

El Dorado Water Utilities

500 NORTH WASHINGTON □ P.O. BOX 1587 □ EL DORADO, AR 71731 (870) 862-6451

July 24, 2013

ADEQ Permits Branch, Water Division
5301 Northshore Drive
North Little Rock, AR 72118

Certified Mail: 7008 1300 0002 4985 9297

RE: NPDES Permit Numbers AR0033723, AR0033936, AR0049743 & AR0050296

Dear Ms. Bolenbaugh:

Attached are the completed Discharge Monitoring Reports and SSO Report for the above referenced permits covering the period April 1, 2013 through June 30, 2013 and the SSO report for March 1, 2013 through March 31, 2013.

Also included are the results of the WET retests for the south plant collected in April, May and June 2013.

In reference to the Fathead minnow lethal failure experienced at the South Plant in June, 2013 please be advised of the following:

1. Most of the problem occurred with the first day's sample, which may indicate some kind of container contamination.
2. The sample had very little hardness. This could result in the minnows not doing well early in the test while acclimating to the extremely soft effluent.
3. The first required retest is now underway.
- 4.

If you have any questions or comments, you can contact me at 870 862-6451.

Sincerely,



T. Harold Baker
Treatment Superintendent

Enclosures

May 9, 2013

Test Results of
Second Quarter
Chronic 7-Day Renewal
Biomonitoring Testing
for
South Effluent
El Dorado, AR

Control No. 166953-1

Prepared for:

Mr. Harold Baker
El Dorado Water Utilities
Post Office Box 1587
El Dorado, AR 71731

Prepared by:

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



El Dorado Water Utilities
ATTN: Mr. Harold Baker
Post Office Box 1587
El Dorado, AR 71731

Re: Chronic 7-Day Renewal utilizing *Ceriodaphnia dubia*
South Effluent - El Dorado, AR
NPDES Permit No. AR0033723 AFIN No. 70-00341

Dear Mr. Harold Baker:

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 1002.0 Chronic *Ceriodaphnia dubia* Survival and Reproduction 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 reproduction 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 *Ceriodaphnia dubia* 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: El Dorado Water Utilities
ATTN: Mr. Harold Baker
harold@eldoradowater.com

El Dorado Water Utilities
ATTN: Mr. John Peppers
lab@eldoradowater.com

GBMc & Associates, Inc.
ATTN: Mr. Roland McDaniel
rmcdaniel@gbmcassoc.com

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I. Control Acceptance Criteria

Ceriodaphnia dubia Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	17.1	PASS
Control CV < or = 40% per Surviving Female	23.9	PASS
Reproduction Minimum Significant Difference 13 to 47%	22.1	PASS
Critical Dilution CV < or = 40%	16.1	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0033723 AFIN No. 70-00341
2. Test Requirements: Chronic Biomonitoring, Quarterly Test Method 1002.0
3. Receiving Stream: Bayou de Loutre

B. Source of Effluent/Dilution Water

1. Effluent Samples:

- a. Sampling Point: South Effluent
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.0	7.9	8.0
pH (standard units)	7.2	7.4	7.6
Alkalinity (mg/l as CaCO ₃)	61	55	51
Hardness (mg/l as CaCO ₃)	27	26	28
Conductivity (umhos/cm)	730	740	720
Residual Chlorine (mg/l)	<0.05	<0.05	<0.05
Ammonia as N (mg/l)	7.1	4.5	3.6

2. Dilution Water Samples: Synthetic Soft Water #3984

- a. Dates Prepared: April 29 through May 13, 2013
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.1	7.8	7.9
pH (standard units)	7.6	7.8	8.0
Alkalinity (mg/l as CaCO ₃)	31	31	31
Hardness (mg/l as CaCO ₃)	41	43	44
Conductivity (umhos/cm)	160	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 1002.0, *Ceriodaphnia dubia* Survival and Reproduction.

2. Endpoint: No Observable Effects Concentration (NOEC)

3. Test Conditions:

Ceriodaphnia dubia Survival and Growth Method 1002.0

Date & Time Test Initiated: April 30, 2013 at 1320
Date & Time Test Terminated: May 7, 2013 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 1002.0 *Ceriodaphnia dubia*

III. Data Analysis

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

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

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.

Ceriodaphnia dubia

Chronic reference tests are performed monthly.

A chronic reference test was performed on April 2, 2013 at 1510 to April 9, 2013 at 1500

The results were as follows: (Control No. 166187-2.)

Survival LC-50: 1732 mg/l

Growth IC-25: 1250 mg/l

Growth PMSD: 8.05

V. Chemical Analysis/Quality Control

Parameter	Method	% Recovery	Relative % Difference
Alkalinity	SM 2320 B	NA	0.00
Hardness	EPA 200.7	99.5	0.152
pH	SM 4500-H+ B	101	0.134
Conductivity	EPA 120.1	105	0.660

VI. Organism History

Ceriodaphnia dubia

Date: April 30, 2013

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 *Ceriodaphnia dubia*, Cladoceran Survival and Reproduction Test -- Method 1002.0

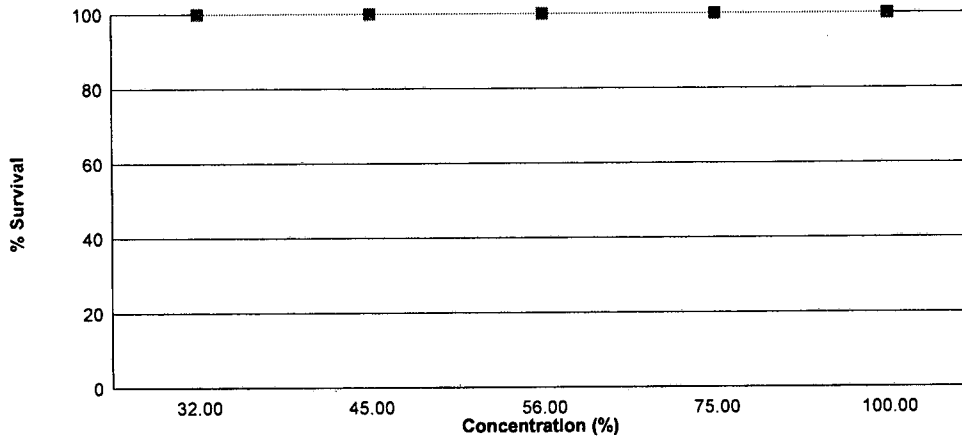
Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring with an average of at least 15 young per female.

Effluent dilutions for this test were 32 %, 45 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

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

The test was initiated on April 30, 2013 at 1320 and continued through May 7, 2013 at 1300. 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



Concentration	Percent Survival	Mean Reproduction
Control	100	17.1
32 %	100	16.9
45 %	100	17.5
56 %	100	17.5
75 %	100	17.3
100 %	100	18.3

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: April 30, 2013 at 1320

Date and Time Test Terminated: May 7, 2013 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	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	0	0	1	2	2	4	2	4	2	3	20	10	2.00	
5	4	5	3	4	4	5	7	6	3	5	46	10	4.60	
6	5	6	0	0	0	0	0	0	0	0	11	10	1.10	
7	0	10	12	11	9	10	12	13	9	8	94	10	9.40	
8														
TOTAL	9	21	16	17	15	19	21	23	14	16	171	10	17.1	

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	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	2	2	2	4	4	2	2	4	2	3	27	10	2.70	
5	4	4	4	0	8	4	6	6	5	4	45	10	4.50	
6	0	0	0	4	0	0	0	0	0	0	4	10	0.400	
7	12	10	8	12	11	9	0	8	12	11	93	10	9.30	
8														
TOTAL	18	16	14	20	23	15	8	18	19	18	169	10	16.9	

Concentration: 45 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	0	0	2	2	0	2	4	2	3	4	19	10	1.90	
5	5	5	4	3	5	4	6	5	6	7	50	10	5.00	
6	5	4	0	0	6	0	0	0	0	0	15	10	1.50	
7	0	12	11	8	13	9	12	8	8	10	91	10	9.10	
8														
TOTAL	10	21	17	13	24	15	22	15	17	21	175	10	17.5	

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: April 30, 2013 at 1320

Date and Time Test Terminated: May 7, 2013 at 1300

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	0	0	0	0	0	0	0	0	0	0	10	0.00
4	2	2	0	2	4	0	4	4	2	2	22	10	2.20
5	4	4	5	3	8	5	8	7	6	5	55	10	5.50
6	0	0	6	0	0	4	0	0	0	0	10	10	1.00
7	12	9	10	11	8	0	7	9	12	10	88	10	8.80
8													
TOTAL	18	15	21	16	20	9	19	20	20	17	175	10	17.5

Concentration: 75 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	0	4	0	2	2	2	4	2	3	3	22	10	2.20
5	4	0	4	3	4	4	6	4	6	8	43	10	4.30
6	8	6	5	0	0	0	0	0	0	0	19	10	1.90
7	0	9	8	12	13	9	8	10	11	9	89	10	8.90
8													
TOTAL	12	19	17	17	19	15	18	16	20	20	173	10	17.3

Concentration: 100 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	4	2	4	2	5	5	4	4	4	4	38	10	3.80
5	0	4	6	4	0	0	6	8	6	7	41	10	4.10
6	5	0	0	0	4	7	0	0	0	0	16	10	1.60
7	10	12	10	9	11	9	8	0	12	7	88	10	8.80
8													
TOTAL	19	18	20	15	20	21	18	12	22	18	183	10	18.3

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
45 %	10	0	10
Total	20	0	20

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

Fisher's Exact Test

Identification	Alive	Dead	Total Animals
Control	10	0	10
56 %	10	0	10
Total	20	0	20

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

Fisher's Exact Test

Identification	Alive	Dead	Total Animals
Control	10	0	10
75 %	10	0	10
Total	20	0	20

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

Appendix A2: Statistics

Ceriodaphnia dubia Survival

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

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

Summary of Fisher's Exact Test				
Group	Identification	Exposed	Dead	Sig 0.05
0	Control	10	0	
1	32 %	10	0	
2	45 %	10	0	
3	56 %	10	0	
4	75 %	10	0	
5	100 %	10	0	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

Kolmogorov Test for Normality	No Transformation
D = 0.0943 D* = 0.7398 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 = 3.835 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	11.73	2.346	0.1752
Within (Error)	54	723	13.39	
Total	59	734.7		
Critical F = 3.38 (alpha = 0.01, df = 5,54)				
2.38 (alpha = 0.05, df = 5,54)				
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	17.1	17.1		
2	32 %	16.9	16.9	0.1222	
3	45 %	17.5	17.5	-0.2444	
4	56 %	17.5	17.5	-0.2444	
5	75 %	17.3	17.3	-0.1222	
6	100 %	18.3	18.3	-0.7333	
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	3.78	22.1	0.2
3	45 %	10	3.78	22.1	-0.4
4	56 %	10	3.78	22.1	-0.4
5	75 %	10	3.78	22.1	-0.2
6	100 %	10	3.78	22.1	-1.2

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: April 30, 2013 at 0844

Date and Time Test Terminated: May 7, 2013 at 1300

Effluent Conc.: Control	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.1	7.7	7.8	8.1	7.9	7.8	8.1
	Final	8.0	7.9	7.9	7.7	7.9	7.6	8.2
pH, units	Initial	7.6	7.8	7.8	7.9	8.0	7.6	7.6
	Final	7.9	7.6	7.5	7.7	8.0	6.4	8.0
Alkalinity, mg CaCO ₃ /l	31	NA	31	NA	31	NA	NA	
Hardness, mg CaCO ₃ /l	41	NA	43	NA	44	NA	NA	
Conductivity, umhos/cm	160	160	160	160	170	180	160	
Res. Chlorine, mg/l	<0.05	NA	<0.05	NA	<0.05	NA	NA	

Effluent Conc.: 32 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	7.8	7.7	7.9	8.3	7.9	7.8	8.0
	Final	7.8	7.8	8.0	7.8	7.7	7.8	8.0
pH, units	Initial	7.4	7.6	7.6	7.6	7.8	7.7	7.5
	Final	7.8	7.6	7.5	7.5	7.8	6.7	7.8

Effluent Conc.: 45 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.1	7.7	7.9	8.1	7.7	7.8	7.8
	Final	7.9	7.9	8.0	7.7	7.8	7.8	8.1
pH, units	Initial	7.3	7.6	7.5	7.6	7.7	7.7	7.5
	Final	7.7	7.5	7.6	7.6	7.7	6.9	7.8

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: April 30, 2013 at 0844

Date and Time Test Terminated: May 7, 2013 at 1300

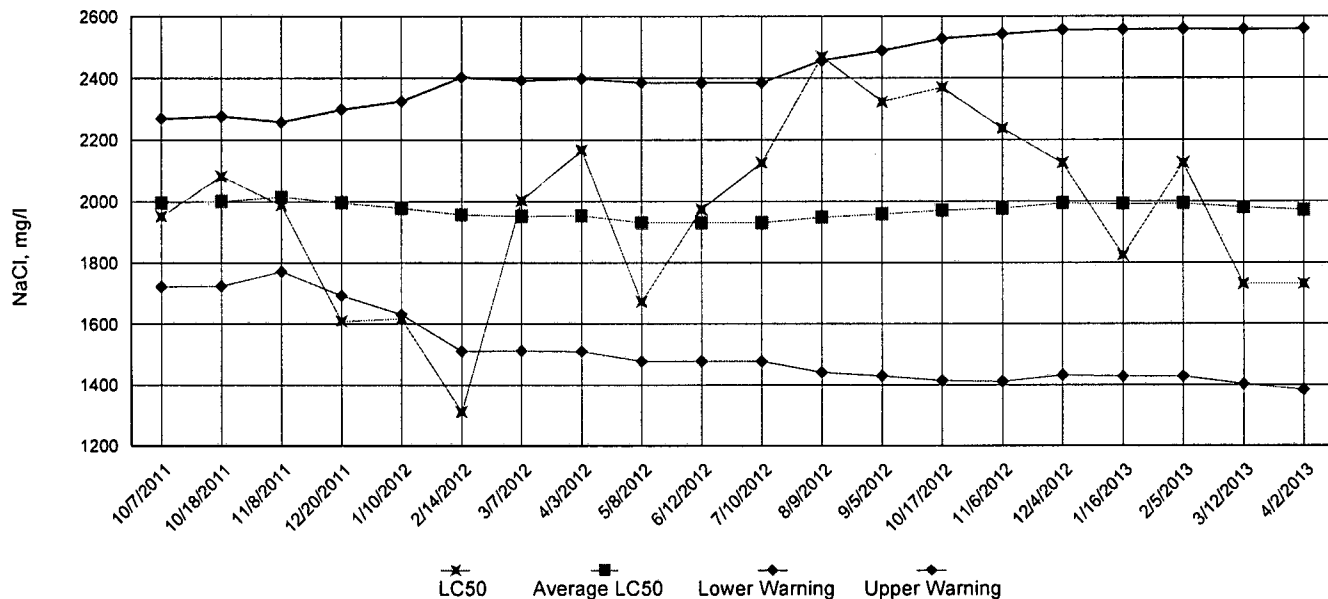
Effluent Conc.: 56 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.2	7.9	7.8	8.1	7.6	7.7	7.9
	Final	7.9	7.9	8.0	7.6	7.7	7.6	8.2
pH, units	Initial	7.3	7.5	7.5	7.5	7.7	7.7	7.4
	Final	7.7	7.5	7.6	7.7	7.7	7.0	7.7

Effluent Conc.: 75 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.0	7.7	7.8	8.1	7.7	7.6	7.9
	Final	7.8	7.7	8.1	7.9	8.0	7.7	8.0
pH, units	Initial	7.2	7.6	7.4	7.4	7.7	7.8	7.4
	Final	7.7	7.5	7.7	7.7	7.8	7.1	7.8

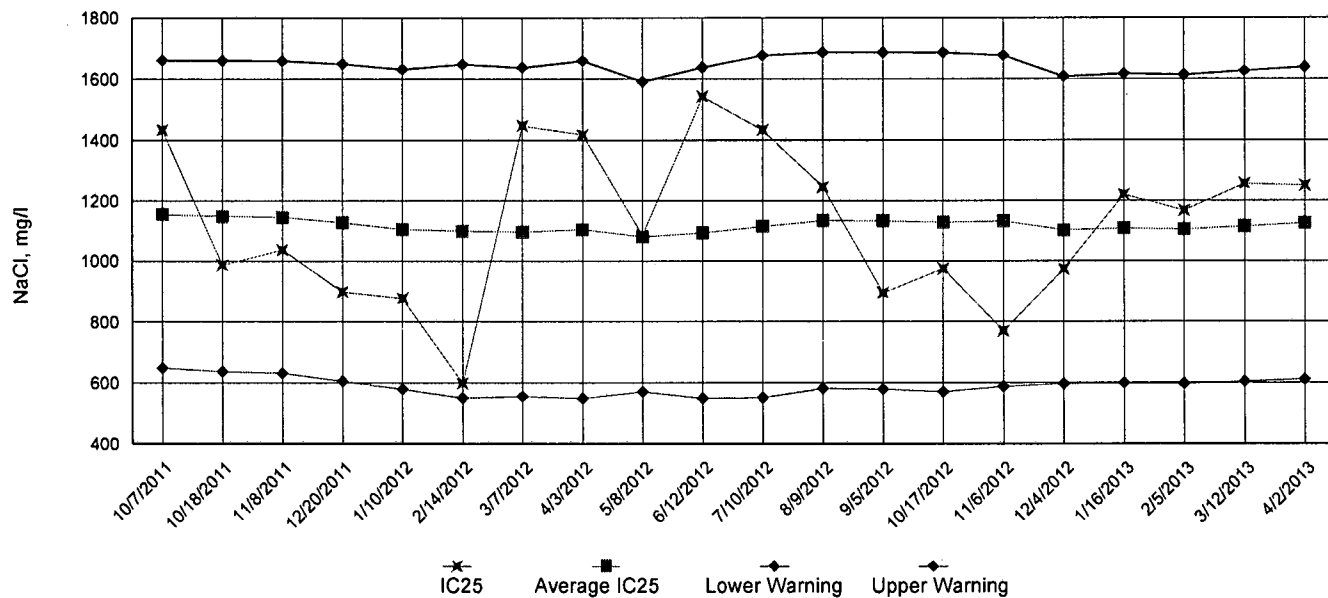
Effluent Conc.: 100 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.0	7.8	7.9	8.3	8.0	7.4	7.9
	Final	7.8	7.4	8.0	7.9	7.9	7.5	7.9
pH, units	Initial	7.2	7.4	7.4	7.5	7.6	7.8	7.4
	Final	7.8	7.7	7.8	7.6	7.8	7.4	7.9
Alkalinity, mg CaCO ₃ /l		61	NA	55	NA	51	NA	NA
Hardness, mg CaCO ₃ /l		27	NA	26	NA	28	NA	NA
Conductivity, umhos/cm		730	740	740	690	720	730	720
Res. Chlorine, mg/l		<0.05	NA	<0.05	NA	<0.05	NA	NA

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

LC50 Survival Data



IC25 Reproduction Data



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

Permittee: El Dorado Water Utilities

NPDES No.: AR0033723 AFIN No. 70-00341

Date and Time Test Initiated: April 30, 2013 at 1320

Date and Time Test Terminated: May 7, 2013 at 1300

Dilution water used: Synthetic Soft Water #3984

PERCENT SURVIVAL

Time of Reading	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
24 hour	100	100	100	100	100	100
48 hour	100	100	100	100	100	100
7 day	100	100	100	100	100	100

NUMBER OF YOUNG PRODUCED PER FEMALE @ 7 DAYS

Replicates	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
A	9	18	10	18	12	19
B	21	16	21	15	19	18
C	16	14	17	21	17	20
D	17	20	13	16	17	15
E	15	23	24	20	19	20
F	19	15	15	9	15	21
G	21	8	22	19	18	18
H	23	18	15	20	16	12
I	14	19	17	20	20	22
J	16	18	21	17	20	18
Mean per Adult	17.1	16.9	17.5	17.5	17.3	18.3
Mean per Surviving Adult	17.1	16.9	17.5	17.5	17.3	18.3
CV %	23.9	23.9	25.3	20.4	14.4	16.1

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	(100 %)	_____ YES	_____ X NO
b.) 1/2 LOW FLOW DILUTION	(NA)	_____ YES	_____ NO

2. Dunnett's 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	(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 (TLP3B)
- 4. If you answered NO to 2.a) enter [0] otherwise enter [1]: 0 (TGP3B)
- 5. NOEC Ceriodaphnia Lethality: 100 % (TOP3B)
- 6. LOEC Ceriodaphnia Lethality: 100 % (TXP3B)
- 7. NOEC Ceriodaphnia Sublethality: 100 % (TPP3B)
- 8. LOEC Ceriodaphnia Sublethality: 100 % (TYP3B)
- 9. Coefficient of variation for Ceriodaphnia Reproduction: 23.9 (TQP3B)

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

PERMITTEE: El Dorado Water Utilities
NPDES NO.: AR0033723 AFIN No. 70-00341
CONTACT: Mr. Harold Baker
ANALYST: 280, 298, 304, 307

Test Initiated: DATE: April 30, 2013 TIME: 1320
Test Terminated: DATE: May 7, 2013 TIME: 1300

DILUTION Control	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.1	7.7	7.8	8.1	7.9	7.8	8.1
Final	8.0	7.9	7.9	7.7	7.9	7.6	8.2
pH Initial	7.6	7.8	7.8	7.9	8.0	7.6	7.6
Final	7.9	7.6	7.5	7.7	8.0	6.4	8.0
Alkalinity	31	NA	31	NA	31	NA	NA
Hardness	41	NA	43	NA	44	NA	NA
Conductivity	160	160	160	160	170	180	160
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION 32 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	7.8	7.7	7.9	8.3	7.9	7.8	8.0
Final	7.8	7.8	8.0	7.8	7.7	7.8	8.0
pH Initial	7.4	7.6	7.6	7.6	7.8	7.7	7.5
Final	7.8	7.6	7.5	7.5	7.8	6.7	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	340	340	350	330	350	360	290
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 45 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.1	7.7	7.9	8.1	7.7	7.8	7.8
Final	7.9	7.9	8.0	7.7	7.8	7.8	8.1
pH Initial	7.3	7.6	7.5	7.6	7.7	7.7	7.5
Final	7.7	7.5	7.6	7.6	7.7	6.9	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	400	400	400	380	410	410	320
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 56 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.2	7.9	7.8	8.1	7.6	7.7	7.9
Final	7.9	7.9	8.0	7.6	7.7	7.6	8.2
pH Initial	7.3	7.5	7.5	7.5	7.7	7.7	7.4
Final	7.7	7.5	7.6	7.7	7.7	7.0	7.7
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	470	470	480	440	480	480	370
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 75 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.0	7.7	7.8	8.1	7.7	7.6	7.9
Final	7.8	7.7	8.1	7.9	8.0	7.7	8.0
pH Initial	7.2	7.6	7.4	7.4	7.7	7.8	7.4
Final	7.7	7.5	7.7	7.7	7.8	7.1	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	570	580	580	550	590	590	440
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 100 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.0	7.8	7.9	8.3	8.0	7.4	7.9
Final	7.8	7.4	8.0	7.9	7.9	7.5	7.9
pH Initial	7.2	7.4	7.4	7.5	7.6	7.8	7.4
Final	7.8	7.7	7.8	7.6	7.8	7.4	7.9
Alkalinity	61	NA	55	NA	51	NA	NA
Hardness	27	NA	26	NA	28	NA	NA
Conductivity	730	740	740	690	720	730	720
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

PAGE 1 OF 1

Client: <u>EL DORADO WATER UTILITIES</u>			PO No.		NO OF BOTTLES	ANALYSES REQUESTED										AIC CONTROL NO: <u>166953</u>		
Project Reference: <u>SOUTH EFFLUENT</u>			SAMPLE MATRIX			CD ONLY BIO-MONITORING											AIC PROPOSAL NO:	
Project Manager: <u>HAROLD BAKER</u>			WATER														Carrier: <u>Fed-x</u>	
Sampled By: <u>JOHN M. PEPPERS</u>			G R A M B	C O M P	A T E R	S O I L											Received Temperature C <u>4</u>	
AIC No.	Sample Identification	Date/Time Collected															Remarks	
<u>3</u>	<u>SE-1650</u>	<u>5-3-13</u>																
			Container Type												Field pH calibration			
			Preservative												on _____ @ _____			
			P NO												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							
Turnaround Time Requested: (Please circle) <u>(NORMAL)</u> or EXPEDITED IN _____ DAYS					Relinquished By: <u>JOHN M. PEPPERS</u> <u>John M. Peppers</u>		Date/Time <u>1615</u> <u>5-3-13</u>		Received By: <u>FED EX</u>		Date/Time <u>1625</u> <u>5-3-13</u>							
Expedited results requested by: _____					Relinquished By:		Date/Time		Received in Lab By: <u>[Signature]</u>		Date/Time <u>5/4/13 0830</u>							
Who should AIC contact with questions: <u>JOHN M. PEPPERS</u>					Comments:													
Phone: <u>870-814-1764</u> LAB # <u>870-862-0421</u>																		
Report Attention to: <u>HAROLD BAKER</u>																		
Report Address to: <u>P.O. Box 1587</u> <u>EL DORADO, AR 71731</u>																		

June 10, 2013

Test Results of
Second Quarter
Chronic 7-Day Renewal
Biomonitoring Testing
for
Outfall 001
South Effluent

Control No. 167770-1

Prepared for:

Mr. Harold Baker
El Dorado Water Utilities
Post Office Box 1587
El Dorado, AR 71731

Prepared by:

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

El Dorado Water Utilities
ATTN: Mr. Harold Baker
Post Office Box 1587
El Dorado, AR 71731

Re: Chronic 7-Day Renewal utilizing *Ceriodaphnia dubia*
Outfall 001 - South Effluent
NPDES Permit No. AR0033723 AFIN No. 70-00341

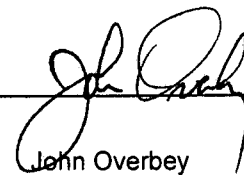
Dear Mr. Harold Baker:

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 1002.0 Chronic *Ceriodaphnia dubia* Survival and Reproduction 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 reproduction occurred at 75 % effluent, which is below the critical dilution of 100 %. **The sample PASSED lethal effects, however, FAILED sub-lethal effects for the *Ceriodaphnia dubia* test.**

AMERICAN INTERPLEX CORPORATION



John Overbey
Laboratory Director

PDF cc: El Dorado Water Utilities
ATTN: Mr. Harold Baker
harold@eldoradowater.com

El Dorado Water Utilities
ATTN: Mr. John Peppers
lab@eldoradowater.com

GBMc & Associates, Inc.
ATTN: Mr. Roland McDaniel
rmcdaniel@gbmcassoc.com

Table of Contents

- I. Control Acceptance Criteria
- II. Outlined Report
- III. Data Analysis
- IV. Standard Reference Toxicants
- V. Chemical Analysis/Quality Control
- VI. Organism History
- VII. Results Summary
Ceriodaphnia dubia

Appendix A: Raw Data

- A1: Test 1002.0
Ceriodaphnia dubia Survival and Reproduction
- A2: Statistics
- A3: Water Chemistry
- A4: Reference Toxicant

Appendix B: Chains of Custody

I. Control Acceptance Criteria

Ceriodaphnia dubia Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	23.7	PASS
Control CV < or = 40% per Surviving Female	14.8	PASS
Reproduction Minimum Significant Difference 13 to 47%	19.3	PASS
Critical Dilution CV < or = 40%	26.5	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0033723 AFIN No. 70-00341
2. Test Requirements: Chronic Biomonitoring, Quarterly Test Method 1002.0
3. Receiving Stream: Bayou de Loutre

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	8.0	7.7
pH (standard units)	6.4	6.5	6.6
Alkalinity (mg/l as CaCO ₃)	8.2	8.2	14
Hardness (mg/l as CaCO ₃)	10	9.4	11
Conductivity (umhos/cm)	670	670	690
Residual Chlorine (mg/l)	<0.05	<0.05	<0.05
Ammonia as N (mg/l)	0.25	0.054	0.77

2. Dilution Water Samples: Synthetic Soft Water #3993

- a. Dates Prepared: May 21 through June 4, 2013
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	7.1	7.8	7.6
pH (standard units)	7.5	7.6	7.5
Alkalinity (mg/l as CaCO ₃)	30	30	30
Hardness (mg/l as CaCO ₃)	47	47	47
Conductivity (umhos/cm)	170	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 1002.0, *Ceriodaphnia dubia* Survival and Reproduction.

2. Endpoint: No Observable Effects Concentration (NOEC)

3. Test Conditions:

Ceriodaphnia dubia Survival and Growth Method 1002.0

Date & Time Test Initiated: May 28, 2013 at 1300
Date & Time Test Terminated: June 4, 2013 at 1430
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 1002.0 *Ceriodaphnia dubia*

III. Data Analysis

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

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. 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.

Ceriodaphnia dubia

Chronic reference tests are performed monthly.

A chronic reference test was performed on May 28, 2013 at 1700 to June 4, 2013 at 1500

The results were as follows: (Control No. 167778-2.)

Survival LC-50: 2236 mg/l

Growth IC-25: 1573 mg/l

Growth PMSD: 13.9

V. Chemical Analysis/Quality Control

Parameter	Method	% Recovery	Relative % Difference
Alkalinity	SM 2320 B	NA	0.00
Hardness	EPA 200.7	93.1	3.04
pH	SM 4500-H+ B	100	0.134
Conductivity	EPA 120.1	98.0	0.656

VI. Organism History

Ceriodaphnia dubia

Date: May 28, 2013

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 *Ceriodaphnia dubia*, Cladoceran Survival and Reproduction Test -- Method 1002.0

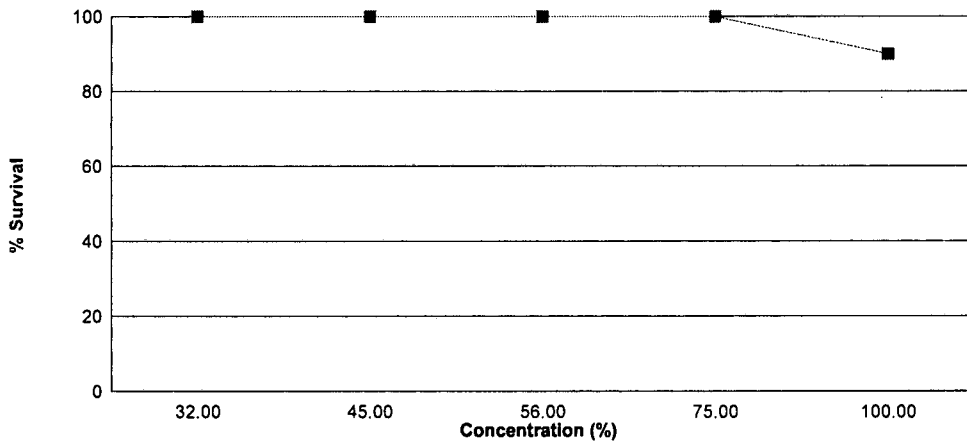
Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring with an average of at least 15 young per female.

Effluent dilutions for this test were 32 %, 45 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

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

The test was initiated on May 28, 2013 at 1300 and continued through June 4, 2013 at 1430. 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 = 75 % effluent



Concentration	Percent Survival	Mean Reproduction
Control	100	23.7
32 %	100	24.2
45 %	100	25.0
56 %	100	23.1
75 %	100	22.2
100 %	90.0	17.0 *

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

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: May 28, 2013 at 1300

Date and Time Test Terminated: June 4, 2013 at 1430

Concentration: Control														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	3	4	4	3	3	4	6	3	4	6	40	10	4.00	
5	9	7	8	8	7	9	9	8	10	8	83	10	8.30	
6	0	0	0	9	0	0	0	0	0	0	9	10	0.900	
7	10	10	10	0	9	13	15	13	13	12	105	10	10.5	
8														
TOTAL	22	21	22	20	19	26	30	24	27	26	237	10	23.7	

Concentration: 32 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	4	4	5	4	4	0	4	4	4	6	39	10	3.90	
5	8	8	7	8	10	8	9	9	8	9	84	10	8.40	
6	0	0	8	0	13	13	0	0	0	0	34	10	3.40	
7	9	10	0	9	0	0	15	14	13	15	85	10	8.50	
8														
TOTAL	21	22	20	21	27	21	28	27	25	30	242	10	24.2	

Concentration: 45 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	3	4	4	4	4	3	4	5	5	5	41	10	4.10	
5	9	7	8	7	9	9	10	9	9	0	77	10	7.70	
6	0	0	10	9	0	13	0	0	0	11	43	10	4.30	
7	11	10	0	0	13	0	14	14	13	14	89	10	8.90	
8														
TOTAL	23	21	22	20	26	25	28	28	27	30	250	10	25.0	

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: May 28, 2013 at 1300

Date and Time Test Terminated: June 4, 2013 at 1430

Concentration: 56 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	2	4	2	4	4	5	4	4	5	5	39	10	3.90	
5	8	8	7	8	7	9	10	10	8	11	86	10	8.60	
6	0	0	12	8	0	0	10	0	0	0	30	10	3.00	
7	10	10	0	0	0	14	0	13	14	15	76	10	7.60	
8														
TOTAL	20	22	21	20	11	28	24	27	27	31	231	10	23.1	

Concentration: 75 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	3	3	2	2	4	0	5	4	5	5	33	10	3.30	
5	7	8	5	6	7	7	8	9	9	8	74	10	7.40	
6	0	0	0	0	0	14	0	0	0	0	14	10	1.40	
7	10	9	10	9	9	0	13	15	11	15	101	10	10.1	
8														
TOTAL	20	20	17	17	20	21	26	28	25	28	222	10	22.2	

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	X	0	0	0	0	0	0	9	0.00	
3	0	0	0	0	X	0	0	0	0	0	0	9	0.00	
4	2	2	3	0	X	0	5	3	5	4	24	9	2.67	
5	4	6	5	6	X	3	7	7	6	8	52	9	5.78	
6	0	0	0	0	X	11	0	0	0	0	11	9	1.22	
7	8	9	7	8	X	0	10	14	14	13	83	9	9.22	
8														
TOTAL	14	17	15	14	0	14	22	24	25	25	170	10	17.0	

Appendix A2: Statistics

Ceriodaphnia dubia Survival

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

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

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
45 %	10	0	10
Total	20	0	20

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

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

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

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

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

Appendix A2: Statistics

Ceriodaphnia dubia Survival

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
100 %	9	1	10
Total	19	1	20

Critical Fisher's value (10,10,10) ($\alpha=0.05$) is 6. b value is 9. 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	45 %	10	0	
3	56 %	10	0	
4	75 %	10	0	
5	100 %	10	1	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

Kolmogorov Test for Normality	No Transformation
D = 0.1073 D* = 0.8418 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 = 10.19 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	412.7	82.54	3.429	
Within (Error)	54	1300	24.07		
Total	59	1713			
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	23.7	23.7			
2	32 %	24.2	24.2	-0.2279		
3	45 %	25	25	-0.5925		
4	56 %	23.1	23.1	0.2735		
5	75 %	22.2	22.2	0.6837		
6	100 %	17	17	3.054	*	
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	5.068	21.4	-0.5		
3	45 %	10	5.068	21.4	-1.3		
4	56 %	10	5.068	21.4	0.6		
5	75 %	10	5.068	21.4	1.5		
6	100 %	10	5.068	21.4	6.7		

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	217.5	43.5	2.354	
Within (Error)	53	979.5	18.48		
Total	58	1197			
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	23.7	23.7			
2	32 %	24.2	24.2	-0.2601		
3	45 %	25	25	-0.6762		
4	56 %	23.1	23.1	0.3121		
5	75 %	22.2	22.2	0.7802		
6	100 %	18.889	18.889	2.436	*	
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	32 %	10	4.441	18.7	-0.5	
3	45 %	10	4.441	18.7	-1.3	
4	56 %	10	4.441	18.7	0.6	
5	75 %	10	4.441	18.7	1.5	
6	100 %	9	4.563	19.3	4.811	

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: May 28, 2013 at 0826

Date and Time Test Terminated: June 4, 2013 at 1430

Effluent Conc.: Control	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	7.1	7.2	7.8	7.6	7.6	7.6	7.7
	Final	7.8	8.2	8.7	7.4	7.9	8.1	8.1
pH, units	Initial	7.5	7.4	7.6	7.6	7.5	7.6	7.5
	Final	7.8	8.0	8.0	7.7	7.9	7.7	7.8
Alkalinity, mg CaCO ₃ /l	30	NA	30	NA	30	NA	NA	
Hardness, mg CaCO ₃ /l	47	NA	47	NA	47	NA	NA	
Conductivity, umhos/cm	170	150	160	160	170	170	180	
Res. Chlorine, mg/l	<0.05	NA	<0.05	NA	<0.05	NA	NA	

Effluent Conc.: 32 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.3	7.8	7.9	8.0	7.7	7.7	7.7
	Final	8.0	8.3	8.9	8.0	7.7	8.1	8.2
pH, units	Initial	7.3	7.3	7.5	7.7	7.4	7.5	7.4
	Final	7.9	8.1	8.0	7.8	7.8	7.7	7.8

Effluent Conc.: 45 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.3	7.8	7.7	7.9	7.5	7.7	8.0
	Final	7.9	8.3	8.6	7.8	7.9	8.1	8.1
pH, units	Initial	7.2	7.2	7.3	7.6	7.2	7.4	7.1
	Final	7.8	8.0	8.0	7.8	7.9	7.7	7.8

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: May 28, 2013 at 0826

Date and Time Test Terminated: June 4, 2013 at 1430

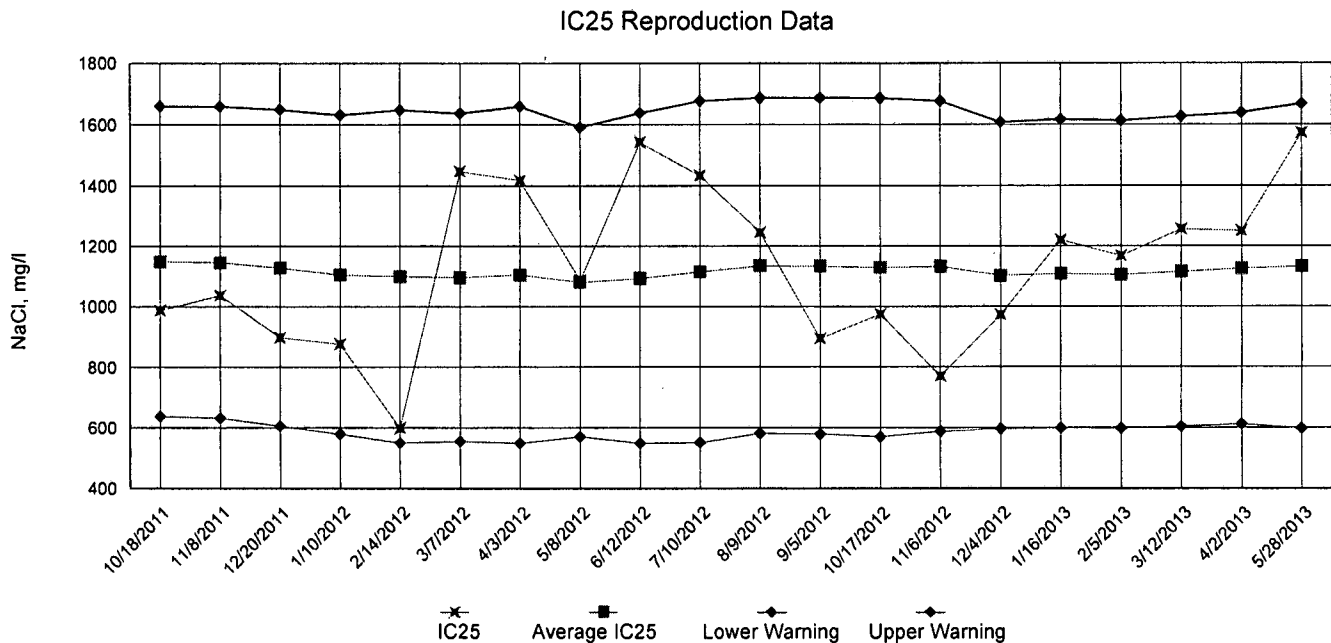
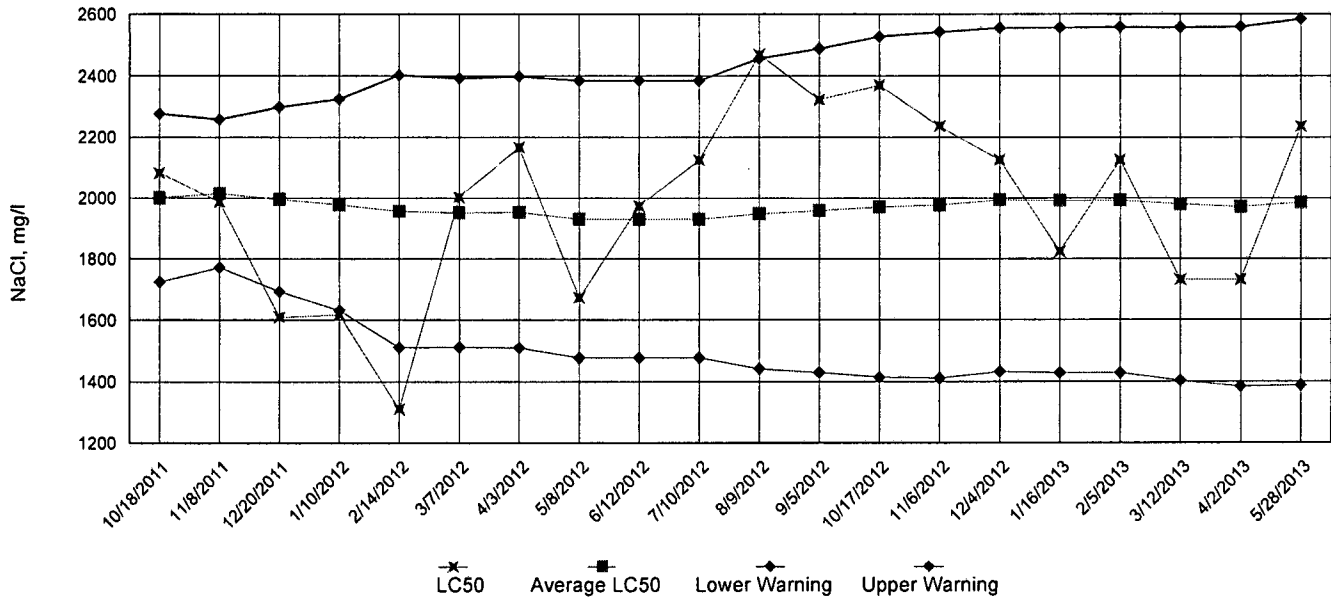
Effluent Conc.: 56 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.0	7.7	7.6	7.8	7.7	7.7	7.5
	Final	7.7	8.3	8.9	7.9	7.7	8.1	8.0
pH, units	Initial	7.0	7.0	7.2	7.5	7.1	7.3	7.0
	Final	7.8	7.9	8.0	7.7	7.8	7.6	7.7

Effluent Conc.: 75 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.4	7.7	7.5	7.7	7.8	7.4	8.0
	Final	7.8	8.3	8.9	7.7	7.9	8.1	7.7
pH, units	Initial	6.8	6.9	7.0	7.4	6.9	7.1	6.9
	Final	7.6	7.7	7.8	7.7	7.7	7.5	7.7

Effluent Conc.: 100 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.7	7.8	8.0	9.0	7.7	7.6	7.9
	Final	7.6	8.4	8.6	7.6	7.6	8.0	8.1
pH, units	Initial	6.4	6.6	6.5	6.4	6.6	6.8	6.7
	Final	7.4	7.5	7.5	7.5	7.5	7.3	7.6
Alkalinity, mg CaCO ₃ /l		8.2	NA	8.2	NA	14	NA	NA
Hardness, mg CaCO ₃ /l		10	NA	9.4	NA	11	NA	NA
Conductivity, umhos/cm		670	620	670	660	690	690	680
Res. Chlorine, mg/l		<0.05	NA	<0.05	NA	<0.05	NA	NA

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

LC50 Survival Data



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

Permittee: El Dorado Water Utilities

NPDES No.: AR0033723 AFIN No. 70-00341

Date and Time Test Initiated: May 28, 2013 at 1300

Date and Time Test Terminated: June 4, 2013 at 1430

Dilution water used: Synthetic Soft Water #3993

PERCENT SURVIVAL

Time of Reading	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
24 hour	100	100	100	100	100	100
48 hour	100	100	100	100	100	90.0
7 day	100	100	100	100	100	90.0

NUMBER OF YOUNG PRODUCED PER FEMALE @ 7 DAYS

Replicates	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
A	22	21	23	20	20	14
B	21	22	21	22	20	17
C	22	20	22	21	17	15
D	20	21	20	20	17	14
E	19	27	26	11	20	0
F	26	21	25	28	21	14
G	30	28	28	24	26	22
H	24	27	28	27	28	24
I	27	25	27	27	25	25
J	26	30	30	31	28	25
Mean per Adult	23.7	24.2	25.0	23.1	22.2	17.0
Mean per Surviving Adult	23.7	24.2	25.0	23.1	22.2	18.9
CV %	14.8	14.9	13.5	24.5	19.0	26.5

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	(100 %)	<input type="checkbox"/> YES	<input checked="" type="checkbox"/> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<input type="checkbox"/> YES	<input type="checkbox"/> NO

2. Dunnett's 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	(100 %)	<input checked="" type="checkbox"/> YES	<input type="checkbox"/> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<input type="checkbox"/> YES	<input type="checkbox"/> 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]: 1 (TGP3B)

5. NOEC Ceriodaphnia Lethality: 100 % (TOP3B)

6. LOEC Ceriodaphnia Lethality: 100 % (TXP3B)

7. NOEC Ceriodaphnia Sublethality: 75 % (TPP3B)

8. LOEC Ceriodaphnia Sublethality: 100 % (TYP3B)

9. Coefficient of variation for Ceriodaphnia Reproduction: 26.5 (TQP3B)

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

PERMITTEE: El Dorado Water Utilities
NPDES NO.: AR0033723 AFIN No. 70-00341
CONTACT: Mr. Harold Baker
ANALYST: 280, 298, 304, 307

Test Initiated: DATE: May 28, 2013 TIME: 1300
Test Terminated: DATE: June 4, 2013 TIME: 1430

DILUTION Control	DAY						
	1	2	3	4	5	6	7
D.O. Initial	7.1	7.2	7.8	7.6	7.6	7.6	7.7
Final	7.8	8.2	8.7	7.4	7.9	8.1	8.1
pH Initial	7.5	7.4	7.6	7.6	7.5	7.6	7.5
Final	7.8	8.0	8.0	7.7	7.9	7.7	7.8
Alkalinity	30	NA	30	NA	30	NA	NA
Hardness	47	NA	47	NA	47	NA	NA
Conductivity	170	150	160	160	170	170	180
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION 32 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.3	7.8	7.9	8.0	7.7	7.7	7.7
Final	8.0	8.3	8.9	8.0	7.7	8.1	8.2
pH Initial	7.3	7.3	7.5	7.7	7.4	7.5	7.4
Final	7.9	8.1	8.0	7.8	7.8	7.7	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	320	300	320	300	340	340	350
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 45 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.3	7.8	7.7	7.9	7.5	7.7	8.0
Final	7.9	8.3	8.6	7.8	7.9	8.1	8.1
pH Initial	7.2	7.2	7.3	7.6	7.2	7.4	7.1
Final	7.8	8.0	8.0	7.8	7.9	7.7	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	380	360	370	290	380	390	390
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 56 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.0	7.7	7.6	7.8	7.7	7.7	7.5
Final	7.7	8.3	8.9	7.9	7.7	8.1	8.0
pH Initial	7.0	7.0	7.2	7.5	7.1	7.3	7.0
Final	7.8	7.9	8.0	7.7	7.8	7.6	7.7
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	450	420	440	300	450	460	450
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 75 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.4	7.7	7.5	7.7	7.8	7.4	8.0
Final	7.8	8.3	8.9	7.7	7.9	8.1	7.7
pH Initial	6.8	6.9	7.0	7.4	6.9	7.1	6.9
Final	7.6	7.7	7.8	7.7	7.7	7.5	7.7
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	540	510	540	290	550	550	550
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 100 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.7	7.8	8.0	9.0	7.7	7.6	7.9
Final	7.6	8.4	8.6	7.6	7.6	8.0	8.1
pH Initial	6.4	6.6	6.5	6.4	6.6	6.8	6.7
Final	7.4	7.5	7.5	7.5	7.5	7.3	7.6
Alkalinity	8.2	NA	8.2	NA	14	NA	NA
Hardness	10	NA	9.4	NA	11	NA	NA
Conductivity	670	620	670	660	690	690	680
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

June 19, 2013

Test Results of
Second Quarter
Chronic 7-Day Renewal
Biomonitoring Testing
for
Outfall 001
South Effluent

Control No. 167908-1

Prepared for:

Mr. Harold Baker
El Dorado Water Utilities
Post Office Box 1587
El Dorado, AR 71731

Prepared by:

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

El Dorado Water Utilities
ATTN: Mr. Harold Baker
Post Office Box 1587
El Dorado, AR 71731

Re: Chronic 7-Day Renewal utilizing *Pimephales promelas* (Fathead minnow) and *Ceriodaphnia dubia*
Outfall 001 - South Effluent
NPDES Permit No. AR0033723 AFIN No. 70-00341

Dear Mr. Harold Baker:

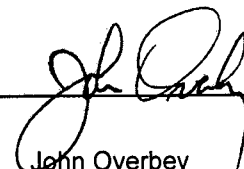
This report is the analytical results and supporting information for the samples submitted to American Interplex Corporation (AIC). The following results are applicable only to the sample identified by the control number referenced above. Accurate assessment of the data requires access to the entire 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 75 % effluent, which is below the critical dilution of 100 %. The NOEC for growth occurred at 75 % effluent, which is below the critical dilution of 100 %. **The sample, therefore, FAILED both lethal and 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 100 % effluent, which is equal to the critical dilution of 100 %. The NOEC for reproduction occurred at 75 % effluent, which is below the critical dilution of 100 %. **The sample PASSED lethal effects, however, FAILED sub-lethal effects for the *Ceriodaphnia dubia* test.**

AMERICAN INTERPLEX CORPORATION



John Overbey
Laboratory Director

PDF cc: El Dorado Water Utilities
ATTN: Mr. Harold Baker
harold@eldoradowater.com

El Dorado Water Utilities
ATTN: Mr. John Peppers
lab@eldoradowater.com

GBMc & Associates, Inc.
ATTN: Mr. Roland McDaniel
rmcdaniel@gbmcassoc.com

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Pimephales promelas (Fathead minnow)

Ceriodaphnia dubia

Appendix A: Raw Data

A1: Test 1000.0

Pimephales promelas (Fathead minnow) Survival and Growth

Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

A2: Statistics

A3: Water Chemistry

A4: Reference Toxicant

Appendix B: Chains of Custody

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	0.293	PASS
Control Growth CV < or = 40%	6.94	PASS
Growth Minimum Significant Difference 12 to 30%	17.4	PASS
Critical Dilution CV < or = 40%	113	FAIL

Ceriodaphnia dubia Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	19.4	PASS
Control CV < or = 40% per Surviving Female	26.0	PASS
Reproduction Minimum Significant Difference 13 to 47%	26.9	PASS
Critical Dilution CV < or = 40%	19.5	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0033723 AFIN No. 70-00341
2. Test Requirements: Chronic Biomonitoring, Quarterly Test Methods 1000.0 and 1002.0
3. Receiving Stream: Bayou de Loutre

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	8.5	8.5
pH (standard units)	6.4	6.6	6.8
Alkalinity (mg/l as CaCO ₃)	16	12	11
Hardness (mg/l as CaCO ₃)	8.9	9.6	7.0
Conductivity (umhos/cm)	640	650	640
Residual Chlorine (mg/l)	<0.05	0.050	<0.05
Ammonia as N (mg/l)	0.42	0.19	<0.1

2. Dilution Water Samples: Synthetic Soft Water #3995

- a. Dates Prepared: May 29 through June 12, 2013
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	7.9	7.6	8.6
pH (standard units)	7.7	7.6	7.7
Alkalinity (mg/l as CaCO ₃)	30	30	30
Hardness (mg/l as CaCO ₃)	47	47	44
Conductivity (umhos/cm)	170	160	160
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: June 4, 2013 at 1600
Date & Time Test Terminated: June 11, 2013 at 1400
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: June 4, 2013 at 1420
Date & Time Test Terminated: June 11, 2013 at 1620
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 and Bartlett's test. 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. 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 May 28, 2013 at 1645 to June 4, 2013 at 1520

The results were as follows: (Control No. 167778-1.)

Survival LC-50: 6598 mg/l

Growth IC-25: 5369 mg/l

Growth PMSD: 22.6

Ceriodaphnia dubia

Chronic reference tests are performed monthly.

A chronic reference test was performed on May 28, 2013 at 1700 to June 4, 2013 at 1500

The results were as follows: (Control No. 167778-2.)

Survival LC-50: 2236 mg/l

Growth IC-25: 1573 mg/l

Growth PMSD: 13.9

V. Chemical Analysis/Quality Control

Parameter	Method	% Recovery	Relative % Difference
Alkalinity	SM 2320 B	NA	0.00
Hardness	EPA 200.7	101	0.750
pH	SM 4500-H+ B	101	0.00
Conductivity	EPA 120.1	110	1.04

VI. Organism History

Pimephales promelas (Fathead minnow)

Date: June 4, 2013

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: June 4, 2013

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

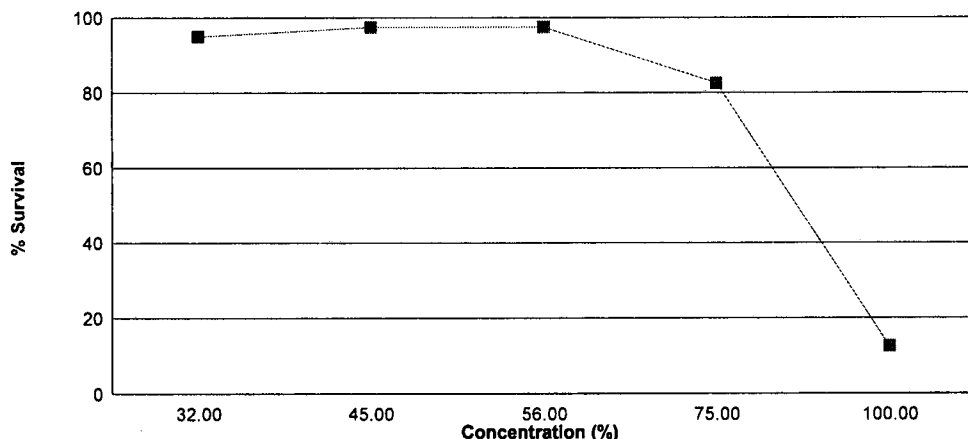
Larvae are exposed in a static renewal system for seven days to different concentrations of effluent with dilution water. Test results are based on the survival and growth (increase in weight) of the larvae.

Effluent dilutions for this test were 32 %, 45 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

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

The test was initiated on June 4, 2013 at 1600 and continued through June 11, 2013 at 1400. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

- a.) NOEC survival = 75 % effluent
- b.) NOEC growth = 75 % effluent



Summary of the 7-day Fathead Minnow Survival and Growth		
Concentration	Percent Survival	Mean Growth (mg)
Control	100	0.293
32 %	95.0	0.316
45 %	97.5	0.296
56 %	97.5	0.273
75 %	82.5	0.247
100 %	12.5 *	--

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

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

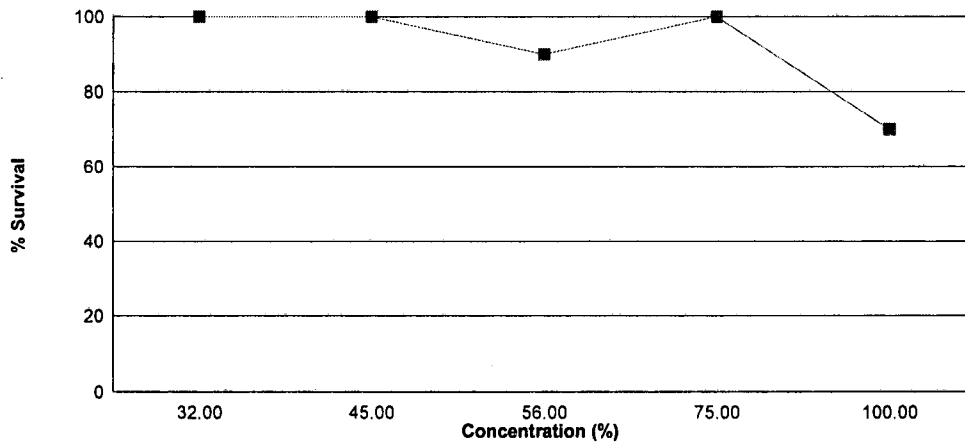
Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring with an average of at least 15 young per female.

Effluent dilutions for this test were 32 %, 45 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

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

The test was initiated on June 4, 2013 at 1420 and continued through June 11, 2013 at 1620. 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 = 75 % effluent



Summary of the 7-day <i>Ceriodaphnia dubia</i> Survival and Reproduction Data		
Concentration	Percent Survival	Mean Reproduction
Control	100	19.4
32 %	100	18.7
45 %	100	20.3
56 %	90.0	19.2
75 %	100	16.4
100 %	70.0	11.7 *

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

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Survival

Date and Time Test Initiated: June 4, 2013 at 1600

Date and Time Test Terminated: June 11, 2013 at 1400

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	7	7
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	7	7	7	7	7
45 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	7	7	7	7	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	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
75 %	A	8	8	6	6	6	6	5
	B	8	8	7	7	7	6	6
	C	8	8	8	8	8	8	8
	D	8	8	7	7	7	7	7
	E	8	7	7	7	7	7	7
100 %	A	8	3	3	3	2	2	2
	B	8	3	1	0	0	0	0
	C	8	2	1	1	1	1	1
	D	8	3	1	0	0	0	0
	E	8	5	3	2	2	2	2

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Growth

Test Initiated: June 4, 2013 at 1600
Test Terminated: June 11, 2013 at 1400

Drying Started: June 10, 2013 at 0900
Drying Ended: June 12, 2013 at 1515

Concentration	Replicate	Weight of pan	Weight of pan + fish	Total weight of fish (g)	Original # of fish	Mean dry weight (mg)
Control	A	.91275	.91499	0.00224	8	0.280
	B	.91074	.91286	0.00212	8	0.265
	C	.90949	.91201	0.00252	8	0.315
	D	.90989	.91234	0.00245	8	0.306
	E	.90806	.91046	0.00240	8	0.300
32 %	A	.90787	.91021	0.00234	8	0.292
	B	.90626	.90855	0.00229	8	0.286
	C	.90641	.90912	0.00271	8	0.339
	D	.90534	.90780	0.00246	8	0.308
	E	.90236	.90520	0.00284	8	0.355
45 %	A	.90345	.90564	0.00219	8	0.274
	B	.90533	.90787	0.00254	8	0.318
	C	.90619	.90835	0.00216	8	0.270
	D	.90611	.90833	0.00222	8	0.278
	E	.90749	.91022	0.00273	8	0.341
56 %	A	.91317	.91476	0.00159	8	0.199
	B	.91497	.91703	0.00206	8	0.258
	C	.91300	.91528	0.00228	8	0.285
	D	.91234	.91473	0.00239	8	0.299
	E	.91103	.91362	0.00259	8	0.324
75 %	A	.91085	.91233	0.00148	8	0.185
	B	.91003	.91190	0.00187	8	0.234
	C	.90945	.91153	0.00208	8	0.260
	D	.91001	.91230	0.00229	8	0.286
	E	.91190	.91404	0.00214	8	0.268
100 %	A	.91295	.91333	0.00038	8	0.047
	B	.90860	.90860	0.00000	8	0.000
	C	.90792	.90812	0.00020	8	0.025
	D	.90633	.90633	0.00000	8	0.000
	E	.90603	.90669	0.00066	8	0.082

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: June 4, 2013 at 1420
Date and Time Test Terminated: June 11, 2013 at 1620

Concentration: Control														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	0	4	0	2	2	2	2	0	4	6	22	10	2.20	
5	4	0	4	0	0	4	1	4	7	0	24	10	2.40	
6	9	10	8	6	7	0	7	10	0	11	68	10	6.80	
7	12	14	0	10	10	10	12	0	12	0	80	10	8.00	
8														
TOTAL	25	28	12	18	19	16	22	14	23	17	194	10	19.4	

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	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	0	4	2	0	2	3	0	0	3	2	16	10	1.60	
5	4	0	4	6	0	6	3	4	6	5	38	10	3.80	
6	8	8	0	10	8	0	10	10	0	0	54	10	5.40	
7	0	12	11	0	14	15	0	0	14	13	79	10	7.90	
8														
TOTAL	12	24	17	16	24	24	13	14	23	20	187	10	18.7	

Concentration: 45 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
4	0	0	0	2	2	2	2	0	2	6	16	10	1.60	
5	4	4	5	5	0	8	6	6	7	0	45	10	4.50	
6	10	9	10	0	8	0	0	10	0	12	59	10	5.90	
7	9	12	0	11	14	13	11	0	13	0	83	10	8.30	
8														
TOTAL	23	25	15	18	24	23	19	16	22	18	203	10	20.3	

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: June 4, 2013 at 1420

Date and Time Test Terminated: June 11, 2013 at 1620

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	X	0	0	0	0	0	0	0	0	0	0	9	0.00
4	X	3	0	2	2	2	3	0	4	6	22	9	2.44
5	X	0	5	6	0	9	0	4	5	0	29	9	3.22
6	X	9	8	8	8	0	8	8	0	12	61	9	6.78
7	X	10	0	12	11	12	12	0	11	12	80	9	8.89
8													
TOTAL	0	22	13	28	21	23	23	12	20	30	192	10	19.2

Concentration: 75 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	0	0	0	0	0	0	0	0	0	0	0	10	0.00
4	2	4	0	0	2	2	3	2	0	0	15	10	1.50
5	0	0	2	5	0	7	0	6	5	3	28	10	2.80
6	8	3	0	10	6	0	9	0	9	12	57	10	5.70
7	10	11	5	0	10	9	10	9	0	0	64	10	6.40
8													
TOTAL	20	18	7	15	18	18	22	17	14	15	164	10	16.4

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	X	X	X	0	0	0	0	0	0	0	7	0.00
3	0	X	X	X	0	0	0	0	0	0	0	7	0.00
4	0	X	X	X	2	0	2	0	2	0	6	7	0.857
5	3	X	X	X	0	6	8	2	3	6	28	7	4.00
6	8	X	X	X	8	8	0	9	3	10	46	7	6.57
7	9	X	X	X	9	0	9	0	10	0	37	7	5.29
8													
TOTAL	20	0	0	0	19	14	19	11	18	16	117	10	11.7

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	0.87500	1.20940
2	32 %	3	1.00000	1.39310
2	32 %	4	1.00000	1.39310
2	32 %	5	0.87500	1.20940
3	45 %	1	1.00000	1.39310
3	45 %	2	1.00000	1.39310
3	45 %	3	0.87500	1.20940
3	45 %	4	1.00000	1.39310
3	45 %	5	1.00000	1.39310
4	56 %	1	1.00000	1.39310
4	56 %	2	0.87500	1.20940
4	56 %	3	1.00000	1.39310
4	56 %	4	1.00000	1.39310
4	56 %	5	1.00000	1.39310
5	75 %	1	0.62500	0.91174
5	75 %	2	0.75000	1.04720
5	75 %	3	1.00000	1.39310
5	75 %	4	0.87500	1.20940
5	75 %	5	0.87500	1.20940
6	100 %	1	0.25000	0.52360
6	100 %	2	0.00000	0.17771
6	100 %	3	0.12500	0.36137
6	100 %	4	0.00000	0.17771
6	100 %	5	0.25000	0.52360

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Survival

Shapiro - Wilk's Test for Normality		Transform: Arc Sin(Square Root(Y))
<p>D = 0.3476 W = 0.9277 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		Transform: Arc Sin(Square Root(Y))
<p>Test can not be performed because at least one group has zero variance. Data FAIL to meet homogeneity of variance assumption.</p>		

Steel's Many-One Rank Test			Transform: Arc Sin(Square Root(Y))		
Ho: Control < Treatment					
Group	Identification	Rank Sum	Critical Value	DF	Sig 0.05
1	Control				
2	32 %	22.50	16.00	5.00	
3	45 %	25.00	16.00	5.00	
4	56 %	25.00	16.00	5.00	
5	75 %	17.50	16.00	5.00	
6	100 %	15.00	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 = 0.0245 W = 0.9551 Critical W = 0.888 (alpha = 0.01, N = 25) Critical W = 0.918 (alpha = 0.05, N = 25)</p> <p>Data PASS normality test (alpha = 0.01).</p>	

Bartlett's Test for Homogeneity of Variance	No Transformation
<p>Calculated B1 statistic = 2.743 Critical B = 13.277 (alpha = 0.01, df = 4)</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	4	0.01386	0.003465	2.829
Within (Error)	20	0.0245	0.001225	
Total	24	0.03836		
Critical F = 4.43 (alpha = 0.01, df = 4,20) 2.87 (alpha = 0.05, df = 4,20)				
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	0.2932	0.2932		
2	32 %	0.316	0.316	-1.03	
3	45 %	0.2962	0.2962	-0.1355	
4	56 %	0.273	0.273	0.9125	
5	75 %	0.2466	0.2466	2.105	
Dunnett's critical value = 2.3 (1 Tailed, alpha = 0.05, df = 4,20) (Actual df = 4,20)					

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	0.05091	17.4	-0.0228	
3	45 %	5	0.05091	17.4	-0.003	
4	56 %	5	0.05091	17.4	0.0202	
5	75 %	5	0.05091	17.4	0.0466	

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
45 %	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 %	9	1	10
Total	19	1	20

Critical Fisher's value (10,10,10) (alpha=0.05) is 6. b value is 9. Since b is greater than 6 there is NO SIGNIFICANT DIFFERENCE between CONTROL and TREATMENT at the 0.05 level.

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

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

Appendix A2: Statistics

Ceriodaphnia dubia Survival

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
100 %	7	3	10
Total	17	3	20

Critical Fisher's value (10,10,10) ($\alpha=0.05$) is 6. b value is 7. 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	45 %	10	0	
3	56 %	10	1	
4	75 %	10	0	
5	100 %	10	3	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

Kolmogorov Test for Normality	No Transformation
<p>D = 0.1064 D* = 0.8348 Critical D* = 1.035 (alpha = 0.01, N = 60)</p> <p>Data PASS normality test (alpha = 0.01).</p>	

Bartlett's Test for Homogeneity of Variance	No Transformation
<p>Calculated B1 statistic = 12.51 Critical B = 15.086 (alpha = 0.01, df = 5)</p> <p>Data PASS B1 homogeneity test at 0.01 level.</p>	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

ANOVA Table				No Transformation
SOURCE	DF	SS	MS	F
Between	5	505.5	101.1	2.665
Within (Error)	54	2048	37.93	
Total	59	2554		
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	19.4	19.4		
2	32 %	18.7	18.7	0.2542	
3	45 %	20.3	20.3	-0.3268	
4	56 %	19.2	19.2	0.07261	
5	75 %	16.4	16.4	1.089	
6	100 %	11.7	11.7	2.796	*
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	6.362	32.8	0.7	
3	45 %	10	6.362	32.8	-0.9	
4	56 %	10	6.362	32.8	0.2	
5	75 %	10	6.362	32.8	3	
6	100 %	10	6.362	32.8	7.7	

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	171.7	34.34	1.632	
Within (Error)	50	1052	21.04		
Total	55	1224			
Critical F = 3.41 (alpha = 0.01, df = 5,50) 2.4 (alpha = 0.05, df = 5,50)					
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	19.4	19.4		
2	32 %	18.7	18.7	0.3412	
3	45 %	20.3	20.3	-0.4387	
4	56 %	21.333	21.333	-0.9172	
5	75 %	16.4	16.4	1.462	
6	100 %	16.714	16.714	1.188	
Dunnett's critical value = 2.31 (1 Tailed, alpha = 0.05, df [used] = 5,40) (Actual df = 5,50) 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	32 %	10	4.739	24.4	0.7	
3	45 %	10	4.739	24.4	-0.9	
4	56 %	9	4.868	25.1	-1.933	
5	75 %	10	4.739	24.4	3	
6	100 %	7	5.222	26.9	2.686	

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: June 4, 2013 at 1044

Date and Time Test Terminated: June 11, 2013 at 1620

Effluent Conc.: Control	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	7.9	7.8	7.6	7.8	8.6	8.0	8.0
	Final *1	7.6	7.2	6.9	8.2	7.1	7.6	6.8
	Final *2	7.9	7.8	8.0	8.1	7.9	8.6	7.8
pH, units	Initial	7.7	7.6	7.6	7.6	7.7	7.5	7.4
	Final *1	7.5	7.3	7.2	7.3	7.3	7.4	7.1
	Final *2	8.0	8.0	7.9	7.9	8.0	7.6	7.8
Alkalinity, mg CaCO ₃ /l	30	NA	30	NA	30	NA	NA	
Hardness, mg CaCO ₃ /l	47	NA	47	NA	44	NA	NA	
Conductivity, umhos/cm	170	160	160	160	160	170	170	
Res. Chlorine, mg/l	<0.05	NA	<0.05	NA	<0.05	NA	NA	

Effluent Conc.: 32 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.0	7.7	7.9	7.5	8.8	8.1	7.8
	Final *1	7.4	6.9	6.6	7.6	7.1	7.3	6.5
	Final *2	8.2	8.1	8.3	8.2	8.1	8.5	8.4
pH, units	Initial	7.3	7.3	7.2	7.3	7.4	7.5	7.2
	Final *1	7.4	7.3	7.1	7.3	7.3	7.3	7.0
	Final *2	8.0	8.1	8.1	8.0	8.0	7.6	7.8

Effluent Conc.: 45 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.1	7.9	7.9	7.6	8.9	7.9	7.9
	Final *1	7.4	7.0	6.7	7.3	6.9	7.4	6.5
	Final *2	8.4	8.0	8.0	8.1	8.1	8.5	8.0
pH, units	Initial	7.2	7.2	7.1	7.2	7.3	7.5	7.1
	Final *1	7.4	7.3	7.1	7.2	7.2	7.2	7.0
	Final *2	8.2	8.1	8.1	7.9	8.0	7.6	7.9

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: June 4, 2013 at 1044

Date and Time Test Terminated: June 11, 2013 at 1620

Effluent Conc.: 56 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.2	7.8	8.2	7.6	7.7	7.7	7.7
	Final *1	7.5	7.2	6.8	7.4	6.9	7.5	6.9
	Final *2	8.1	7.8	8.2	8.1	8.4	8.2	8.2
pH, units	Initial	6.9	7.0	7.0	7.1	6.9	7.2	7.0
	Final *1	7.2	7.3	7.1	7.2	7.2	7.3	7.0
	Final *2	8.1	8.0	8.2	7.9	8.0	7.6	7.8

Effluent Conc.: 75 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.3	7.8	8.4	8.0	8.3	7.7	8.0
	Final *1	7.4	7.1	7.0	7.6	7.2	7.3	7.0
	Final *2	8.1	7.8	8.2	8.1	8.3	8.2	8.2
pH, units	Initial	6.7	6.9	6.8	6.9	7.0	7.2	6.8
	Final *1	7.2	7.2	7.0	7.2	7.2	7.2	6.9
	Final *2	7.9	7.9	8.0	7.8	7.9	7.5	7.7

Effluent Conc.: 100 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.5	7.8	8.5	8.3	8.5	7.9	8.9
	Final *1	7.4	7.0	6.9	7.4	7.1	7.3	7.0
	Final *2	7.9	7.8	8.2	8.1	8.4	8.3	8.3
pH, units	Initial	6.4	6.6	6.6	6.5	6.8	7.2	6.3
	Final *1	7.0	7.0	6.9	7.1	7.1	7.0	6.8
	Final *2	7.6	7.8	8.0	7.8	7.8	7.4	7.6
Alkalinity, mg CaCO ₃ /l	16	NA	12	NA	11	NA	NA	NA
Hardness, mg CaCO ₃ /l	8.9	NA	9.6	NA	7.0	NA	NA	NA
Conductivity, umhos/cm	640	650	650	650	640	660	650	650
Res. Chlorine, mg/l	<0.05	NA	0.050	NA	<0.05	NA	NA	NA

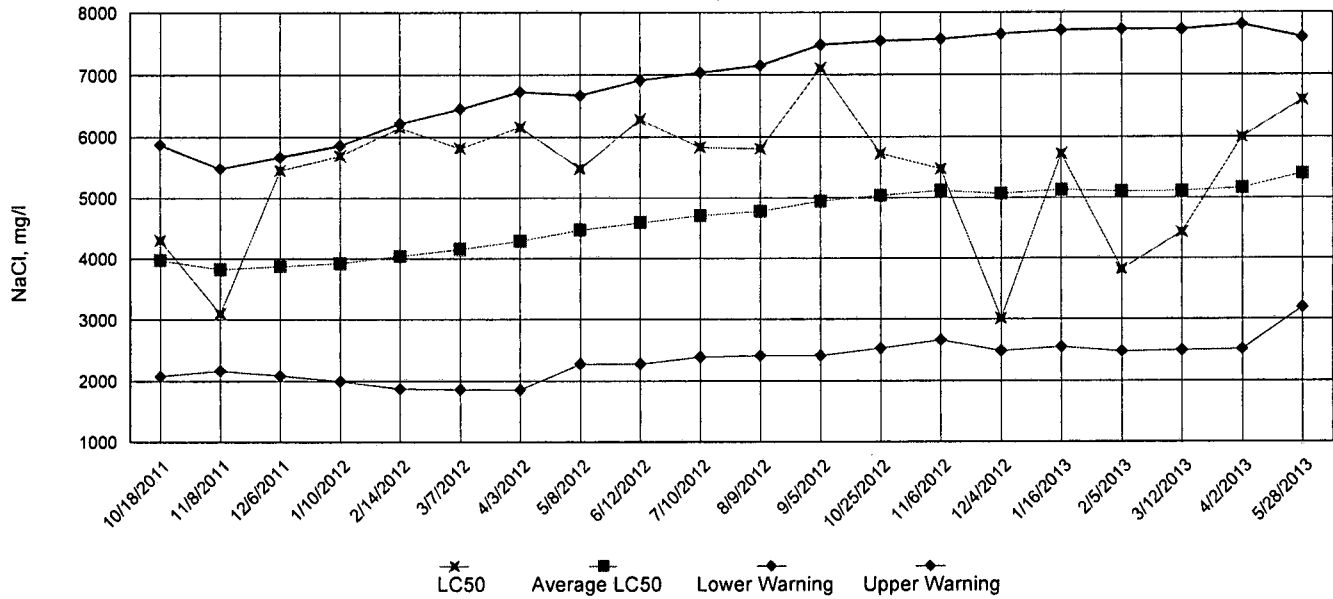
*1 = data from the *Pimephales promelas* (Fathead Minnow) test

*2 = data from the *Ceriodaphnia dubia* test

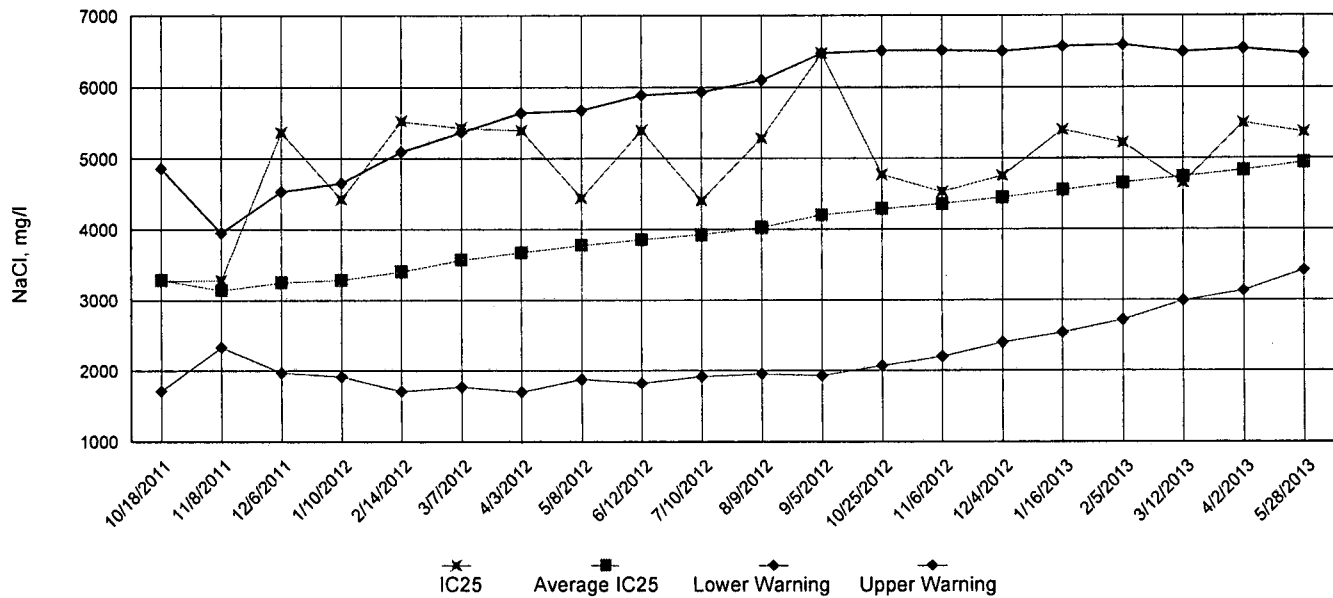
Appendix A4: Test 1000.0

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

LC50 Survival Data

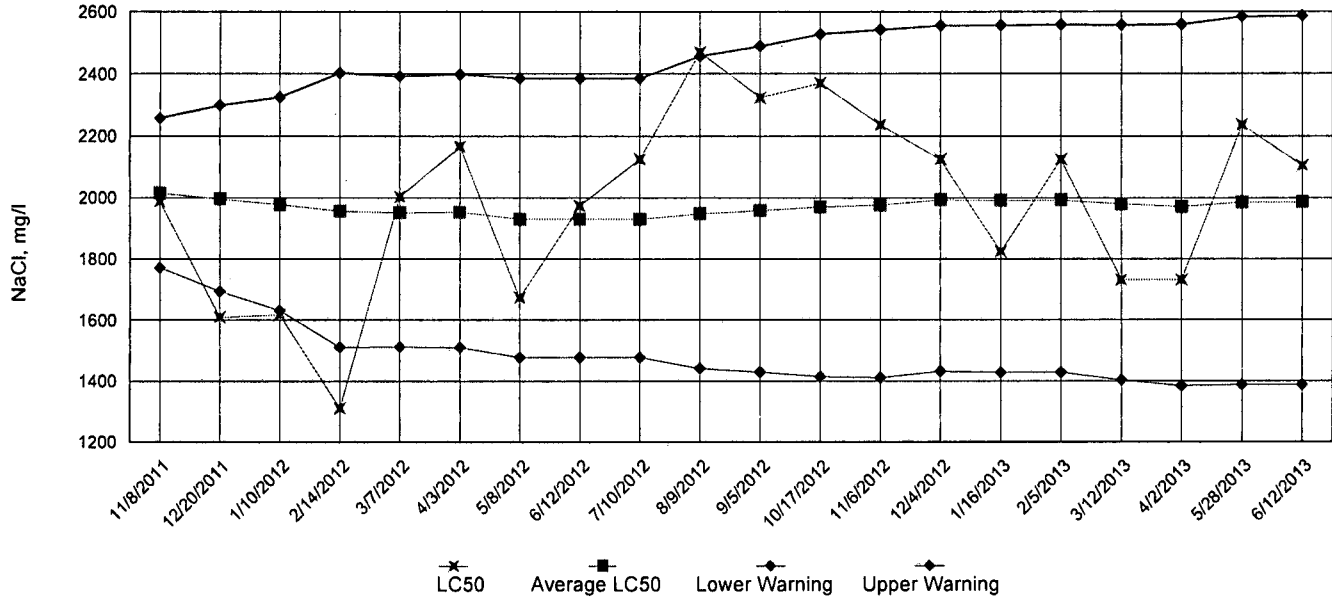


IC25 Growth Data

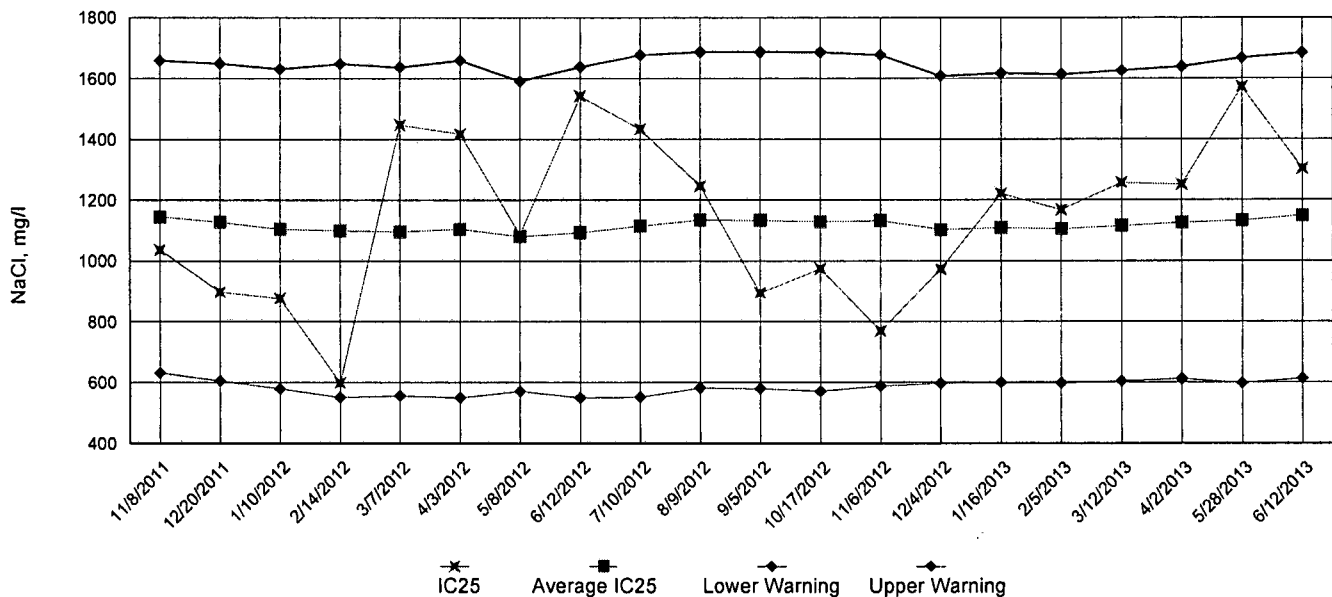


Appendix A4: Test 1002.0
Chronic Reference Toxicant, *Ceriodaphnia dubia*

LC50 Survival Data



IC25 Reproduction Data



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

Permittee: El Dorado Water Utilities

NPDES No.: AR0033723 AFIN No. 70-00341

Date and Time Test Initiated: June 4, 2013 at 1600

Date and Time Test Terminated: June 11, 2013 at 1400

Dilution water used: Synthetic Soft Water #3995

DATA TABLE FOR SURVIVAL

Effluent Conc. %	Percent Survival in replicate chambers					Mean percent survival			CV%
	A	B	C	D	E	24 hr	48 hr	7 days	
Control	100	100	100	100	100	100	100	100	0.00
32 %	100	87.5	100	100	87.5	100	100	95.0	7.21
45 %	100	100	87.5	100	100	100	100	97.5	5.73
56 %	100	87.5	100	100	100	100	100	97.5	5.73
75 %	62.5	75.0	100	87.5	87.5	100	97.5	82.5	17.3
100 %	25.0	0.00	12.5	0.00	25.0	100	40.0	12.5	100

DATA TABLE FOR GROWTH

Effluent Conc. %	Average dry weight, mg replicate chambers					Mean dry weight, mg	CV%
	A	B	C	D	E		
Control	0.280	0.265	0.315	0.306	0.300	0.293	6.94
32 %	0.292	0.286	0.339	0.308	0.355	0.316	9.48
45 %	0.274	0.318	0.270	0.278	0.341	0.296	10.7
56 %	0.199	0.258	0.285	0.299	0.324	0.273	17.5
75 %	0.185	0.234	0.260	0.286	0.268	0.247	15.9
100 %	0.047	0.000	0.025	0.000	0.082	0.0308	113

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

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

1. Steel's Many-One Rank Test:

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

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

2. Dunnett's Test:

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

a.) LOW FLOW OR CRITICAL DILUTION	(100 %)	<u> X </u> YES	<u> </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]: 1 (TLP6C)
4. If you answered NO to 2.a) enter [0] otherwise enter [1]: 1 (TGP6C)
5. NOEC *Pimephales* Lethality: 75 % (TOP6C)
6. LOEC *Pimephales* Lethality: 100 % (TXP6C)
7. NOEC *Pimephales* Sublethality: 75 % (TPP6C)
8. LOEC *Pimephales* Sublethality: 75 % (TYP6C)
9. Coefficient of variation for *Pimephales* growth: 113 (TQP6C)

Appendix B: Test 1000.0

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

PERMITTEE: El Dorado Water Utilities
NPDES NO.: AR0033723 AFIN No. 70-00341
CONTACT: Mr. Harold Baker
ANALYST: 280, 298, 304, 307

Test Initiated: DATE: June 4, 2013 TIME: 1600
Test Terminated: DATE: June 11, 2013 TIME: 1400

DILUTION Control	DAY						
	1	2	3	4	5	6	7
D.O. Initial	7.9	7.8	7.6	7.8	8.6	8.0	8.0
Final	7.6	7.2	6.9	8.2	7.1	7.6	6.8
pH Initial	7.7	7.6	7.6	7.6	7.7	7.5	7.4
Final	7.5	7.3	7.2	7.3	7.3	7.4	7.1
Alkalinity	30	NA	30	NA	30	NA	NA
Hardness	47	NA	47	NA	44	NA	NA
Conductivity	170	160	160	160	160	170	170
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION 32 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.0	7.7	7.9	7.5	8.8	8.1	7.8
Final	7.4	6.9	6.6	7.6	7.1	7.3	6.5
pH Initial	7.3	7.3	7.2	7.3	7.4	7.5	7.2
Final	7.4	7.3	7.1	7.3	7.3	7.3	7.0
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	330	330	320	320	320	340	330
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 45 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.1	7.9	7.9	7.6	8.9	7.9	7.9
Final	7.4	7.0	6.7	7.3	6.9	7.4	6.5
pH Initial	7.2	7.2	7.1	7.2	7.3	7.5	7.1
Final	7.4	7.3	7.1	7.2	7.2	7.2	7.0
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	380	380	380	380	380	390	390
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 56 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.2	7.8	8.2	7.6	7.7	7.7	7.7
Final	7.5	7.2	6.8	7.4	6.9	7.5	6.9
pH Initial	6.9	7.0	7.0	7.1	6.9	7.2	7.0
Final	7.2	7.3	7.1	7.2	7.2	7.3	7.0
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	440	440	430	440	430	450	430
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 75 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.3	7.8	8.4	8.0	8.3	7.7	8.0
Final	7.4	7.1	7.0	7.6	7.2	7.3	7.0
pH Initial	6.7	6.9	6.8	6.9	7.0	7.2	6.8
Final	7.2	7.2	7.0	7.2	7.2	7.2	6.9
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	530	530	520	520	520	520	520
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION 100 %	DAY						
	1	2	3	4	5	6	7
D.O. Initial	8.5	7.8	8.5	8.3	8.5	7.9	8.9
Final	7.4	7.0	6.9	7.4	7.1	7.3	7.0
pH Initial	6.4	6.6	6.6	6.5	6.8	7.2	6.3
Final	7.0	7.0	6.9	7.1	7.1	7.0	6.8
Alkalinity	16	NA	12	NA	11	NA	NA
Hardness	8.9	NA	9.6	NA	7.0	NA	NA
Conductivity	640	650	650	650	640	660	650
Chlorine	<0.05	NA	0.050	NA	<0.05	NA	NA

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

Permittee: El Dorado Water Utilities

NPDES No.: AR0033723 AFIN No. 70-00341

Date and Time Test Initiated: June 4, 2013 at 1420

Date and Time Test Terminated: June 11, 2013 at 1620

Dilution water used: Synthetic Soft Water #3995

PERCENT SURVIVAL

Time of Reading	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
24 hour	100	100	100	100	100	100
48 hour	100	100	100	100	100	70.0
7 day	100	100	100	90.0	100	70.0

NUMBER OF YOUNG PRODUCED PER FEMALE @ 7 DAYS

Replicates	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
A	25	12	23	0	20	20
B	28	24	25	22	18	0
C	12	17	15	13	7	0
D	18	16	18	28	15	0
E	19	24	24	21	18	19
F	16	24	23	23	18	14
G	22	13	19	23	22	19
H	14	14	16	12	17	11
I	23	23	22	20	14	18
J	17	20	18	30	15	16
Mean per Adult	19.4	18.7	20.3	19.2	16.4	11.7
Mean per Surviving Adult	19.4	18.7	20.3	21.3	16.4	16.7
CV %	26.0	26.1	17.4	27.9	24.9	19.5

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

2. Dunnett's Test:

Is the mean 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	(100 %)	<u> X </u> YES	<u> </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]: 1 (TGP3B)

5. NOEC Ceriodaphnia Lethality: 100 % (TOP3B)

6. LOEC Ceriodaphnia Lethality: 100 % (TXP3B)

7. NOEC Ceriodaphnia Sublethality: 75 % (TPP3B)

8. LOEC Ceriodaphnia Sublethality: 100 % (TYP3B)

9. Coefficient of variation for Ceriodaphnia Reproduction: 26 (TQP3B)

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

PERMITTEE: El Dorado Water Utilities
NPDES NO.: AR0033723 AFIN No. 70-00341
CONTACT: Mr. Harold Baker
ANALYST: 280, 298, 304, 307

Test Initiated: DATE: June 4, 2013 TIME: 1420
Test Terminated: DATE: June 11, 2013 TIME: 1620

DILUTION	DAY						
	1	2	3	4	5	6	7
Control							
D.O. Initial	7.9	7.8	7.6	7.8	8.6	8.0	8.0
Final	7.9	7.8	8.0	8.1	7.9	8.6	7.8
pH Initial	7.7	7.6	7.6	7.6	7.7	7.5	7.4
Final	8.0	8.0	7.9	7.9	8.0	7.6	7.8
Alkalinity	30	NA	30	NA	30	NA	NA
Hardness	47	NA	47	NA	44	NA	NA
Conductivity	170	160	160	160	160	170	170
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
32 %							
D.O. Initial	8.0	7.7	7.9	7.5	8.8	8.1	7.8
Final	8.2	8.1	8.3	8.2	8.1	8.5	8.4
pH Initial	7.3	7.3	7.2	7.3	7.4	7.5	7.2
Final	8.0	8.1	8.1	8.0	8.0	7.6	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	330	330	320	320	320	340	330
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
45 %							
D.O. Initial	8.1	7.9	7.9	7.6	8.9	7.9	7.9
Final	8.4	8.0	8.0	8.1	8.1	8.5	8.0
pH Initial	7.2	7.2	7.1	7.2	7.3	7.5	7.1
Final	8.2	8.1	8.1	7.9	8.0	7.6	7.9
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	380	380	380	380	380	390	390
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
56 %							
D.O. Initial	8.2	7.8	8.2	7.6	7.7	7.7	7.7
Final	8.1	7.8	8.2	8.1	8.4	8.2	8.2
pH Initial	6.9	7.0	7.0	7.1	6.9	7.2	7.0
Final	8.1	8.0	8.2	7.9	8.0	7.6	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	440	440	430	440	430	450	430
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
75 %							
D.O. Initial	8.3	7.8	8.4	8.0	8.3	7.7	8.0
Final	8.1	7.8	8.2	8.1	8.3	8.2	8.2
pH Initial	6.7	6.9	6.8	6.9	7.0	7.2	6.8
Final	7.9	7.9	8.0	7.8	7.9	7.5	7.7
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	530	530	520	520	520	520	520
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
100 %							
D.O. Initial	8.5	7.8	8.5	8.3	8.5	7.9	8.9
Final	7.9	7.8	8.2	8.1	8.4	8.3	8.3
pH Initial	6.4	6.6	6.6	6.5	6.8	7.2	6.3
Final	7.6	7.8	8.0	7.8	7.8	7.4	7.6
Alkalinity	16	NA	12	NA	11	NA	NA
Hardness	8.9	NA	9.6	NA	7.0	NA	NA
Conductivity	640	650	650	650	640	660	650
Chlorine	<0.05	NA	0.050	NA	<0.05	NA	NA

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

PAGE 1 OF 1

Client: <u>EL DORADO WATER UTILITIES</u>			PO No.		NO OF	ANALYSES REQUESTED										AIC CONTROL NO: <u>167908</u>																						
Project Reference: <u>SOUTH EFFLUENT</u>			SAMPLE MATRIX			BOTTLES	<table border="1"> <tr><td>BIOMONITORING</td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td></tr> </table>										BIOMONITORING																					AIC PROPOSAL NO:
BIOMONITORING																																						
Project Manager: <u>HAROLD BAKER</u>			G R A B	C O M P	W A T E R	S O I L	NO OF											Carrier: <u>Fed-Ex</u>																				
Sampled By: <u>JOHN M. PEPPERS</u>																		Date/Time Collected		Received Temperature C <u>2</u>																		
AIC No.	Sample Identification	Date/Time Collected																Remarks																				
<u>3</u>	<u>SE-</u>	<u>0930 6-7-13</u>	<u>OK</u>	<input checked="" type="checkbox"/>			<u>3</u>											<u>ID: SE-1658</u>																				
			Container Type				<u>P</u>											Field pH calibration on _____ @ _____																				
			Preservative				<u>No</u>											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) <u>NORMAL</u> or EXPEDITED IN _____ DAYS							Relinquished By: <u>JOHN M. PEPPERS</u> <u>John M. Pepp</u>		Date/Time <u>1545</u> <u>6-7-13</u>		Received By: <u>FED EX</u>		Date/Time <u>1600</u> <u>6-7-13</u>																									
Expedited results requested by: _____							Relinquished By: <u>FEDEX</u>		Date/Time <u>0830</u> <u>6-8-13</u>		Received in Lab By: <u>Anthony</u>		Date/Time <u>6-8-13</u> <u>0830</u>																									
Who should AIC contact with questions: <u>JOHN M. PEPPERS</u>							Comments:																															
Phone: <u>870-814-1764</u> LAB # <u>870-862-0421</u>																																						
Report Attention to: <u>HAROLD BAKER</u>																																						
Report Address to: <u>P.O. Box 1587</u> <u>EL DORADO, AR 71731</u>																																						



June 19, 2013
Control No. 168124-1
Page 1 of 30

June 19, 2013

Test Results of
Second Quarter
Chronic 7-Day Renewal
Biomonitoring Testing
for
North Effluent
El Dorado, AR

Control No. 168124-1

Prepared for:

Mr. Harold Baker
El Dorado Water Utilities
Post Office Box 1587
El Dorado, AR 71731

Prepared by:

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



El Dorado Water Utilities
ATTN: Mr. Harold Baker
Post Office Box 1587
El Dorado, AR 71731

Re: Chronic 7-Day Renewal utilizing *Pimephales promelas* (Fathead minnow) and *Ceriodaphnia dubia*
North Effluent - El Dorado, AR
NPDES Permit No. AR0033936 AFIN#70-00341

Dear Mr. Harold Baker:

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 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: The No Observable Effects Concentration (NOEC) for survival occurred at 100 % effluent, which is equal to the critical dilution of 100 %. 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 *Ceriodaphnia dubia* test.**

AMERICAN INTERPLEX CORPORATION

John Overbey
Laboratory Director

PDF cc: El Dorado Water Utilities
ATTN: Mr. Harold Baker
harold@eldoradowater.com

El Dorado Water Utilities
ATTN: Mr. John Peppers
lab@eldoradowater.com

GBMc & Associates, Inc.
ATTN: Mr. Roland McDaniel
rmcdaniel@gbmcassoc.com

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Pimephales promelas (Fathead minnow) Survival and Growth

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Ceriodaphnia dubia Survival and Reproduction

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Appendix B: Chains of Custody

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	0.423	PASS
Control Growth CV < or = 40%	6.22	PASS
Growth Minimum Significant Difference 12 to 30%	18.0	PASS
Critical Dilution CV < or = 40%	19.5	PASS

Ceriodaphnia dubia Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	25.3	PASS
Control CV < or = 40% per Surviving Female	5.29	PASS
Reproduction Minimum Significant Difference 13 to 47%	11.8	BELOW
Critical Dilution CV < or = 40%	12.6	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0033936 AFIN#70-00341
2. Test Requirements: Chronic Biomonitoring, Quarterly Test Methods 1000.0 and 1002.0
3. Receiving Stream: Flat Creek to Ouachita River Basin

B. Source of Effluent/Dilution Water

1. Effluent Samples:

- a. Sampling Point: North Effluent
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.1	7.8	8.6
pH (standard units)	7.4	7.3	6.8
Alkalinity (mg/l as CaCO ₃)	49	25	34
Hardness (mg/l as CaCO ₃)	14	15	15
Conductivity (umhos/cm)	390	550	590
Residual Chlorine (mg/l)	0.060	<0.05	0.050
Ammonia as N (mg/l)	0.39	0.62	0.63

2. Dilution Water Samples: Synthetic Soft Water #3998

- a. Dates Prepared: June 10 through 24, 2013
- b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.0	8.1	8.7
pH (standard units)	7.6	7.6	7.7
Alkalinity (mg/l as CaCO ₃)	30	30	30
Hardness (mg/l as CaCO ₃)	43	42	42
Conductivity (umhos/cm)	170	170	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 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: June 11, 2013 at 1415
Date & Time Test Terminated: June 18, 2013 at 1320
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: June 11, 2013 at 1205
Date & Time Test Terminated: June 18, 2013 at 1005
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 Bartlett's test and analyzed with Dunnett's Test to determine the No Observable Effects Concentration (NOEC) for Reproduction.

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 May 28, 2013 at 1645 to June 4, 2013 at 1520

The results were as follows: (Control No. 167778-1.)

Survival LC-50: 6598 mg/l

Growth IC-25: 5369 mg/l

Growth PMSD: 22.6

Ceriodaphnia dubia

Chronic reference tests are performed monthly.

A chronic reference test was performed on May 28, 2013 at 1700 to June 4, 2013 at 1500

The results were as follows: (Control No. 167778-2.)

Survival LC-50: 2236 mg/l

Growth IC-25: 1573 mg/l

Growth PMSD: 13.9

V. Chemical Analysis/Quality Control

Parameter	Method	% Recovery	Relative % Difference
Alkalinity	SM 2320 B	NA	0.00
Hardness	EPA 200.7	99.9	0.300
pH	SM 4500-H+ B	100	0.336
Conductivity	EPA 120.1	103	4.50

VI. Organism History

Pimephales promelas (Fathead minnow)

Date: June 11, 2013

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: June 11, 2013

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

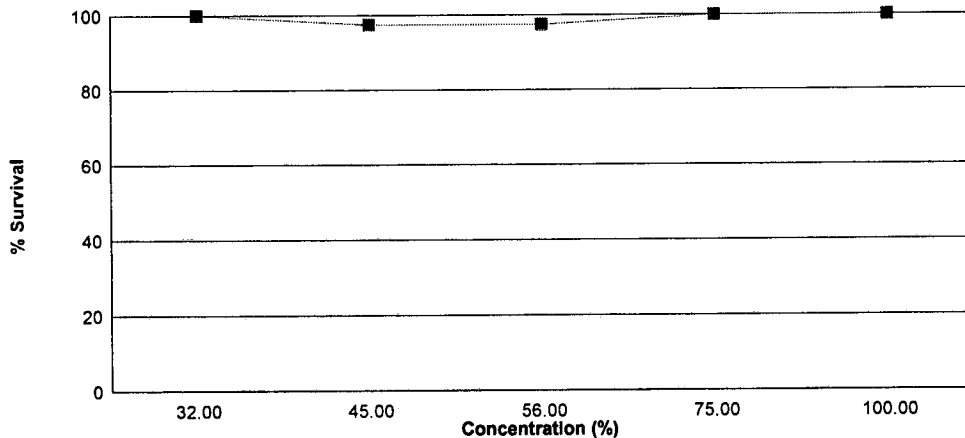
Larvae are exposed in a static renewal system for seven days to different concentrations of effluent with dilution water. Test results are based on the survival and growth (increase in weight) of the larvae.

Effluent dilutions for this test were 32 %, 45 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

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

The test was initiated on June 11, 2013 at 1415 and continued through June 18, 2013 at 1320. 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	0.423
32 %	100	0.371
45 %	97.5	0.449
56 %	97.5	0.386
75 %	100	0.430
100 %	100	0.396

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

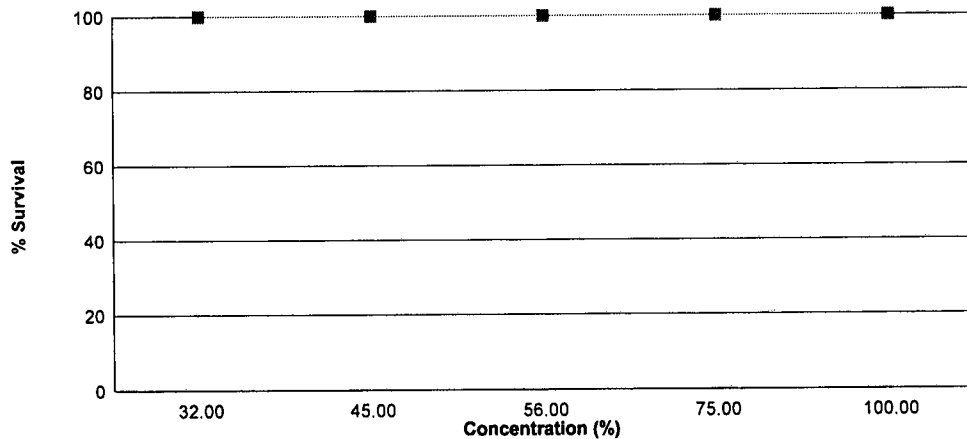
Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring with an average of at least 15 young per female.

Effluent dilutions for this test were 32 %, 45 %, 56 %, 75 %, 100 % in accordance with the NPDES permit.

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

The test was initiated on June 11, 2013 at 1205 and continued through June 18, 2013 at 1005. 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



Concentration	Percent Survival	Mean Reproduction
Control	100	25.3
32 %	100	23.9
45 %	100	24.2
56 %	100	25.0
75 %	100	25.9
100 %	100	24.0

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Survival

Date and Time Test Initiated: June 11, 2013 at 1415

Date and Time Test Terminated: June 18, 2013 at 1320

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
45 %	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	7	7	7	7	7	7	7
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	7	7
75 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
100 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Growth

Test Initiated: June 11, 2013 at 1415
Test Terminated: June 18, 2013 at 1320

Drying Started: June 17, 2013 at 1424
Drying Ended: June 19, 2013 at 1515

Concentration	Replicate	Weight of pan	Weight of pan + fish	Total weight of fish (g)	Original # of fish	Mean dry weight (mg)
Control	A	.91668	.91995	0.00327	8	0.409
	B	.91734	.92108	0.00374	8	0.468
	C	.91781	.92114	0.00333	8	0.416
	D	.91889	.92210	0.00321	8	0.401
	E	.91893	.92229	0.00336	8	0.420
32 %	A	.91763	.92114	0.00351	8	0.439
	B	.92199	.92497	0.00298	8	0.372
	C	.92051	.92320	0.00269	8	0.336
	D	.91742	.92007	0.00265	8	0.331
	E	.91777	.92080	0.00303	8	0.379
45 %	A	.92267	.92626	0.00359	8	0.449
	B	.92502	.92860	0.00358	8	0.448
	C	.92695	.93047	0.00352	8	0.440
	D	.92797	.93196	0.00399	8	0.499
	E	.92473	.92799	0.00326	8	0.408
56 %	A	.92437	.92755	0.00318	8	0.398
	B	.91945	.92262	0.00317	8	0.396
	C	.92100	.92413	0.00313	8	0.391
	D	.92285	.92641	0.00356	8	0.445
	E	.92171	.92410	0.00239	8	0.299
75 %	A	.92677	.92956	0.00279	8	0.349
	B	.92485	.92830	0.00345	8	0.431
	C	.92369	.92714	0.00345	8	0.431
	D	.91985	.92333	0.00348	8	0.435
	E	.92051	.92456	0.00405	8	0.506
100 %	A	.92033	.92249	0.00216	8	0.270
	B	.93472	.93853	0.00381	8	0.476
	C	.93609	.93927	0.00318	8	0.398
	D	.91833	.92182	0.00349	8	0.436
	E	.93677	.93998	0.00321	8	0.401

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: June 11, 2013 at 1205

Date and Time Test Terminated: June 18, 2013 at 1005

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	2	4	0	4	4	0	4	0	0	18	10	1.80
4	4	0	0	4	0	0	4	0	3	3	18	10	1.80
5	10	10	7	9	9	8	9	10	11	9	92	10	9.20
6	0	0	0	0	0	0	0	0	0	0	0	10	0.00
7	12	13	14	10	13	12	14	13	12	12	125	10	12.5
8													
TOTAL	26	25	25	23	26	24	27	27	26	24	253	10	25.3

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	3	4	3	0	3	4	0	4	3	0	24	10	2.40
4	0	0	0	2	0	0	5	0	0	4	11	10	1.10
5	9	10	10	5	10	9	8	9	11	9	90	10	9.00
6	0	0	0	0	0	0	0	0	0	0	0	10	0.00
7	14	13	12	6	12	11	12	13	11	10	114	10	11.4
8													
TOTAL	26	27	25	13	25	24	25	26	25	23	239	10	23.9

Concentration: 45 %													
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	3	0	3	2	3	3	4	4	2	0	24	10	2.40
4	0	4	0	0	0	0	0	0	0	4	8	10	0.800
5	9	10	9	10	12	10	9	8	8	7	92	10	9.20
6	0	0	0	0	0	0	0	0	0	0	0	10	0.00
7	13	11	13	12	13	12	10	11	13	10	118	10	11.8
8													
TOTAL	25	25	25	24	28	25	23	23	23	21	242	10	24.2

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: June 11, 2013 at 1205

Date and Time Test Terminated: June 18, 2013 at 1005

Concentration: 56 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	4	0	3	3	4	2	3	4	3	0	26	10	2.60	
4	0	4	0	0	0	0	0	0	0	4	8	10	0.800	
5	10	8	8	9	10	9	10	12	10	9	95	10	9.50	
6	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
7	14	9	13	14	13	12	11	16	10	9	121	10	12.1	
8														
TOTAL	28	21	24	26	27	23	24	32	23	22	250	10	25.0	

Concentration: 75 %														
Day	Replicate										No. of Young	No. of Adults	Young per Adult	
	1	2	3	4	5	6	7	8	9	10				
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
3	4	2	2	2	3	2	3	3	0	0	21	10	2.10	
4	0	4	0	0	0	0	0	0	5	5	14	10	1.40	
5	10	12	11	9	10	12	10	11	9	9	103	10	10.3	
6	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
7	10	12	14	10	15	14	11	15	8	12	121	10	12.1	
8														
TOTAL	24	30	27	21	28	28	24	29	22	26	259	10	25.9	

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	2	0	3	4	5	3	4	2	2	0	25	10	2.50	
4	0	4	0	0	0	0	0	0	0	2	6	10	0.600	
5	9	9	11	9	10	11	9	9	8	8	93	10	9.30	
6	0	0	0	0	0	0	0	0	0	0	0	10	0.00	
7	13	11	11	10	15	9	14	13	9	11	116	10	11.6	
8														
TOTAL	24	24	25	23	30	23	27	24	19	21	240	10	24.0	

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	45 %	1	1.00000	1.39310
3	45 %	2	1.00000	1.39310
3	45 %	3	1.00000	1.39310
3	45 %	4	1.00000	1.39310
3	45 %	5	0.87500	1.20940
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	0.87500	1.20940
5	75 %	1	1.00000	1.39310
5	75 %	2	1.00000	1.39310
5	75 %	3	1.00000	1.39310
5	75 %	4	1.00000	1.39310
5	75 %	5	1.00000	1.39310
6	100 %	1	1.00000	1.39310
6	100 %	2	1.00000	1.39310
6	100 %	3	1.00000	1.39310
6	100 %	4	1.00000	1.39310
6	100 %	5	1.00000	1.39310

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Survival

Shapiro - Wilk's Test for Normality		Transform: Arc Sin(Square Root(Y))
<p>D = 0.05399 W = 0.5466 Critical W = 0.9 (alpha = 0.01, N = 30) Critical W = 0.927 (alpha = 0.05, N = 30)</p> <p>Data FAIL normality test (alpha = 0.01).</p>		

Steel's Many-One Rank Test				Transform: Arc Sin(Square Root(Y))	
Ho: Control < Treatment					
Group	Identification	Rank Sum	Critical Value	DF	Sig 0.05
1	Control				
2	32 %	27.50	16.00	5.00	
3	45 %	25.00	16.00	5.00	
4	56 %	25.00	16.00	5.00	
5	75 %	27.50	16.00	5.00	
6	100 %	27.50	16.00	5.00	
Critical values are 1 tailed (k=5)					

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Growth

Shapiro - Wilk's Test for Normality	No Transformation
<p>D = 0.06213 W = 0.9288 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 = 5.200 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	0.02174	0.004348	1.679	
Within (Error)	24	0.06213	0.002589		
Total	29	0.08387			
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	0.4228	0.4228			
2	32 %	0.3714	0.3714	1.597		
3	45 %	0.4488	0.4488	-0.8079		
4	56 %	0.3858	0.3858	1.15		
5	75 %	0.4304	0.4304	-0.2362		
6	100 %	0.3962	0.3962	0.8266		
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	0.07595	18	0.0514		
3	45 %	5	0.07595	18	-0.026		
4	56 %	5	0.07595	18	0.037		
5	75 %	5	0.07595	18	-0.0076		
6	100 %	5	0.07595	18	0.0266		

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
45 %	10	0	10
Total	20	0	20

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

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

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

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

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

Appendix A2: Statistics

Ceriodaphnia dubia Survival

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

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

Summary of Fisher's Exact Test				
Group	Identification	Exposed	Dead	Sig 0.05
0	Control	10	0	
1	32 %	10	0	
2	45 %	10	0	
3	56 %	10	0	
4	75 %	10	0	
5	100 %	10	0	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

Kolmogorov Test for Normality	No Transformation
D = 0.1025 D* = 0.8042 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 = 11.63 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	32.68	6.536	0.7783
Within (Error)	54	453.5	8.398	
Total	59	486.2		
Critical F = 3.38 (alpha = 0.01, df = 5,54)				
2.38 (alpha = 0.05, df = 5,54)				
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	25.3	25.3		
2	32 %	23.9	23.9	1.08	
3	45 %	24.2	24.2	0.8488	
4	56 %	25	25	0.2315	
5	75 %	25.9	25.9	-0.463	
6	100 %	24	24	1.003	
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	2.994	11.8	1.4
3	45 %	10	2.994	11.8	1.1
4	56 %	10	2.994	11.8	0.3
5	75 %	10	2.994	11.8	-0.6
6	100 %	10	2.994	11.8	1.3

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: June 11, 2013 at 0841

Date and Time Test Terminated: June 18, 2013 at 1320

Effluent Conc.: Control	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.0	8.0	8.1	8.1	8.7	7.9	7.8
	Final *1	7.4	7.2	7.9	8.4	7.5	7.2	6.5
	Final *2	8.0	8.4	8.0	7.9	8.4	8.3	8.1
pH, units	Initial	7.6	7.6	7.6	7.5	7.7	7.6	7.6
	Final *1	7.4	7.3	7.4	7.6	7.7	7.3	7.1
	Final *2	8.0	7.9	7.8	8.0	8.0	7.8	7.8
Alkalinity, mg CaCO ₃ /l	30	NA	30	NA	30	NA	NA	
Hardness, mg CaCO ₃ /l	43	NA	42	NA	42	NA	NA	
Conductivity, umhos/cm	170	170	170	300	170	170	160	
Res. Chlorine, mg/l	<0.05	NA	<0.05	NA	<0.05	NA	NA	

Effluent Conc.: 32 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.5	7.8	8.0	8.4	8.6	7.9	7.8
	Final *1	7.5	7.3	7.9	8.5	7.9	7.0	6.1
	Final *2	8.0	8.5	7.5	7.9	8.3	8.3	8.2
pH, units	Initial	7.6	7.5	7.5	7.1	7.5	7.6	7.2
	Final *1	7.6	7.4	7.3	7.5	7.7	7.2	7.1
	Final *2	8.1	8.0	7.9	7.9	8.0	7.8	7.8

Effluent Conc.: 45 %	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	
DO, mg/l	Initial	8.6	8.0	7.9	8.5	8.5	8.0	7.8
	Final *1	7.4	6.9	7.6	8.2	7.9	7.0	6.1
	Final *2	8.4	8.7	7.3	8.0	8.2	8.1	8.1
pH, units	Initial	7.5	7.5	7.5	7.0	7.4	7.6	7.0
	Final *1	7.6	7.4	7.3	7.5	7.7	7.3	7.0
	Final *2	8.2	8.1	7.9	7.9	8.0	7.7	7.8

Appendix A3: Water Chemistry

Routine Chemical and Physical Data

Date and Time Test Initiated: June 11, 2013 at 0841

Date and Time Test Terminated: June 18, 2013 at 1320

Effluent Conc.: 56 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.8	8.0	7.9	8.7	8.6	8.0	7.8
	Final *1	7.6	7.7	7.8	8.3	7.8	7.3	5.7
	Final *2	8.4	8.5	7.2	8.1	8.2	8.2	8.2
pH, units	Initial	7.5	7.5	7.5	6.9	7.5	7.6	6.9
	Final *1	7.6	7.6	7.2	7.5	7.7	7.2	7.0
	Final *2	8.2	8.0	7.9	7.9	7.9	7.7	7.8

Effluent Conc.: 75 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.9	7.9	7.9	8.8	8.7	8.0	7.8
	Final *1	7.7	7.0	7.5	8.1	7.8	7.4	6.1
	Final *2	8.1	8.1	7.1	8.0	8.0	8.1	8.2
pH, units	Initial	7.4	7.4	7.5	6.6	7.4	7.6	6.8
	Final *1	7.6	7.4	7.2	7.4	7.7	7.3	7.0
	Final *2	8.2	8.1	7.9	7.9	7.9	7.7	7.8

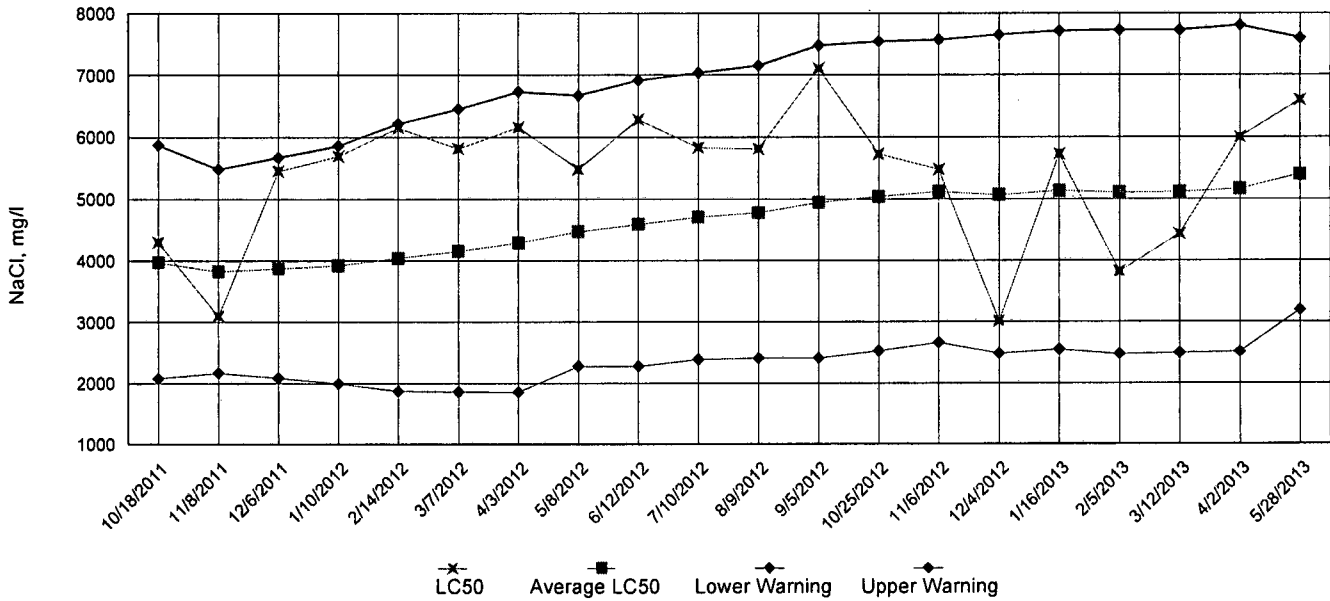
Effluent Conc.: 100 %		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
DO, mg/l	Initial	8.1	8.2	7.8	8.6	8.6	8.0	8.0
	Final *1	7.5	7.5	8.1	8.4	7.9	7.3	6.4
	Final *2	8.1	8.4	7.2	8.1	8.4	8.4	8.0
pH, units	Initial	7.4	7.3	7.3	7.9	6.8	7.5	6.5
	Final *1	7.6	7.6	7.4	7.5	7.7	7.2	7.0
	Final *2	8.2	8.2	8.0	7.9	7.9	7.7	7.7
Alkalinity, mg CaCO ₃ /l	49	NA	25	NA	34	NA	NA	NA
Hardness, mg CaCO ₃ /l	14	NA	15	NA	15	NA	NA	NA
Conductivity, umhos/cm	390	550	550	310	590	590	580	580
Res. Chlorine, mg/l	0.060	NA	<0.05	NA	0.050	NA	NA	NA

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

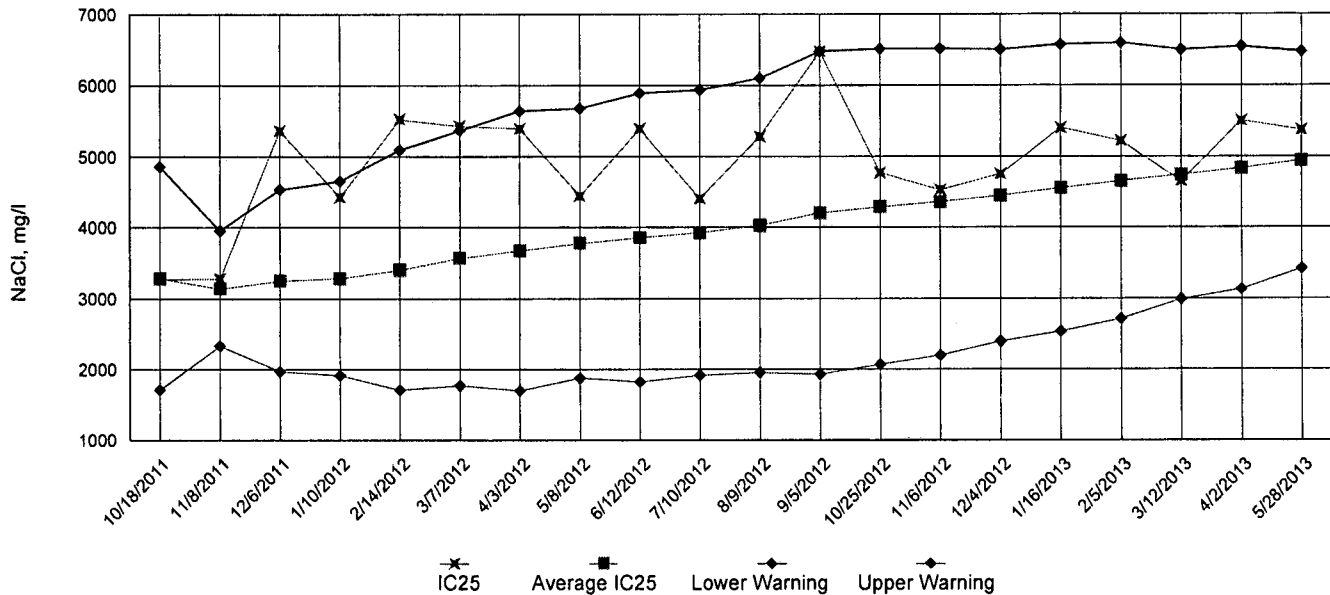
*2 = data from the *Ceriodaphnia dubia* test

Appendix A4: Test 1000.0
Chronic Reference Toxicant, *Pimephales promelas* (Fathead Minnow)

LC50 Survival Data

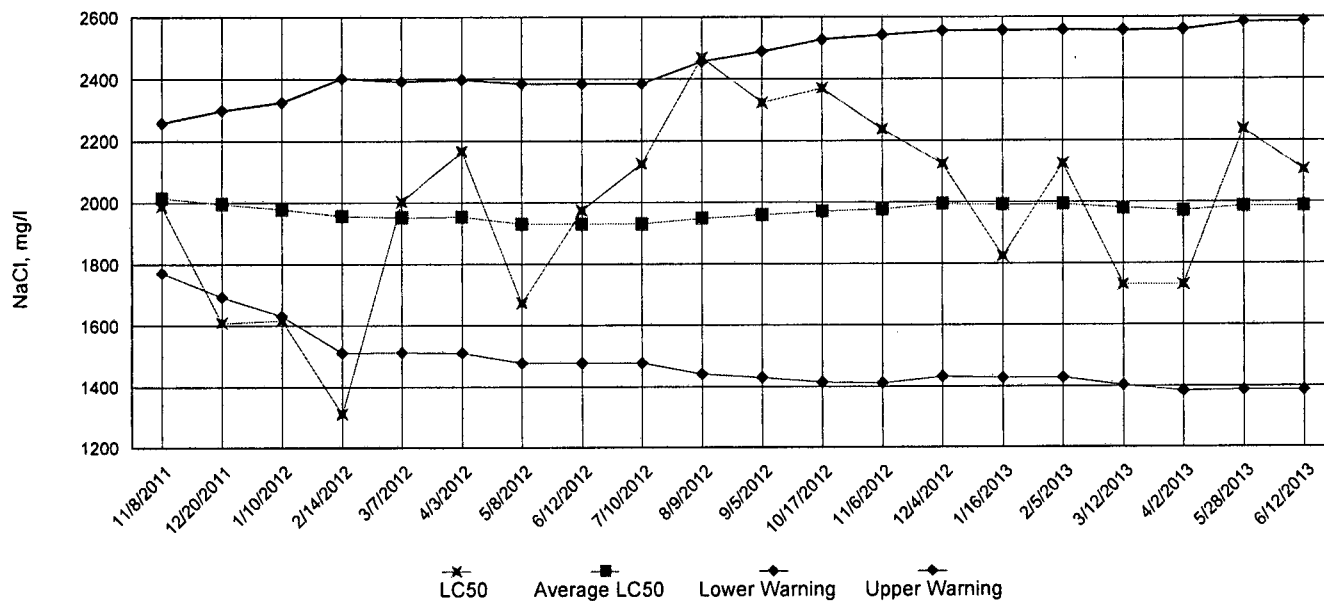


IC25 Growth Data

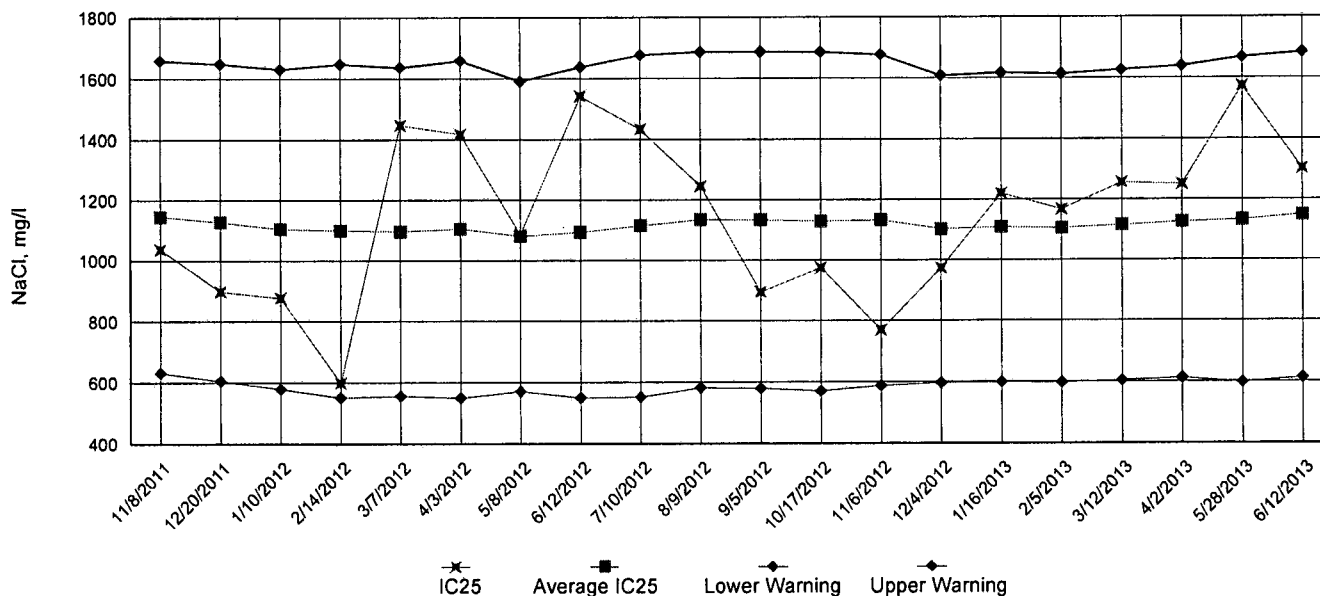


Appendix A4: Test 1002.0
Chronic Reference Toxicant, *Ceriodaphnia dubia*

LC50 Survival Data



IC25 Reproduction Data



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

Permittee: El Dorado Water Utilities

NPDES No.: AR0033936 AFIN#70-00341

Date and Time Test Initiated: June 11, 2013 at 1415

Date and Time Test Terminated: June 18, 2013 at 1320

Dilution water used: Synthetic Soft Water #3998

DATA TABLE FOR SURVIVAL

Effluent Conc. %	Percent Survival in replicate chambers					Mean percent survival			CV%
	A	B	C	D	E	24 hr	48 hr	7 days	
Control	100	100	100	100	100	100	100	100	0.00
32 %	100	100	100	100	100	100	100	100	0.00
45 %	100	100	100	100	87.5	97.5	97.5	97.5	5.73
56 %	100	100	100	100	87.5	100	100	97.5	5.73
75 %	100	100	100	100	100	100	100	100	0.00
100 %	100	100	100	100	100	100	100	100	0.00

DATA TABLE FOR GROWTH

Effluent Conc. %	Average dry weight, mg replicate chambers					Mean dry weight, mg	CV%
	A	B	C	D	E		
Control	0.409	0.468	0.416	0.401	0.420	0.423	6.22
32 %	0.439	0.372	0.336	0.331	0.379	0.371	11.7
45 %	0.449	0.448	0.440	0.499	0.408	0.449	7.27
56 %	0.398	0.396	0.391	0.445	0.299	0.386	13.8
75 %	0.349	0.431	0.431	0.435	0.506	0.43	12.9
100 %	0.270	0.476	0.398	0.436	0.401	0.396	19.5

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 %)	<input type="checkbox"/> YES	<input checked="" type="checkbox"/> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<input type="checkbox"/> YES	<input type="checkbox"/> 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 %)	<input type="checkbox"/> YES	<input checked="" type="checkbox"/> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<input type="checkbox"/> YES	<input type="checkbox"/> 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: 19.5 (TQP6C)

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

PERMITTEE: El Dorado Water Utilities
NPDES NO.: AR0033936 AFIN#70-00341
CONTACT: Mr. Harold Baker
ANALYST: 280, 298, 304, 307

Test Initiated: DATE: June 11, 2013 TIME: 1415
Test Terminated: DATE: June 18, 2013 TIME: 1320

DILUTION	DAY						
	1	2	3	4	5	6	7
Control							
D.O. Initial	8.0	8.0	8.1	8.1	8.7	7.9	7.8
Final	7.4	7.2	7.9	8.4	7.5	7.2	6.5
pH Initial	7.6	7.6	7.6	7.5	7.7	7.6	7.6
Final	7.4	7.3	7.4	7.6	7.7	7.3	7.1
Alkalinity	30	NA	30	NA	30	NA	NA
Hardness	43	NA	42	NA	42	NA	NA
Conductivity	170	170	170	300	170	170	160
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
32 %							
D.O. Initial	8.5	7.8	8.0	8.4	8.6	7.9	7.8
Final	7.5	7.3	7.9	8.5	7.9	7.0	6.1
pH Initial	7.6	7.5	7.5	7.1	7.5	7.6	7.2
Final	7.6	7.4	7.3	7.5	7.7	7.2	7.1
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	290	290	290	350	300	300	290
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
45 %							
D.O. Initial	8.6	8.0	7.9	8.5	8.5	8.0	7.8
Final	7.4	6.9	7.6	8.2	7.9	7.0	6.1
pH Initial	7.5	7.5	7.5	7.0	7.4	7.6	7.0
Final	7.6	7.4	7.3	7.5	7.7	7.3	7.0
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	340	340	330	390	350	360	350
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
56 %							
D.O. Initial	8.8	8.0	7.9	8.7	8.6	8.0	7.8
Final	7.6	7.7	7.8	8.3	7.8	7.3	5.7
pH Initial	7.5	7.5	7.5	6.9	7.5	7.6	6.9
Final	7.6	7.6	7.2	7.5	7.7	7.2	7.0
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	380	380	380	470	400	400	390
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
75 %							
D.O. Initial	8.9	7.9	7.9	8.8	8.7	8.0	7.8
Final	7.7	7.0	7.5	8.1	7.8	7.4	6.1
pH Initial	7.4	7.4	7.5	6.6	7.4	7.6	6.8
Final	7.6	7.4	7.2	7.4	7.7	7.3	7.0
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	450	450	450	570	470	470	470
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
100 %							
D.O. Initial	8.1	8.2	7.8	8.6	8.6	8.0	8.0
Final	7.5	7.5	8.1	8.4	7.9	7.3	6.4
pH Initial	7.4	7.3	7.3	7.9	6.8	7.5	6.5
Final	7.6	7.6	7.4	7.5	7.7	7.2	7.0
Alkalinity	49	NA	25	NA	34	NA	NA
Hardness	14	NA	15	NA	15	NA	NA
Conductivity	390	550	550	310	590	590	580
Chlorine	0.060	NA	<0.05	NA	0.050	NA	NA

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

Permittee: El Dorado Water Utilities

NPDES No.: AR0033936 AFIN#70-00341

Date and Time Test Initiated: June 11, 2013 at 1205

Date and Time Test Terminated: June 18, 2013 at 1005

Dilution water used: Synthetic Soft Water #3998

PERCENT SURVIVAL

Time of Reading	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
24 hour	100	100	100	100	100	100
48 hour	100	100	100	100	100	100
7 day	100	100	100	100	100	100

NUMBER OF YOUNG PRODUCED PER FEMALE @ 7 DAYS

Replicates	Control	Percent Effluent				
		32 %	45 %	56 %	75 %	100 %
A	26	26	25	28	24	24
B	25	27	25	21	30	24
C	25	25	25	24	27	25
D	23	13	24	26	21	23
E	26	25	28	27	28	30
F	24	24	25	23	28	23
G	27	25	23	24	24	27
H	27	26	23	32	29	24
I	26	25	23	23	22	19
J	24	23	21	22	26	21
Mean per Adult	25.3	23.9	24.2	25.0	25.9	24.0
Mean per Surviving Adult	25.3	23.9	24.2	25.0	25.9	24.0
CV %	5.29	16.7	7.74	13.2	11.7	12.6

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

2. Dunnett's Test:

Is the mean 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	(100 %)	<u> </u> YES	<u> X </u> NO
b.) 1/2 LOW FLOW DILUTION	(NA)	<u> </u> YES	<u> </u> NO

3. If you answered NO to 1.a) enter [0] otherwise enter [1]: 0 (TLP3B)

4. If you answered NO to 2.a) enter [0] otherwise enter [1]: 0 (TGP3B)

5. NOEC Ceriodaphnia Lethality: 100 % (TOP3B)

6. LOEC Ceriodaphnia Lethality: 100 % (TXP3B)

7. NOEC Ceriodaphnia Sublethality: 100 % (TPP3B)

8. LOEC Ceriodaphnia Sublethality: 100 % (TYP3B)

9. Coefficient of variation for Ceriodaphnia Reproduction: 12.6 (TQP3B)

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

PERMITTEE: El Dorado Water Utilities
NPDES NO.: AR0033936 AFIN#70-00341
CONTACT: Mr. Harold Baker
ANALYST: 280, 298, 304, 307

Test Initiated: DATE: June 11, 2013 TIME: 1205
Test Terminated: DATE: June 18, 2013 TIME: 1005

DILUTION	DAY						
	1	2	3	4	5	6	7
Control							
D.O. Initial	8.0	8.0	8.1	8.1	8.7	7.9	7.8
Final	8.0	8.4	8.0	7.9	8.4	8.3	8.1
pH Initial	7.6	7.6	7.6	7.5	7.7	7.6	7.6
Final	8.0	7.9	7.8	8.0	8.0	7.8	7.8
Alkalinity	30	NA	30	NA	30	NA	NA
Hardness	43	NA	42	NA	42	NA	NA
Conductivity	170	170	170	300	170	170	160
Chlorine	<0.05	NA	<0.05	NA	<0.05	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
32 %							
D.O. Initial	8.5	7.8	8.0	8.4	8.6	7.9	7.8
Final	8.0	8.5	7.5	7.9	8.3	8.3	8.2
pH Initial	7.6	7.5	7.5	7.1	7.5	7.6	7.2
Final	8.1	8.0	7.9	7.9	8.0	7.8	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	290	290	290	350	300	300	290
Chlorine	NA	NA	NA	NA	NA	NA	NA

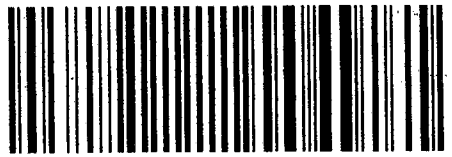
DILUTION	DAY						
	1	2	3	4	5	6	7
45 %							
D.O. Initial	8.6	8.0	7.9	8.5	8.5	8.0	7.8
Final	8.4	8.7	7.3	8.0	8.2	8.1	8.1
pH Initial	7.5	7.5	7.5	7.0	7.4	7.6	7.0
Final	8.2	8.1	7.9	7.9	8.0	7.7	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	340	340	330	390	350	360	350
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
56 %							
D.O. Initial	8.8	8.0	7.9	8.7	8.6	8.0	7.8
Final	8.4	8.5	7.2	8.1	8.2	8.2	8.2
pH Initial	7.5	7.5	7.5	6.9	7.5	7.6	6.9
Final	8.2	8.0	7.9	7.9	7.9	7.7	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	380	380	380	470	400	400	390
Chlorine	NA	NA	NA	NA	NA	NA	NA

DILUTION	DAY						
	1	2	3	4	5	6	7
75 %							
D.O. Initial	8.9	7.9	7.9	8.8	8.7	8.0	7.8
Final	8.1	8.1	7.1	8.0	8.0	8.1	8.2
pH Initial	7.4	7.4	7.5	6.6	7.4	7.6	6.8
Final	8.2	8.1	7.9	7.9	7.9	7.7	7.8
Alkalinity	NA	NA	NA	NA	NA	NA	NA
Hardness	NA	NA	NA	NA	NA	NA	NA
Conductivity	450	450	450	570	470	470	470
Chlorine	NA	NA	NA	NA	NA	NA	NA

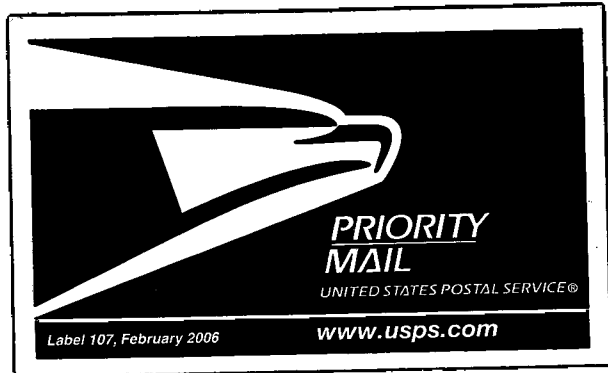
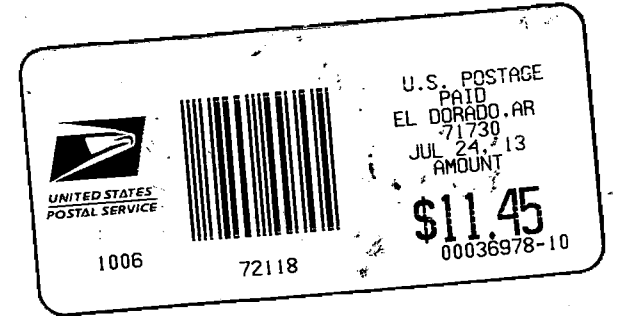
DILUTION	DAY						
	1	2	3	4	5	6	7
100 %							
D.O. Initial	8.1	8.2	7.8	8.6	8.6	8.0	8.0
Final	8.1	8.4	7.2	8.1	8.4	8.4	8.0
pH Initial	7.4	7.3	7.3	7.9	6.8	7.5	6.5
Final	8.2	8.2	8.0	7.9	7.9	7.7	7.7
Alkalinity	49	NA	25	NA	34	NA	NA
Hardness	14	NA	15	NA	15	NA	NA
Conductivity	390	550	550	310	590	590	580
Chlorine	0.060	NA	<0.05	NA	0.050	NA	NA

CERTIFIED MAIL™



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RETURN RECEIPT
REQUESTED



FROM
El Dorado Water Utilities
P.O. BOX 1587 EL DORADO, ARKANSAS 71731

*ADEA Permitto Branch,
Water Division
5301 Northshore Drive
W. Little Rock, AR 72118*