July 30, 2020

Biomonitoring Testing
for
East Effluent

Control No. 247076-1

Prepared for:

Ms. Whitney Young
City Water & Light of Jonesboro
5205 Ingels Road
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Prepared by:

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City Water & Light of Jonesboro
ATTN: Ms. Whitney Young
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Jonesboro, AR 72401

Re: *Pimephales promelas* (Fathead minnow) and *Ceriodaphnia dubia*
East Effluent
NPDES Permit No. AR0043401 AFIN16-00936

Dear Ms. Whitney Young:

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 NOEC for growth occurred at 100 % effluent, which is equal to the critical dilution of 100 %. **The sample, therefore, PASSED both lethal and sub-lethal effects for the Fathead minnow test.**

Method 1002.0 Chronic *Ceriodaphnia dubia* Survival and Reproduction Test: Due to a control failure for reproduction, the test is invalid and will need to be repeated.

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	0.593	PASS
Control Growth CV < or = 40%	7.39	PASS
Growth Minimum Significant Difference 12 to 30%	16.5	PASS
Critical Dilution CV < or = 40%	11.2	PASS

Ceriodaphnia dubia Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	13.2	FAIL
Control CV < or = 40% per Surviving Female	22.0	PASS
Reproduction Minimum Significant Difference 13 to 47%	26.3	PASS
Critical Dilution CV < or = 40%	17.0	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0043401 AFIN16-00936
2. Test Requirements:
   Test Methods 1000.0 and 1002.0

B. Source of Effluent/Dilution Water:

1. Effluent Samples:
   a. Sampling Point: East Effluent
   b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	7.3	8.1	7.4
pH (standard units)	8.2	8.0	8.2
Alkalinity (mg/l as CaCO3)	160	160	160
Hardness (mg/l as CaCO3)	140	130	120
Conductivity (umhos/cm)	730	720	740
Residual Chlorine (mg/l)	<0.05	<0.05	<0.05
Ammonia as N (mg/l)	0.16	0.21	0.17

2. Dilution Water Samples:
   Moderately Hard

Analysis	246811-1
Dissolved oxygen (mg/l)	7.1
pH (standard units)	8.2
Alkalinity (mg/l as CaCO3)	61
Hardness (mg/l as CaCO3)	84
Conductivity (umhos/cm)	290
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 21, 2020 at 0924
Date & Time Test Terminated:	July 28, 2020 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 21, 2020 at 1025
Date & Time Test Terminated:	July 28, 2020 at 1045
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 June 02, 2020 at 1418 to June 09, 2020 at 1310
The results were as follows: (Control No. 245673-1.)

Survival LC-50: 3610.4 mg/l
Growth IC-25: 2364 mg/l
Growth PMSD: 0

Ceriodaphnia dubia

A chronic reference test was performed on June 02, 2020 at 1155 to June 08, 2020 at 1255
The results were as follows: (Control No. 245673-2.)

Survival LC-50: 1673.1 mg/l
Reproduction IC-25: 1091 mg/l
Reproduction PMSD: 23.3

V. Organism History

Pimephales promelas (Fathead minnow)

Date: July 21, 2020
Age: <24 hours
Source: In-house culture
Water: Moderately hard synthetic
Temperature: 25 deg.C

Ceriodaphnia dubia

Date: July 21, 2020
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

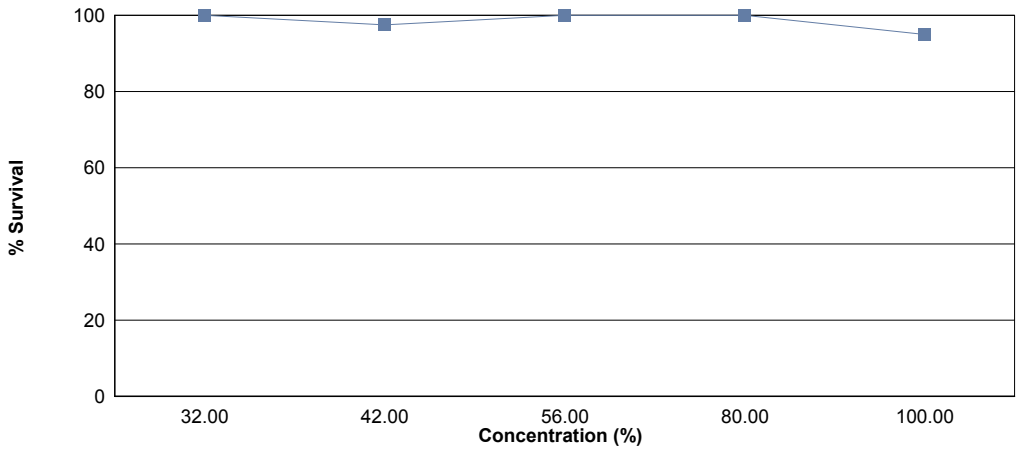
Larvae are exposed in a static renewal system for seven days to different concentrations of effluent with dilution water. Test results are based on the survival and growth (weight) of the larvae.

Effluent dilutions for this test were 32 %, 42 %, 56 %, 80 %, 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 21, 2020 at 0924 and continued through July 28, 2020 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



Summary of the 7-day Fathead Minnow Survival and Growth		
Concentration	Percent Survival	Mean Growth (mg)
Control	100	0.593
32 %	100	0.634
42 %	97.5	0.514
56 %	100	0.603
80 %	100	0.548
100 %	95.0	0.582

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

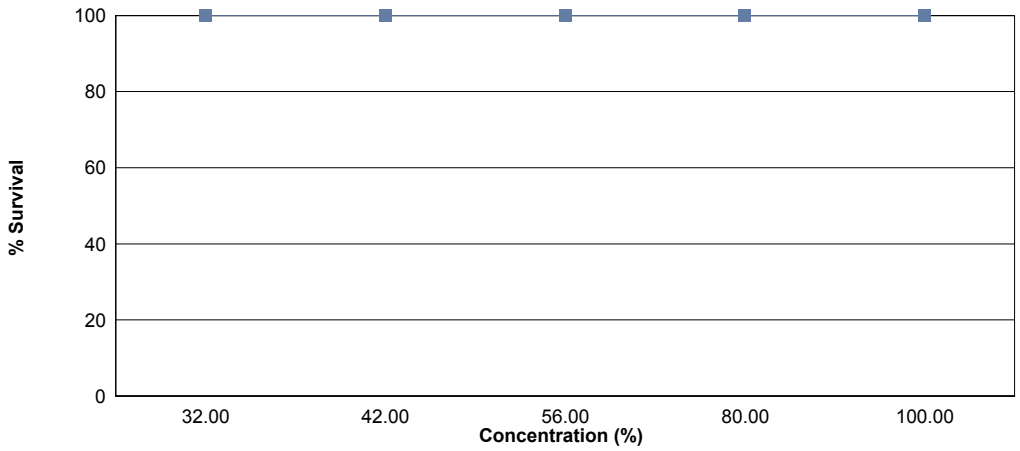
Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring or a maximum of eight test days.

Effluent dilutions for this test were 32 %, 42 %, 56 %, 80 %, 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 21, 2020 at 1025 and continued through July 28, 2020 at 1045. 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 = 100 % effluent



Summary of the 7-day *Ceriodaphnia dubia* Survival and Reproduction Data		
Concentration	Percent Survival	Mean Reproduction
Control	100	13.2
32 %	100	20.9
42 %	100	20.6
56 %	100	19.2
80 %	100	24.7
100 %	100	22.9

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Survival

Date and Time Test Initiated: July 21, 2020 at 0924
Date and Time Test Terminated: July 28, 2020 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	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
42 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	7
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
56 %	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
80 %	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
100 %	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	7	7	7
	E	8	8	8	8	8	8	8

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Growth

Test Initiated: July 21, 2020 at 0924
Test Terminated: July 28, 2020 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	.65622	.66100	0.00478	8	0.598
	B	.65677	.66183	0.00506	8	0.632
	C	.65545	.65974	0.00429	8	0.536
	D	.66265	.66774	0.00509	8	0.636
	E	.65829	.66278	0.00449	8	0.561
32 %	A	.65295	.65783	0.00488	8	0.610
	B	.65925	.66474	0.00549	8	0.686
	C	.65951	.66461	0.00510	8	0.638
	D	.65914	.66338	0.00424	8	0.530
	E	.65807	.66372	0.00565	8	0.706
42 %	A	.66060	.66546	0.00486	8	0.608
	B	.65096	.65506	0.00410	8	0.512
	C	.66215	.66550	0.00335	8	0.419
	D	.66709	.67116	0.00407	8	0.509
	E	.66135	.66552	0.00417	8	0.521
56 %	A	.65723	.66218	0.00495	8	0.619
	B	.66740	.67132	0.00392	8	0.490
	C	.65851	.66333	0.00482	8	0.602
	D	.65156	.65611	0.00455	8	0.569
	E	.64303	.64890	0.00587	8	0.734
80 %	A	.65454	.65907	0.00453	8	0.566
	B	.66755	.67227	0.00472	8	0.590
	C	.65776	.66230	0.00454	8	0.568
	D	.66004	.66443	0.00439	8	0.549
	E	.65433	.65806	0.00373	8	0.466
100 %	A	.65586	.65986	0.00400	8	0.500
	B	.65465	.65996	0.00531	8	0.664
	C	.66559	.67003	0.00444	8	0.555
	D	.65052	.65500	0.00448	8	0.560
	E	.64555	.65060	0.00505	8	0.631

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: July 21, 2020 at 1025
Date and Time Test Terminated: July 28, 2020 at 1045

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	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	3	0	4	7	10	0.700
4	2	3	2	2	3	2	0	0	2	0	16	10	1.60
5	3	4	3	5	0	5	4	6	4	4	38	10	3.80
6	0	8	0	6	5	8	4	8	6	7	52	10	5.20
7	6	0	5	0	8	0	0	0	0	6E	19	10	1.90
8													
TOTAL	11	15	10	13	16	15	8	17	12	15	132	10	13.2

E = Excluded fourth brood neonates

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	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	3	0	0	4	0	0	4	3	4	18	10	1.80
4	2	0	4	4	0	4	4	0	0	0	18	10	1.80
5	8	5	7	9	2	6	5	7	7	0	56	10	5.60
6	11	10	11	10	10	8	14	10	11	11	106	10	10.6
7	0	0	0	0	13E	0	0	12E	13E	11	11	10	1.10
8													
TOTAL	21	18	22	23	16	18	23	21	21	26	209	10	20.9

E = Excluded fourth brood neonates

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	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	3	0	0	3	0	0	5	4	4	19	10	1.90
4	2	0	3	3	0	1	4	0	0	0	13	10	1.30
5	8	7	8	7	7	6	6	8	8	2	67	10	6.70
6	12	4	14	11	10	8	10	12	11	14	106	10	10.6
7	0	0	0	0	1	0	0	15E	13E	13E	1	10	0.100
8													
TOTAL	22	14	25	21	21	15	20	25	23	20	206	10	20.6

E = Excluded fourth brood neonates

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: July 21, 2020 at 1025
Date and Time Test Terminated: July 28, 2020 at 1045

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	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	3	2	0	4	0	0	4	4	5	22	10	2.20
4	3	0	1	0	0	4	3	0	0	0	11	10	1.10
5	8	4	8	7	2	7	9	4	3	7	59	10	5.90
6	13	8	11	8	10	9	10	13	8	10	100	10	10.0
7	0	0	0	0	0	0	0	0	16E	17E	0	10	0.00
8													
TOTAL	24	15	22	15	16	20	22	21	15	22	192	10	19.2

E = Excluded fourth brood neonates

Concentration: 80 %													
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	4	4	0	0	4	3	4	5	3	4	31	10	3.10
4	0	0	4	4	0	0	0	0	0	0	8	10	0.800
5	7	7	9	8	0	8	9	9	9	9	75	10	7.50
6	14	10	12	12	11	12	11	15	10	10	117	10	11.7
7	0	0	0	0	16	11E	17E	0	15E	14E	16	10	1.60
8													
TOTAL	25	21	25	24	31	23	24	29	22	23	247	10	24.7

E = Excluded fourth brood neonates

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	4	4	0	4	3	4	5	4	4	32	10	3.20
4	4	0	0	0	0	0	0	0	0	0	4	10	0.400
5	8	8	8	8	8	7	7	3	8	9	74	10	7.40
6	12	7	14	11	14	9	16	9	14	13	119	10	11.9
7	0	0	0	0	13E	0	0	14E	15E	18E	0	10	0.00
8													
TOTAL	24	19	26	19	26	19	27	17	26	26	229	10	22.9

E = Excluded fourth brood neonates

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	1.00000	1.39310
2	32 %	2	1.00000	1.39310
2	32 %	3	1.00000	1.39310
2	32 %	4	1.00000	1.39310
2	32 %	5	1.00000	1.39310
3	42 %	1	1.00000	1.39310
3	42 %	2	1.00000	1.39310
3	42 %	3	0.87500	1.20940
3	42 %	4	1.00000	1.39310
3	42 %	5	1.00000	1.39310
4	56 %	1	1.00000	1.39310
4	56 %	2	1.00000	1.39310
4	56 %	3	1.00000	1.39310
4	56 %	4	1.00000	1.39310
4	56 %	5	1.00000	1.39310
5	80 %	1	1.00000	1.39310
5	80 %	2	1.00000	1.39310
5	80 %	3	1.00000	1.39310
5	80 %	4	1.00000	1.39310
5	80 %	5	1.00000	1.39310
6	100 %	1	0.87500	1.20940
6	100 %	2	1.00000	1.39310
6	100 %	3	1.00000	1.39310
6	100 %	4	0.87500	1.20940
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))
D = 0.06749		
W = 0.7138		
Critical W = 0.9	(alpha = 0.01, N = 30)	
Critical W = 0.927	(alpha = 0.05, N = 30)	

Data FAIL normality test (alpha = 0.01).

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 %	27.50	16.00	5.00	
3	42 %	25.00	16.00	5.00	
4	56 %	27.50	16.00	5.00	
5	80 %	27.50	16.00	5.00	
6	100 %	22.50	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
D = 0.1025		
W = 0.9769		
Critical W = 0.9	(alpha = 0.01, N = 30)	
Critical W = 0.927	(alpha = 0.05, N = 30)	
Data PASS normality test (alpha = 0.01).		

Bartlett's Test for Homogeneity of Variance		No Transformation
Calculated B1 statistic = 2.346		
Critical B = 15.086	(alpha = 0.01, df = 5)	
Data PASS B1 homogeneity test at 0.01 level.		

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Growth

ANOVA Table				No Transformation
SOURCE	DF	SS	MS	F
Between	5	0.04505	0.00901	2.11
Within (Error)	24	0.1025	0.004271	
Total	29	0.1476		

Critical F = 3.9 (alpha = 0.01, df = 5,24)
2.62 (alpha = 0.05, df = 5,24)

Since F < Critical F FAIL TO 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	0.5926	0.5926		
2	32 %	0.634	0.634	-1.002	
3	42 %	0.5138	0.5138	1.906	
4	56 %	0.6028	0.6028	-0.2468	
5	80 %	0.5478	0.5478	1.084	
6	100 %	0.582	0.582	0.2565	

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	0.09755	16.5	-0.0414
3	42 %	5	0.09755	16.5	0.0788
4	56 %	5	0.09755	16.5	-0.0102
5	80 %	5	0.09755	16.5	0.0448
6	100 %	5	0.09755	16.5	0.0106

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
80 %	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	80 %	10	0	
5	100 %	10	0	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

Kolmogorov Test for Normality	No Transformation
D = 0.0956 D* = 0.75 Critical D* = 1.035 (alpha = 0.01, N = 60) Data PASS normality test (alpha = 0.01).	

Bartlett's Test for Homogeneity of Variance	No Transformation
Calculated B1 statistic = 1.400 Critical B = 15.086 (alpha = 0.01, df = 5) Data PASS B1 homogeneity test at 0.01 level.	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

ANOVA Table				No Transformation
SOURCE	DF	SS	MS	F
Between	5	781.8	156.4	13.82
Within (Error)	54	611.5	11.32	
Total	59	1393		

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	13.2	13.2		
2	32 %	20.9	20.9	-5.117	
3	42 %	20.6	20.6	-4.918	
4	56 %	19.2	19.2	-3.988	
5	80 %	24.7	24.7	-7.643	
6	100 %	22.9	22.9	-6.447	

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

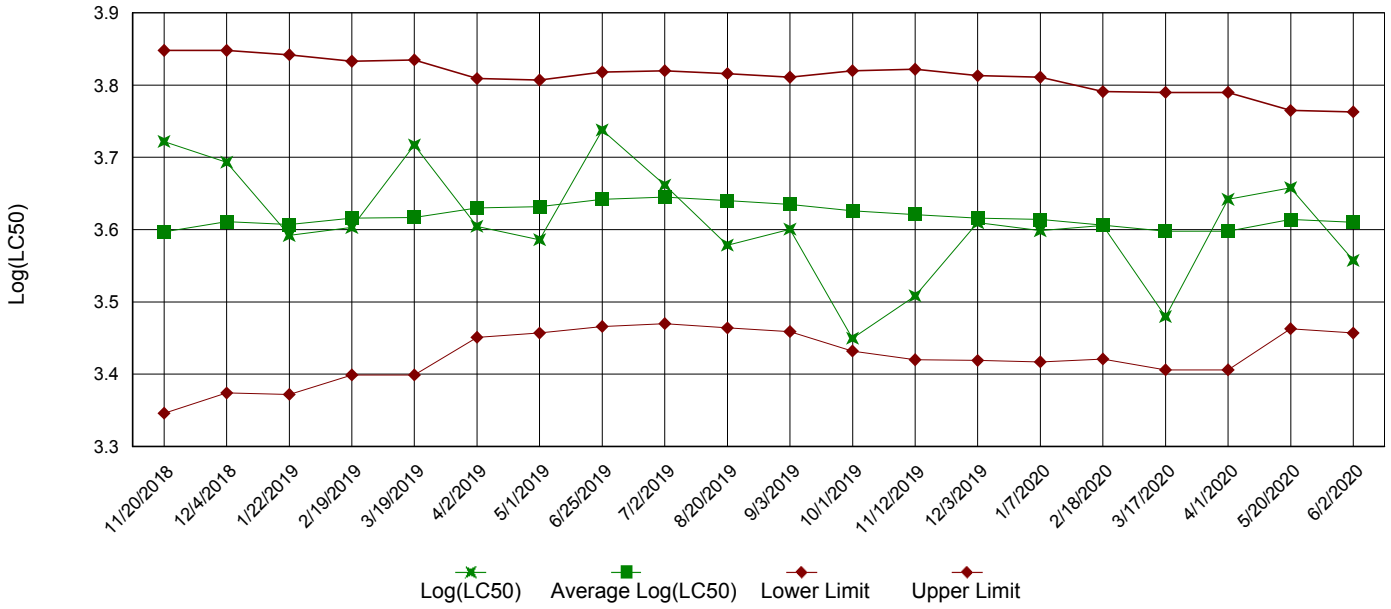
Ho:Control<Treatment

Group	Identification	Num of Reps	Min Sig Diff (In Orig. Units)	% of Control	Difference From Control
1	Control	10			
2	32 %	10	3.476	26.3	-7.7
3	42 %	10	3.476	26.3	-7.4
4	56 %	10	3.476	26.3	-6
5	80 %	10	3.476	26.3	-11.5
6	100 %	10	3.476	26.3	-9.7

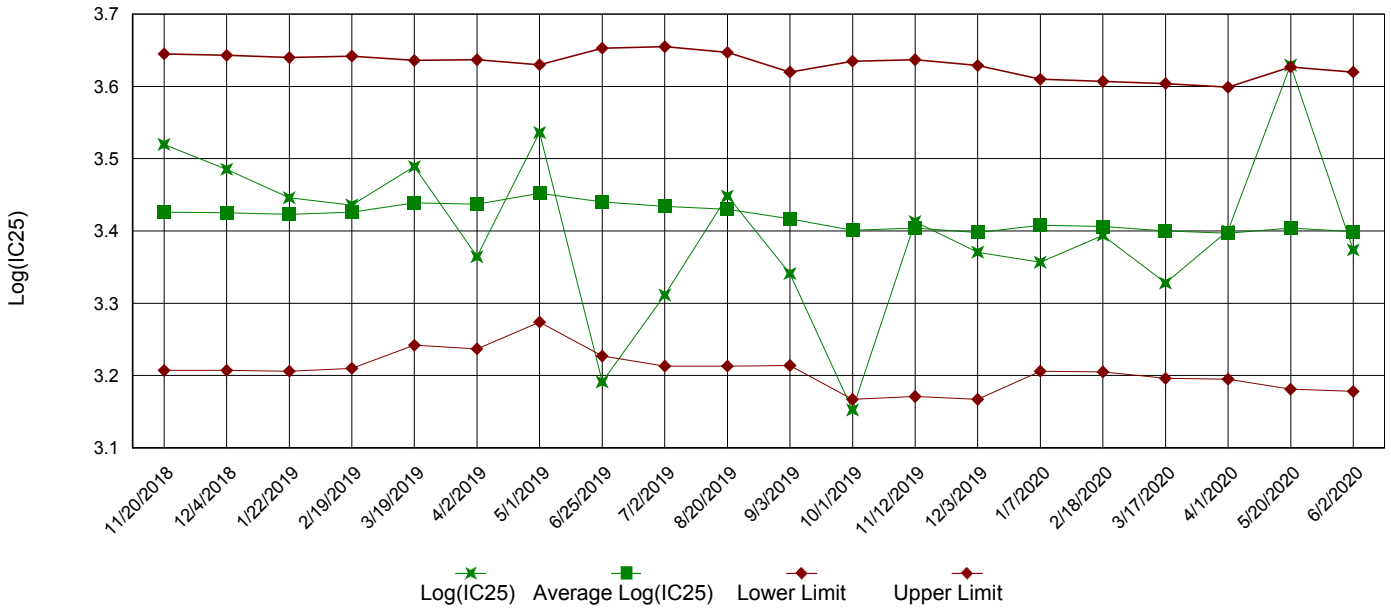
Appendix A3: Test 1000.0

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

LC50 Survival Data



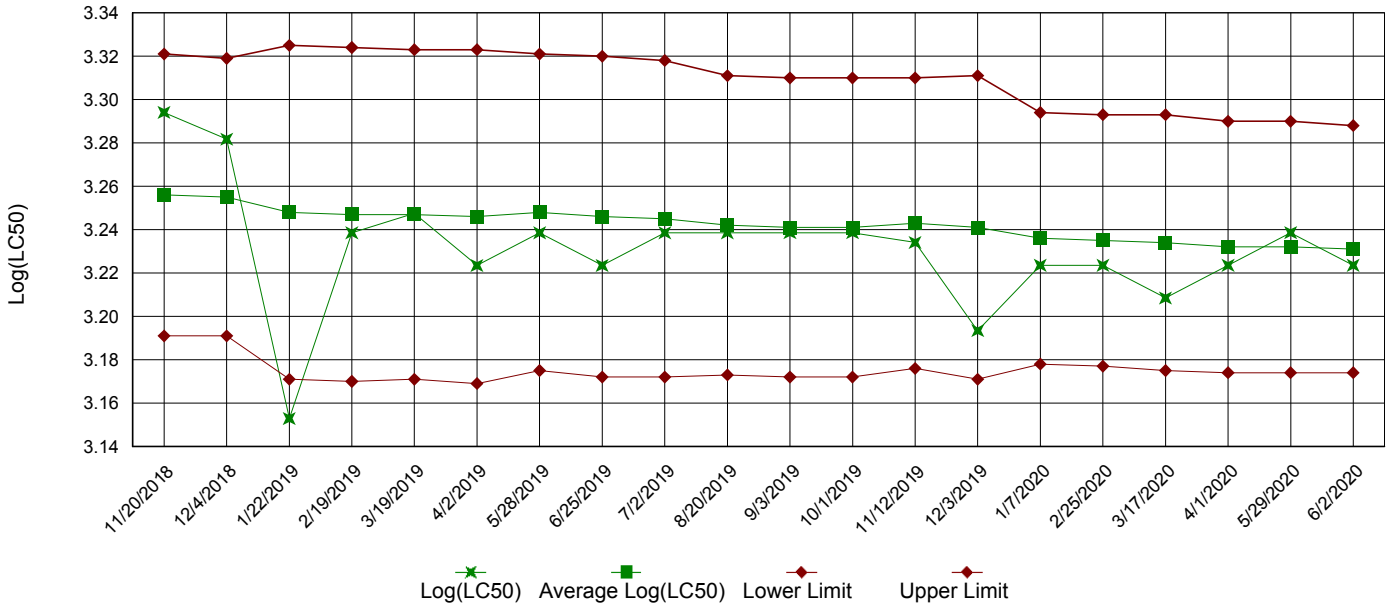
IC25 Growth Data



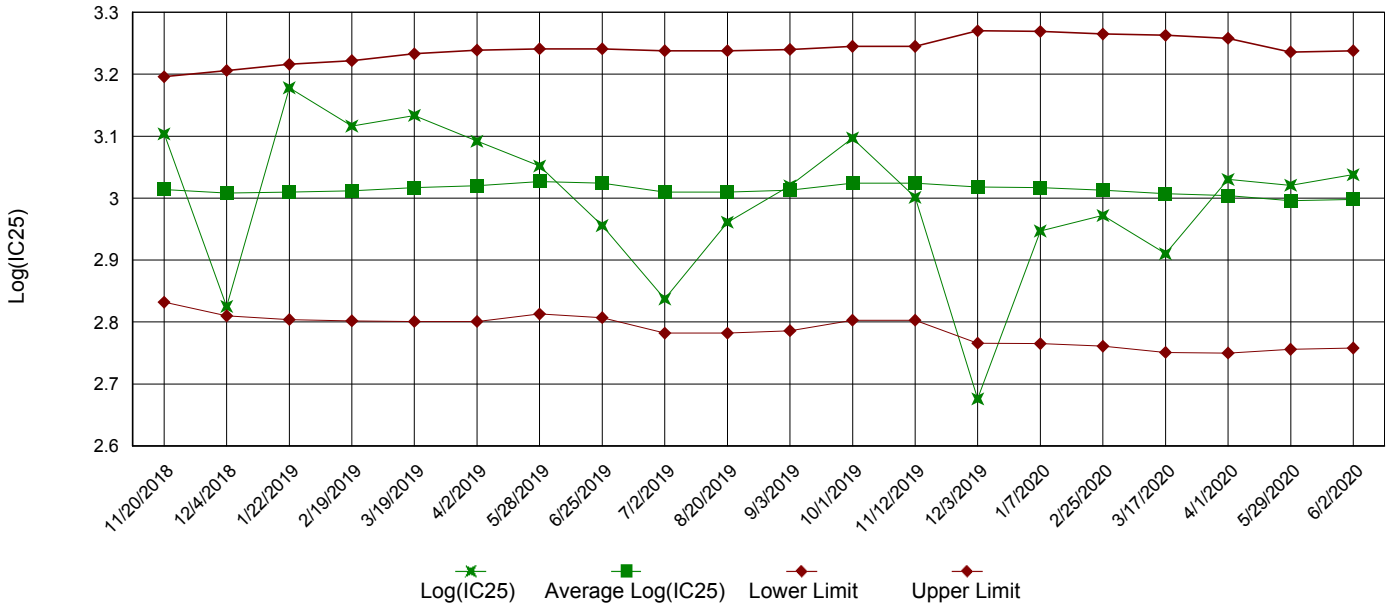
Appendix A3: Test 1002.0

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: City Water & Light of Jonesboro

NPDES No.: AR0043401 AFIN16-00936

Date and Time Test Initiated: July 21, 2020 at 0924
Date and Time Test Terminated: July 28, 2020 at 0830
Dilution water used: Moderately Hard

DATA TABLE FOR SURVIVAL

Effluent Conc. %	Percent Survival in replicate chambers A	B	C	D	E	Mean percent survival 24 hr	48 hr	7 days	CV%
Control	100	100	100	100	100	100	100	100	0.00
32 %	100	100	100	100	100	100	100	100	0.00
42 %	100	100	87.5	100	100	100	100	97.5	5.73
56 %	100	100	100	100	100	100	100	100	0.00
80 %	100	100	100	100	100	100	100	100	0.00
100 %	87.5	100	100	87.5	100	100	100	95.0	7.21

DATA TABLE FOR GROWTH

Effluent Conc. %	Average dry weight, mg replicate chambers A	B	C	D	E	Mean dry weight, mg	CV%
Control	0.598	0.632	0.536	0.636	0.561	0.593	7.39
32 %	0.610	0.686	0.638	0.530	0.706	0.634	11.0
42 %	0.608	0.512	0.419	0.509	0.521	0.514	13.0
56 %	0.619	0.490	0.602	0.569	0.734	0.603	14.7
80 %	0.566	0.590	0.568	0.549	0.466	0.548	8.76
100 %	0.500	0.664	0.555	0.560	0.631	0.582	11.2

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 %) ____YES __X__ NO
   b.) 1/2 LOW FLOW DILUTION (NA) ____YES ____ 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 %) ____YES __X__ NO
   b.) 1/2 LOW FLOW DILUTION (NA) ____YES ____ 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: 11.2 (TQP6C)

10. Lethality for this test: 100 % (51714 or 51714R)

11. Sublethality for this test: 100 % (51714 or 51714S)

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Appendix B: Test 1000.0

CHRONIC TOXICITY SUMMARY FORM
Pimephales promelas (Fathead minnow)
CHEMICAL PARAMETERS CHART

PERMITTEE: City Water & Light of Jonesboro
NPDES NO.: AR0043401 AFIN16-00936
CONTACT: Ms. Whitney Young
ANALYST: 280, 310, 343

Test Initiated: DATE: July 21, 2020 TIME: 0924
Test Terminated: DATE: July 28, 2020 TIME: 0830

DILUTION Control		DAY 1	2	3	4	5	6	7
D.O.	Initial	7.1	7.6	7.3	7.3	7.4	7.3	7.2
	Final	6.5	6.4	6.4	5.9	6.2	6.4	6.2
pH	Initial	8.2	8.2	8.2	8.1	8.1	8.0	8.1
	Final	7.9	7.8	7.7	7.6	7.7	7.7	7.7

DILUTION 32 %		DAY 1	2	3	4	5	6	7
D.O.	Initial	7.2	7.4	7.6	7.3	7.5	7.2	7.2
	Final	5.6	6.0	6.4	5.9	5.9	6.2	6.2
pH	Initial	8.1	8.3	8.1	8.0	8.0	8.0	8.1
	Final	8.0	7.9	7.8	7.8	7.8	7.9	7.9

DILUTION 42 %		DAY 1	2	3	4	5	6	7
D.O.	Initial	7.5	7.2	7.9	7.7	7.8	7.5	7.4
	Final	5.7	7.0	6.4	6.2	6.0	6.5	6.3
pH	Initial	8.1	8.2	8.1	8.0	8.0	8.0	8.0
	Final	8.0	8.1	7.9	7.8	7.8	7.9	7.9

DILUTION 56 %		DAY 1	2	3	4	5	6	7
D.O.	Initial	7.8	7.2	7.6	7.3	7.6	7.2	6.9
	Final	6.2	6.3	6.5	6.2	6.0	6.6	6.8
pH	Initial	8.1	8.2	8.1	8.0	8.0	8.0	8.1
	Final	8.2	8.0	8.0	7.9	8.0	8.1	8.1

DILUTION 80 %		DAY 1	2	3	4	5	6	7
D.O.	Initial	7.8	7.2	7.8	7.5	7.8	7.1	7.3
	Final	6.0	6.3	6.4	5.9	6.1	6.7	6.6
pH	Initial	8.1	8.2	8.1	8.0	8.0	8.0	8.1
	Final	8.3	8.1	8.0	8.0	8.0	8.2	8.2

DILUTION 100 %		DAY 1	2	3	4	5	6	7
D.O.	Initial	7.3	7.4	8.1	7.7	7.4	7.3	7.4
	Final	6.4	6.5	6.5	6.2	6.2	6.7	6.6
pH	Initial	8.2	8.2	8.0	8.0	8.2	8.0	8.0
	Final	8.4	8.3	8.1	8.1	8.2	8.3	8.3

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
160	140	730	<0.05	East Effluent 20-JUL-20
160	130	720	<0.05	East Effluent 22-JUL-20
160	120	740	<0.05	East Effluent 24-JUL-20

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
61	84	290	<0.05	246811-1

Appendix B: Test 1002.0

SUMMARY REPORTING FORMS
CHRONIC BIOMONITORING
Ceriodaphnia dubia
SURVIVAL AND REPRODUCTION

Permittee: City Water & Light of Jonesboro

NPDES No.: AR0043401 AFIN16-00936

Date and Time Test Initiated: July 21, 2020 at 1025
Date and Time Test Terminated: July 28, 2020 at 1045
Dilution water used: Moderately Hard

PERCENT SURVIVAL

		Percent Effluent				
Time of Reading	Control	32 %	42 %	56 %	80 %	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

		Percent Effluent				
Replicates	Control	32 %	42 %	56 %	80 %	100 %
A	11	21	22	24	25	24
B	15	18	14	15	21	19
C	10	22	25	22	25	26
D	13	23	21	15	24	19
E	16	16	21	16	31	26
F	15	18	15	20	23	19
G	8	23	20	22	24	27
H	17	21	25	21	29	17
I	12	21	23	15	22	26
J	15	26	20	22	23	26
Mean per Adult	13.2	20.9	20.6	19.2	24.7	22.9
Mean per Surviving Adult	13.2	20.9	20.6	19.2	24.7	22.9
CV %	22.0	14.0	17.9	18.5	12.5	17.0

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

AMERICAN INTERPLEX CORPORATION LABORATORIES

Appendix B: Test 1002.0

CHRONIC TOXICITY SUMMARY FORM
Ceriodaphnia dubia
CHEMICAL PARAMETERS CHART

PERMITTEE: City Water & Light of Jonesboro
NPDES NO.: AR0043401 AFIN16-00936
CONTACT: Ms. Whitney Young
ANALYST: 280, 310, 343

Test Initiated: DATE: July 21, 2020 TIME: 1025
Test Terminated: DATE: July 28, 2020 TIME: 1045

DILUTION Control	DAY 1	2	3	4	5	6	7
D.O. Initial	7.1	7.6	7.3	7.3	7.4	7.3	7.2
Final	7.4	8.0	7.9	7.7	7.6	7.4	7.2
pH Initial	8.2	8.2	8.2	8.1	8.1	8.0	8.1
Final	8.6	8.4	8.5	8.3	8.4	8.4	8.3

DILUTION 32 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.2	7.4	7.6	7.3	7.5	7.2	7.2
Final	7.2	5.9	7.0	7.4	7.4	7.3	7.2
pH Initial	8.1	8.3	8.1	8.0	8.0	8.0	8.1
Final	8.7	8.0	8.3	8.4	8.5	8.6	8.5

DILUTION 42 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.5	7.2	7.9	7.7	7.8	7.5	7.4
Final	7.6	6.0	7.1	7.8	7.6	7.8	7.4
pH Initial	8.1	8.2	8.1	8.0	8.0	8.0	8.0
Final	8.9	8.2	8.4	8.5	8.6	8.6	8.6

DILUTION 56 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.8	7.2	7.6	7.3	7.6	7.2	6.9
Final	7.4	5.5	7.1	7.6	7.4	7.6	7.4
pH Initial	8.1	8.2	8.1	8.0	8.0	8.0	8.1
Final	8.9	8.1	8.4	8.5	8.6	8.6	8.6

DILUTION 80 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.8	7.2	7.8	7.5	7.8	7.1	7.3
Final	7.6	5.1	7.4	7.8	7.7	7.5	7.2
pH Initial	8.1	8.2	8.1	8.0	8.0	8.0	8.1
Final	9.0	8.2	8.6	8.6	8.7	8.7	8.7

DILUTION 100 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.3	7.4	8.1	7.7	7.4	7.3	7.4
Final	7.5	5.3	7.6	7.6	7.8	7.9	7.0
pH Initial	8.2	8.2	8.0	8.0	8.2	8.0	8.0
Final	9.0	8.3	8.7	8.7	8.8	8.8	8.8

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
160	140	730	<0.05	East Effluent 20-JUL-20
160	130	720	<0.05	East Effluent 22-JUL-20
160	120	740	<0.05	East Effluent 24-JUL-20

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
61	84	290	<0.05	246811-1

AMERICAN INTERPLEX CORPORATION
LABORATORIES

COPY

8600 Kanis Road
Little Rock, AR 72204-2322
(501) 224-5060
FAX (501) 224-5072

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: Jonesboro CWL
Project Reference: WET TESTing
Project Manager: Whitney Young
Sampled By: RS/JH

PO No.

AIC No.	Sample Identification	Date/Time Collected	GRAB	COMP	SAMPLE MATRIX: WATER	SOIL	NO OF BOTTLES	ANALYSES REQUESTED: C. Dubia	P. promelas
1	East Effluent	10:00am-9:00am 7/19-20/20		X	X		3	X	X
	West Effluent	10:00am-9:00am 7/19-20/20		X	X		3	X	X
		Container Type							
		Preservative							

AIC CONTROL NO: 247076
AIC PROPOSAL NO:
Carrier:
Received on Ice (4°C)? (YES) 1.1 NO
Remarks
USE WITH UV Treatment
Field pH calibration on _____ @ _____
Buffer:

G = Glass P = Plastic V = VOA vials H = HCl to pH2 T = Sodium Thiosulfate
NO = none S = Sulfuric acid pH2 N = Nitric acid pH2 B = NaOH to pH12 Z = Zinc acetate

Turnaround Time Requested: (Please circle)
NORMAL or EXPEDITED IN ____ DAYS
Expedited results requested by: _____
Who should AIC contact with questions: _____
Phone: _____ Fax: _____
Report Attention to: _____
Report Address to:

Relinquished By:	Date/Time	Received By:	Date/Time
	7/20/20 12:52pm		
Relinquished By:	Date/Time	Received in Lab By: P. BROWN	Date/Time 7-20-20 1252

Comments:

5/01

FORM 0060

AMERICAN INTERPLEX CORPORATION
LABORATORIES

8600 Kanis Road
Little Rock, AR 72204-2322
(501) 224-5060
FAX (501) 224-5072

COPY

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: Jonesboro CWL
Project Reference: WET TESTING
Project Manager: Whitney Young
Sampled By: JH/RS

PO No.

AIC No.	Sample Identification	Date/Time Collected	GRAB	COMP	SAMPLE MATRIX: WATER	SAMPLE MATRIX: SOIL	NO OF BOTTLES	ANALYSES REQUESTED: C. Dubia	ANALYSES REQUESTED: P. promelas
2	East Effluent	10:00AM-9:00AM 7/21-22/20		X	X		3	X	X
	West Effluent	10:00AM-9:00AM 7/21-22/20		X	X		3	X	X
		Container Type							
		Preservative							

AIC CONTROL NO: 247076
AIC PROPOSAL NO:
Carrier:
Received on Ice (4°C)? (YES) NO
1.1/1.5
Remarks
use UV treatment
247078-2
Field pH calibration
on _____ @ _____
Buffer:

G = Glass P = Plastic V = VOA vials H = HCl to pH2 T = Sodium Thiosulfate
NO = none S = Sulfuric acid pH2 N = Nitric acid pH2 B = NaOH to pH12 Z = Zinc acetate

Turnaround Time Requested: (Please circle)
NORMAL or EXPEDITED IN ____ DAYS
Expedited results requested by: _____
Who should AIC contact with questions: _____
Phone: _____ Fax: _____
Report Attention to: _____
Report Address to:

Relinquished By:	Date/Time	Received By:	Date/Time
Robert Bolton	7/22/20 12:49		
Relinquished By:	Date/Time	Received in Lab By: D. Brown	Date/Time 7-22-20 1249

Comments:

5/01

FORM 0060

AMERICAN INTERPLEX CORPORATION
LABORATORIES

COPY

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: Jonesboro CWL
Project Reference: WET TESTING
Project Manager: Whitney Young
Sampled By: RS/JH

PO No.

MATRIX

AIC No.	Sample Identification	Date/Time Collected	GRAB	COMP	WATER	SOIL		NO OF BOTTLES	ANALYSES REQUESTED: C.dubia	ANALYSES REQUESTED: P. promelas	Remarks
3	EAST EFFLUENT	7/23-24/20 10:00am-9:00am		X	X			3	X	X	
	WEST Effluent	7/23-24/20 10:00am-9:00am		X	X			3	X	X	USE U/V Treatment
		Container Type									Field pH calibration
		Preservative									on _____ @ _____
											Buffer:

AIC CONTROL NO: 247076
AIC PROPOSAL NO:
Carrier: CWL
Received Temperature C: 0.7

G = Glass P = Plastic V = VOA vials H = HCl to pH2 T = Sodium Thiosulfate
NO = none S = Sulfuric acid pH2 N = Nitric acid pH2 B = NaOH to pH12 Z = Zinc acetate A=$(NH_4)_2SO_4$, NH_4OH

Turnaround Time Requested: (Please circle)
NORMAL or EXPEDITED IN ____ DAYS
Expedited results requested by: _____
Who should AIC contact with questions:
Phone:_____ Fax:
Report Attention to:
Report Address to:

Email Address:

Relinquished By:	Date/Time	Received By:	Date/Time
Matthew Bamett Matthew Hammett	7-24-20 12:38PM		
Relinquished By:	Date/Time	Received in Lab By:	Date/Time
		D. BROWN	7-24-20 1238

Comments:

9/2014

FORM 0060