

November 12, 2014

Test Results of  
Fourth Quarter  
Chronic 7-Day Renewal  
Biomonitoring Testing  
for  
Outfall 001

Control No. 184032-1

Prepared for:

Mr. Paul Abernathy  
Searcy Water and Sewer System  
Post Office Box 1319  
Searcy, AR 72145

Prepared by:

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Searcy Water and Sewer System  
ATTN: Mr. Paul Abernathy  
Post Office Box 1319  
Searcy, AR 72145

Re: Chronic 7-Day Renewal utilizing *Pimephales promelas* (Fathead minnow) and *Ceriodaphnia dubia*  
Outfall 001  
NPDES Permit No. AR0021601 AFIN# 73-00055

Dear Mr. Paul Abernathy:

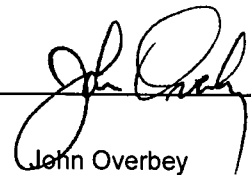
This report is the analytical results and supporting information for the samples submitted to American Interplex Corporation (AIC). The following results are applicable only to the sample identified by the control number referenced above. Accurate assessment of the data requires access to the entire document. Each section of the report has been reviewed and approved by the laboratory director 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 25 % effluent, which is above the critical dilution of 19 %. The NOEC for growth occurred at 11 % effluent, which is below the critical dilution of 19 %. **The sample PASSED lethal effects, however, FAILED sub-lethal effects for the Fathead minnow test.**

Method 1002.0 Chronic *Ceriodaphnia dubia* Survival and Reproduction Test: The No Observable Effects Concentration (NOEC) for survival occurred at 25 % effluent, which is above the critical dilution of 19 %. The NOEC for reproduction occurred at 25 % effluent, which is above the critical dilution of 19 %. **The sample, therefore, PASSED both lethal and sub-lethal effects for the *Ceriodaphnia dubia* test.**

AMERICAN INTERPLEX CORPORATION

  
\_\_\_\_\_  
John Overbey  
Laboratory Director

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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	0.291	PASS
Control Growth CV < or = 40%	10.3	PASS
Growth Minimum Significant Difference 12 to 30%	14.8	PASS
Critical Dilution CV < or = 40%	12.9	PASS

*Ceriodaphnia dubia* Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	24.8	PASS
Control CV < or = 40% per Surviving Female	22.9	PASS
Reproduction Minimum Significant Difference 13 to 47%	17.6	PASS
Critical Dilution CV < or = 40%	5.99	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0021601 AFIN# 73-00055
2. Test Requirements: Chronic Biomonitoring, Quarterly  
Test Methods 1000.0 and 1002.0
3. Receiving Stream: Little Red River

B. Source of Effluent/Dilution Water

1. Effluent Samples:
  - a. Sampling Point: Outfall 001
  - b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.5	7.7	7.9
pH (standard units)	7.0	7.0	6.8
Alkalinity (mg/l as CaCO <sub>3</sub> )	57	59	43
Hardness (mg/l as CaCO <sub>3</sub> )	42	54	57
Conductivity (umhos/cm)	340	340	340
Residual Chlorine (mg/l)	<0.05	<0.05	<0.05
Ammonia as N (mg/l)	0.89	0.89	<0.1

2. Dilution Water Samples: Synthetic Soft Water #4050

- a. Dates Prepared: October 22 through November 5, 2014
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	7.8	7.9	7.5
pH (standard units)	7.2	7.2	7.6
Alkalinity (mg/l as CaCO <sub>3</sub> )	30	30	31
Hardness (mg/l as CaCO <sub>3</sub> )	45	48	48
Conductivity (umhos/cm)	160	150	180
Residual Chlorine (mg/l)	<0.05	<0.05	<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: October 29, 2014 at 1320  
Date & Time Test Terminated: November 5, 2014 at 1500  
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 Growth Method 1002.0

Date & Time Test Initiated: October 29, 2014 at 1315  
Date & Time Test Terminated: November 4, 2014 at 1300  
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. Acclimation 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.

*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 analyzed with Wilcoxon's Rank Sum with Bonferroni Adjustment to determine the No Observable Effects Concentration (NOEC) for reproduction. Dunnett's Test was used to calculate the PMSD.

IV. Standard Reference Toxicants

American Interplex Corporation has an ongoing test organism culturing program. The sensitivity of the offspring is determined by performing a standard reference toxicant test with each effluent test. Sodium chloride in synthetic moderately hard water is used as prescribed in EPA-821-R-02-013.

*Pimephales promelas* (Fathead minnow)

Chronic reference tests are performed monthly.

A chronic reference test was performed on October 22, 2014 at 1030 to October 29, 2014 at 1230

The results were as follows: (Control No. 183793-1.)

Survival LC-50: 4815 mg/l

Growth IC-25: 3427 mg/l

Growth PMSD: 19.3

*Ceriodaphnia dubia*

Chronic reference tests are performed monthly.

A chronic reference test was performed on October 22, 2014 at 1315 to October 28, 2014 at 1500

The results were as follows: (Control No. 183793-2.)

Survival LC-50: 2236 mg/l

Growth IC-25: 993.1 mg/l

Growth PMSD: 13:3

V. Chemical Analysis/Quality Control

Parameter	Method	% Recovery	Relative % Difference
Alkalinity	SM 2320 B	NA	0.00
Hardness	EPA 200.7	101	0.447
pH	SM 4500-H+ B	101	0.676
Conductivity	EPA 120.1	94.0	6.32

VI. Organism History

*Pimephales promelas* (Fathead minnow)

Date: October 29, 2014

Age: <24 hours

Source: In-house culture

Water Chemistry Record:

Alkalinity: 57-64 mg/l

Hardness: 80-100 mg/l

Temperature: 25 deg.C

*Ceriodaphnia dubia*

Date: October 29, 2014

Age: <24 hours

Source: In-house culture

Water Chemistry Record:

Alkalinity: 57-64 mg/l

Hardness: 80-100 mg/l

Temperature: 25 deg.C

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

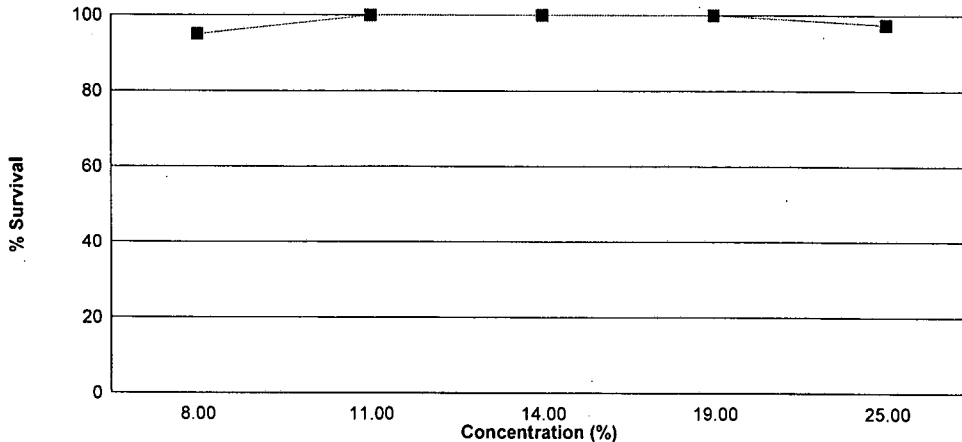
Larvae are exposed in a static renewal system for seven days to different concentrations of effluent with dilution water. Test results are based on the survival and growth (increase in weight) of the larvae.

Effluent dilutions for this test were 8 %, 11 %, 14 %, 19 %, 25 % in accordance with the NPDES permit.

The low flow or 'critical' dilution is specified in the NPDES permit as 19 % effluent.

The test was initiated on October 29, 2014 at 1320 and continued through November 5, 2014 at 1500. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

- a.) NOEC survival = 25 % effluent
- b.) NOEC growth = 11 % effluent



Summary of the 7-day Fathead Minnow Survival and Growth		
Concentration	Percent Survival	Mean Growth (mg)
Control	100	0.291
8 %	95.0	0.251
11 %	100	0.255
14 %	100	0.238 *
19 %	100	0.221 *
25 %	97.5	0.185 *

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

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

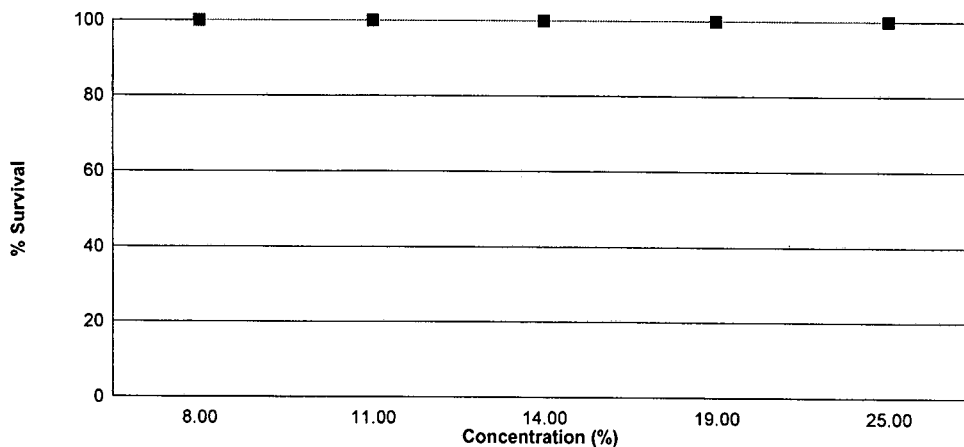
Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring with an average of at least 15 young per female.

Effluent dilutions for this test were 8 %, 11 %, 14 %, 19 %, 25 % in accordance with the NPDES permit.

The low flow or 'critical' dilution is specified in the NPDES permit as 19 % effluent.

The test was initiated on October 29, 2014 at 1315 and continued through November 4, 2014 at 1300. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

- a.) NOEC survival = 25 % effluent
- b.) NOEC reproduction = 25 % effluent



Summary of the 6-day <i>Ceriodaphnia dubia</i> Survival and Reproduction Data		
Concentration	Percent Survival	Mean Reproduction
Control	100	24.8
8 %	100	27.6
11 %	100	27.6
14 %	100	27.3
19 %	100	28.6
25 %	100	25.9



Appendix A1: Test 1000.0

*Pimephales promelas* (Fathead Minnow) 7-Day Survival

Date and Time Test Initiated: October 29, 2014 at 1320

Date and Time Test Terminated: November 5, 2014 at 1500

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
8 %	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	7	7	7	7	7	7
11 %	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
14 %	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
19 %	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
25 %	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	7	7	7	7

Appendix A1: Test 1000.0

*Pimephales promelas* (Fathead Minnow) 7-Day Growth

Test Initiated: October 29, 2014 at 1320  
Test Terminated: November 5, 2014 at 1500

Drying Started: November 5, 2014 at 1501  
Drying Ended: November 11, 2014 at 0945

Concentration	Replicate	Weight of pan	Weight of pan + fish	Total weight of fish (g)	Original # of fish	Mean dry weight (mg)
Control	A	.94950	.95182	0.00232	8	0.290
	B	.94910	.95124	0.00214	8	0.268
	C	.95057	.95331	0.00274	8	0.342
	D	.95667	.95884	0.00217	8	0.271
	E	.95642	.95869	0.00227	8	0.284
8 %	A	.95624	.95799	0.00175	8	0.219
	B	.95462	.95654	0.00192	8	0.240
	C	.95558	.95766	0.00208	8	0.260
	D	.95698	.95938	0.00240	8	0.300
	E	.95817	.96005	0.00188	8	0.235
11 %	A	.95932	.96160	0.00228	8	0.285
	B	.95764	.95953	0.00189	8	0.236
	C	.95497	.95703	0.00206	8	0.258
	D	.95519	.95702	0.00183	8	0.229
	E	.95271	.95484	0.00213	8	0.266
14 %	A	.95327	.95485	0.00158	8	0.198
	B	.95526	.95727	0.00201	8	0.251
	C	.95609	.95830	0.00221	8	0.276
	D	.95775	.95947	0.00172	8	0.215
	E	.95935	.96133	0.00198	8	0.248
19 %	A	.95617	.95823	0.00206	8	0.258
	B	.95993	.96152	0.00159	8	0.199
	C	.96011	.96167	0.00156	8	0.195
	D	.95882	.96078	0.00196	8	0.245
	E	.96110	.96276	0.00166	8	0.208
25 %	A	.95935	.96052	0.00117	8	0.146
	B	.96032	.96197	0.00165	8	0.206
	C	.95907	.96084	0.00177	8	0.221
	D	.95732	.95876	0.00144	8	0.180
	E	.95864	.96002	0.00138	8	0.172

Appendix A1: Test 1002.0

*Ceriodaphnia dubia* Survival and Reproduction

Date and Time Test Initiated: October 29, 2014 at 1315  
Date and Time Test Terminated: November 4, 2014 at 1300

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	5	0	4	4	5	0	0	5	0	23	10	2.30
4	9	0	4	0	0	0	2	0	0	4	19	10	1.90
5	10	11	8	11	12	10	10	11	10	0	93	10	9.30
6	0	13	11	12	16	14	12	12	14	9	113	10	11.3
7													
8													
TOTAL	19	29	23	27	32	29	24	23	29	13	248	10	24.8

Concentration: 8 %													
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	5	4	0	0	4	0	0	0	4	0	17	10	1.70
4	0	0	4	5	0	5	3	2	0	5	24	10	2.40
5	10	9	11	10	12	9	9	7	8	11	96	10	9.60
6	14	13	14	15	14	13	14	15	14	13	139	10	13.9
7													
8													
TOTAL	29	26	29	30	30	27	26	24	26	29	276	10	27.6

Concentration: 11 %													
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	LIA	0	0	0	0	0	0	9	0.00
2	0	0	0	0	LIA	0	0	0	0	0	0	9	0.00
3	0	5	0	4	LIA	0	0	0	5	0	14	9	1.56
4	6	0	4	0	LIA	4	4	3	0	4	25	9	2.78
5	12	8	9	8	LIA	8	9	7	10	11	82	9	9.11
6	14	15	14	13	LIA	14	15	15	14	13	127	9	14.1
7													
8													
TOTAL	32	28	27	25		26	28	25	29	28	248	9	27.6

LIA = Lost in Analysis

Appendix A1: Test 1002.0

*Ceriodaphnia dubia* Survival and Reproduction

Date and Time Test Initiated: October 29, 2014 at 1315

Date and Time Test Terminated: November 4, 2014 at 1300

Concentration: 14 %														
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	4	0	4	5	0	4	0	0	0	17	10	1.70	
4	6	0	5	0	0	4	0	4	3	5	27	10	2.70	
5	12	9	10	8	7	7	8	9	10	12	92	10	9.20	
6	15	14	13	15	14	13	12	14	13	14	137	10	13.7	
7														
8														
TOTAL	33	27	28	27	26	24	24	27	26	31	273	10	27.3	

Concentration: 19 %														
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	4	5	4	0	4	5	22	10	2.20	
4	6	4	4	5	0	0	0	4	0	0	23	10	2.30	
5	11	10	10	11	9	10	9	11	10	11	102	10	10.2	
6	14	13	14	14	15	14	13	14	13	15	139	10	13.9	
7														
8														
TOTAL	31	27	28	30	28	29	26	29	27	31	286	10	28.6	

Concentration: 25 %														
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	5	4	3	4	0	5	0	29	10	2.90	
4	0	0	4	0	0	0	0	7	0	7	18	10	1.80	
5	11	13	0	12	8	12	10	0	10	7	83	10	8.30	
6	14	16	13	18	17	15	11	13	12	0	129	10	12.9	
7														
8														
TOTAL	29	33	17	35	29	30	25	20	27	14	259	10	25.9	

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	8 %	1	0.87500	1.20940	
2	8 %	2	1.00000	1.39310	
2	8 %	3	1.00000	1.39310	
2	8 %	4	1.00000	1.39310	
2	8 %	5	0.87500	1.20940	
3	11 %	1	1.00000	1.39310	
3	11 %	2	1.00000	1.39310	
3	11 %	3	1.00000	1.39310	
3	11 %	4	1.00000	1.39310	
3	11 %	5	1.00000	1.39310	
4	14 %	1	1.00000	1.39310	
4	14 %	2	1.00000	1.39310	
4	14 %	3	1.00000	1.39310	
4	14 %	4	1.00000	1.39310	
4	14 %	5	1.00000	1.39310	
5	19 %	1	1.00000	1.39310	
5	19 %	2	1.00000	1.39310	
5	19 %	3	1.00000	1.39310	
5	19 %	4	1.00000	1.39310	
5	19 %	5	1.00000	1.39310	
6	25 %	1	1.00000	1.39310	
6	25 %	2	1.00000	1.39310	
6	25 %	3	1.00000	1.39310	
6	25 %	4	1.00000	1.39310	
6	25 %	5	0.87500	1.20940	

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	8 %	22.50	16.00	5.00	
3	11 %	27.50	16.00	5.00	
4	14 %	27.50	16.00	5.00	
5	19 %	27.50	16.00	5.00	
6	25 %	25.00	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 = 0.0201 W = 0.9405 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 = 0.4526 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	0.03164	0.006328	7.556	
Within (Error)	24	0.0201	0.0008375		
Total	29	0.05174			
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	0.291	0.291		
2	8 %	0.2508	0.2508	2.196	
3	11 %	0.2548	0.2548	1.978	
4	14 %	0.2376	0.2376	2.918	*
5	19 %	0.221	0.221	3.825	*
6	25 %	0.185	0.185	5.791	*
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	8 %	5	0.0432	14.8	0.0402	
3	11 %	5	0.0432	14.8	0.0362	
4	14 %	5	0.0432	14.8	0.0534	
5	19 %	5	0.0432	14.8	0.07	
6	25 %	5	0.0432	14.8	0.106	



Appendix A2: Statistics

*Ceriodaphnia dubia* Survival

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
8 %	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
11 %	9	0	9
Total	19	0	19

Critical Fisher's value (10,9,10) (alpha=0.05) is 5. b value is 9. Since b is greater than 5 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
14 %	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
19 %	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
25 %	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	8 %	10	0	
2	11 %	9	0	
3	14 %	10	0	
4	19 %	10	0	
5	25 %	10	0	

Appendix A2: Statistics

*Ceriodaphnia dubia* Reproduction

Kolmogorov Test for Normality		No Transformation
<p>D = 0.1364  D* = 1.061  Critical D* = 1.035 (alpha = 0.01, N = 59)</p> <p>Data FAIL normality test (alpha = 0.01).</p>		

Wilcoxon's Rank Sum w/ Bonferroni Adjustment					No Transformation
Ho: Control < Treatment					
Group	Identification	Rank Sum	Critical Value	Reps	Sig 0.05
1	Control				
2	8 %	120.50	74.00	10	
3	11 %	100.50	61.00	9	
4	14 %	115.50	74.00	10	
5	19 %	125.00	74.00	10	
6	25 %	113.50	74.00	10	
Critical values are 1 tailed (k=5)					

Appendix A2: Statistics

*Ceriodaphnia dubia* Reproduction

Dunnett's Test for PMSD Calculation (excluding deaths if applicable)

ANOVA Table				No Transformation	
SOURCE	DF	SS	MS	F	
Between	5	93.23	18.65	1.109	
Within (Error)	53	891.6	16.82		
Total	58	984.8			

Critical F = 3.39 (alpha = 0.01, df = 5,53)  
2.39 (alpha = 0.05, df = 5,53)

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	24.8	24.8			
2	8 %	27.6	27.6	-1.527		
3	11 %	27.556	27.556	-1.463		
4	14 %	27.3	27.3	-1.363		
5	19 %	28.6	28.6	-2.072		
6	25 %	25.9	25.9	-0.5997		

Dunnett's critical value = 2.31 (1 Tailed, alpha = 0.05, df [used] = 5,40) (Actual df = 5,53)  
WARNING - Unequal replicate sizes. Critical values assuming equal replicate sizes have been used.

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	8 %	10	4.237	17.1	-2.8	
3	11 %	9	4.353	17.6	-2.756	
4	14 %	10	4.237	17.1	-2.5	
5	19 %	10	4.237	17.1	-3.8	
6	25 %	10	4.237	17.1	-1.1	

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: October 29, 2014 at 1101

Date and Time Test Terminated: November 5, 2014 at 1500

Effluent Conc.: Control		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	7.8	7.5	7.9	8.1	7.5	6.9	7.4
	Final *1	7.8	8.1	8.0	7.9	7.0	7.8	7.4
	Final *2	8.7	8.0	8.2	7.9	7.7	8.4	
pH, units	Initial	7.2	7.2	7.2	7.0	7.6	7.8	7.2
	Final *1	7.2	7.1	7.1	7.8	7.8	7.5	7.7
	Final *2	7.4	7.2	7.0	7.7	7.6	7.7	
Alkalinity, mg CaCO <sub>3</sub> /l	30	NA	30	NA	31	NA	NA	NA
Hardness, mg CaCO <sub>3</sub> /l	45	NA	48	NA	48	NA	NA	NA
Conductivity, umhos/cm	160	150	150	160	180	200	160	
Res. Chlorine, mg/l	<0.05	NA	<0.05	NA	<0.05	NA	NA	NA

Effluent Conc.: 8 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.0	7.6	8.0	8.2	7.5	6.9	7.6
	Final *1	7.5	8.3	8.0	7.9	6.8	7.7	7.2
	Final *2	8.2	8.1	8.1	7.8	7.7	8.3	
pH, units	Initial	7.2	7.2	7.2	7.1	7.6	7.7	7.1
	Final *1	7.2	7.2	7.1	7.8	7.7	7.5	7.6
	Final *2	7.4	7.2	7.0	7.7	7.5	7.8	

Effluent Conc.: 11 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.0	7.6	8.0	8.2	7.5	6.8	7.5
	Final *1	7.4	8.1	8.1	7.9	7.0	7.7	7.3
	Final *2	8.6	8.1	8.0	7.8	7.7	8.4	
pH, units	Initial	7.2	7.2	7.2	7.2	7.6	7.7	7.1
	Final *1	7.2	7.2	7.1	7.8	7.8	7.5	7.7
	Final *2	7.4	7.2	7.0	7.7	7.5	7.7	

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: October 29, 2014 at 1101

Date and Time Test Terminated: November 5, 2014 at 1500

Effluent Conc.: 14 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.2	7.5	8.0	8.1	7.6	6.8	7.2
	Final *1	7.7	8.1	8.0	4.5	6.8	8.0	7.3
	Final *2	8.5	8.1	8.2	7.9	7.9	8.3	
pH, units	Initial	7.2	7.2	7.2	7.1	7.7	7.6	7.0
	Final *1	7.2	7.2	7.1	7.9	7.7	7.6	7.6
	Final *2	7.4	7.2	7.0	7.7	7.6	7.7	

Effluent Conc.: 19 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.0	7.6	8.0	8.2	7.4	6.8	7.2
	Final *1	6.9	8.1	7.9	7.9	6.9	7.5	7.2
	Final *2	8.4	7.8	7.9	7.8	7.8	8.4	
pH, units	Initial	7.2	7.2	7.2	7.2	7.7	7.6	7.0
	Final *1	7.1	7.2	7.1	7.8	7.9	7.5	7.6
	Final *2	7.5	7.2	7.0	7.7	7.6	7.7	
Alkalinity, mg CaCO <sub>3</sub> /l	38	NA	43	NA	39	NA	NA	NA
Hardness, mg CaCO <sub>3</sub> /l	41	NA	52	NA	48	NA	NA	NA
Conductivity, umhos/cm	200	200	190	190	210	220	200	200
Res. Chlorine, mg/l	<0.05	NA	<0.05	NA	<0.05	NA	NA	NA

Effluent Conc.: 25 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.2	7.8	8.0	8.2	7.4	6.7	7.5
	Final *1	7.3	8.0	7.9	7.8	6.9	7.9	7.2
	Final *2	8.5	7.9	7.9	7.7	7.8	8.5	
pH, units	Initial	7.2	7.2	7.3	7.2	7.7	7.6	7.0
	Final *1	7.2	7.2	7.1	7.9	7.9	7.7	7.6
	Final *2	7.5	7.2	7.1	7.8	7.6	7.8	

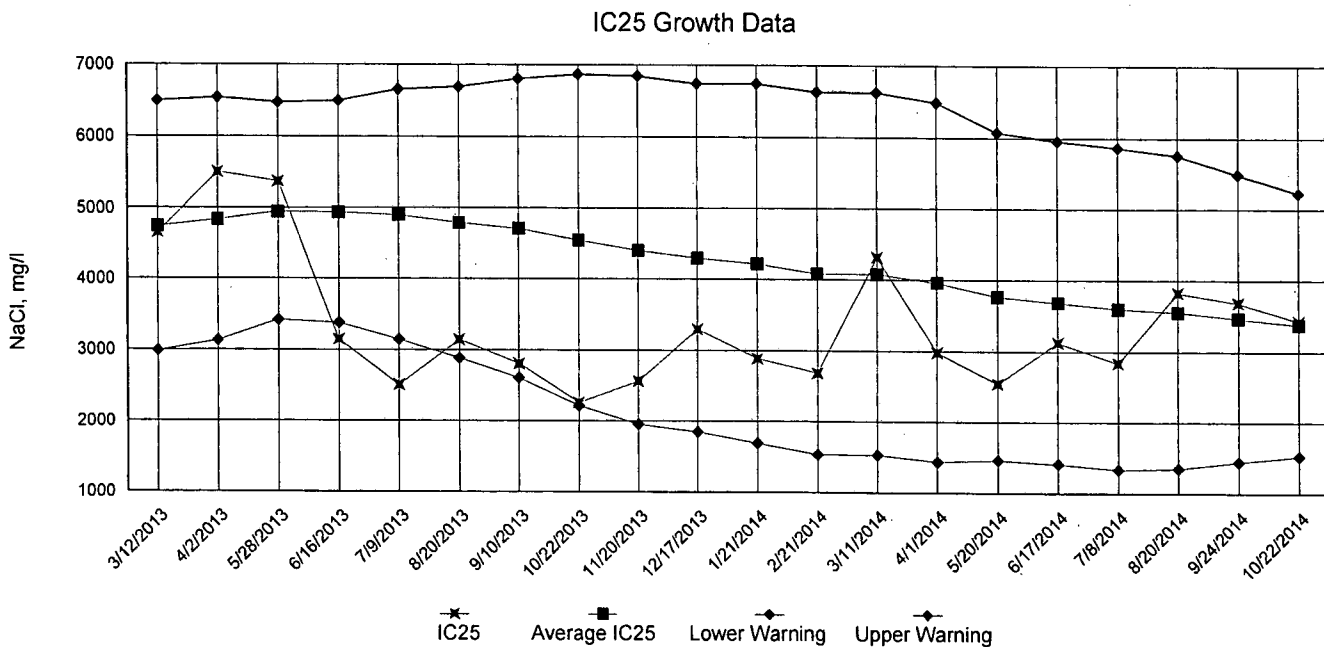
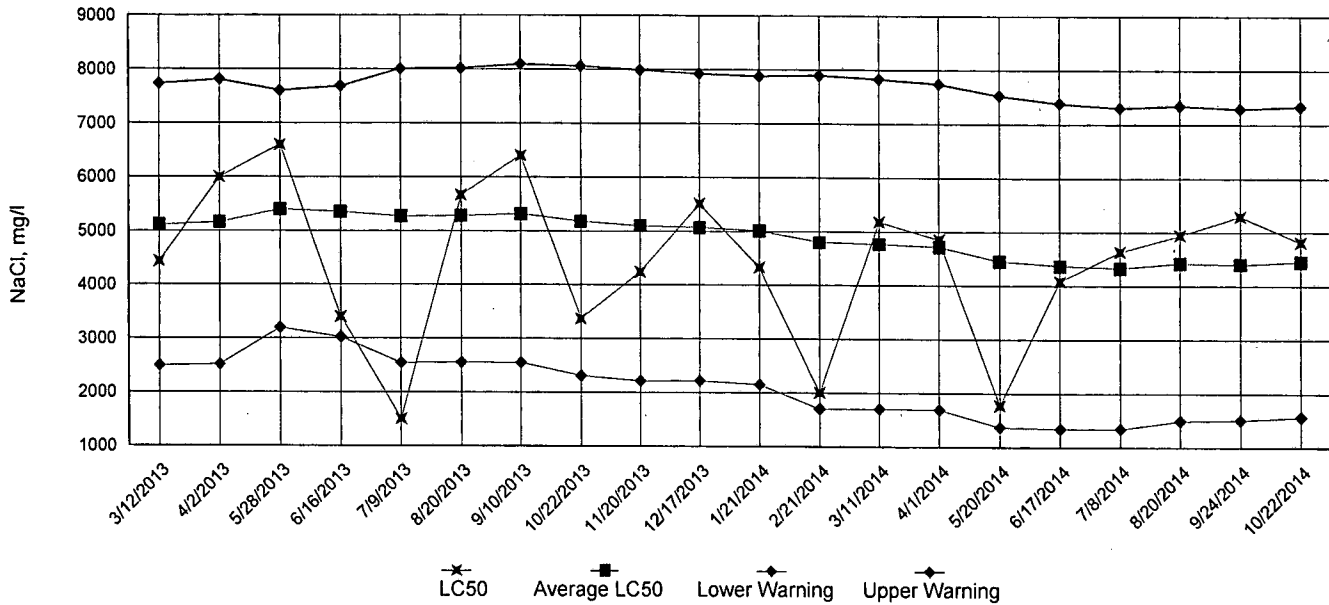
\*1 = data from the *Pimephales promelas* (Fathead Minnow) test

\*2 = data from the *Ceriodaphnia dubia* test

Appendix A4: Test 1000.0

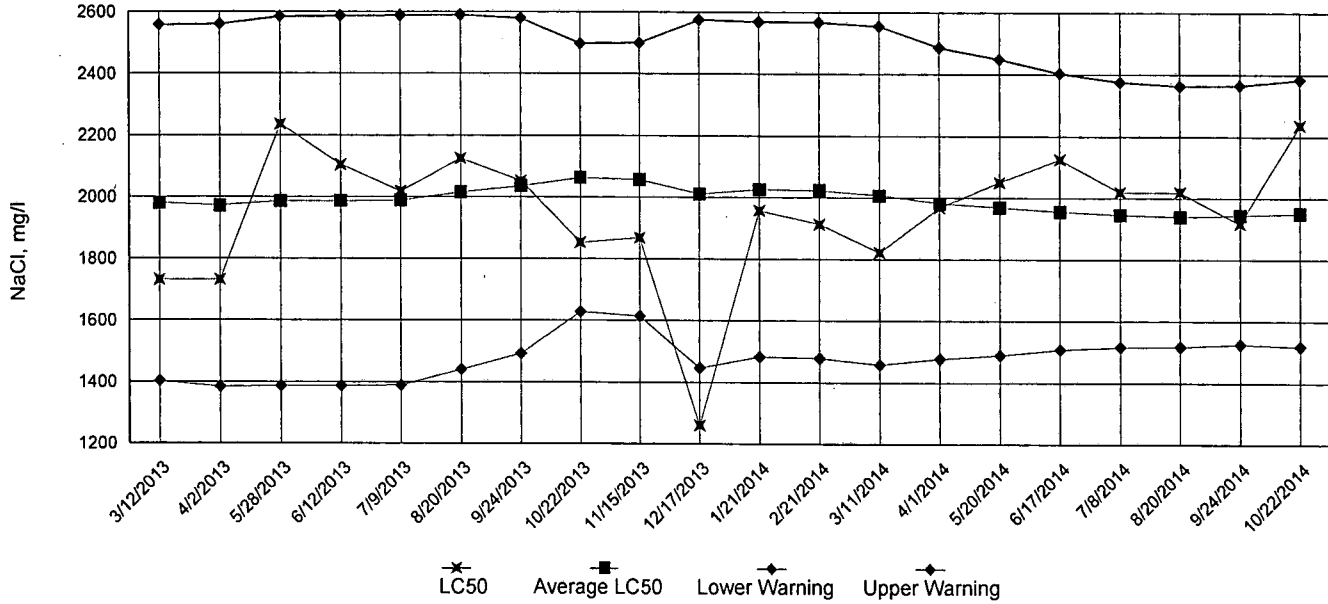
Chronic Reference Toxicant, *Pimephales promelas* (Fathead Minnow)

LC50 Survival Data

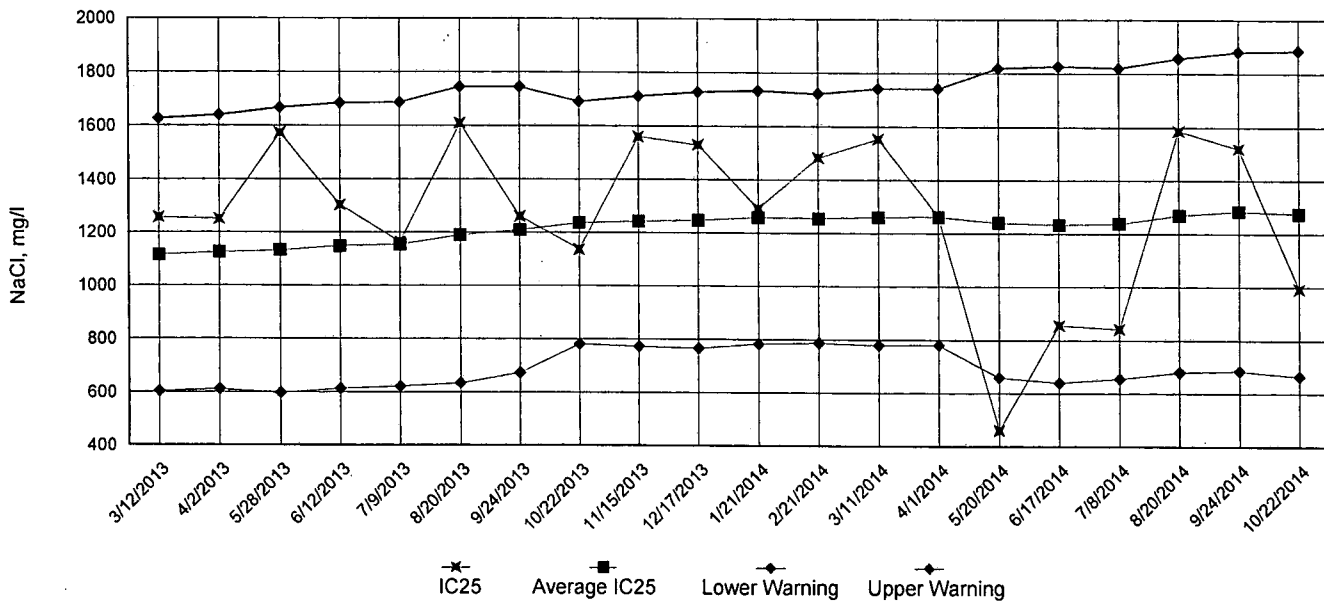


Appendix A4: Test 1002.0  
Chronic Reference Toxicant, *Ceriodaphnia dubia*

LC50 Survival Data



IC25 Reproduction Data





Appendix B: Test 1000.0

SUMMARY REPORTING FORMS  
CHRONIC BIOMONITORING  
*Pimephales promelas* (Fathead Minnow)  
SURVIVAL AND GROWTH

Permittee: Searcy Water and Sewer System

NPDES No.: AR0021601 AFIN# 73-00055

Date and Time Test Initiated: October 29, 2014 at 1320

Date and Time Test Terminated: November 5, 2014 at 1500

Dilution water used: Synthetic Soft Water #4050

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
8 %	87.5	100	100	100	87.5	100	97.5	95.0	7.21
11 %	100	100	100	100	100	100	100	100	0.00
14 %	100	100	100	100	100	100	100	100	0.00
19 %	100	100	100	100	100	100	100	100	0.00
25 %	100	100	100	100	87.5	100	100	97.5	5.73

DATA TABLE FOR GROWTH

Effluent Conc. %	Average dry weight, mg replicate chambers					Mean dry weight, mg	CV%
	A	B	C	D	E		
Control	0.290	0.268	0.342	0.271	0.284	0.291	10.3
8 %	0.219	0.240	0.260	0.300	0.235	0.251	12.4
11 %	0.285	0.236	0.258	0.229	0.266	0.255	8.92
14 %	0.198	0.251	0.276	0.215	0.248	0.238	13.0
19 %	0.258	0.199	0.195	0.245	0.208	0.221	12.9
25 %	0.146	0.206	0.221	0.180	0.172	0.185	15.9

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	(19 %)	_____ YES	<u>  X  </u> 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	(19 %)	<u>  X  </u> YES	_____ 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]:   1   (TGP6C)

5. NOEC Pimephales Lethality:   25 %   (TOP6C)

6. LOEC Pimephales Lethality:   25 %   (TXP6C)

7. NOEC Pimephales Sublethality:   11 %   (TPP6C)

8. LOEC Pimephales Sublethality:   14 %   (TYP6C)

9. Coefficient of variation for Pimephales growth:   12.9   (TQP6C)

Appendix B: Test 1000.0

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

PERMITTEE: Searcy Water and Sewer System SAMPLE No. 1 COLLECTED ending: DATE: October 28, 2014 TIME: 1140  
 NPDES NO.: AR0021601 AFIN# 73-00055 SAMPLE No. 2 COLLECTED ending: DATE: October 30, 2014 TIME: 1145  
 CONTACT: Mr. Paul Abernathy SAMPLE No. 3 COLLECTED ending: DATE: November 2, 2014 TIME: 1145  
 ANALYST: 280, 304, 310 Test Initiated: DATE: October 29, 2014 TIME: 1320  
 Test Terminated: DATE: November 5, 2014 TIME: 1500

DILUTION Control	DAY						
	1	2	3	4	5	6	7
D.O. Initial	7.8	7.5	7.9	8.1	7.5	6.9	7.4
Final	7.8	8.1	8.0	7.9	7.0	7.8	7.4
pH Initial	7.2	7.2	7.2	7.0	7.6	7.8	7.2
Final	7.2	7.1	7.1	7.8	7.8	7.5	7.7
Alkalinity	30	NA	30	NA	31	NA	NA
Hardness	45	NA	48	NA	48	NA	NA
Conductivity	160	150	150	160	180	200	160
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION 8 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.0	7.6	8.0	8.2	7.5	6.9	7.6
Final	7.5	8.3	8.0	7.9	6.8	7.7	7.2
pH Initial	7.2	7.2	7.2	7.1	7.6	7.7	7.1
Final	7.2	7.2	7.1	7.8	7.7	7.5	7.6
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	180	170	170	170	190	200	170
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 11 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.0	7.6	8.0	8.2	7.5	6.8	7.5
Final	7.4	8.1	8.1	7.9	7.0	7.7	7.3
pH Initial	7.2	7.2	7.2	7.2	7.6	7.7	7.1
Final	7.2	7.2	7.1	7.8	7.8	7.5	7.7
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	180	170	170	170	200	200	180
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 14 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.2	7.5	8.0	8.1	7.6	6.8	7.2
Final	7.7	8.1	8.0	4.5	6.8	8.0	7.3
pH Initial	7.2	7.2	7.2	7.1	7.7	7.6	7.0
Final	7.2	7.2	7.1	7.9	7.7	7.6	7.6
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	190	180	180	180	200	210	180
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 19 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.0	7.6	8.0	8.2	7.4	6.8	7.2
Final	6.9	8.1	7.9	7.9	6.9	7.5	7.2
pH Initial	7.2	7.2	7.2	7.2	7.7	7.6	7.0
Final	7.1	7.2	7.1	7.8	7.9	7.5	7.6
Alkalinity	38	NA	43	NA	39	NA	NA
Hardness	41	NA	52	NA	48	NA	NA
Conductivity	200	200	190	190	210	220	200
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION 25 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.2	7.8	8.0	8.2	7.4	6.7	7.5
Final	7.3	8.0	7.9	7.8	6.9	7.9	7.2
pH Initial	7.2	7.2	7.3	7.2	7.7	7.6	7.0
Final	7.2	7.2	7.1	7.9	7.9	7.7	7.6
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	210	200	200	200	230	240	210
Chlorine	NA	NA	NA	NA	NA	NA	NA

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

Permittee: Searcy Water and Sewer System

NPDES No.: AR0021601 AFIN# 73-00055

Date and Time Test Initiated: October 29, 2014 at 1315

Date and Time Test Terminated: November 4, 2014 at 1300

Dilution water used: Synthetic Soft Water #4050

PERCENT SURVIVAL

Time of Reading	Control	Percent Effluent				
		8 %	11 %	14 %	19 %	25 %
24 hour	100	100	100	100	100	100
48 hour	100	100	100	100	100	100
6 day	100	100	100	100	100	100

NUMBER OF YOUNG PRODUCED PER FEMALE @ 6 DAYS

Replicates	Control	Percent Effluent				
		8 %	11 %	14 %	19 %	25 %
A	19	29	32	33	31	29
B	29	26	28	27	27	33
C	23	29	27	28	28	17
D	27	30	25	27	30	35
E	32	30		26	28	29
F	29	27	26	24	29	30
G	24	26	28	24	26	25
H	23	24	25	27	29	20
I	29	26	29	26	27	27
J	13	29	28	31	31	14
Mean per Adult	24.8	27.6	27.6	27.3	28.6	25.9
Mean per Surviving Adult	24.8	27.6	27.6	27.3	28.6	25.9
CV %	22.9	7.48	7.93	10.4	5.99	26.6

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

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

1. Fisher's Exact 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	(19 %)	<u>          </u> YES	<u>  X  </u> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<u>          </u> YES	<u>          </u> NO

2. Wilcoxon's Rank Sum with Bonferroni Adjustment Test:

Is the mean number of young produced per female significantly different ( $p=0.05$ ) than the control's number of young per female for the % effluent corresponding to (significant non-lethal effects):

a.) LOW FLOW OR CRITICAL DILUTION	(19 %)	<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     (TLP3B)
- 4. If you answered NO to 2.a) enter [0] otherwise enter [1]:     0     (TGP3B)
- 5. NOEC *Ceriodaphnia* Lethality:   25 %   (TOP3B)
- 6. LOEC *Ceriodaphnia* Lethality:   25 %   (TXP3B)
- 7. NOEC *Ceriodaphnia* Sublethality:   25 %   (TPP3B)
- 8. LOEC *Ceriodaphnia* Sublethality:   25 %   (TYP3B)
- 9. Coefficient of variation for *Ceriodaphnia* Reproduction:   22.9   (TQP3B)

Appendix B: Test 1002.0

CHRONIC TOXICITY SUMMARY FORM  
*Ceriodaphnia dubia*  
CHEMICAL PARAMETERS CHART

PERMITTEE: Searcy Water and Sewer System SAMPLE No. 1 COLLECTED ending: DATE: October 28, 2014 TIME: 1140  
 NPDES NO.: AR0021601 AFIN# 73-00055 SAMPLE No. 2 COLLECTED ending: DATE: October 30, 2014 TIME: 1145  
 CONTACT: Mr. Paul Abernathy SAMPLE No. 3 COLLECTED ending: DATE: November 2, 2014 TIME: 1145  
 ANALYST: 280, 304, 310 Test Initiated: DATE: October 29, 2014 TIME: 1315  
 Test Terminated: DATE: November 4, 2014 TIME: 1300

DILUTION	DAY						
	1	2	3	4	5	6	7
Control							
D.O. Initial	7.8	7.5	7.9	8.1	7.5	6.9	7.4
Final	8.7	8.0	8.2	7.9	7.7	8.4	--
pH Initial	7.2	7.2	7.2	7.0	7.6	7.8	7.2
Final	7.4	7.2	7.0	7.7	7.6	7.7	--
Alkalinity	30	NA	30	NA	31	NA	NA
Hardness	45	NA	48	NA	48	NA	NA
Conductivity	160	150	150	160	180	200	160
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
8 %							
D.O. Initial	8.0	7.6	8.0	8.2	7.5	6.9	7.6
Final	8.2	8.1	8.1	7.8	7.7	8.3	--
pH Initial	7.2	7.2	7.2	7.1	7.6	7.7	7.1
Final	7.4	7.2	7.0	7.7	7.5	7.8	--
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	180	170	170	170	190	200	170
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
11 %							
D.O. Initial	8.0	7.6	8.0	8.2	7.5	6.8	7.5
Final	8.6	8.1	8.0	7.8	7.7	8.4	--
pH Initial	7.2	7.2	7.2	7.2	7.6	7.7	7.1
Final	7.4	7.2	7.0	7.7	7.5	7.7	--
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	180	170	170	170	200	200	180
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
14 %							
D.O. Initial	8.2	7.5	8.0	8.1	7.6	6.8	7.2
Final	8.5	8.1	8.2	7.9	7.9	8.3	--
pH Initial	7.2	7.2	7.2	7.1	7.7	7.6	7.0
Final	7.4	7.2	7.0	7.7	7.6	7.7	--
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	190	180	180	180	200	210	180
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
19 %							
D.O. Initial	8.0	7.6	8.0	8.2	7.4	6.8	7.2
Final	8.4	7.8	7.9	7.8	7.8	8.4	--
pH Initial	7.2	7.2	7.2	7.2	7.7	7.6	7.0
Final	7.5	7.2	7.0	7.7	7.6	7.7	--
Alkalinity	38	NA	43	NA	39	NA	NA
Hardness	41	NA	52	NA	48	NA	NA
Conductivity	200	200	190	190	210	220	200
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
25 %							
D.O. Initial	8.2	7.8	8.0	8.2	7.4	6.7	7.5
Final	8.5	7.9	7.9	7.7	7.8	8.5	--
pH Initial	7.2	7.2	7.3	7.2	7.7	7.6	7.0
Final	7.5	7.2	7.1	7.8	7.6	7.8	--
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	210	200	200	200	230	240	210
Chlorine	NA	NA	NA	NA	NA	NA	NA

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: <u>SEARCS</u>			PO No.		NO OF BOTTLES	ANALYSES REQUESTED										AIC CONTROL NO: <u>184032</u>								
Project Reference:			MATRIX			Biomonitoring											AIC PROPOSAL NO:							
Project Manager: <u>Paul Abeworth</u>																	Carrier:							
Sampled By: <u>Johny Frank</u>			G	C	W	S											Received Temperature C <u>0.9 C</u>							
AIC No.	Sample Identification	Date/Time Collected	A	O	E	I	L											Remarks						
①	<u>ETP</u>	<u>5/20/10-27/14 U.A.P.A.</u> <u>5/27/10-28/14 U.A.P.A.</u>						2	✓															
Container Type															Field pH calibration									
Preservative															on _____ @ _____									
G = Glass			P = Plastic		V = VOA vials			H = HCl to pH2			T = Sodium Thiosulfate				A = (NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> , NH <sub>4</sub> OH									
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					Relinquished By: <u>[Signature]</u>					Date/Time <u>10-29-14</u> <u>6:00 AM</u>					Received By: <u>[Signature]</u>					Date/Time <u>10-29-14</u> <u>6:00 AM</u>				
Expedited results requested by: _____					Relinquished By: <u>[Signature]</u>					Date/Time <u>10-29-14</u> <u>9:40 AM</u>					Received in Lab By: <u>[Signature]</u>					Date/Time <u>10/29/14</u> <u>0940</u>				
Who should AIC contact with questions: Phone: _____ Fax: _____					Comments:																			
Report Attention to: Report Address to: <u>Searcy Water Util. Office</u> <u>P.O. Box 1319</u> <u>Searcy, MD 22145</u>																								



CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: <u>SEANCY</u>		PO No.		ANALYSES REQUESTED												AIC CONTROL NO: <u>184032</u>							
Project Reference:		MATRIX		<div style="display: flex; justify-content: space-between;"> <span>Bio-monitoring</span> <span>Pest</span> <span>BNA 675</span> </div>												AIC PROPOSAL NO:							
Project Manager: <u>Paul Asensio</u>		WATER SOIL														Carrier:							
Sampled By: <u>Tom Hartfield</u>		GRA B		Received Temperature C <u>19.2</u>																			
AIC No.		Sample Identification		Date/Time Collected		G		C		A		S		O		I		L		NO OF BOTTLES		Remarks	
②		<del>EFF</del> EFF		START 10-29-14 11:50 AM STOP 10-30-14 11:50 AM		✓		✓												2			
		<del>EFF</del> EFF		START 10-29-14 11:50 AM STOP 10-30-14 11:50 AM		✓		✓												3		ATE # 184131	
		<del>EFF</del> EFF		START 10-29-14 11:50 AM STOP 10-30-14 11:50 AM		✓		✓												3			
		INP START STOP		10-29-14 11:50 AM 10-30-14 11:50 AM		✓		✓												3			
		INP START STOP		10-29-14 11:50 AM 10-30-14 11:50 AM		✓		✓												3			
		Container Type		Preservative		P		M		B										NO		NO	
		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 <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> . NH <sub>4</sub> OH	
Turnaround Time Requested: (Please circle) NORMAL or EXPEDITED IN _____ DAYS				Relinquished By: <u>Tom Hartfield</u>				Date/Time <u>10-31-14</u> <u>6:00 AM</u>				Received By: <u>[Signature]</u>				Date/Time <u>10-31-14</u> <u>6:00 AM</u>							
Expedited results requested by: _____				Relinquished By: <u>[Signature]</u>				Date/Time <u>10-31-14</u> <u>9:07</u>				Received in Lab By: <u>[Signature]</u>				Date/Time <u>10/31/14</u> <u>9:07</u>							
Who should AIC contact with questions: Phone: _____ Fax: _____				Comments:																			
Report Attention to:																							
Report Address to:																							



CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: <u>Seawater</u>			PO No.		NO OF BOTTLES	ANALYSES REQUESTED										AIC CONTROL NO: <u>184032</u>								
Project Reference:			MATRIX			BOTTLES											AIC PROPOSAL NO:							
Project Manager: <u>Paul Abernethy</u>							BOTTLES											Carrier:						
Sampled By: <u>Johnny Paul</u>			GRA	COMP	WATER	SOIL		BOTTLES	BOTTLES	BOTTLES	BOTTLES	BOTTLES	BOTTLES	BOTTLES	BOTTLES	BOTTLES	BOTTLES	Received Temperature C <u>0.3°C</u>						
AIC No.	Sample Identification	Date/Time Collected																Remarks						
③	<u>FF</u>	<u>STATION 1-14 - 11/12/14 - Stop 11-2-14/11:45Z</u>					<u>2</u>	<u>✓</u>																
																	Field pH calibration on _____ @ _____ Buffer:							
			Container Type <u>P</u>																					
			Preservative <u>NO</u>																					
			G = Glass NO = none		P = Plastic S = Sulfuric acid pH2		V = VOA vials N = Nitric acid pH2		H = HCl to pH2 B = NaOH to pH12		T = Sodium Thiosulfate Z = Zinc acetate		A = (NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> , NH <sub>4</sub> OH											
Turnaround Time Requested: (Please circle) NORMAL or EXPEDITED IN _____ DAYS					Relinquished By: <u>JE</u>					Date/Time <u>11-3-14 6:00 AM</u>					Received By: <u>[Signature]</u>					Date/Time <u>11-3-14 6:00 AM</u>				
Expedited results requested by: _____					Relinquished By: <u>[Signature]</u>					Date/Time <u>11-3-14 9:04 AM</u>					Received in Lab By: <u>[Signature]</u>					Date/Time <u>03/NOV/14 0904</u>				
Who should AIC contact with questions: Phone: _____ Fax: _____					Report Attention to:					Report Address to: <u>Seawater UT. Lit. Inc. P.O. Box 1349 Seawater, AZ 85455</u>					Comments:									

December 18, 2014

Test Results of  
Fourth Quarter  
Chronic 7-Day Renewal  
Biomonitoring Testing  
for  
Outfall 001

Control No. 185554-1

Prepared for:

Mr. Paul Abernathy  
Searcy Water and Sewer System  
Post Office Box 1319  
Searcy, AR 72145

Prepared by:

AMERICAN INTERPLEX CORPORATION  
8600 Kanis Road  
Little Rock, AR 72204-2322



Searcy Water and Sewer System  
ATTN: Mr. Paul Abernathy  
Post Office Box 1319  
Searcy, AR 72145

Re: Chronic 7-Day Renewal utilizing *Pimephales promelas* (Fathead minnow)  
Outfall 001  
NPDES Permit No. AR0021601 AFIN# 73-00055

Dear Mr. Paul Abernathy:

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 laboratory director 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 25 % effluent, which is above the critical dilution of 19 %. Any statistical difference with sublethal effects cannot be considered toxic due to the minimum significant difference (PMSD) calculated result being below the lower PMSD bounds. **The sample, therefore PASSED both lethal and sub-lethal effects for the Fathead minnow test.**

AMERICAN INTERPLEX CORPORATION

A handwritten signature in black ink, appearing to read 'John Overbey', is written over a horizontal line.

John Overbey  
Laboratory Director

PDF cc: Searcy Water and Sewer System  
ATTN: Mr. Paul Abernathy  
pabernathy1@hotmail.com

Searcy Water and Sewer System  
ATTN: Mr. Dan Dawson  
d.dawson@cablelynx.com

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*Pimephales promelas* (Fathead minnow) Survival and Growth

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I. Control Acceptance Criteria

*Pimephales promelas* (Fathead minnow) Method 1000.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	97.5	PASS
Control Growth > or = 0.25 mg per Surviving minnow	0.275	PASS
Control Growth CV < or = 40%	8.95	PASS
Growth Minimum Significant Difference 12 to 30%	11.2	BELOW
Critical Dilution CV < or = 40%	6.03	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0021601 AFIN# 73-00055
2. Test Requirements: Chronic Biomonitoring, Quarterly  
Test Method 1000.0
3. Receiving Stream: Little Red River

B. Source of Effluent/Dilution Water

1. Effluent Samples:
  - a. Sampling Point: Outfall 001
  - b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.7	8.2	7.7
pH (standard units)	6.4	7.1	6.5
Alkalinity (mg/l as CaCO <sub>3</sub> )	33	39	40
Hardness (mg/l as CaCO <sub>3</sub> )	54	52	54
Conductivity (umhos/cm)	310	370	360
Residual Chlorine (mg/l)	<0.05	<0.05	<0.05
Ammonia as N (mg/l)	0.12	<0.1	0.36

2. Dilution Water Samples: Synthetic Soft Water #4166

- a. Dates Prepared: December 4 through December 18, 2014
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.9	8.7	8.5
pH (standard units)	7.4	7.4	7.5
Alkalinity (mg/l as CaCO <sub>3</sub> )	31	31	31
Hardness (mg/l as CaCO <sub>3</sub> )	48	48	48
Conductivity (umhos/cm)	150	160	170
Residual Chlorine (mg/l)	<0.05	<0.05	<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 Method 1000.0, Fathead Minnow Survival and Growth.

2. Endpoint: No Observable Effects Concentration (NOEC)

3. Test Conditions:

*Pimephales promelas* (Fathead minnow) Survival and Growth Method 1000.0

Date & Time Test Initiated: December 9, 2014 at 1627  
Date & Time Test Terminated: December 17, 2014 at 1500  
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

4. Acclimation 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*

III. Data Analysis

The data was analyzed using American Interplex Corporation's Laboratory Information Management Software based on Toxstat.

*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.

IV. Standard Reference Toxicants

American Interplex Corporation has an ongoing test organism culturing program. The sensitivity of the offspring is determined by performing a standard reference toxicant test with each effluent test. Sodium chloride in synthetic moderately hard water is used as prescribed in EPA-821-R-02-013.

*Pimephales promelas* (Fathead minnow)

Chronic reference tests are performed monthly.

A chronic reference test was performed on November 18, 2014 at 1600 to November 25, 2014 at 1415

The results were as follows: (Control No. 184773-1.)

Survival LC-50: 4807 mg/l

Growth IC-25: 3075 mg/l

Growth PMSD: 13.5

V. Chemical Analysis/Quality Control

Parameter	Method	% Recovery	Relative % Difference
Alkalinity	SM 2320 B	NA	0.00
Hardness	EPA 200.7	101	1.50
pH	SM 4500-H+ B	101	0.268
Conductivity	EPA 120.1	104	2.65

VI. Organism History

*Pimephales promelas* (Fathead minnow)

Date: December 9, 2014

Age: <24 hours

Source: In-house culture

Water Chemistry Record:

Alkalinity: 57-64 mg/l

Hardness: 80-100 mg/l

Temperature: 25 deg.C

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

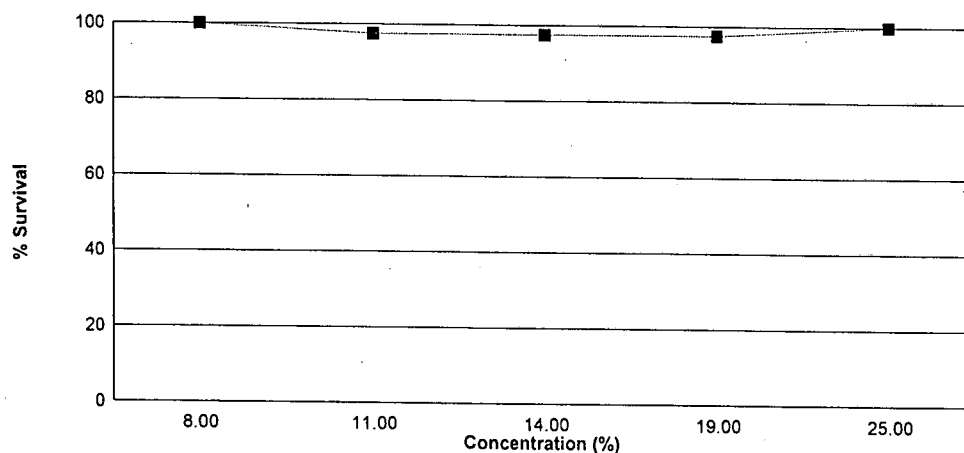
Larvae are exposed in a static renewal system for seven days to different concentrations of effluent with dilution water. Test results are based on the survival and growth (increase in weight) of the larvae.

Effluent dilutions for this test were 8 %, 11 %, 14 %, 19 %, 25 % in accordance with the NPDES permit.

The low flow or 'critical' dilution is specified in the NPDES permit as 19 % effluent.

The test was initiated on December 9, 2014 at 1627 and continued through December 17, 2014 at 1500. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

- a.) NOEC survival = 25 % effluent
- b.) NOEC growth = 25 % effluent



Summary of the 7-day Fathead Minnow Survival and Growth		
Concentration	Percent Survival	Mean Growth (mg)
Control	97.5	0.268
8 %	100	0.248
11 %	97.5	0.242
14 %	97.5	0.259
19 %	97.5	0.244
25 %	100	0.245



Appendix A1: Test 1000.0

*Pimephales promelas* (Fathead Minnow) 7-Day Survival

Date and Time Test Initiated: December 9, 2014 at 1627  
Date and Time Test Terminated: December 17, 2014 at 1500

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	7	7	7	7	7	7
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
8 %	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
11 %	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	7	7	7	7	7	7	7
	E	8	8	8	8	8	8	8
14 %	A	8	8	8	8	8	8	8
	B	8	7	7	7	7	7	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
19 %	A	8	8	8	8	8	8	8
	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
25 %	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: December 9, 2014 at 1627  
Test Terminated: December 17, 2014 at 1500

Drying Started: December 17, 2014 at 0939  
Drying Ended: December 18, 2014 at 1200

Concentration	Replicate	Weight of pan	Weight of pan + fish	Total weight of fish (g)	Original # of fish	Mean dry weight (mg)
Control	A	.93723	.93907	0.00184	8	0.230
	B	.93611	.93820	0.00209	8	0.261
	C	.93825	.94053	0.00228	8	0.285
	D	.93498	.93717	0.00219	8	0.274
	E	.93720	.93952	0.00232	8	0.290
8 %	A	.93394	.93576	0.00182	8	0.228
	B	.93525	.93741	0.00216	8	0.270
	C	.93705	.93893	0.00188	8	0.235
	D	.93757	.93943	0.00186	8	0.232
	E	.93810	.94031	0.00221	8	0.276
11 %	A	.94300	.94487	0.00187	8	0.234
	B	.94152	.94322	0.00170	8	0.212
	C	.94220	.94403	0.00183	8	0.229
	D	.93617	.93840	0.00223	8	0.279
	E	.93723	.93926	0.00203	8	0.254
14 %	A	.93839	.94051	0.00212	8	0.265
	B	.93425	.93657	0.00232	8	0.290
	C	.93364	.93551	0.00187	8	0.234
	D	.92996	.93197	0.00201	8	0.251
	E	.93833	.94037	0.00204	8	0.255
19 %	A	.94089	.94303	0.00214	8	0.268
	B	.93904	.94094	0.00190	8	0.238
	C	.93857	.94042	0.00185	8	0.231
	D	.93087	.93276	0.00189	8	0.236
	E	.93590	.93789	0.00199	8	0.249
25 %	A	.93300	.93493	0.00193	8	0.241
	B	.93984	.94181	0.00197	8	0.246
	C	.94092	.94292	0.00200	8	0.250
	D	.94086	.94284	0.00198	8	0.248
	E	.93556	.93747	0.00191	8	0.239

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	0.87500	1.20940
1	Control	4	1.00000	1.39310
1	Control	5	1.00000	1.39310
2	8 %	1	1.00000	1.39310
2	8 %	2	1.00000	1.39310
2	8 %	3	1.00000	1.39310
2	8 %	4	1.00000	1.39310
2	8 %	5	1.00000	1.39310
3	11 %	1	1.00000	1.39310
3	11 %	2	1.00000	1.39310
3	11 %	3	1.00000	1.39310
3	11 %	4	0.87500	1.20940
3	11 %	5	1.00000	1.39310
4	14 %	1	1.00000	1.39310
4	14 %	2	0.87500	1.20940
4	14 %	3	1.00000	1.39310
4	14 %	4	1.00000	1.39310
4	14 %	5	1.00000	1.39310
5	19 %	1	1.00000	1.39310
5	19 %	2	0.87500	1.20940
5	19 %	3	1.00000	1.39310
5	19 %	4	1.00000	1.39310
5	19 %	5	1.00000	1.39310
6	25 %	1	1.00000	1.39310
6	25 %	2	1.00000	1.39310
6	25 %	3	1.00000	1.39310
6	25 %	4	1.00000	1.39310
6	25 %	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 = 0.108		
W = 0.5958		
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	8 %	30.00	16.00	5.00	
3	11 %	27.50	16.00	5.00	
4	14 %	27.50	16.00	5.00	
5	19 %	27.50	16.00	5.00	
6	25 %	30.00	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 = 0.009698 W = 0.9782 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 = 8.707 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	0.002656	0.0005312	1.315
Within (Error)	24	0.009694	0.0004039	
Total	29	0.01235		
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	0.268	0.268		
2	8 %	0.2482	0.2482	1.558	
3	11 %	0.2416	0.2416	2.077	
4	14 %	0.259	0.259	0.7081	
5	19 %	0.2444	0.2444	1.857	
6	25 %	0.2448	0.2448	1.825	
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	8 %	5	0.03	11.2	0.0198
3	11 %	5	0.03	11.2	0.0264
4	14 %	5	0.03	11.2	0.009
5	19 %	5	0.03	11.2	0.0236
6	25 %	5	0.03	11.2	0.0232

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: December 9, 2014 at 1039  
Date and Time Test Terminated: December 16, 2014 at 1500

Effluent Conc.: Control		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.9	8.0	8.7	8.6	8.5	8.4	8.6
	Final	7.0	7.6	7.5	7.7	7.7	7.6	7.3
pH, units	Initial	7.4	7.4	7.4	7.3	7.5	7.4	7.2
	Final	7.2	7.4	7.5	7.5	7.4	7.4	7.2
Alkalinity, mg CaCO <sub>3</sub> /l		31	NA	31	NA	31	NA	NA
Hardness, mg CaCO <sub>3</sub> /l		48	NA	48	NA	48	NA	NA
Conductivity, umhos/cm		150	180	160	170	170	160	170
Res. Chlorine, mg/l		<0.05	NA	<0.05	NA	<0.05	NA	NA

Effluent Conc.: 8 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.9	8.1	8.6	8.6	8.4	8.4	8.5
	Final	7.0	7.2	7.3	8.1	7.7	7.8	7.5
pH, units	Initial	7.2	7.3	7.3	7.2	7.4	7.3	7.2
	Final	7.1	7.2	7.3	7.5	7.4	7.4	7.3

Effluent Conc.: 11 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.9	8.3	8.6	8.6	8.4	8.5	8.5
	Final	7.4	7.5	7.5	8.0	7.7	7.4	7.1
pH, units	Initial	7.1	7.3	7.3	7.2	7.4	7.2	7.2
	Final	7.2	7.3	7.5	7.5	7.4	7.4	7.3

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: December 9, 2014 at 1039

Date and Time Test Terminated: December 16, 2014 at 1500

Effluent Conc.: 14 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.8	8.2	8.5	8.6	8.5	8.3	8.2
	Final	6.9	7.3	7.1	8.0	7.7	7.5	7.5
pH, units	Initial	7.1	7.3	7.2	7.2	7.4	7.2	7.1
	Final	7.1	7.3	7.4	7.5	7.4	7.4	7.4

Effluent Conc.: 19 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.4	7.9	8.6	8.7	8.5	8.4	8.0
	Final	6.9	7.3	7.1	8.0	7.7	7.4	7.5
pH, units	Initial	7.0	7.2	7.2	7.2	7.3	7.2	7.1
	Final	7.1	7.2	7.4	7.5	7.4	7.4	7.3
Alkalinity, mg CaCO <sub>3</sub> /l		33	NA	34	NA	33	NA	NA
Hardness, mg CaCO <sub>3</sub> /l		51	NA	50	NA	49	NA	NA
Conductivity, umhos/cm		180	210	190	200	200	200	190
Res. Chlorine, mg/l		<0.05	NA	<0.05	NA	<0.05	NA	NA

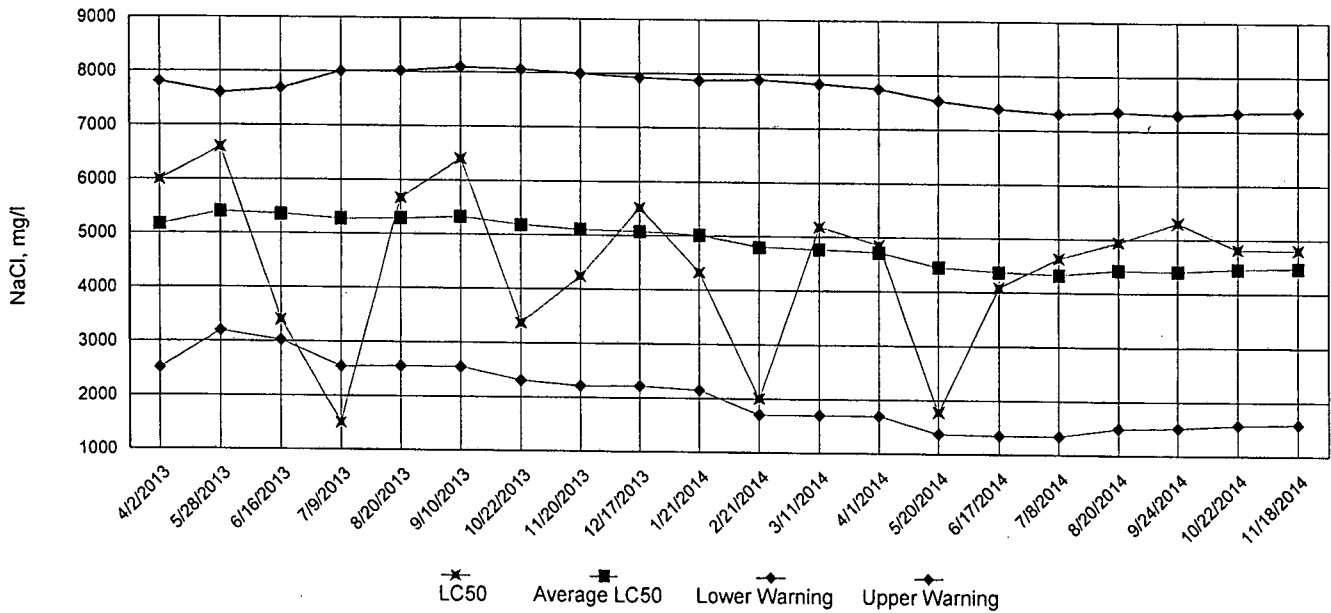
Effluent Conc.: 25 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.3	8.2	8.6	9.0	8.6	8.4	8.5
	Final	6.9	7.2	7.2	7.8	7.8	7.6	7.2
pH, units	Initial	6.9	7.2	7.2	7.1	7.3	7.1	7.1
	Final	7.1	7.2	7.3	7.5	7.4	7.4	7.2



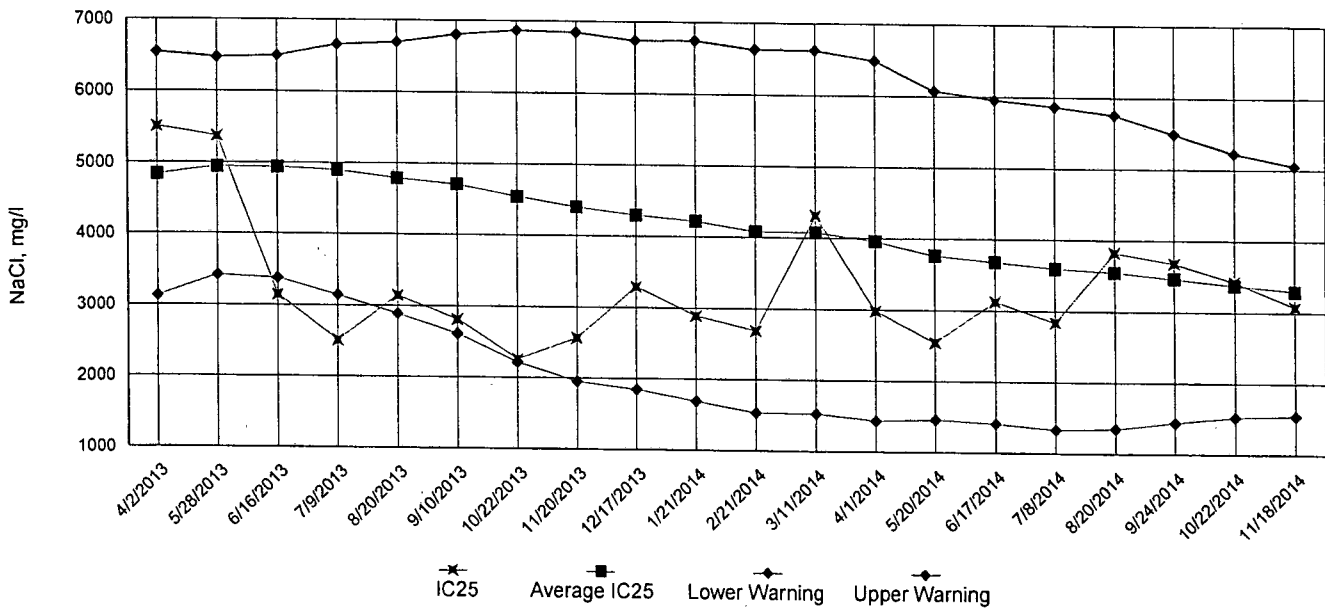
Appendix A4: Test 1000.0

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

LC50 Survival Data



IC25 Growth Data



Appendix B: Test 1000.0

SUMMARY REPORTING FORMS  
CHRONIC BIOMONITORING  
*Pimephales promelas* (Fathead Minnow)  
SURVIVAL AND GROWTH

Permittee: Searcy Water and Sewer System

NPDES No.: AR0021601 AFIN# 73-00055

Date and Time Test Initiated: December 9, 2014 at 1627

Date and Time Test Terminated: December 17, 2014 at 1500

Dilution water used: Synthetic Soft Water #4166

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	87.5	100	100	100	97.5	97.5	5.73
8 %	100	100	100	100	100	100	100	100	0.00
11 %	100	100	100	87.5	100	97.5	97.5	97.5	5.73
14 %	100	87.5	100	100	100	100	97.5	97.5	5.73
19 %	100	87.5	100	100	100	100	100	97.5	5.73
25 %	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	0.230	0.261	0.285	0.274	0.290	0.268	8.95
8 %	0.228	0.270	0.235	0.232	0.276	0.248	9.22
11 %	0.234	0.212	0.229	0.279	0.254	0.242	10.6
14 %	0.265	0.290	0.234	0.251	0.255	0.259	7.96
19 %	0.268	0.238	0.231	0.236	0.249	0.244	6.03
25 %	0.241	0.246	0.250	0.248	0.239	0.245	1.90

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	(19 %)	<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	(19 %)	<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:   25 %   (TOP6C)
6. LOEC *Pimephales* Lethality:   25 %   (TXP6C)
7. NOEC *Pimephales* Sublethality:   25 %   (TPP6C)
8. LOEC *Pimephales* Sublethality:   25 %   (TYP6C)
9. Coefficient of variation for *Pimephales* growth:   8.95   (TQP6C)

Appendix B: Test 1000.0

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

PERMITTEE: Searcy Water and Sewer System SAMPLE No. 1 COLLECTED ending: DATE: December 9, 2014 TIME: 1140  
 NPDES NO.: AR0021601 AFIN# 73-00055 SAMPLE No. 2 COLLECTED ending: DATE: December 11, 2014 TIME: 1140  
 CONTACT: Mr. Paul Abernathy SAMPLE No. 3 COLLECTED ending: DATE: December 14, 2014 TIME: 1145  
 ANALYST: 280, 304, 310 Test Initiated: DATE: December 9, 2014 TIME: 1627  
 Test Terminated: DATE: December 17, 2014 TIME: 1500

DILUTION Control	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.9	8.0	8.7	8.6	8.5	8.4	8.6
Final	7.0	7.6	7.5	7.7	7.7	7.6	7.3
pH Initial	7.4	7.4	7.4	7.3	7.5	7.4	7.2
Final	7.2	7.4	7.5	7.5	7.4	7.4	7.2
Alkalinity	31	NA	31	NA	31	NA	NA
Hardness	48	NA	48	NA	48	NA	NA
Conductivity	150	180	160	170	170	160	170
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION 8 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.9	8.1	8.6	8.6	8.4	8.4	8.5
Final	7.0	7.2	7.3	8.1	7.7	7.8	7.5
pH Initial	7.2	7.3	7.3	7.2	7.4	7.3	7.2
Final	7.1	7.2	7.3	7.5	7.4	7.4	7.3
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	160	190	180	180	180	170	180
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 11 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.9	8.3	8.6	8.6	8.4	8.5	8.5
Final	7.4	7.5	7.5	8.0	7.7	7.4	7.1
pH Initial	7.1	7.3	7.3	7.2	7.4	7.2	7.2
Final	7.2	7.3	7.5	7.5	7.4	7.4	7.3
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	170	200	180	180	180	180	180
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 14 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.8	8.2	8.5	8.6	8.5	8.3	8.2
Final	6.9	7.3	7.1	8.0	7.7	7.5	7.5
pH Initial	7.1	7.3	7.2	7.2	7.4	7.2	7.1
Final	7.1	7.3	7.4	7.5	7.4	7.4	7.4
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	170	200	180	190	190	180	180
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 19 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.4	7.9	8.6	8.7	8.5	8.4	8.0
Final	6.9	7.3	7.1	8.0	7.7	7.4	7.5
pH Initial	7.0	7.2	7.2	7.2	7.3	7.2	7.1
Final	7.1	7.2	7.4	7.5	7.4	7.4	7.3
Alkalinity	33	NA	34	NA	33	NA	NA
Hardness	51	NA	50	NA	49	NA	NA
Conductivity	180	210	190	200	200	200	190
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION 25 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.3	8.2	8.6	9.0	8.6	8.4	8.5
Final	6.9	7.2	7.2	7.8	7.8	7.6	7.2
pH Initial	6.9	7.2	7.2	7.1	7.3	7.1	7.1
Final	7.1	7.2	7.3	7.5	7.4	7.4	7.2
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	190	220	200	210	220	200	210
Chlorine	NA	NA	NA	NA	NA	NA	NA



**CHAIN OF CUSTODY / ANALYSIS REQUEST FORM**

Client: <i>Seary</i>			PO No.		<b>NO OF BOTTLES</b>	<b>ANALYSES REQUESTED</b>												AIC CONTROL NO: <i>185554</i>						
Project			MATRIX			<i>Biomonitoring</i>													AIC PROPOSAL NO:					
Reference:							<i>Biomonitoring</i>													Carrier:				
Project Manager: <i>Paul Oberlin</i>								<i>Biomonitoring</i>													Received Temperature C <i>3.3</i>			
Sampled By: <i>Johnny Sauter</i>			G	R	A				B	W	A	T	E	R	S	O	I	L	Remarks					
AIC No.			Sample Identification			Date/Time Collected																		
<i>(1)</i>							<i>11-24-12-8-11/11.4A</i>																	
				<i>Stop 12-5-14/11.9A</i>																				
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=(NH4)2SO4, NH4OH														
Turnaround Time Requested: (Please circle) NORMAL or EXPEDITED IN ____ DAYS					Relinquished By: <i>Johnny Sauter</i>					Date/Time <i>12-10-14</i> <i>6:00 AM</i>					Received By: <i>Raymond Dy</i>					Date/Time <i>12-10-14</i> <i>6:00 AM</i>				
Expedited results requested by: _____					Relinquished By: <i>Raymond Dy</i>					Date/Time <i>12-10-14</i> <i>8:52 AM</i>					Received in Lab By: <i>Troy Williams</i>					Date/Time <i>12-10-14</i> <i>8:52</i>				
Who should AIC contact with questions: Phone: _____ Fax: _____					Comments:																			
Report Attention to: Report Address to: <i>Seary Water Utilities</i> <i>P.O. Box 1319</i> <i>Seary AR 72145</i>																								
Email Address: _____																								



CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: <u>Seaway</u>			PO No.		NO OF BOTTLES	ANALYSES REQUESTED										AIC CONTROL NO: <u>185554</u>				
Project Reference:			MATRIX			BOTTLES											AIC PROPOSAL NO:			
Project Manager: <u>Paul Abernethy</u>			G R A B	C O M P	W A T E R		S O I L	BOTTLES											Carrier:	
Sampled <u>Galaxy South</u>																Received Temperature C <u>20°C</u>				
By:													Remarks							
AIC No.	Sample Identification	Date/Time Collected																		
(2)	<u>EFF</u>	<u>STAN 12-10-14 / 11:40 AM</u> <u>STAP 12-11-14 / 11:40 AM</u>		<input checked="" type="checkbox"/>			2	<input checked="" type="checkbox"/>												
Container Type													Field pH calibration							
Preservative													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			A = (NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> , NH <sub>4</sub> OH					
Turnaround Time Requested: (Please circle) NORMAL or EXPEDITED IN _____ DAYS						Relinquished By: <u>Galaxy South</u>		Date/Time <u>12-12-14</u> <u>6:00 AM</u>		Received By: <u>Lynn Brewer</u>		Date/Time <u>12-12-14</u> <u>6:00 AM</u>								
Expedited results requested by: _____						Relinquished By: <u>Lynn Brewer</u>		Date/Time <u>12-12-14</u> <u>9:20 AM</u>		Received in Lab By: <u>Jimmy Day</u>		Date/Time <u>12/12/14</u> <u>0920</u>								
Who should AIC contact with questions: Phone: _____ Fax: _____						Comments:														
Report Attention to: Report Address to: <u>Seaway Water Utilities</u> <u>P.O. Box 12A</u> <u>Seaway, WA 99214</u>																				
Email Address: _____																				



CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client:			PO No.		NO OF BOTTLES	ANALYSES REQUESTED												AIC CONTROL NO:			
Project Reference:																		185554			
Project Manager:			MATRIX		BOTTLES													AIC PROPOSAL NO:			
Sampled By:			GRA	COMP														Carrier:			
AIC No.	Sample Identification	Date/Time Collected			WATER	SOIL													Received Temperature C		
③	Plant EFF	12-13-14 11:45pm 12-14-14 11:42pm		✓			2	✓	Bio-Munitals												4.4
																					Remarks
																					Field pH calibration
			Container Type																		on _____ @ _____
			Preservative																		Buffer:
			G = Glass NO = none	P = Plastic S = Sulfuric acid pH2	V = VOA vials N = Nitric acid pH2	H = HCl to pH2 B = NaOH to pH12	T = Sodium Thiosulfate Z = Zinc acetate													A=(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> , NH <sub>4</sub> OH	
Turnaround Time Requested: (Please circle) NORMAL or EXPEDITED IN ___ DAYS					Relinquished By: <i>Raymond J. Williams</i>				Date/Time: 12-15-14/6:00AM				Received By: <i>Raymond J. Williams</i>				Date/Time: 12-15-14 6:00AM				
Expedited results requested by: _____					Relinquished By: <i>Raymond J. Williams</i>				Date/Time: 12-15-14/9:00AM				Received in Lab By: <i>TROY WILLIAMS</i>				Date/Time: 12-15-14				
Who should AIC contact with questions: _____					Comments:																
Phone: _____ Fax: _____																					
Report Attention to: _____																					
Report Address to: <i>Searcy Water Utilities P.O. Box 1319</i>																					
Email Address: <i>Searcy, AIC 7145</i>																					

PLACE STICKER AT TOP OF ENVELOPE TO THE RIGHT OF THE RETURN ADDRESS, FOLD AT DOTTED LINE

**CERTIFIED MAIL™**



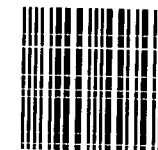
7010 2780 0002 5135 3374

Searcy, Water, Mill  
300  
SEA



UNITED STATES  
POSTAL SERVICE

1000



72118

U.S. POSTAGE  
PAID  
SEARCY, AR  
72143  
JAN 16, 15  
AMOUNT

**\$9.08**

00051413-05

RETURN RECEIPT  
REQUESTED

NPDES Enforcement Division  
A.D.E.Q.  
5301 Northshore Dr.  
North Little Rock, AR 72118-5317