

August 10, 2017

Biomonitoring Testing
for
Cabot

Control No. 214760-1

Prepared for:

Mr. Matt Bienvenu
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Prepared by:

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McClelland Consulting Engineers, Inc.
ATTN: Mr. Matt Bienvenu
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Re: Chronic 7-Day Renewal utilizing *Pimephales promelas* (Fathead minnow) and *Ceriodaphnia dubia*
Cabot
NPDES Permit No. AR0021661 AFIN:43-00059

Dear Mr. Matt Bienvenu:

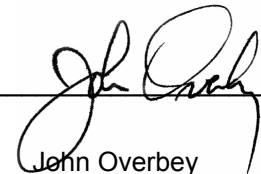
This report is the analytical results and supporting information for the samples submitted to American Interplex Corporation (AIC). The following results are applicable only to the sample identified by the control number referenced above. Accurate assessment of the data requires access to the entire document. Each section of the report has been reviewed and approved by the Chief Operating Officer or qualified designee.

Testing procedures and Quality Assurance were in accordance with "Short Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms" EPA-821-R-02-013, Fourth Edition, October 2002. Test results are summarized below:

Method 1000.0 Chronic *Pimephales promelas* (Fathead minnow) Survival and Growth Test: The No Observable Effects Concentration (NOEC) for survival occurred at 100 % effluent, which is equal to the critical dilution of 100 %. The percent minimum significant difference (PMSD) was below the limit of 12. Following additional calculations provided in the EPA document "Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications under the National Pollutant Discharge Elimination Systems Program", the NOEC for sublethal effects was calculated to be 100 %. **The sample, therefore, PASSED both lethal and sub-lethal effects for the Fathead minnow test.**

Chronic *Ceriodaphnia dubia* Survival and Reproduction test: Due to a high coefficient of variance in the critical dilution, the test is invalid and will need to be repeated. The data is attached for your review.

AMERICAN INTERPLEX CORPORATION



John Overbey
Chief Operating Officer

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I. Control Acceptance Criteria

Pimephales promelas (Fathead minnow) Method 1000.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Growth > or = 0.25 mg per Surviving minnow	0.480	PASS
Control Growth CV < or = 40%	6.46	PASS
Growth Minimum Significant Difference 12 to 30%	9.45	BELOW
Critical Dilution CV < or = 40%	5.61	PASS

Ceriodaphnia dubia Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	25.7	PASS
Control CV < or = 40% per Surviving Female	18.7	PASS
Reproduction Minimum Significant Difference 13 to 47%	30.3	PASS
Critical Dilution CV < or = 40%	48.4	FAIL

II. Outlined Report

A. Introduction

1. Permit Number: AR0021661 AFIN:43-00059
2. Test Requirements: Test Methods 1000.0 and 1002.0

B. Source of Effluent/Dilution Water:

1. Effluent Samples:
 - a. Sampling Point: Cabot
 - b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.0	7.6	8.6
pH (standard units)	7.7	7.5	7.7
Alkalinity (mg/l as CaCO ₃)	79	100	96
Hardness (mg/l as CaCO ₃)	96	95	100
Conductivity (umhos/cm)	450	440	440
Residual Chlorine (mg/l)	<0.05	<0.05	<0.05
Ammonia as N (mg/l)	<0.1	<0.1	<0.1

2. Dilution Water Samples:
Moderately Hard

Analysis	214582
Dissolved oxygen (mg/l)	7.9
pH (standard units)	7.9
Alkalinity (mg/l as CaCO ₃)	59
Hardness (mg/l as CaCO ₃)	91
Conductivity (umhos/cm)	320
Residual Chlorine (mg/l)	<0.05

C. Test Methods

1. Test methods used:

Short Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, EPA-821-R-02-013; test Methods 1000.0 and 1002.0, Fathead Minnow Survival and Growth and *Ceriodaphnia dubia* Survival and Reproduction.

2. Endpoint: No Observable Effects Concentration (NOEC)

3. Test Conditions:

Pimephales promelas (Fathead minnow) Survival and Growth Method 1000.0

Date & Time Test Initiated: July 25, 2017 at 1315
Date & Time Test Terminated: Aug 1, 2017 at 0830
Type & Volume of Test Chamber: 500 ml disposable beaker
Volume of Sample: 250 ml
Number of Organisms per replicate: 8
Number of Replicates per dilution: 5

Ceriodaphnia dubia Survival and Reproduction Method 1002.0

Date & Time Test Initiated: July 25, 2017 at 1350
Date & Time Test Terminated: Aug 1, 2017 at 1315
Type & Volume of Test Chamber: 30 ml disposable beaker
Volume of Sample: 15 ml
Number of Organisms per replicate: 1
Number of Replicates per dilution: 10

4. Source of test organisms: Obtained from in-house cultures

5. Test Temperature: 25 +/- 1 degree Celsius

D. Test Organisms

1. Scientific Name

- a. Test 1000.0 *Pimephales promelas*
- b. Test 1002.0 *Ceriodaphnia dubia*

III. Data Analysis

The data was analyzed using American Interplex Corporation's Laboratory Information Management Software based on Toxstat and following EPA method criteria.

Pimephales promelas (Fathead minnow) survival data was transformed using the Arc Sine transformation. Normality and homogeneity of variance were checked using Shapiro-Wilk's. The survival data was then analyzed using Steel's Many-One Rank Test to determine the No Observable Effects Concentration (NOEC).

Fathead minnow growth data was analyzed for normality and homogeneity of variance using Shapiro-Wilk's and Bartlett's test. Dunnett's Test was used to determine the No Observable Effects Concentration (NOEC) for growth.

Ceriodaphnia dubia survival data was analyzed with Fisher's Exact Test. Reproduction data was analyzed using Kolmogorov's Test for Normality and Bartlett's test and analyzed with Dunnett's Test to determine the No Observable Effects Concentration (NOEC) for Reproduction.

IV. Standard Reference Toxicants

The sensitivity of the offspring is determined by performing a standard reference toxicant test monthly. Sodium chloride in synthetic moderately hard water is used as prescribed in EPA-821-R-02-013.

Pimephales promelas (Fathead minnow)

A chronic reference test was performed on July 3, 2017 at 1215 to July 10, 2017 at 1200

The results were as follows: (Control No. 214133-1.)

Survival LC-50: 5056 mg/l

Growth IC-25: 1701 mg/l

Growth PMSD: 16.2

Ceriodaphnia dubia

A chronic reference test was performed on July 3, 2017 at 1245 to July 10, 2017 at 1520

The results were as follows: (Control No. 214133-2.)

Survival LC-50: 1823 mg/l

Growth IC-25: 1114 mg/l

Growth PMSD: 14.6

V. Organism History

Pimephales promelas (Fathead minnow)

Date: July 25, 2017

Age: <24 hours

Source: In-house culture

Water: Moderately hard synthetic

Temperature: 25 deg.C

Ceriodaphnia dubia

Date: July 25, 2017

Age: <24 hours

Source: In-house culture

Water: Moderately hard synthetic

Temperature: 25 deg.C

VII. Results Summary *Pimephales promelas*, Fathead minnow Larval Survival and Growth Test -- Method 1000.0

Larvae are exposed in a static renewal system for seven days to different concentrations of effluent with dilution water. Test results are based on the survival and growth (weight) of the larvae.

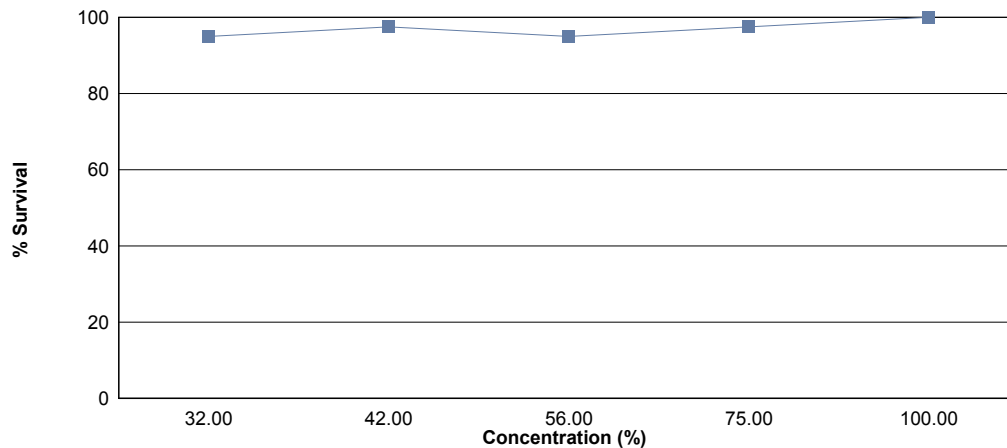
Effluent dilutions for this test were 32 %, 42 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

The low flow or 'critical' dilution is specified in the NPDES permit as 100 % effluent.

The test was initiated on July 25, 2017 at 1315 and continued through Aug 1, 2017 at 0830. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

- a.) NOEC survival = 100 % effluent
- b.) NOEC growth = 100 % effluent

(NOEC for sublethal effects was determined by Lower PMSD Bound Test.)



Summary of the 7-day Fathead Minnow Survival and Growth		
Concentration	Percent Survival	Mean Growth (mg)
Control	100	0.480
32 %	95.0	0.459
42 %	97.5	0.435
56 %	95.0	0.431 *
75 %	97.5	0.441
100 %	100	0.480

*Significant difference when compared to the control (p=0.05)

VII. Results Summary *Ceriodaphnia dubia*, Cladoceran Survival and Reproduction Test -- Method 1002.0

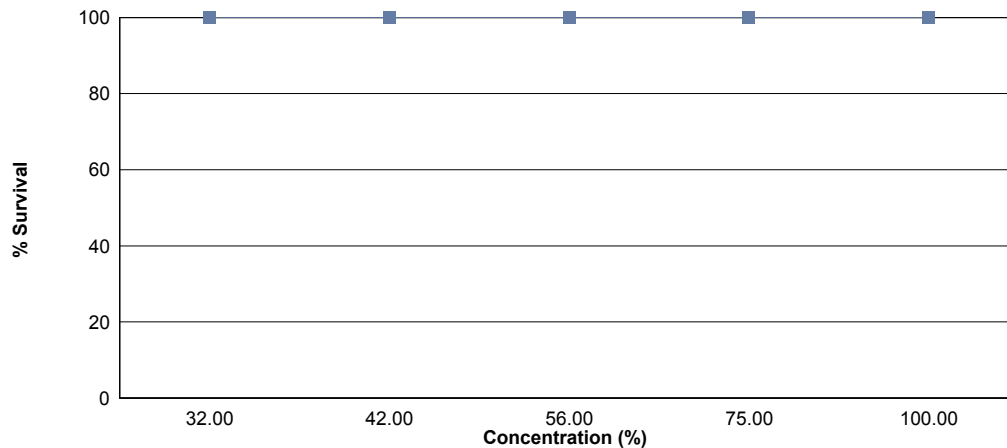
Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring or a maximum of eight test days.

Effluent dilutions for this test were 32 %, 42 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

The low flow or 'critical' dilution is specified in the NPDES permit as 100 % effluent.

The test was initiated on July 25, 2017 at 1350 and continued through Aug 1, 2017 at 1315. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

- a.) NOEC survival = 100 % effluent
- b.) NOEC reproduction = 32 % effluent



Concentration	Percent Survival	Mean Reproduction
Control	100	25.7
32 %	100	21.8
42 %	100	11.5 *
56 %	100	16.3 *
75 %	100	15.3 *
100 %	100	21.5

*Significant difference when compared to the control (p=0.05)

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Survival

Date and Time Test Initiated: July 25, 2017 at 1315

Date and Time Test Terminated: Aug 1, 2017 at 0830

Concentration	Replicate	Number of Survivors						
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
32 %	A	8	8	8	8	8	7	7
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	7	7
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
42 %	A	8	8	8	8	8	8	7
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
56 %	A	8	8	8	8	8	8	7
	B	8	8	8	8	8	8	7
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
75 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	7
	E	8	8	8	8	8	8	8
100 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Growth

Test Initiated: July 25, 2017 at 1315

Test Terminated: Aug 1, 2017 at 0830

Concentration	Replicate	Weight of pan	Weight of pan + fish	Total weight of fish (g)	Original # of fish	Mean dry weight (mg)
Control	A	.93150	.93548	0.00398	8	0.498
	B	.93593	.93946	0.00353	8	0.441
	C	.93691	.94107	0.00416	8	0.520
	D	.93731	.94099	0.00368	8	0.460
	E	.93900	.94285	0.00385	8	0.481
32 %	A	.93702	.94069	0.00367	8	0.459
	B	.93772	.94180	0.00408	8	0.510
	C	.93843	.94220	0.00377	8	0.471
	D	.93336	.93698	0.00362	8	0.452
	E	.93856	.94179	0.00323	8	0.404
42 %	A	.93365	.93722	0.00357	8	0.446
	B	.93377	.93706	0.00329	8	0.411
	C	.93423	.93770	0.00347	8	0.434
	D	.93324	.93663	0.00339	8	0.424
	E	.93510	.93879	0.00369	8	0.461
56 %	A	.94119	.94440	0.00321	8	0.401
	B	.93842	.94162	0.00320	8	0.400
	C	.94163	.94539	0.00376	8	0.470
	D	.93815	.94170	0.00355	8	0.444
	E	.93791	.94141	0.00350	8	0.438
75 %	A	.93929	.94292	0.00363	8	0.454
	B	.93895	.94238	0.00343	8	0.429
	C	.94064	.94385	0.00321	8	0.401
	D	.94061	.94406	0.00345	8	0.431
	E	.94382	.94775	0.00393	8	0.491
100 %	A	.93896	.94276	0.00380	8	0.475
	B	.94307	.94673	0.00366	8	0.458
	C	.94337	.94722	0.00385	8	0.481
	D	.94161	.94530	0.00369	8	0.461
	E	.94110	.94530	0.00420	8	0.525

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: July 25, 2017 at 1350

Date and Time Test Terminated: Aug 1, 2017 at 1315

Concentration: Control														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	3	4	4	3	2	4	4	6	6	0	36	10	3.60	
5	8	8	8	8	8	10	0	0	0	6	56	10	5.60	
6	11	0	16	0	13	11	7	12	11	13	94	10	9.40	
7	0	10	0	12	0	0	8	16	13	12	71	10	7.10	
8														
TOTAL	22	22	28	23	23	25	19	34	30	31	257	10	25.7	

Concentration: 32 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	4	4	5	4	2	4	3	3	2	2	33	10	3.30	
5	8	7	6	8	8	10	6	2	0	11	66	10	6.60	
6	1	0	7	0	11	14	0	0	11	13	57	10	5.70	
7	2	12	0	14	1	2	13	5	13	0	62	10	6.20	
8														
TOTAL	15	23	18	26	22	30	22	10	26	26	218	10	21.8	

Concentration: 42 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	0	0	4	4	2	3	6	0	5	0	24	10	2.40	
5	7	8	2	2	1	0	0	0	0	0	20	10	2.00	
6	7	1	11	0	2	1	12	0	1	1	36	10	3.60	
7	0	6	0	7	0	0	15	0	1	6	35	10	3.50	
8														
TOTAL	14	15	17	13	5	4	33	0	7	7	115	10	11.5	

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: July 25, 2017 at 1350

Date and Time Test Terminated: Aug 1, 2017 at 1315

Concentration: 56 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	1	1	4	3	0	2	3	4	0	6	24	10	2.40	
5	10	8	2	3	0	0	0	8	0	0	31	10	3.10	
6	0	0	5	0	10	6	4	3	5	9	42	10	4.20	
7	10	8	1	4	13	10	4	0	4	12	66	10	6.60	
8														
TOTAL	21	17	12	10	23	18	11	15	9	27	163	10	16.3	

Concentration: 75 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	4	2	4	3	6	4	4	1	8	3	39	10	3.90	
5	2	6	2	8	0	0	8	4	0	3	33	10	3.30	
6	0	0	5	0	8	9	3	2	14	9	50	10	5.00	
7	2	6	0	12	0	2	0	0	9	0	31	10	3.10	
8														
TOTAL	8	14	11	23	14	15	15	7	31	15	153	10	15.3	

Concentration: 100 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	0	0	1	0	0	5	0	2	0	0	8	10	0.800	
5	1	6	1	3	0	0	0	0	0	1	12	10	1.20	
6	0	0	3	0	12	12	13	8	11	17	76	10	7.60	
7	12	16	0	17	15	20	21	0	18	0	119	10	11.9	
8														
TOTAL	13	22	5	20	27	37	34	10	29	18	215	10	21.5	

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Survival

Transformation of Data			Transform: Arc Sin(Square Root(Y))	
Group	Identification	Rep	Value	Transformed
1	Control	1	1.00000	1.39310
1	Control	2	1.00000	1.39310
1	Control	3	1.00000	1.39310
1	Control	4	1.00000	1.39310
1	Control	5	1.00000	1.39310
2	32 %	1	0.87500	1.20940
2	32 %	2	1.00000	1.39310
2	32 %	3	0.87500	1.20940
2	32 %	4	1.00000	1.39310
2	32 %	5	1.00000	1.39310
3	42 %	1	0.87500	1.20940
3	42 %	2	1.00000	1.39310
3	42 %	3	1.00000	1.39310
3	42 %	4	1.00000	1.39310
3	42 %	5	1.00000	1.39310
4	56 %	1	0.87500	1.20940
4	56 %	2	0.87500	1.20940
4	56 %	3	1.00000	1.39310
4	56 %	4	1.00000	1.39310
4	56 %	5	1.00000	1.39310
5	75 %	1	1.00000	1.39310
5	75 %	2	1.00000	1.39310
5	75 %	3	1.00000	1.39310
5	75 %	4	0.87500	1.20940
5	75 %	5	1.00000	1.39310
6	100 %	1	1.00000	1.39310
6	100 %	2	1.00000	1.39310
6	100 %	3	1.00000	1.39310
6	100 %	4	1.00000	1.39310
6	100 %	5	1.00000	1.39310

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Survival

Shapiro - Wilk's Test for Normality		Transform: Arc Sin(Square Root(Y))
<p>D = 0.135 W = 0.8143 Critical W = 0.9 (alpha = 0.01, N = 30) Critical W = 0.927 (alpha = 0.05, N = 30)</p> <p>Data FAIL normality test (alpha = 0.01).</p>		

Steel's Many-One Rank Test				Transform: Arc Sin(Square Root(Y))	
Ho:Control<Treatment					
Group	Identification	Rank Sum	Critical Value	DF	Sig 0.05
1	Control				
2	32 %	22.50	16.00	5.00	
3	42 %	25.00	16.00	5.00	
4	56 %	22.50	16.00	5.00	
5	75 %	25.00	16.00	5.00	
6	100 %	27.50	16.00	5.00	
Critical values are 1 tailed (k=5)					

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Growth

Shapiro - Wilk's Test for Normality	No Transformation
<p>D = 0.02217 W = 0.9712 Critical W = 0.9 (alpha = 0.01, N = 30) Critical W = 0.927 (alpha = 0.05, N = 30)</p> <p>Data PASS normality test (alpha = 0.01).</p>	

Bartlett's Test for Homogeneity of Variance	No Transformation
<p>Calculated B1 statistic = 1.750 Critical B = 15.086 (alpha = 0.01, df = 5)</p> <p>Data PASS B1 homogeneity test at 0.01 level.</p>	

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Growth

ANOVA Table				No Transformation	
SOURCE	DF	SS	MS	F	
Between	5	0.01222	0.002444	2.647	
Within (Error)	24	0.02216	0.0009233		
Total	29	0.03438			
Critical F = 3.9 (alpha = 0.01, df = 5,24) 2.62 (alpha = 0.05, df = 5,24)					
Since F > Critical F REJECT Ho: All equal (alpha = 0.05)					

Dunnett's Test - Table 1 of 2					No Transformation	
Ho:Control<Treatment						
Group	Identification	Transformed Mean	Mean In Original Units	T Stat	Sig 0.05	
1	Control	0.48	0.48			
2	32 %	0.4592	0.4592	1.082		
3	42 %	0.4352	0.4352	2.331		
4	56 %	0.4306	0.4306	2.571	*	
5	75 %	0.4412	0.4412	2.019		
6	100 %	0.48	0.48	0		
Dunnett's critical value = 2.36 (1 Tailed, alpha = 0.05, df = 5,24)						

Dunnett's Test - Table 2 of 2					No Transformation	
Ho:Control<Treatment						
Group	Identification	Num of Reps	Min Sig Diff (In Orig. Units)	% of Control	Difference From Control	
1	Control	5				
2	32 %	5	0.04535	9.45	0.0208	
3	42 %	5	0.04535	9.45	0.0448	
4	56 %	5	0.04535	9.45	0.0494	
5	75 %	5	0.04535	9.45	0.0388	
6	100 %	5	0.04535	9.45	0	

Appendix A2: Statistics

Ceriodaphnia dubia Survival

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
32 %	10	0	10
Total	20	0	20

Critical Fisher's value (10,10,10) (alpha=0.05) is 6. b value is 10. Since b is greater than 6 there is NO SIGNIFICANT DIFFERENCE between CONTROL and TREATMENT at the 0.05 level.

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
42 %	10	0	10
Total	20	0	20

Critical Fisher's value (10,10,10) (alpha=0.05) is 6. b value is 10. Since b is greater than 6 there is NO SIGNIFICANT DIFFERENCE between CONTROL and TREATMENT at the 0.05 level.

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
56 %	10	0	10
Total	20	0	20

Critical Fisher's value (10,10,10) (alpha=0.05) is 6. b value is 10. Since b is greater than 6 there is NO SIGNIFICANT DIFFERENCE between CONTROL and TREATMENT at the 0.05 level.

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
75 %	10	0	10
Total	20	0	20

Critical Fisher's value (10,10,10) (alpha=0.05) is 6. b value is 10. Since b is greater than 6 there is NO SIGNIFICANT DIFFERENCE between CONTROL and TREATMENT at the 0.05 level.

Appendix A2: Statistics

Ceriodaphnia dubia Survival

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
100 %	10	0	10
Total	20	0	20

Critical Fisher's value (10,10,10) (alpha=0.05) is 6. b value is 10. Since b is greater than 6 there is NO SIGNIFICANT DIFFERENCE between CONTROL and TREATMENT at the 0.05 level.

Summary of Fisher's Exact Test				
Group	Identification	Exposed	Dead	Sig 0.05
0	Control	10	0	
1	32 %	10	0	
2	42 %	10	0	
3	56 %	10	0	
4	75 %	10	0	
5	100 %	10	0	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

ANOVA Table				No Transformation	
SOURCE	DF	SS	MS	F	
Between	5	1356	271.2	4.778	
Within (Error)	54	3065	56.76		
Total	59	4421			
Critical F = 3.38 (alpha = 0.01, df = 5,54)					
2.38 (alpha = 0.05, df = 5,54)					
Since F > Critical F REJECT Ho: All equal (alpha = 0.05)					

Dunnett's Test - Table 1 of 2					No Transformation	
Ho:Control<Treatment						
Group	Identification	Transformed Mean	Mean In Original Units	T Stat	Sig 0.05	
1	Control	25.7	25.7			
2	32 %	21.8	21.8	1.158		
3	42 %	11.5	11.5	4.215	*	
4	56 %	16.3	16.3	2.79	*	
5	75 %	15.3	15.3	3.087	*	
6	100 %	21.5	21.5	1.247		
Dunnett's critical value = 2.31 (1 Tailed, alpha = 0.05, df [used] = 5,40) (Actual df = 5,54)						

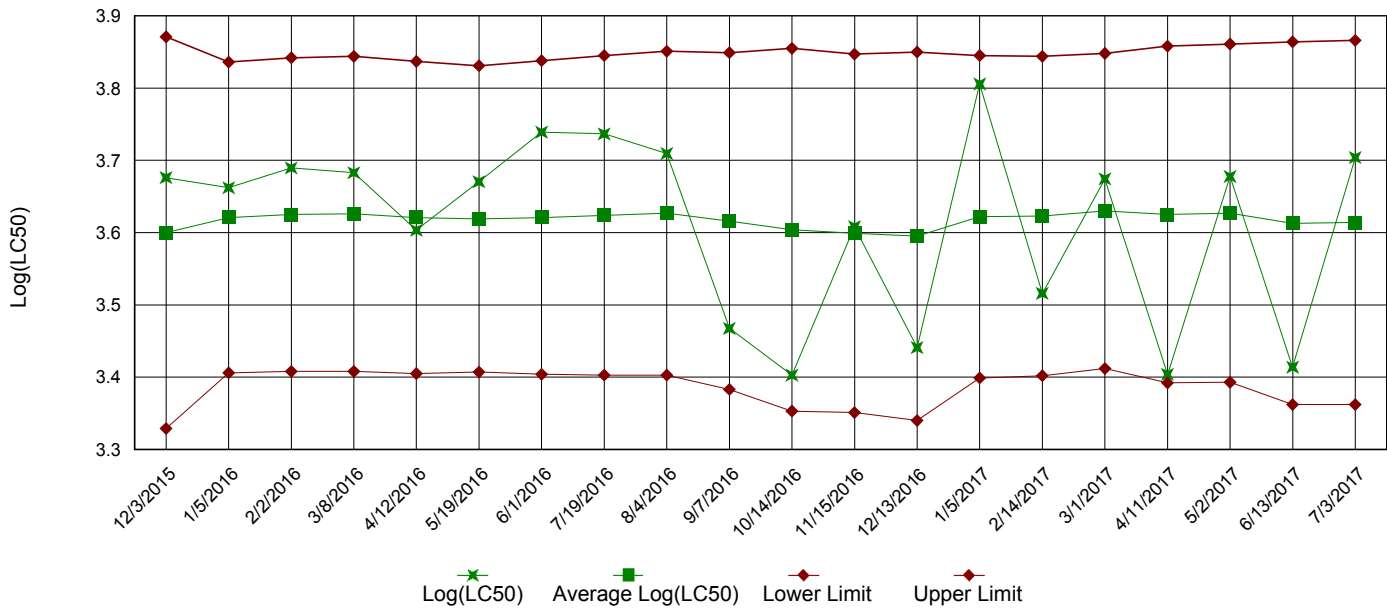
Dunnett's Test - Table 2 of 2					No Transformation	
Ho:Control<Treatment						
Group	Identification	Num of Reps	Min Sig Diff (In Orig. Units)	% of Control	Difference From Control	
1	Control	10				
2	32 %	10	7.783	30.3	3.9	
3	42 %	10	7.783	30.3	14.2	
4	56 %	10	7.783	30.3	9.4	
5	75 %	10	7.783	30.3	10.4	
6	100 %	10	7.783	30.3	4.2	

Lower PMSD Bound Test			
Concentration	Growth	Relative Difference from Control	Pass/Fail
Control	0.480	-	
32 %	0.459	4.4	PASS
42 %	0.435	9.4	PASS
56 %	0.431	10	PASS
75 %	0.441	8.1	PASS
100 %	0.480	0.0	PASS
		Limit = 12	
		NOEC = 100 %	
		LOEC = 100 %	

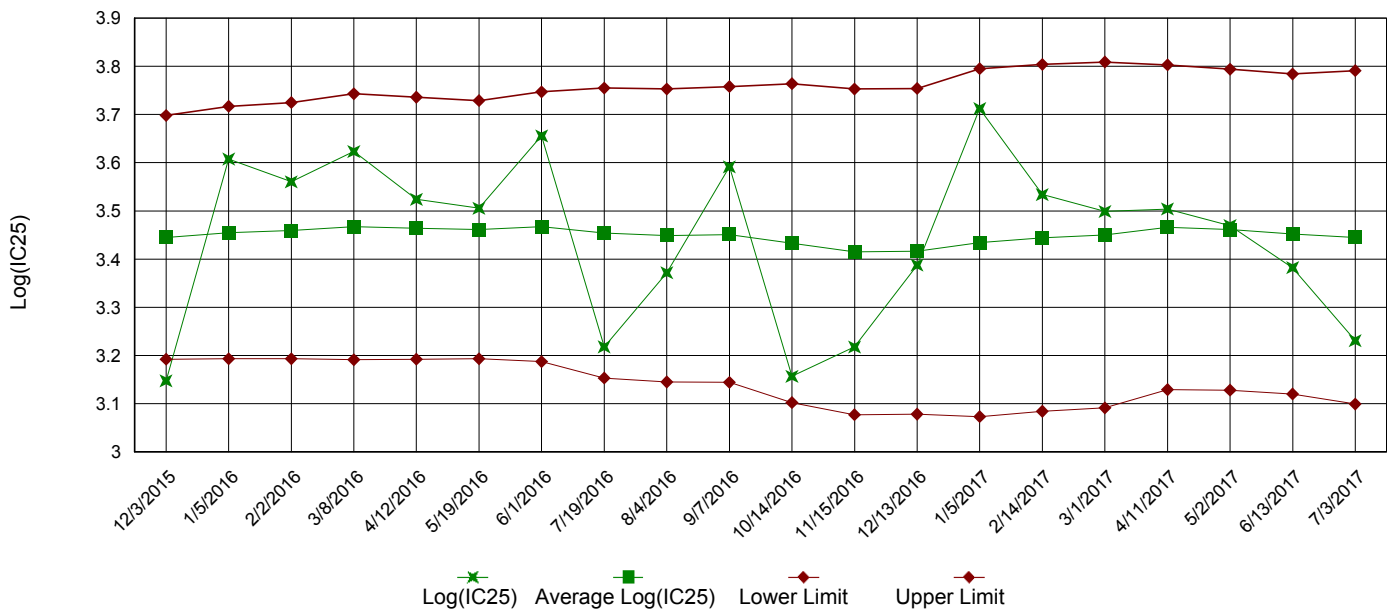
Appendix A3: Test 1000.0

Chronic Reference Toxicant, *Pimephales promelas* (Fathead Minnow)

LC50 Survival Data

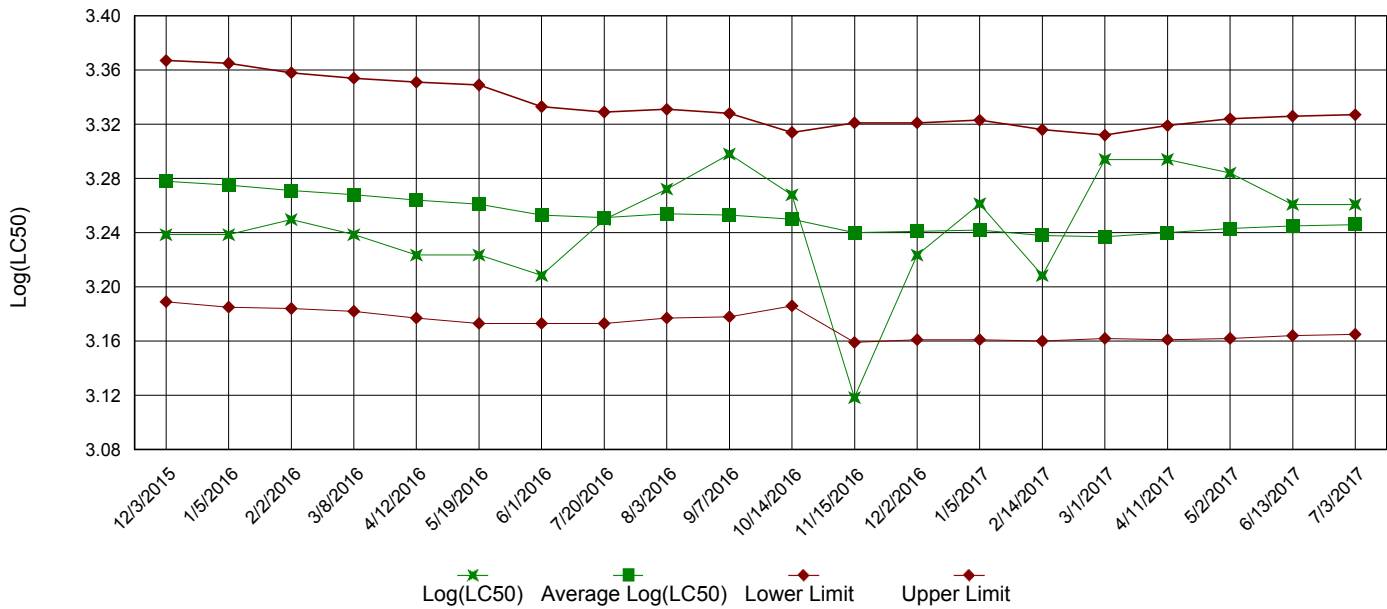


IC25 Growth Data

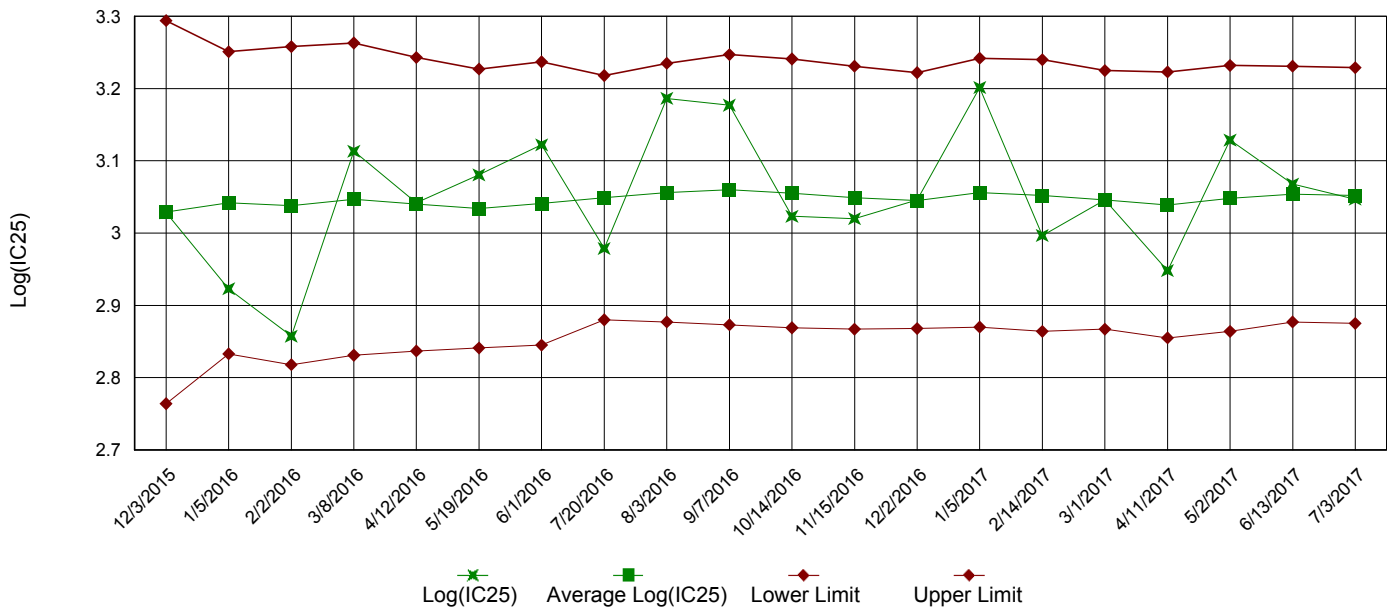


Appendix A3: Test 1002.0
Chronic Reference Toxicant, *Ceriodaphnia dubia*

LC50 Survival Data



IC25 Reproduction Data



Appendix B: Test 1000.0
SUMMARY REPORTING FORMS
CHRONIC BIOMONITORING
Pimephales promelas (Fathead Minnow)
SURVIVAL AND GROWTH

Permittee: McClelland Consulting Engineers, Inc.

NPDES No.: AR0021661 AFIN:43-00059

Date and Time Test Initiated: July 25, 2017 at 1315

Date and Time Test Terminated: Aug 1, 2017 at 0830

Dilution water used: Moderately Hard

DATA TABLE FOR SURVIVAL

Effluent Conc. %	Percent Survival in replicate chambers					Mean percent survival			CV%
	A	B	C	D	E	24 hr	48 hr	7 days	
Control	100	100	100	100	100	100	100	100	0.00
32 %	87.5	100	87.5	100	100	100	100	95.0	7.21
42 %	87.5	100	100	100	100	100	100	97.5	5.73
56 %	87.5	87.5	100	100	100	100	100	95.0	7.21
75 %	100	100	100	87.5	100	100	100	97.5	5.73
100 %	100	100	100	100	100	100	100	100	0.00

DATA TABLE FOR GROWTH

Effluent Conc. %	Average dry weight, mg replicate chambers					Mean dry weight, mg	CV%
	A	B	C	D	E		
Control	0.498	0.441	0.520	0.460	0.481	0.48	6.46
32 %	0.459	0.510	0.471	0.452	0.404	0.459	8.31
42 %	0.446	0.411	0.434	0.424	0.461	0.435	4.44
56 %	0.401	0.400	0.470	0.444	0.438	0.431	6.97
75 %	0.454	0.429	0.401	0.431	0.491	0.441	7.61
100 %	0.475	0.458	0.481	0.461	0.525	0.48	5.61

CV = Coefficient of variation = standard deviation * 100 / mean

Appendix B: Test 1000.0
SUMMARY REPORTING FORMS
CHRONIC BIOMONITORING
Pimephales promelas (Fathead Minnow)
SURVIVAL AND GROWTH

1. Steel's Many-One Rank Test:

Is the mean survival significantly different ($p=0.05$) than the control survival for the % effluent corresponding to (lethality):

a.) LOW FLOW OR CRITICAL DILUTION	(100 %)	<u> </u> YES	<u> X </u> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<u> </u> YES	<u> </u> NO

2. Dunnett's Test:

Is the mean dry weight (growth) significantly different ($p=0.05$) than the control's dry weight (growth) for the % effluent corresponding to (significant non-lethal effects):

a.) LOW FLOW OR CRITICAL DILUTION	(100 %)	<u> </u> YES	<u> X </u> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<u> </u> YES	<u> </u> NO

3. If you answered NO to 1.a) enter [0] otherwise enter [1]: 0 (TLP6C)
4. If you answered NO to 2.a) enter [0] otherwise enter [1]: 0 (TGP6C)
5. NOEC *Pimephales* Lethality: 100 % (TOP6C)
6. LOEC *Pimephales* Lethality: 100 % (TXP6C)
7. NOEC *Pimephales* Sublethality: 100 % (TPP6C)
8. LOEC *Pimephales* Sublethality: 100 % (TYP6C)
9. Coefficient of variation for *Pimephales* growth: 6.46 (TQP6C)

Appendix B: Test 1000.0
CHRONIC TOXICITY SUMMARY FORM
Pimephales promelas (Fathead minnow)
CHEMICAL PARAMETERS CHART

PERMITTEE: McClelland Consulting Engineers,
NPDES NO.: AR0021661 AFIN:43-00059
CONTACT: Mr. Matt Bienvenu
ANALYST: 280, 310, 322

Test Initiated: DATE: July 25, 2017 TIME: 1315
Test Terminated: DATE: Aug 1, 2017 TIME: 0830

DILUTION	DAY						
	1	2	3	4	5	6	7
Control							
D.O. Initial	7.9	7.9	7.8	7.9	7.6	7.5	8.0
Final	8.0	7.4	7.4	7.3	8.0	7.4	6.7
pH Initial	7.9	7.9	7.9	7.8	8.1	8.1	7.6
Final	7.5	7.9	7.3	7.8	8.1	7.6	7.2

DILUTION	DAY						
	1	2	3	4	5	6	7
32 %							
D.O. Initial	7.8	7.8	8.1	8.0	8.1	7.7	8.1
Final	7.8	6.8	7.1	7.1	7.7	7.6	6.5
pH Initial	7.8	7.9	7.8	7.8	7.9	8.2	7.6
Final	7.6	7.7	7.4	7.8	8.2	7.7	7.3

DILUTION	DAY						
	1	2	3	4	5	6	7
42 %							
D.O. Initial	8.1	7.6	7.9	8.0	8.2	8.0	8.0
Final	8.0	6.9	7.6	7.2	8.1	7.4	6.7
pH Initial	7.9	7.9	7.7	7.7	7.9	8.2	7.6
Final	7.7	7.8	7.6	7.8	8.2	7.7	7.4

DILUTION	DAY						
	1	2	3	4	5	6	7
56 %							
D.O. Initial	7.9	7.9	8.1	7.6	8.2	8.1	7.9
Final	7.8	7.2	7.8	7.4	7.9	7.3	7.4
pH Initial	7.8	7.8	7.7	7.7	7.8	8.2	7.6
Final	7.7	7.9	7.6	7.9	8.2	7.8	7.6

DILUTION	DAY						
	1	2	3	4	5	6	7
75 %							
D.O. Initial	7.9	7.8	7.8	7.8	8.2	8.0	8.0
Final	8.0	6.9	7.5	7.3	8.0	7.6	7.6
pH Initial	7.8	7.8	7.6	7.7	7.8	8.3	7.5
Final	7.8	7.8	7.7	7.9	8.2	7.9	7.7

DILUTION	DAY						
	1	2	3	4	5	6	7
100 %							
D.O. Initial	8.0	7.7	7.6	7.9	8.6	7.9	7.8
Final	8.0	7.1	7.4	7.4	7.7	7.7	6.8
pH Initial	7.7	7.7	7.5	7.6	7.7	8.3	7.6
Final	7.8	7.9	7.7	8.0	8.3	8.0	7.6

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
79	96	450	<0.05	Cabot Bio 24-JUL-17
100	95	440	<0.05	Cabot Bio 26-JUL-17
96	100	440	<0.05	Cabot Bio 28-JUL-17

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
59	91	320	<0.05	214582

Appendix B: Test 1002.0
SUMMARY REPORTING FORMS
CHRONIC BIOMONITORING
Ceriodaphnia dubia
SURVIVAL AND REPRODUCTION

Permittee: McClelland Consulting Engineers, Inc.

NPDES No.: AR0021661 AFIN:43-00059

Date and Time Test Initiated: July 25, 2017 at 1350

Date and Time Test Terminated: Aug 1, 2017 at 1315

Dilution water used: Moderately Hard

PERCENT SURVIVAL

Time of Reading	Control	Percent Effluent				
		32 %	42 %	56 %	75 %	100 %
24 hour	100	100	100	100	100	100
48 hour	100	100	100	100	100	100
7 day	100	100	100	100	100	100

NUMBER OF YOUNG PRODUCED PER FEMALE @ 7 DAYS

Replicates	Control	Percent Effluent				
		32 %	42 %	56 %	75 %	100 %
A	22	15	14	21	8	13
B	22	23	15	17	14	22
C	28	18	17	12	11	5
D	23	26	13	10	23	20
E	23	22	5	23	14	27
F	25	30	4	18	15	37
G	19	22	33	11	15	34
H	34	10	0	15	7	10
I	30	26	7	9	31	29
J	31	26	7	27	15	18
Mean per Adult	25.7	21.8	11.5	16.3	15.3	21.5
Mean per Surviving Adult	25.7	21.8	11.5	16.3	15.3	21.5
CV %	18.7	27.4	81.2	36.9	46.2	48.4

CV = Coefficient of variation = standard deviation * 100 / mean
(calculated based on young produced by surviving females)

Appendix B: Test 1002.0
CHRONIC TOXICITY SUMMARY FORM
Ceriodaphnia dubia
CHEMICAL PARAMETERS CHART

PERMITTEE: McClelland Consulting Engineers,
NPDES NO.: AR0021661 AFIN:43-00059
CONTACT: Mr. Matt Bienvenu
ANALYST: 280, 310, 322

Test Initiated: DATE: July 25, 2017 TIME: 1350
Test Terminated: DATE: Aug 1, 2017 TIME: 1315

DILUTION	DAY						
	1	2	3	4	5	6	7
Control							
D.O. Initial	7.9	7.9	7.8	7.9	7.6	7.5	8.0
Final	7.8	8.4	8.2	7.3	8.0	8.0	7.9
pH Initial	7.9	7.9	7.9	7.8	8.1	8.1	7.6
Final	8.3	8.2	8.3	8.3	8.4	8.1	8.0

DILUTION	DAY						
	1	2	3	4	5	6	7
32 %							
D.O. Initial	7.8	7.8	8.1	8.0	8.1	7.7	8.1
Final	8.1	8.4	8.5	7.9	8.3	8.4	7.9
pH Initial	7.8	7.9	7.8	7.8	7.9	8.2	7.6
Final	8.5	8.4	8.6	8.6	8.7	8.5	8.3

DILUTION	DAY						
	1	2	3	4	5	6	7
42 %							
D.O. Initial	8.1	7.6	7.9	8.0	8.2	8.0	8.0
Final	8.0	8.9	8.4	8.4	8.2	8.4	8.0
pH Initial	7.9	7.9	7.7	7.7	7.9	8.2	7.6
Final	8.5	8.6	8.7	8.6	8.8	8.6	8.3

DILUTION	DAY						
	1	2	3	4	5	6	7
56 %							
D.O. Initial	7.9	7.9	8.1	7.6	8.2	8.1	7.9
Final	8.1	8.4	8.0	7.9	8.2	8.2	8.0
pH Initial	7.8	7.8	7.7	7.7	7.8	8.2	7.6
Final	8.5	8.5	8.6	8.6	8.7	8.5	8.4

DILUTION	DAY						
	1	2	3	4	5	6	7
75 %							
D.O. Initial	7.9	7.8	7.8	7.8	8.2	8.0	8.0
Final	8.2	8.8	8.4	8.1	8.4	8.0	8.2
pH Initial	7.8	7.8	7.6	7.7	7.8	8.3	7.5
Final	8.5	8.6	8.7	8.7	8.7	8.6	8.4

DILUTION	DAY						
	1	2	3	4	5	6	7
100 %							
D.O. Initial	8.0	7.7	7.6	7.9	8.6	7.9	7.8
Final	8.1	8.7	8.5	7.9	8.4	7.8	7.7
pH Initial	7.7	7.7	7.5	7.6	7.7	8.3	7.6
Final	8.5	8.5	8.6	8.7	8.7	8.6	8.4

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
79	96	450	<0.05	Cabot Bio 24-JUL-17
100	95	440	<0.05	Cabot Bio 26-JUL-17
96	100	440	<0.05	Cabot Bio 28-JUL-17

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
59	91	320	<0.05	214582



CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

PAGE 1 OF 1

Client: MCE		NO. OF BOTTLES: 3		ANALYSES REQUESTED:		AIC CONTROL NO: 214760	
Project Reference: MCE		MATRIX: WATER		V = VOA vials N = Nitric acid pH2		AIC PROPSAL NO:	
Project Manager: Tim Joyner		WATER		H = HCl to pH2 B = NaOH for pH2		Carrier: MCE	
Sampled By: James Johnson		SOIL		Relinquished By: James Johnson		Received Temperature: °C	
AIC No: 7/23/17		CDMP		Date/Time: 7/24/17		Remarks:	
Sample Identification: Cabot bio		G.R.A.B.		Date/Time: 7-24-17		Field pH calibration on: @	
Date/Time Collected: 7/23/17		Containers/Preservative: Plastic		Date/Time: 0950		Buffer:	
Date/Time: 8:20 AM 2015		NO = none G = Glass P = Plastic S = Sulphuric acid pH2		Date/Time: 0950		Y = Sodium Thiosulfate Z = Zinc acetate	
Turnaround Time Requested: (Please circle) NORMAL or EXPEDITED IN _____ DAYS		Relinquished By: James Johnson		Date/Time: 7/24/17		A = NH4SO4 NH4OH	
Expedited results requested by:		Relinquished By: JESSICA		Date/Time: 7-24-17		Received @ Lab	
Who should AIC contact with questions:		By: JESSICA		Date/Time: 7-24-17/235		Received by: D.K.	
Phone: _____ Fax: _____		Comments:					
Report Alterations to:							
Report Address to:							
Email Address:							

FORM 9069

9/2014



CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

PAGE: 2 OF 3

Client: MCE		NO. OF BOTTLES: 3		ANALYSES REQUESTED		AIC CONTROL NO.: 214760	
Project Reference: Tim Jayner		MATRIX: SOIL				AIC PROPOSAL NO.:	
Project Manager: James Johnson		WATER: <input checked="" type="checkbox"/>				Carrier:	
Sampled By: James Johnson		CDMP: <input checked="" type="checkbox"/>				Requested Temperature: 1.6	
AIC No.:		G.R.A.B.:				Remarks:	
Sample Identification: 2 Cabot		Date/Time Collected: 7/25-7/26/80				Field pH calibration on: @	
Container Type: Plastic		Preservative: Silicic acid pH2				Buffer:	
NO = Glass		P = Plastic				A = (NH ₄) ₂ SO ₄ NH ₄ OH	
NO = none		S = Silicic acid pH2				T = Sodium Thiosulfate	
		V = VOA vials				Z = Zinc acetate	
		N = Nitric acid pH2				Date/Time: 7-26-17 10:35	
		Relinquished By: James Johnson				Date/Time: 7-26-17 12:41	
		Relinquished By: Aboufah				Date/Time: 7-26-17 12:41	
		Comments:					

FORM 0063

9/2014

