X, August 3 through 5, 2010.**

% Total Effluent	Survival (%)
Laboratory Control <sup>(a)</sup>	100
52.5	100
105	100
210	95
420	90
840	100
<b>NOEC <sup>(b)</sup> (ppm)</b>	<b>840</b>
<b>LC50 <sup>(c)</sup> (ppm)</b>	<b>&gt;840</b>

Source: MACTEC, 2010

Prepared by: RDD

Checked by: AWW

<sup>(a)</sup> Laboratory Water Control; Dilute Mineral Water Batch #PD2010-21.

<sup>(b)</sup> NOEC = No Observable Effect Concentration.

<sup>(c)</sup> LC50 = Lethal Concentration at 50%

**Table 2. Survival and Growth Data for Rainbow Trout Exposed for Four Days to Floc Log 708X, August 3 through 7, 2010.**

% Total Effluent	Survival (%)
Laboratory Control <sup>(a)</sup>	100
210	100
420	100
840	100
1680	100
3360	100
<b>NOEC <sup>(b)</sup> (ppm)</b>	<b>840</b>
<b>LC50 <sup>(c)</sup> (ppm)</b>	<b>&gt;840</b>

Source: MACTEC, 2010

Prepared by: RDD

Checked by: AWW

<sup>(a)</sup> Laboratory Water Control, Moderately Hard Synthetic Freshwater, Batch #MHW073010.

<sup>(b)</sup> NOEC = No Observable Effect Concentration.

<sup>(c)</sup> LC50 = Lethal Concentration at 50%

## V. Summary

The results at the conclusion of the acute toxicity test conducted with the water flea during the test period August 3 through 5, 2010 are: survival NOEC was 840 ppm and the LC50 was >840 ppm of Floc Log 708X for water flea survival.

The results at the conclusion of the acute toxicity test conducted with the rainbow trout during the test period August 3 through 7, 2010 are: survival NOEC was 3360 ppm and the LC50 was >3360 ppm of Floc Log 708X for water flea survival.

## VI. Quality Assurance

*Daphnia magna*, KCl Reference Toxicant, RT48HDM20100803, 08/03/10

Survival: 48-hour LC50 = 4702 mg/L NaCl  
 48-hour LC50 Range of Acceptability = 2860 to 6950 mg/L NaCl (Acceptable)

*Oncorhynchus mykiss*, KCl Reference Toxicant, RT96HOM20100803, 08/03/10

Survival: 48-hour LC50 = 31,620 mg/L KCl  
 48-hour LC50 Range of Acceptability = 24,722 to 35,726 mg/L KCl (Acceptable)

## GLOSSARY AND ABBREVIATIONS

Acute	Involving a stimulus severe enough to rapidly induce a response; in toxicity tests, a response observed in 96 hours or less typically is considered acute.
Chronic	Involving a stimulus that lingers or continues for a relatively long period of time, often one-tenth of the life span or more. A chronic effect can be lethality, growth, reduced reproduction, etc.
Chronic Value	A numeric value representing the geometric mean of the numeric values of concentrations analyzed as the NOEC (No Observed Effect Concentration) and the LOEC (Lowest Observed Effect Concentration) by chronic toxicity testing. The chronic value is an estimate of the toxicant concentration that will be the actual no effect concentration based on the chronic effect tested.
Critical Value	Minimum numeric value for a toxicity test endpoint (i.e., survival, growth, or reproduction) below which a given test result will be statistically significantly different from the control value.
DMW	Diluted Mineral Water
EC	Effective concentration, a point estimate of the toxicant concentration that would cause an adverse response such as death, immobilization, or serious incapacitation.
Ft-c	Foot candles - a measure of <u>light</u> intensity
Graphical Method	Log concentration versus percent mortality method. Toxicity test data are plotted on 2-cycle semi-log graph paper. The logarithmic axis (y axis) is used for percent effluent concentration, and the linear axis (x axis) is used for percent mortality. The graph provides a reasonably accurate estimate of the LC <sub>50</sub> , but does not provide a confidence interval.
IC	Inhibition Concentration, a point estimate of the toxicant concentration that would cause a given percent reduction in a biological measurement such as fecundity or growth.
LC	Lethal Concentration, identical to EC when the observed response is death.
LC <sub>50</sub>	The toxicant concentration that is lethal to 50 percent of exposed organisms at a specific time of observation.
LCL	Lower 95% Confidence Limit
LOEC	Lowest-Observed-Effect-Concentration, the lowest concentration of toxicant to which organisms are exposed that causes adverse effects.
LWC	Lab Water Control, moderately hard synthetic freshwater prepared from MILLIPORE MILLI-Q <sup>R</sup> water and reagent grade chemicals.

NOEC	No-Observed-Effect-Concentration, the highest concentration of toxicant to which organisms are exposed that causes no observable adverse effects.
Probit Analysis	Probit Analysis consists of a group of statistical methods used to analyze data from concentration-response experiments, and provides an estimate of the LC <sub>50</sub> and the precision of this estimate. In Probit Analysis, the percentages of affected organisms are converted to Probits (probability units), and the effluent concentrations are converted to logarithms. The relationship between the Probits and the logarithmic values of the concentrations is approximately linear. A Probit regression line drawn through the data points is used to estimate the LC <sub>50</sub> and its precision estimate. To use Probit Analysis, at least two partial mortalities must be obtained in the toxicity test.
RWC	Receiving Water Control
UCL	Upper 95% Confidence Limit
μE/m <sup>2</sup> /s	Micro-ergs per square meter per second - a measure of <u>light</u> intensity

## REFERENCES

- Gulley, D.D., and WEST, Inc. 1996. TOXSTAT version 3.5. Fish Physiology and Toxicology Laboratory. Department of Zoology and Physiology. University of Wyoming. Laramie, Wyoming.
- U.S. Environmental Protection Agency. 1979. Methods for Chemical Analysis of Water And Wastes. Environmental Monitoring Laboratory, U.S. Environmental Protection Agency, Cincinnati, OH. EPA 600/4-79-020.
- U.S. Environmental Protection Agency. 1999. Errata for Effluent and Receiving Water Toxicity Testing Manuals: Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms; Short-Term Methods for Estimating the Chronic Toxicity of Effluent and Receiving Waters to Freshwater Organisms; and Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Marine and Estuarine Organisms. U.S. Environmental Protection Agency, Office of Research and Development, Duluth, MN. EPA/600/R-98/182.
- U.S. Environmental Protection Agency. 2002. Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms. Fourth Edition. Office of Water (4303T), U.S. Environmental Protection Agency, Washington, DC. EPA-821-R-02-013.

**Table 3. Summary of Effluent Toxicity Test Conditions for the Water Flea (*Daphnia magna*) Acute Survival Test (EPA 800-R-02-012)**

---

1.	Test type:	48-hr Static Acute
2.	Temperature:	(°C): 24.5-25.0°C
3.	Light quality:	Ambient laboratory illumination
4.	Light intensity:	10-20 uE/m <sup>2</sup> /s (50-100 ft-c) (ambient laboratory levels)
5.	Photoperiod:	16-hr light, 8-hr dark
6.	Test chamber size:	30 mL
7.	Test solution volume:	15 mL/replicate
8.	Age of test organisms:	<24-hr
9.	No. neonates per test chamber:	5
10.	No. replicate chambers per concentration:	4
11.	No. neonates per concentration:	20
12.	Feeding regime:	Fed 0.2 mL each of <i>Selenastrum</i> /YCT prior to test start
13.	Aeration:	None
14.	Dilution water:	Moderately hard synthetic water prepared using MILLIPORE MILLI-Q <sup>R</sup> water and 20% Perrier
15.	Effluent concentrations:	Control and 17%
16.	Dilution factor:	17%
17.	Test duration:	48-hr
18.	Effect measured:	Mortality - no movement on gentle prodding
19.	Test acceptability:	90% or greater survival in controls
20.	Sample handling:	Samples are to be iced during shipment and held at 4°C until used. Test must be initiated within 36-hr from time of sampling.
21.	Sample volume required:	2 L
22.	Chemical parameters:	pH, DO, total residual chlorine, total alkalinity, total hardness, conductivity, temperature

**Table 4. Summary of Effluent Toxicity Test Conditions for the Rainbow Trout (*Oncorhynchus mykiss*) Survival Test (EPA 821-R-02-012)**

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1. Test type:	Static non-renewal definitive
2. Temperature:	11.5-13.4°C
3. Light quality:	Cool White Fluorescent
4. Light intensity:	10-20 $\mu\text{E}/\text{m}^2/\text{s}$ (50-100 ft-c)
5. Photoperiod:	16-hr light, 8-hr dark
6. Test chamber size:	5L
7. Test solution volume:	4L
8. Age of rainbow trout:	15-30 days
9. No. fish per test chamber:	10
10. No. replicate chambers per concentration:	2
11. No. fish per concentration:	20
12. Feeding regime:	None.
13. Aeration:	None.
14. Dilution water:	Moderately hard synthetic freshwater (reagent grade chemicals and Di-ionized)
15. Effluent concentrations (ppm):	Control, 210, 420, 840, 1,680, and 3,360
16. Dilution factor:	0.5
17. Test duration:	96-hours
18. Effect measured:	Survival/mortality - no movement on gentle prodding
19. Test acceptability:	90% or greater survival in controls
20. Sample handling:	Samples held at room temperature
21. Sample volume required:	17.8 L of 3360 ppm
22. Chemical parameters:	pH, DO, conductivity, temperature

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## **APPENDICES**

**APPENDIX A**  
**Chemical and Physical Data**

**Table 5. Initial Chemical Characterization of Applied Polymer Systems, Inc. Floc Log 708x and Control Water Used in Chronic Toxicity Tests, August 3-7, 2010.**

<b>Parameter</b>	<b>FLOC LOG 708x (3,360 ppm)</b>	<b>FLOC LOG 708x (840 ppm)</b>	<b>DMW<sup>(a)</sup></b>	<b>MHW<sup>(b)</sup></b>
Dissolved Oxygen (mg/L)	8.4	7.4	7.9	7.9
pH	7.7	7.9	8.0	7.9
Total Alkalinity as CaCO <sub>3</sub> (mg/L)	NR <sup>(c)</sup>	NR <sup>(c)</sup>	68	56
Total Hardness as CaCO <sub>3</sub> (mg/L)	NR <sup>(c)</sup>	NR <sup>(c)</sup>	75	75
Conductivity @ 25°C (µmhos/cm)	1389	455	192	271

Source: MACTEC, 2010

Prepared by: RDD

Checked by: NMG

Determined according to EPA 600/4-79-020, 1979.

<sup>a</sup> DWM = Dilute Mineral Water, (20% Perrier Water and 80% DI Water), Batch #PD2010-21

<sup>b</sup> MHW=Moderately hard synthetic freshwater, (reagent grade chemicals and Di-ionized)

<sup>c</sup> NR=Not Recorded

**APPENDIX B**  
**Organism Document**

**MACTEC Engineering and Consulting, Inc.**  
**BioTox Laboratory**  
 404 SW 140<sup>th</sup> Terrace  
 Newberry, FL 32669

*Daphnia magna* Culture Source Document

MACTEC Organism Batch ID No.	Source	Age of Test Organisms	Water Type/Temp. (°C)	Food Type
2010-43	ARO	<24-hr	DMW / 25.0	YCT & <i>Selenastrum</i>

*Oncorhynchus mykiss* Culture Source Document

MACTEC Organism Batch ID No.	Source	Age of Test Organisms	Water Type/Temp. (°C)	Food Type
2010-42	ARO	15 days	MHW / 12.0	None

**APPENDIX C**

**Test Data Sheets**

**Water Flea**  
**Floc Log 708x Test**

**TOXICITY TEST SYSTEM DESCRIPTION SHEET**

SPONSOR: Applied Polymer Systems

Project Number: 6067100022.01

**TEST MATERIAL**

Test Material: Chemical: Floc Log 708x

See Page N/A of Chemical Log for Test Material Information.

**TEST CONDITIONS**

( ) Range Finder (X) Static  
( ) Screening ( ) Flow - Through  
(X) Definitive

**PROTOCOL/GUIDELINES FOLLOWED:**

EPA-821-R-02-012

Lighting: (X) Fluorescent ( ) Incandescent  
Photoperiod: 16 Hours Light  
8 Hours Dark

Test Concentration Based On: ( ) Active Ingredient  
(X) Whole Material

Test Duration: ~~96~~ hrs. 8/3 am  
Test Area Used: ~~Water Bath~~ Incubator  
Test Temperature: ~~20 ± 1°C~~ 25  
Test Salinity: NA

**TEST ORGANISM HISTORY**

Test Species: *Daphnia magna*

Lot Number: 2010-43

Age/Life Stage: < 24 hrs

Date Acclimation/Maintenance Began: 8/3/10

See Page N/A of Cladoceran Neonate Log for Data.  
cultured out for house

Mortality (%) During 48 hours Prior to Test Initiation: < 1%

Replicates per Concentration: 4  
Organisms per Replicate: 5

Dilution Water: chem prepared mod hard,  
Perrier®/milliQ mix

**TEST VESSELS**

Dimensions (cm): 2 length x2 width x4 height  
Test Solution Height (cm): 3  
Test Vessels: ( ) Open (X) Covered  
Test Container Volume: 0.03 L  
Test Solution Volume: 0.025L

Dilution Water Batch # PD2010-21

**TEST SOLUTION PREPARATION**

Test Concentration mg/L	0	52.5	105	210	420	840		
Volume of Dilution Water Added (mL)	200	188	175	150	100	0		
Volume of Effluent/ Stock Added (mL)	0	12.5	25	50	100	200		

ADDITIONAL OBSERVATIONS: \_\_\_\_\_

DATA BY: [Signature]



ACUTE TOXICITY DATA SHEET - WATER QUALITY DATA												
SPONSOR: Applied Polymer Systems		Project Number: 6067100022.01										
TEST SUBSTANCE: Chemical : Floc Log 708x		TEST SPECIES: <i>Daphnia magna</i>										
DATE	8/3/10	8/4/10	8/6/10									
TIME	1345	1540	1855									
INIT.	QWB	ARB	KLZ									
NOMIN CONCN % mg/L µg/L	T	E	M	P	SL or CN	DO	pH	DO	pH	DO	pH	DO
	R	E	M	P	SL or CN	DO	pH	DO	pH	DO	pH	DO
W10	A	25.1	7.9	7.4	USC							
840	B											
JUL-APS-DM	C					25.5	8.1	8.4	8.70	8.2	6.4	7.2
	D											
	A											
	B											
	C											
	D											
	A											
	B											
	C											
	D											
INST/METH												

Key

TEMP - Temperature  
SL - Salinity (ppt)  
pH - Standard units  
DO - Dissolved oxygen (mg/L)  
CN - Conductivity (µmhos/cm)  
INST - Instrument  
NOMIN CONCN - Nominal concentration  
INIT. - Initials  
REP - Replicate  
METH - Method

*Handwritten mark*

**ACUTE TOXICITY DATA SHEET - SURVIVAL**

SPONSOR: Applied Polymer Systems		Project Number: 6067100022.01				
TEST SUBSTANCE: Chemical : Floc Log 708x		TEST SPECIES: <i>D. magna</i>				
TEST HOUR	0 - HOUR	24 - HOURS	48 - HOURS			
DATE	8/3/10	8/4/10	8/5/10			
FEEDING	IP Holdings	NONE	NONE			
TIME	1430	1400	1300			
INIT.	AKMA	DD	KLE			
NOMINAL CONCENTRATION % mg/L	R					
	E					
W10 CTRL JUL-APS-DM	P					
	A	5 loaded	2	5	2	N
	B		2	5	2	2
	C		2	5	2	2
W10 52.5 JUL-APS-DM	D		2	5	2	2
	A		2	5	2	2
	B		2	5	2	2
	C		2	5	2	2
W10 105 JUL-APS-DM	D		2	5	2	2
	A		2	5	2	2
	B		2	5	2	2
	C		2	5	2	2
W10 210 JUL-APS-DM	D		2	5	2	2
	A		2	5	2	2
	B		2	5	2	2
	C		2	5	2	2
W10 420 JUL-APS-DM	D		2	5	2	2
	A		2	5	2	2
	B		2	5	2	2
	C		2	5	2	2

**OBSERVATION KEY**

REP - Replicate  
OBS - Observations  
INIT. - Initials  
No. ALIVE - Number of Organisms Alive (survival)  
N (None) - Normal Conditions  
NF - Not Found  
YTC/ ALG - YTC and Green Algae (daphnids)

PRE - Precipitate  
FOS - Film on Surface of Water  
UC - Undissolved Chemical  
LETH - Lethargic  
LE - Loss of Equilibrium  
HYP - Hyperactive  
BS - Brine Shrimp

AS - Organisms at Surface of Water  
ERR - Erratic  
GY - Gyration  
G - Gulping  
RAR - Rapid Respiration  
LT - Light Pigmentation  
D - Organisms Dead

✓

**ACUTE TOXICITY DATA SHEET - SURVIVAL**

SPONSOR: Applied Polymer Systems		Project Number: 6067100022.01					
TEST SUBSTANCE: Chemical : Floc Log 708x		TEST SPECIES: <i>D. magna</i>					
TEST HOUR	0 - HOUR	24 - HOURS	48 - HOURS				
DATE	8/13/10	8/15/10					
FEEDING	FN Holders	NONE					
TIME	1430	1300					
INIT.	ARM	BB	KLS				
NOMINAL CONCENTRATION % mg/L	R						
	E						
W10 840 JUL-APS-DM	P	No. ALIVE	OBSERV.	No. ALIVE	OBSERV.	No. ALIVE	OBSERV.
	A	5	N	5	N	5	N
	B	5	N	5	N	5	N
	C	5	N	5	N	5	N
	D	5	N	5	N	5	N
	A						
	B						
	C						
	D						
	A						
	B						
	C						
	D						
	A						
	B						
	C						
	D						

**OBSERVATION KEY**

REP - Replicate  
 OBS - Observations  
 INIT. - Initials  
 No. ALIVE - Number of Organisms Alive (survival)  
 N (None) - Normal Conditions  
 NF - Not Found  
 YTC/ALG - YTC and Green Algae (daphnids)

PRE - Precipitate  
 FOS - Film on Surface of Water  
 UC - Undissolved Chemical  
 LETH - Lethargic  
 LE - Loss of Equilibrium  
 HYP - Hyperactive  
 BS - Brine Shrimp

AS - Organisms at Surface of Water  
 ERR - Erratic  
 GY - Gyration  
 G - Gulping  
 RAR - Rapid Respiration  
 LT - Light Pigmentation  
 D - Organisms Dead

**CETIS Summary Report**

Report Date: 25 Aug-10 13:46 (p 1 of 1)  
 Test Code: 06-1052-9727/W10-AUG-APS-DM

**Daphnia magna 48-h Acute Survival Test**

MACTEC Biology-Toxicology LAB

<b>Batch ID:</b> 06-5168-6787	<b>Test Type:</b> Survival (48h)	<b>Analyst:</b>
<b>Start Date:</b> 03 Aug-10	<b>Protocol:</b> EPA/821/R-02-012 (2002)	<b>Diluent:</b> Dilute Mineral Water
<b>Ending Date:</b> 05 Aug-10	<b>Species:</b> Daphnia magna	<b>Brine:</b> Not Applicable
<b>Duration:</b> 48h	<b>Source:</b> Marinco, FL	<b>Age:</b>
<b>Sample ID:</b> 19-1099-0216	<b>Code:</b> 71E76588	<b>Client:</b> Applied Polymer Systems
<b>Sample Date:</b> 03 Aug-10	<b>Material:</b> Flocc Log	<b>Project:</b> 6067100022
<b>Receive Date:</b> 03 Aug-10	<b>Source:</b> Applied Polymer Systems	
<b>Sample Age:</b> N/A	<b>Station:</b> 708X	

**Comparison Summary**

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
09-5310-3439	48h Survival Rate	840	>840	N/A	11.88%		Steel Many-One Rank Test

**Point Estimate Summary**

Analysis ID	Endpoint	Level	ppm	95% LCL	95% UCL	TU	Method
01-8265-8501	48h Survival Rate	LC5	667.2	N/A	N/A		Linear Regression (MLE)
		LC10	2818	N/A	N/A		
		LC15	7448	N/A	N/A		
		LC20	16130	N/A	N/A		
		LC25	31290	N/A	N/A		
		LC40	166200	N/A	N/A		
		LC50	453900	N/A	N/A		

**Test Acceptability**



Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits	Overlap	Decision
01-8265-8501	48h Survival Rate	Control Resp	1	0.9 - NL	Yes	Result Within Limits
09-5310-3439	48h Survival Rate	Control Resp	1	0.9 - NL	Yes	Result Within Limits

**48h Survival Rate Summary**

Conc-ppm	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Dilution Water	4	1	1	1	1	1	0	0	0.0%	0.0%
52.5		4	1	1	1	1	1	0	0	0.0%	0.0%
105		4	1	1	1	1	1	0	0	0.0%	0.0%
210		4	0.95	0.9127	0.9873	0.8	1	0.01826	0.1	10.53%	5.0%
420		4	0.9	0.8569	0.9431	0.8	1	0.02108	0.1155	12.83%	10.0%
840		4	1	1	1	1	1	0	0	0.0%	0.0%

**48h Survival Rate Detail**

Conc-ppm	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Dilution Water	1	1	1	1
52.5		1	1	1	1
105		1	1	1	1
210		1	0.8	1	1
420		0.8	1	0.8	1
840		1	1	1	1

Analyst:  QA: 

**Rainbow Trout**  
**Floc Log 708x Test**



Project Number: 6067100022.01

Client: Applied Polymer Systems

Test: Acute Definitive, *O. mykiss* and *D. magna*

DAILY LOG

8/3/10 - Received eggs, made dilutions, monitored  
WQ + eggs, loaded eggs - AMW

8/4/10 - Monitored WQ + eggs - AMW

8/5/10 - Monitored WQ + eggs, made new dilutions,  
renewed test - AMW

8/6/10 - Monitored WQ + eggs; AMW

8/7/10 - monitored eggs + WQs  
test terminated DD

8/8/10 - monitored WQ + eggs - AMW





**ACUTE TOXICITY DATA SHEET - SURVIVAL**

SPONSOR: Applied Polymer Systems		Project Number: 6067100022.01									
TEST SUBSTANCE: Chemical : Floc Log 708x		TEST SPECIES: O. mykiss									
TEST HOUR	0 - HOUR	24 - HOURS	48 - HOURS	72 - HOUR	96 - HOUR						
DATE	8/13/10	8/14/10	8/15/10	8/16/10	8/17/10						
FEEDING	No	No	No	No	No						
TIME	1600	1630	1430	1330	1400						
INIT.	Alman	Alman	Alman	Alman	Alman						
NOMINAL CONCENTRATION % mg/L	R	No. ALIVE	OBSERV.	No. ALIVE	OBSERV.	No. ALIVE	OBSERV.	No. ALIVE	OBSERV.		
	E	P	A	B	A	B	A	B	A	B	
JUL-APS-OM	W10	↓	10 loaded	10	22	10	22	10	22	10	22
	CTRL	↓	↓	10	22	10	22	10	22	10	22
JUL-APS-OM	W10	↓	↓	10	22	10	22	10	22	10	22
	210	↓	↓	10	22	10	22	10	22	10	22
JUL-APS-OM	W10	↓	↓	10	22	10	22	10	22	10	22
	420	↓	↓	10	22	10	22	10	22	10	22
JUL-APS-OM	W10	↓	↓	10	22	10	22	10	22	10	22
	840	↓	↓	10	22	10	22	10	22	10	22
JUL-APS-OM	W10	↓	↓	10	22	10	22	10	22	10	22
	1680	↓	↓	10	22	10	22	10	22	10	22

**OBSERVATION KEY**

- REP - Replicate
- OBS - Observations
- INIT. - Initials
- No. ALIVE - Number of Organisms Alive (survival)
- N (None) - Normal Conditions
- NF - Not Found
- YTC/ ALG - YTC and Green Algae (daphnids)
- PRE - Precipitate
- FOS - Film on Surface of Water
- UC - Undissolved Chemical
- LETH - Lethargic
- LE - Loss of Equilibrium
- HYP - Hyperactive
- BS - Brine Shrimp
- AS - Organisms at Surface of Water
- ERR - Erratic
- GY - Gyration
- G - Gulping
- RAR - Rapid Respiration
- LT - Light Pigmentation
- D - Organisms Dead



**Water Flea**  
**Reference Toxicant Data Sheets**



REFERENCE TOXICANT DATA SHEET										
SPONSOR: MACTEC								SPECIES: <i>D. magna</i>		
REF TOXICANT: NaCl										
DAY 0			DATE: 8/3/10			TIME: 1345		ANALYST: KDB		
NOMINAL CONCENTRATION (mg/L)	# ALIVE			OBSERVATION			FS-11	Ref	DO7	PH1
	A	B	C	A	B	C	TEMP (°C)	SALINITY (ppt)	DO (mg/L)	pH
CONTROL	5	5	5	N	N	N	25.5	0.0	7.6	7.9
1	5	5	5	N	N	N	25.1	1.0	7.4	7.7
2	5	5	5	N	N	N	25.3	2.0	7.7	7.8
4	5	5	5	N	N	N	25.3	4.0	7.7	7.7
8	5	5	5	N	N	N	25.3	8.0	7.8	7.6
16	5	5	5	N	N	N	25.4	16.0	7.7	7.6
DAY 1			DATE: 8/4/10			TIME: 1530		ANALYST: (MWW)		
NOMINAL CONCENTRATION (mg/L)	# ALIVE			OBSERVATION			FS-12	Ref 15	DO7	PH1
	A	B	C	A	B	C	TEMP (°C)	SALINITY (ppt)	DO (mg/L)	pH
CONTROL	5	5	5	N	N	N	25.7	0	7.1	8.3
1	5	5	5	N	N	N	25.4	1.0	7.0	8.1
2	5	5	5	N	N	N	25.5	2.0	6.6	8.1
4	4	5	5	1D	N	N	25.5	4.0	7.2	8.1
8	0	0	0	5D	5D	5D	25.6	7.0	6.3	8.0
16	0	0	0	5D	5D	5D	25.6	15.0	6.0	8.0
DAY 2			DATE: 8/5/10			TIME: 1310		ANALYST: ZLE		
NOMINAL CONCENTRATION (mg/L)	# ALIVE			OBSERVATION			FS-12	Ref 15	DO7	PH1
	A	B	C	A	B	C	TEMP (°C)	SALINITY (ppt)	DO (mg/L)	pH
CONTROL	5	5	5	N	N	N	25.2	0	7.8	8.2
1	5	5	5	N	N	N	24.8	1.0	7.3	8.2
2	5	5	5	N	N	N	24.9	2.0	7.9	8.2
4	4	3	4	N	2D	1D	25.1	4.0	7.9	8.2
8										
16										

*[Handwritten signature]*

**REFERENCE TOXICANT DATA SHEET**

SPONSOR: MACTEC

SPECIES: *D. magna*

REF TOXICANT: Na Cl

**SUMMARY**

COMMENTS:

NOMINAL CONCENTRATION	# DEAD			TOTAL DEAD PER CONC.	
	A	B	C		
CONTROL	0	0	0	0/15	0%
1	0	0	0	0/15	0%
2	0	0	0	0/15	0%
4	1	2	1	4/15	26.7%
8	5	5	5	15/15	100%
16	5	5	5	15/15	100%

*J*

**CETIS Summary Report**

Report Date: 25 Aug-10 13:46 (p 1 of 1)  
 Test Code: 04-2080-6285/RT48HDM20100803

**Daphnia magna 48-h Acute Survival Test**

MACTEC Biology-Toxicology LAB

Batch ID: 06-5168-6787	Test Type: Survival (48h)	Analyst:
Start Date: 03 Aug-10	Protocol: EPA/821/R-02-012 (2002)	Diluent: Dilute Mineral Water
Ending Date: 05 Aug-10	Species: Daphnia magna	Brine: Not Applicable
Duration: 48h	Source: Marinco, FL	Age:
Sample ID: 04-6748-3328	Code: 1BDD3AC0	Client: ADMIN
Sample Date: 03 Aug-10	Material: Sodium chloride	Project: ADMIN
Receive Date: 03 Aug-10	Source: Reference Toxicant	
Sample Age: N/A	Station: In House	

**Point Estimate Summary**

Analysis ID	Endpoint	Level	gm/L	95% LCL	95% UCL	TU	Method
21-3240-9708	48h Survival Rate	LC50	4.702	4.014	5.509		Spearman-Kärber

**Test Acceptability**

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits	Overlap	Decision
21-3240-9708	48h Survival Rate	Control Resp	1	0.9 - NL	Yes	Result Within Limits

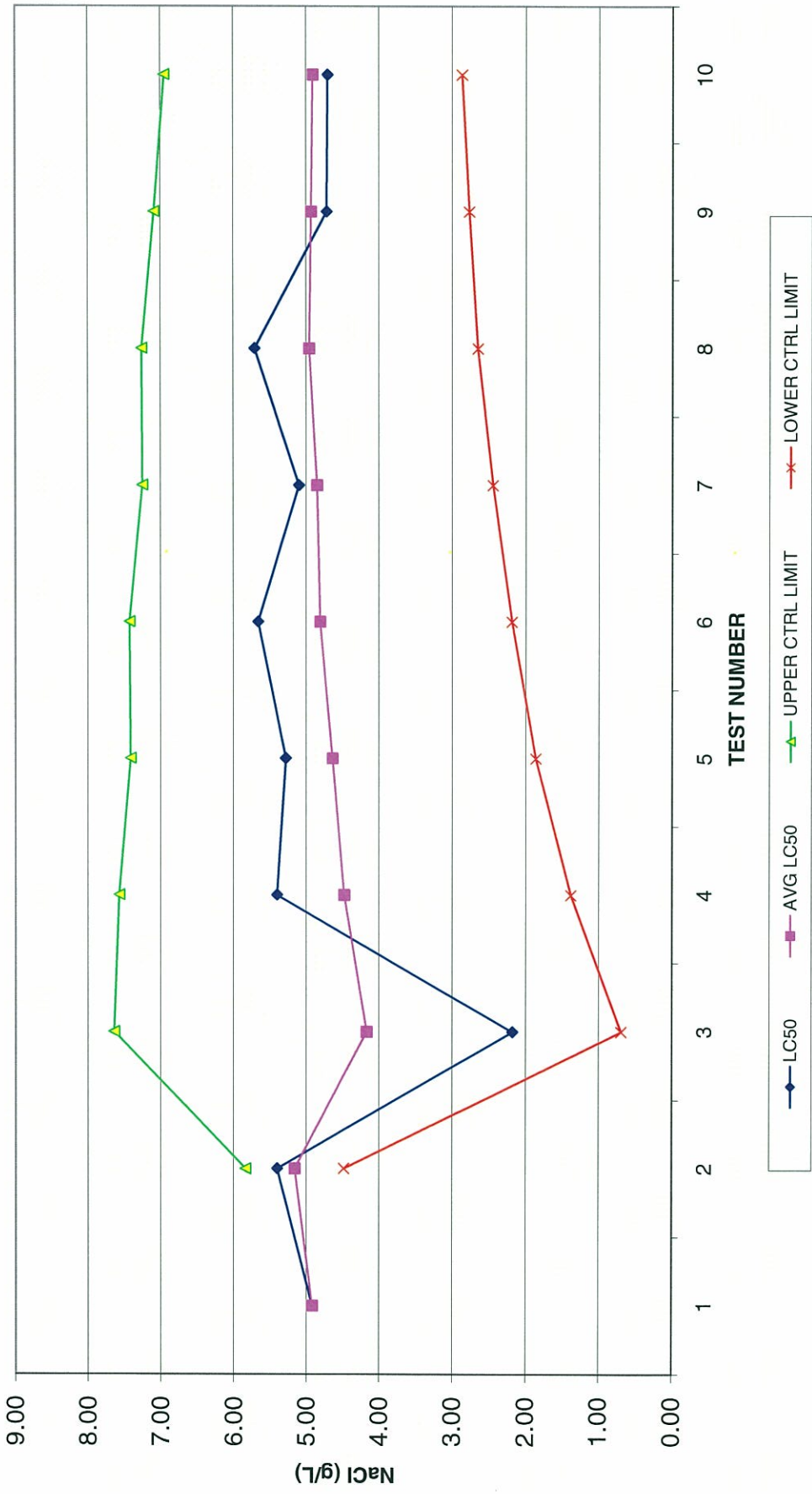
**48h Survival Rate Summary**

Conc-gm/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Dilution Water	3	1	1	1	1	1	0	0	0.0%	0.0%
1		3	1	1	1	1	1	0	0	0.0%	0.0%
2		3	1	1	1	1	1	0	0	0.0%	0.0%
4		3	0.7333	0.6902	0.7765	0.6	0.8	0.02108	0.1155	15.75%	26.67%
8		3	0	0	0	0	0	0	0		100.0%
16		3	0	0	0	0	0	0	0		100.0%

**48h Survival Rate Detail**

Conc-gm/L	Control Type	Rep 1	Rep 2	Rep 3
0	Dilution Water	1	1	1
1		1	1	1
2		1	1	1
4		0.8	0.6	0.8
8		0	0	0
16		0	0	0

MACTEC Engineering and Consulting, Inc.  
*Daphnia Magna*  
 ACUTE REFERENCE TOXICITY TEST: Sodium Chloride



**Rainbow Trout**  
**Reference Toxicant Data Sheets**

**REFERENCE TOXICANT TEST DESCRIPTION SHEET**

SPONSOR: MACTEC PROJECT NUMBER: REF TOX

<p><b>TEST MATERIAL</b></p> <p>Amount of <u>KCl</u> : <u>4</u> (g)</p> <p>Volume of Milli-Q Water: <u>1000</u> (mL)</p> <p>Date Prepared: <u>8/3/10</u></p> <p>Batch Number: <u>K02010-1</u></p>	<p><b>TEST CONDITIONS</b></p> <p>( ) Range Finder (X) Static</p> <p>( ) Screening ( ) Flow - Through</p> <p>(X) Definitive</p>
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<p><b>PROTOCOL/GUIDELINES FOLLOWED:</b></p> <p>EPA-821-R-02-012</p>	<p>Lighting: (X) Fluorescent ( ) Incandescent</p> <p>Photoperiod: 16 Hours Light 8 Hours Dark</p>
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<p>Test Concentration Based On: ( ) Active Ingredient (X) Whole Material</p>	<p>Test Duration: 48 hrs.</p> <p>Test Area Used: Water Bath</p> <p>Test Temperature: 12 ± 1°C</p> <p>Test Salinity: NA</p> <p>Replicates per Concentration: 1</p> <p>Organisms per Replicate: 10</p>
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<p><b>TEST ORGANISM HISTORY</b></p> <p>Test Species: <i>Oncorhynchus mykiss</i></p> <p>Lot Number: <u>2010-42</u></p> <p>Age/Life Stage: <u>15 days</u></p> <p>Date Acclimation/Maintenance Began: <u>8/3/10</u></p> <p>See Page <u>6</u> of Organism Receipt Log for Data.</p> <p>Mortality (%) During 48 hours Prior to Test Initiation: <u>&lt;10%</u></p>	<p>Dilution Water: <u>DDP 8/3/10 MHW013010</u></p> <p><b>TEST VESSELS</b></p> <p>Dimensions (in): 20 length x10 width x12 height</p> <p>Test Solution Height (in):</p> <p>Test Vessels: ( ) Open (X) Covered</p> <p>Test Container Volume: 8 L</p> <p>Test Solution Volume: 4 L</p>
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**TEST SOLUTION PREPARATION**

Test Concentration (Units: mg/L)	Control	250	500	1500	2500	4000		
Volume of Dilution Water Added (L)	8	7.5	7	6.5	5.5	0		
Volume of Effluent/ Stock Added (L)	0	0.5	1	1.5	2.5	8		

ADDITIONAL OBSERVATIONS: ~~.25 .5 .75 1.05 4.0 DDP 8/3/10~~

DATA BY: DDP

**REFERENCE TOXICANT DATA SHEET**

SPONSOR: MACTEC PROJECT NUMBER: REF TOX SPECIES: *O.mykiss*

REF TOXICANT:

DAY 0		DATE: 8/3/10		TIME: 1000		ANALYST: KDB		
NOMINAL CONCENTRATION (mg/L)	# ALIVE <sup>8/20/10</sup>		OBSERVATION <sup>8/24/10</sup>		FS12	Ref14	DO17	PH1
	A	B	A	B	TEMP (°C)	SALINITY (ppt)	DO (mg/L)	pH
CONTROL	10		loaded		12.7	1.0	9.1	8.1
250					12.4	1.0	9.0	8.0
500					12.8	2.0	9.0	7.9
1500					12.9	2.0	9.0	7.9
2500					12.8	3.0	9.0	8.0
4000					12.9	5.0	9.1	7.8

DAY 1		DATE: 8/4/10		TIME: 1615		ANALYST: CMM		
NOMINAL CONCENTRATION (mg/L)	# ALIVE <sup>8/20/10</sup>		OBSERVATION <sup>8/24/10</sup>		FS12	Ref14	DO17	PH1
	A	B	A	B	TEMP (°C)	SALINITY (ppt)	DO (mg/L)	pH
CONTROL	10		N		12.0	1.0	9.0	7.7
250	10		N		12.1	2.0	8.6	7.7
500	10		N		12.4	2.0	8.7	7.7
1500	10		N		12.6	3.0	9.0	7.7
2500	10		N		12.5	4.0	8.9	7.7
4000	0		10DD		12.2	6.0	9.0	7.6

DAY 2		DATE: 8/5/10		TIME:		ANALYST: VLE		
NOMINAL CONCENTRATION (mg/L)	# ALIVE <sup>8/26/10</sup>		OBSERVATION <sup>8/26/10</sup>		FS12	Ref14	DO17	PH1
	A	B	A	B	TEMP (°C)	SALINITY (ppt)	DO (mg/L)	pH
CONTROL	10		N		12.0	2.0	9.6	7.9
250	10		N		11.9	3.0	9.5	7.8
500	10		N		12.2	4.0	9.8	7.8
1500	10		N		12.5	5.0	9.1	7.8
2500	10		N		12.3	5.0	9.1	7.7
4000								

*J*

REFERENCE TOXICANT DATA SHEET		
SPONSOR: KEMIRA	PROJECT NUMBER: REF TOX	SPECIES: O.mykiss
REF TOXICANT: <i>KEL</i>		

SUMMARY			
NOMINAL CONCENTRATION	NUMBER DEAD		TOTAL DEAD PER CONC.
	A	B	
CONTROL	<i>0</i>	<i>0</i>	<i>0</i>
250	<i>0</i>	<i>0</i>	<i>0</i>
500	<i>0</i>	<i>0</i>	<i>0</i>
1500	<i>0</i>	<i>0</i>	<i>0</i>
2500	<i>0</i>		<i>0</i>
4000	<i>10</i>		<i>10</i>

*DD*

COMMENTS:

*J*

**CETIS Summary Report**

*Ref Tox  
O. mykiss*

Report Date: 25 Aug-10 14:18 (p 1 of 1)

Test Code: 08-5771-9813/RT96HOM2100803

**Fish 96-h Acute Survival Test**

**MACTEC Biology-Toxicology LAB**

<b>Batch ID:</b> 16-6227-5614	<b>Test Type:</b> Survival (96h)	<b>Analyst:</b>
<b>Start Date:</b> 03 Aug-10	<b>Protocol:</b> EPA/821/R-02-012 (2002)	<b>Diluent:</b> Mod-Hard Synthetic Water
<b>Ending Date:</b> 07 Aug-10	<b>Species:</b> Oncorhynchus mykiss	<b>Brine:</b> Not Applicable
<b>Duration:</b> 96h	<b>Source:</b> Thomas Fish Co.	<b>Age:</b>
<b>Sample ID:</b> 09-6123-3552	<b>Code:</b> 394B4290	<b>Client:</b> ADMIN
<b>Sample Date:</b> 03 Aug-10	<b>Material:</b> Potassium chloride	<b>Project:</b> ADMIN
<b>Receive Date:</b> 03 Aug-10	<b>Source:</b> Reference Toxicant	
<b>Sample Age:</b> N/A	<b>Station:</b>	

**Point Estimate Summary**

Analysis ID	Endpoint	Level	gm/L	95% LCL	95% UCL	TU	Method
13-5852-1378	96h Survival Rate	LC50	3162 ✓	2726	3669		Binomial/Graphical

**Test Acceptability**

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits	Overlap	Decision
13-5852-1378	96h Survival Rate	Control Resp	1	0.9 - NL	Yes	Result Within Limits

**96h Survival Rate Summary**

Conc-gm/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Dilution Water	1	1			1	1	0	0	0.0%	0.0%
250		1	1			1	1	0	0	0.0%	0.0%
500		1	1			1	1	0	0	0.0%	0.0%
1500		1	1			1	1	0	0	0.0%	0.0%
2500		1	1			1	1	0	0	0.0%	0.0%
4000		1	0			0	0	0	0		100.0%

**96h Survival Rate Detail** ✓

Conc-gm/L	Control Type	Rep 1
0	Dilution Water	1
250		1
500		1
1500		1
2500		1
4000		0

### O mykiss Reference Toxicant Results

