

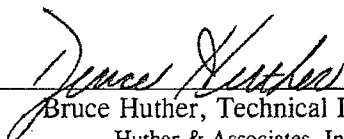
ENVIRONMENTAL ENTERPRISE GROUP
CITY OF CLARKSVILLE WWTP
OUTFALL 001

Chronic Biomonitoring Report
Permit Number NPDES AR0022187
AFIN Number 36-00038

Ceriodaphnia dubia
Pimephales promelas

December 2, 2014

Reviewed by:


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TOXICITY TEST REPORT - CHRONIC

Client Environmental Enterprise Group Sample..... Outfall 001
Facility City of Clarksville WWTP Laboratory I.D. 23040
Permit No. NPDES AR0022187 Begin Date December 2, 2014

Results: Pass *Ceriodaphnia dubia* survival and reproduction and *Pimephales promelas* survival and growth at the critical low flow concentration (100% effluent).

SAMPLE COLLECTION

Composite effluent samples from Environmental Enterprise Group, City of Clarksville WWTP were delivered by United Parcel Service courier to Huthur & Associates on December 2, December 4, and December 6, 2014. Effluent samples were collected and composited from Outfall 001 using an automatic sampler by facility personnel. Two toxicity tests were requested: a seven-day *Ceriodaphnia dubia* survival and reproduction test (EPA Method 1002.0), and a seven-day *Pimephales promelas* larval survival and growth test (EPA Method 1000.0). Test organisms, procedures and quality assurance requirements were in accordance with the EPA manual, "Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, Fourth Edition" (EPA-821-R-02-013).

The effluent and receiving water samples were analyzed for total residual chlorine (Standard Methods, 22nd Edition, 4500-Cl D) and contained <0.01 mg/L, <0.01 mg/L, and <0.01 mg/L, respectively. Effluent and receiving dilution water hardness, alkalinity, conductivity, pH, and dissolved oxygen data were collected and recorded.

TEST SETUP
Ceriodaphnia dubia



The seven-day *Ceriodaphnia dubia* survival and reproduction test was initiated at 1400 hours, December 2, 2014. Five concentrations were prepared (32%, 42%, 56%, 75%, and 100% effluent) utilizing receiving water (Lake Dardanelle) as dilution water. The test was conducted in 25 mL distilled water rinsed plastic beakers containing 15 mL of solution (one neonate per beaker, ten beakers per concentration). *C. dubia* neonates were less than 24-hours old and within eight hours of the same age at test initiation. Neonates were placed in beakers following a randomized block test design. Fresh solutions were prepared and renewed daily. Daily feeding consisted of 0.5 mL *Selenastrum capricornutum* and cerophyll per test chamber. The test proceeded for seven days during which survival, reproduction and water quality data were collected daily.

A true control of ten replicate chambers containing one neonate each in receiving water was conducted concurrently with the test. There was 100% survival in the true control. In addition, a performance control of ten replicate chambers containing one neonate each in synthetic laboratory water was conducted concurrently with the test. The purpose of the performance control was to assess the health of the test organisms and to identify receiving water toxicity. The performance control data was not used in the statistical analysis of the test data. There was 100% survival in the performance control. The test ended at 1400 hours, December 9, 2014. Survival and reproduction data were statistically analyzed ($p = 0.05$) according to EPA procedures to determine the Lowest Observable Effect Concentration (LOEC) and the No Observable Effect Concentration (NOEC).

SURVIVAL
Ceriodaphnia dubia

There was 100% survival to *C. dubia* in all of the effluent concentrations tested. Therefore, statistical analyses were not required to determine a no effect concentration.

LOEC: Not Applicable
NOEC: 100% Effluent

REPRODUCTION
Ceriodaphnia dubia

C. dubia reproduction data were normally distributed at the 0.01 alpha level (13.277) using Chi-Square test for normality. Reproduction data were homogeneous using Bartlett's test at the 0.01 alpha level (15.09) without data transformations. Therefore, a parametric test was performed on the homogeneous data. Dunnett's test on *C. dubia* reproduction data demonstrated that there were no statistically significant differences between the control and any of the effluent concentrations.

LOEC: Not Applicable
NOEC: 100% Effluent

PMSD: 7.5%

TEST SETUP
Pimephales promelas



The seven-day *Pimephales promelas* larval survival and growth test was initiated at 1510 hours, December 2, 2014. Five concentrations were prepared (32%, 42%, 56%, 75%, and 100% effluent) utilizing receiving water (Lake Dardanelle) as dilution water. The test was conducted in 300 mL distilled water rinsed plastic beakers containing 250 mL of solution (eight larvae per beaker, five beakers per concentration). *P. promelas* larvae were less than 24-hours old at test initiation and originated from a minimum of three in-house spawnings. Fresh solutions were prepared and renewed daily. Larvae in each test chamber were fed <24 hour old *Artemia* (brine shrimp) three times per day. The test proceeded for seven days during which survival and water quality data were collected daily.

A true control of five replicate chambers of eight larvae each in receiving water was conducted concurrently with the test. There was 100% survival in the true control. In addition, a performance control of five replicate chambers of eight larvae each in synthetic laboratory water was conducted concurrently with the test. The purpose of the performance control was to assess the health of the test larvae and to identify receiving water toxicity. The performance control data was not used in the statistical analysis of the test data. There was 100% survival in the performance control. At the end of the test, all larvae were sacrificed, dried, and weighed. The test ended at 1510 hours, December 9, 2014. Survival and growth (weight) data were statistically analyzed ($p = 0.05$) according to EPA procedures to determine the Lowest Observable Effect Concentration (LOEC) and the No Observable Effect Concentration (NOEC).

SURVIVAL*Pimephales promelas*

There was 100% survival to *P. promelas* in all of the effluent concentrations tested. Therefore, statistical analyses were not required to determine a no effect concentration.

LOEC: Not Applicable

NOEC: 100% Effluent

GROWTH*Pimephales promelas*

P. promelas growth data were normally distributed at the 0.01 alpha level (0.900) using Shapiro Wilk's test for normality. Growth data were homogeneous using Bartlett's test at the 0.01 alpha level (15.09) without data transformations. Therefore, a parametric test was performed on the homogeneous data. Dunnett's test on *P. promelas* growth data demonstrated that there were no statistically significant differences between the control and any of the effluent concentrations.

LOEC: Not Applicable

NOEC: 100% Effluent

PMSD: 9.9%

SUMMARY

There were no statistically significant differences between the control and the critical low flow concentration (100% effluent) for *C. dubia* survival and reproduction and *P. promelas* survival and growth. Based on biomonitoring requirements for Outfall 001 contained in Permit Number NPDES AR0022187 for Environmental Enterprise Group, City of Clarksville WWTP, Outfall 001 **passed** for this testing period.

Huther and Associates
7-Day/3 Brood *Ceriodaphnia dubia* Survival and Reproduction Chronic Toxicity Test

CLIENT EEG, City of Clarksville WWTP	SAMPLE TYPE 24 Hour Composite
NPDES # AR0022187	DATE COLLECTED 12/01/14 12/03/14 12/05/14
LAB ID # 23040	DATE RECEIVED 12/02/14 12/04/14 12/06/14
TEST TYPE 7 Day Chronic	BEGIN DATE/TIME 12/02/14 1400
TEST ORGANISM <i>Ceriodaphnia dubia</i>	END DATE/TIME 12/09/14 1400
ORGANISM AGE < 24 Hours	TEST TEMPERATURE (°C) 25 ± 1
ORGANISM SOURCE In House	PHOTO PERIOD 16-hr. Light 8-hr. Dark
RECEIVING WATER Lake Dardanelle	LIGHT INTENSITY 50-100 ft. candl.
DILUTION WATER Lake Dardanelle	TECHNICIAN Z. Geiger

SURVIVAL & REPRODUCTION SUMMARY

Performance Control

Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
12/03/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/04/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/05/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/06/14	3	2	2	2	3	4	2	3	4	2
	3	2	2	2	3	4	2	3	4	2
12/07/14	A	A	A	A	A	A	A	A	A	A
	3	2	2	2	3	4	2	3	4	2
12/08/14	6	6	6	7	6	6	7	8	7	6
	9	8	8	9	9	10	9	11	11	8
12/09/14	11	13	12	12	12	13	12	12	13	12
	20	21	20	21	21	23	21	23	24	20
x# Young 21.4 C.V. 6.68%										
x% Survival 100% C.V. 0.00%										

True Control

Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
12/03/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/04/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/05/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/06/14	4	3	3	4	2	4	2	3	4	2
	4	3	3	4	2	4	2	3	4	2
12/07/14	A	A	A	A	A	A	A	A	A	A
	4	3	3	4	2	4	2	3	4	2
12/08/14	8	9	8	7	10	7	6	8	6	9
	12	12	11	11	12	11	8	11	10	11
12/09/14	12	11	12	12	13	14	12	14	13	12
	24	23	23	23	25	25	20	25	23	23
x# Young 23.4 C.V. 6.43%										
x% Survival 100% C.V. 0.00%										

32% Effluent

Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
12/03/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/04/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/05/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/06/14	3	5	3	4	2	4	2	4	3	2
	3	5	3	4	2	4	2	4	3	2
12/07/14	A	A	A	A	A	A	A	A	A	A
	3	5	3	4	2	4	2	4	3	2
12/08/14	8	10	9	6	7	10	11	9	8	8
	11	15	12	10	9	14	13	13	11	10
12/09/14	12	13	13	12	14	13	12	11	12	11
	23	28	25	22	23	27	25	24	23	21
x# Young 24.1 C.V. 9.06%										
x% Survival 100% C.V. 0.00%										

42% Effluent

Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
12/03/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/04/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/05/14	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
12/06/14	2	2	3	4	2	4	3	2	4	3
	2	2	3	4	2	4	3	2	4	3
12/07/14	A	A	A	A	A	A	A	A	A	7
	2	2	3	4	2	4	3	2	4	10
12/08/14	9	10	8	6	9	6	10	7	7	A
	11	12	11	10	11	10	13	9	11	10
12/09/14	13	14	12	13	13	12	12	11	12	13
	24	26	23	23	24	22	25	20	23	23
x# Young 23.3 C.V. 7.02%										
x% Survival 100% C.V. 0.00%										

where: A = Alive
S = Alive, 5 young
D = Dead
D5 = 5 Young, Female died

ex 1:

A
4

 alive today
total young to date

ex 2:

5
12

 alive, 5 young today
total young to date

Huther and Associates
7-Day/3 Brood *Ceriodaphnia dubia* Survival and Reproduction Chronic Toxicity Test

EEG, City of Clarksville WWTP

Lab ID# 23040

Test Date: December 2, 2014

56% Effluent

Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
12/03/14	A	A	A	A	A	A	A	A	A	A
12/03/14	0	0	0	0	0	0	0	0	0	0
12/04/14	A	A	A	A	A	A	A	A	A	A
12/04/14	0	0	0	0	0	0	0	0	0	0
12/05/14	A	A	A	A	A	A	A	A	A	A
12/05/14	0	0	0	0	0	0	0	0	0	0
12/06/14	2	4	3	2	3	4	2	2	2	4
12/06/14	2	4	3	2	3	4	2	2	2	4
12/07/14	A	A	A	A	A	A	A	A	A	A
12/07/14	2	4	3	2	3	4	2	2	2	4
12/08/14	10	7	6	8	9	6	7	10	9	9
12/08/14	12	11	9	10	12	10	9	12	11	13
12/09/14	13	13	12	14	12	12	14	13	12	11
12/09/14	25	24	21	24	24	22	23	25	23	24
x# Young		23.5				C.V.		5.40%		
x% Survival		100%				C.V.		0.00%		

75% Effluent

Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
12/03/14	A	A	A	A	A	A	A	A	A	A
12/03/14	0	0	0	0	0	0	0	0	0	0
12/04/14	A	A	A	A	A	A	A	A	A	A
12/04/14	0	0	0	0	0	0	0	0	0	0
12/05/14	A	A	A	A	A	A	A	A	A	A
12/05/14	0	0	0	0	0	0	0	0	0	0
12/06/14	2	4	4	3	2	3	2	2	3	2
12/06/14	2	4	4	3	2	3	2	2	3	2
12/07/14	A	A	A	A	A	A	A	A	A	A
12/07/14	2	4	4	3	2	3	2	2	3	2
12/08/14	10	10	6	7	9	6	7	6	10	9
12/08/14	12	14	10	10	11	9	9	8	13	11
12/09/14	13	12	11	13	13	11	13	14	13	12
12/09/14	25	26	21	23	24	20	22	22	26	23
x# Young		23.2				C.V.		8.81%		
x% Survival		100%				C.V.		0.00%		

100% Effluent

Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
12/03/14	A	A	A	A	A	A	A	A	A	A
12/03/14	0	0	0	0	0	0	0	0	0	0
12/04/14	A	A	A	A	A	A	A	A	A	A
12/04/14	0	0	0	0	0	0	0	0	0	0
12/05/14	A	A	A	A	A	A	A	A	A	A
12/05/14	0	0	0	0	0	0	0	0	0	0
12/06/14	2	4	3	3	2	2	2	3	4	2
12/06/14	2	4	3	3	2	2	2	3	4	2
12/07/14	A	A	A	A	A	A	A	A	A	A
12/07/14	2	4	3	3	2	2	2	3	4	2
12/08/14	10	8	9	6	10	8	7	10	9	10
12/08/14	12	12	12	9	12	10	9	13	13	12
12/09/14	12	12	13	12	12	12	13	12	11	12
12/09/14	24	24	25	21	24	22	22	25	24	24
x# Young		23.5				C.V.		5.76%		
x% Survival		100%				C.V.		0.00%		

where: A = Alive
5 = Alive, 5 young
D = Dead
D5 = 5 Young, Female died

ex 1:

A
4

 alive today
total young to date

ex 2:

5
12

 alive, 5 young today
total young to date

Huther and Associates
7-Day/3 Brood *Ceriodaphnia dubia* Survival and Reproduction Chronic Toxicity Test

EEG, City of Clarksville WWTP

Lab ID# 23040

Test Date: December 2, 2014

WET CHEMISTRY MEASUREMENTS

Date	Time	Temp	Samp. No.	pH of Solution						Analyst	
				PCON	TCON	32%	42%	56%	75%		100%
12/02/14	Start	25.0	1	8.76	7.40	7.48	7.52	7.53	7.55	7.61	CS
12/03/14	24 Hr.	23.2	1	7.98	7.75	7.62	7.58	7.49	7.43	7.33	TN
12/03/14	Renew	23.2	1	8.75	7.58	7.46	7.31	7.25	7.13	7.00	TN
12/04/14	48 Hr.	24.4	1	8.15	7.75	7.57	7.52	7.48	7.45	7.36	CS
12/04/14	Renew	24.4	2	8.75	7.39	7.36	7.34	7.33	7.24	7.61	CS
12/05/14	72 Hr.	24.1	2	8.21	7.76	7.63	7.63	7.56	7.53	7.55	EMS
12/05/14	Renew	24.1	2	7.74	7.72	7.69	7.64	7.60	7.51	7.49	EMS
12/06/14	96 Hr.	24.1	2	8.11	7.75	7.71	7.64	7.60	7.58	7.55	CS
12/06/14	Renew	24.8	3	7.59	7.34	7.50	7.48	7.46	7.45	7.62	CS
12/07/14	120 Hr.	24.8	3	8.02	7.96	8.00	7.89	7.85	7.80	7.73	EMS
12/07/14	Renew	24.8	3	8.78	8.70	7.58	7.55	7.82	7.89	7.96	EMS
12/08/14	144 Hr.	24.5	3	8.09	8.24	8.19	8.12	8.04	7.92	7.73	EMS
12/08/14	Renew	24.5	3	8.34	7.98	7.95	7.89	7.78	7.68	7.65	EMS
12/09/14	168 Hr.	24.4	3	8.03	7.99	8.04	7.88	7.86	7.81	7.73	CS

Date	Time	Temp	Samp. No.	DO (mg/L) of Solution						Analyst	
				PCON	TCON	32%	42%	56%	75%		100%
12/02/14	Start	25.0	1	7.57	8.88	8.47	8.23	8.94	8.31	8.61	CS
12/03/14	24 Hr.	23.2	1	7.97	8.27	7.91	7.17	7.29	8.07	7.66	TN
12/03/14	Renew	23.2	1	8.64	7.38	7.85	8.75	8.18	8.08	8.05	TN
12/04/14	48 Hr.	24.4	1	7.66	8.15	8.10	7.65	7.81	7.47	8.32	CS
12/04/14	Renew	24.4	2	8.64	8.81	8.46	8.53	8.17	8.23	8.53	CS
12/05/14	72 Hr.	24.1	2	7.66	7.74	7.56	7.68	7.65	7.51	8.32	EMS
12/05/14	Renew	24.1	2	8.07	7.88	7.99	7.78	8.16	7.14	7.53	EMS
12/06/14	96 Hr.	24.1	2	8.42	7.89	7.65	7.48	7.57	7.83	7.18	CS
12/06/14	Renew	24.8	3	8.64	8.87	8.59	8.62	8.47	8.12	8.48	CS
12/07/14	120 Hr.	24.8	3	7.72	7.75	7.95	8.07	8.30	7.51	7.52	EMS
12/07/14	Renew	24.8	3	8.64	7.69	8.83	8.94	8.68	8.53	8.07	EMS
12/08/14	144 Hr.	24.5	3	7.56	7.83	8.40	8.17	8.38	8.34	7.50	EMS
12/08/14	Renew	24.5	3	8.80	8.21	7.74	7.69	8.10	7.83	8.17	EMS
12/09/14	168 Hr.	24.4	3	8.94	8.95	8.96	8.98	8.99	8.97	8.91	CS

Huther and Associates
7-Day/3 Brood *Ceriodaphnia dubia* Survival and Reproduction Chronic Toxicity Test

EEG, City of Clarksville WWTP

Lab ID# 23040

Test Date: December 2, 2014

INITIAL CHEMISTRY MEASUREMENTS @ 100% EFFLUENT

Date	Samp. No.	pH	DO	Hardness mg/L CaCO ₃ ¹	Alkalinity mg/L CaCO ₃ ¹	Conduct. umhos/cm ¹	Resid. Cl ₂ mg/L ¹	Dechlor(mL) Na ₂ S ₂ O ₃ mg/L ¹	Analyst
12/02/14	1	7.61	8.61	80	18	410	<0.01	N/A	TN
12/04/14	2	7.61	8.53	84	20	441	<0.01	N/A	TN
12/06/14	3	7.62	8.48	88	22	444	<0.01	N/A	TN

¹ Measurements taken in 100% solution.

INITIAL CHEMISTRY MEASUREMENTS @ RECEIVING WATER

Date	Samp. No.	Ph	DO	Hardness mg/L CaCO ₃ ¹	Alkalinity mg/L CaCO ₃ ¹	Conduct. umhos/cm ¹	Resid. Cl ₂ mg/L ¹	Dechlor(mL) Na ₂ S ₂ O ₃ mg/L ¹	Analyst
12/02/14	RS1	7.40	8.88	108	58	520	<0.01	N/A	TN
12/04/14	RS2	7.39	8.81	112	56	555	<0.01	N/A	TN
12/06/14	RS3	7.34	8.87	116	58	550	<0.01	N/A	TN

Huther and Associates, Inc.
 Begin Date: December 02, 2014
 Lab I.D.# 23040

CERIODAPHNIA DUBIA STATISTICAL ANALYSES
 Reproduction

Summary Statistics on Transformed Data Table 1 of 2

Grp	Identification	N	Min	Max	Mean
1	Control	10	20.000	25.000	23.400
2	32% Effluent	10	21.000	28.000	24.100
3	42% Effluent	10	20.000	26.000	23.300
4	56% Effluent	10	21.000	25.000	23.500
5	75% Effluent	10	20.000	26.000	23.200
6	100% Effluent	10	21.000	25.000	23.500

Summary Statistics on Transformed Data Table 2 of 2

Grp	Identification	Variance	Sd	Sem	C.V.%
1	Control	2.267	1.506	0.476	6.43
2	32% Effluent	4.767	2.183	0.690	9.06
3	42% Effluent	2.678	1.636	0.517	7.02
4	56% Effluent	1.611	1.269	0.401	5.40
5	75% Effluent	4.178	2.044	0.646	8.81
6	100% Effluent	1.833	1.354	0.428	5.76

Chi-Square Test For Normality: Actual And Expected Frequencies

Interval	< -1.5	-1.5 to -0.5	-0.5 to 0.5	> 0.5 to 1.5	> 1.5
Expected	4.020	14.520	22.920	14.520	4.020
Observed	5	12	29	12	2

Calculated Chi-Square goodness of fit test statistic = 3.7415
 Table Chi-Square value (alpha = 0.01) = 13.277

Data Pass normality test. Continue analysis.

Bartlett's Test For Homogeneity of Variance

Calculated B1 statistic = 4.21

Table Chi-square value = 15.09 (alpha = 0.01, DF = 5)
 Table Chi-square value = 11.07 (alpha = 0.05, DF = 5)

Data Pass B1 homogeneity test at 0.01 level. Continue analysis.

ANOVA Table

SOURCE	DF	SS	MS	F
Between	5	5.000	1.000	0.346
Within (Error)	54	156.000	2.889	
Total	59	161.000		

Critical F value = 2.45 (0.05,5,40)
 Since F < Critical F Fail to Reject Ho: All equal

Dunnett's Test - Table 1 of 2 Ho:Control < Treatment

Grp	Identification	Mean		T Stat	Sig
		Transformed Mean	Calculated In Original Units		
1	Control	23.400	23.400		
2	32% Effluent	24.100	24.100	-0.921	
3	42% Effluent	23.300	23.300	0.132	
4	56% Effluent	23.500	23.500	-0.132	
5	75% Effluent	23.200	23.200	0.263	
6	100% Effluent	23.500	23.500	-0.132	

Dunnett table value = 2.31 (1 Tailed Value, P=0.05, DF=40,5)
 No statistically significant difference

Dunnett's Test - Table 1 of 2 Ho:Control < Treatment

Grp	Identification	Num of Reps	Minimum Sig Diff (In Orig. Units)	Difference	
				% of Control	from Control
1	Control	10			
2	32% Effluent	10	1.756	7.5	-0.700
3	42% Effluent	10	1.756	7.5	0.100
4	56% Effluent	10	1.756	7.5	-0.100
5	75% Effluent	10	1.756	7.5	0.200
6	100% Effluent	10	1.756	7.5	-0.100

Huther and Associates
7-Day *Pimephales promelas* Survival and Growth Chronic Toxicity Test

CLIENT	EEG, City of Clarksville WWTP	SAMPLE TYPE	24 Hour Composite
NPDES #	AR0022187	DATE COLLECTED	12/01/14 12/03/14 12/05/14
LAB ID #	23040	DATE RECEIVED	12/02/14 12/04/14 12/06/14
TEST TYPE	7 Day Chronic	BEGIN DATE/TIME	12/02/14 1510
TEST ORGANISM	<i>Pimephales promelas</i>	END DATE/TIME	12/09/14 1510
ORGANISM AGE	< 24 Hours	TEST TEMPERATURE (°C)	25 ± 1
ORGANISM SOURCE	In House	PHOTO PERIOD	16-hr. Light 8-hr. Dark
RECEIVING WATER	Lake Dardanelle	LIGHT INTENSITY	50-100 ft. cndl.
DILUTION WATER	Lake Dardanelle	TECHNICIAN	M. Horner

SURVIVAL SUMMARY

Conc.	12/03/14					12/04/14					12/05/14					12/06/14					12/07/14				
	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E
Pcon	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
Tcon	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
32%	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
42%	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
56%	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
75%	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
100%	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8

Conc.	12/08/14					12/09/14					x % Survival	C.V. %
	A	B	C	D	E	A	B	C	D	E		
Pcon	8	8	8	8	8	8	8	8	8	8	100.0	0.00
Tcon	8	8	8	8	8	8	8	8	8	8	100.0	0.00
32%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
42%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
56%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
75%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
100%	8	8	8	8	8	8	8	8	8	8	100.0	0.00

MEAN DRY WEIGHT PER REP

% Effluent	Rep A	Rep B	Rep C	Rep D	Rep E	x	C.V. %
Pcon	0.4590	0.4860	0.5020	0.4470	0.4950	0.4778	4.96
Tcon	0.4260	0.4760	0.4930	0.4450	0.4610	0.4602	5.67
32%	0.4820	0.4930	0.4200	0.4750	0.5010	0.4742	6.73
42%	0.4450	0.4720	0.4950	0.4260	0.5040	0.4684	7.03
56%	0.5040	0.4560	0.4810	0.4960	0.4270	0.4728	6.65
75%	0.4650	0.5040	0.4440	0.4860	0.4950	0.4788	5.06
100%	0.5040	0.4210	0.4670	0.4950	0.5050	0.4784	7.44

Huthier and Associates
7-Day *Pimephales promelas* Survival and Growth Chronic Toxicity Test

EEG, City of Clarksville WWTP

Lab ID# 23040

Test Date: December 2, 2014

WET CHEMISTRY MEASUREMENTS

Date	Time	Temp	Samp. No.	pH of Solution						Analyst	
				PCON	TCON	32%	42%	56%	75%		100%
12/02/14	Start	25.0	1	8.76	7.40	7.48	7.52	7.53	7.55	7.61	CS
12/03/14	24 Hr.	23.3	1	8.15	7.82	7.68	7.61	7.52	7.41	7.14	TG
12/03/14	Renew	23.3	1	8.75	7.58	7.46	7.31	7.25	7.13	7.00	TG
12/04/14	48 Hr.	24.6	1	8.13	7.76	7.65	7.53	7.50	7.43	7.08	CS
12/04/14	Renew	24.4	2	8.75	7.39	7.36	7.34	7.33	7.24	7.61	CS
12/05/14	72 Hr.	24.6	2	7.58	7.59	7.61	7.54	7.41	7.40	7.39	EMS
12/05/14	Renew	24.6	2	7.74	7.72	7.69	7.64	7.60	7.51	7.49	EMS
12/06/14	96 Hr.	24.1	2	7.59	7.60	7.64	7.50	7.48	7.45	7.44	CS
12/06/14	Renew	24.8	3	7.59	7.34	7.50	7.48	7.46	7.45	7.62	CS
12/07/14	120 Hr.	24.4	3	7.85	7.55	7.50	7.49	7.46	7.45	7.49	EMS
12/07/14	Renew	24.4	3	8.78	8.70	7.58	7.55	7.82	7.89	7.96	EMS
12/08/14	144 Hr.	24.3	3	7.92	7.68	7.50	7.49	7.45	7.43	7.48	EMS
12/08/14	Renew	24.3	3	8.34	7.98	7.95	7.89	7.78	7.68	7.65	EMS
12/09/14	168 Hr.	24.2	3	8.10	7.53	7.51	7.50	7.47	7.46	7.42	CS

Date	Time	Temp	Samp. No.	DO (mg/L) of Solution						Analyst	
				PCON	TCON	32%	42%	56%	75%		100%
12/02/14	Start	25.0	1	7.57	8.88	8.47	8.23	8.94	8.31	8.61	CS
12/03/14	24 Hr.	23.3	1	8.01	7.05	7.01	7.01	7.59	8.22	8.32	TG
12/03/14	Renew	23.3	1	8.64	7.38	7.85	8.75	8.18	8.08	8.05	TG
12/04/14	48 Hr.	24.6	1	8.49	8.88	8.52	8.72	8.16	8.33	8.66	CS
12/04/14	Renew	24.4	2	8.64	8.81	8.46	8.53	8.17	8.23	8.53	CS
12/05/14	72 Hr.	24.6	2	7.92	7.71	7.91	8.18	7.92	7.98	8.03	EMS
12/05/14	Renew	24.6	2	8.07	7.88	7.99	7.78	8.16	7.14	7.53	EMS
12/06/14	96 Hr.	24.1	2	8.68	8.48	8.57	8.73	8.88	8.17	8.42	CS
12/06/14	Renew	24.8	3	8.64	8.87	8.59	8.62	8.47	8.12	8.48	CS
12/07/14	120 Hr.	24.4	3	8.00	7.45	8.87	8.69	7.89	7.97	7.88	EMS
12/07/14	Renew	24.4	3	8.64	7.69	8.83	8.94	8.68	8.53	8.07	EMS
12/08/14	144 Hr.	24.3	3	8.43	8.32	8.15	8.39	8.58	7.94	7.96	EMS
12/08/14	Renew	24.3	3	8.80	8.21	7.74	7.69	8.10	7.83	8.17	EMS
12/09/14	168 Hr.	24.2	3	8.29	8.47	8.32	8.69	8.10	7.84	8.22	CS

Huther and Associates
7-Day *Pimephales promelas* Survival and Growth Chronic Toxicity Test

EEG, City of Clarksville WWTP

Lab ID# 23040

Test Date: December 2, 2014

INITIAL CHEMISTRY MEASUREMENTS @ 100% EFFLUENT

Date	Samp. No.	pH	DO	Hardness mg/L CaCO ₃ ¹	Alkalinity mg/L CaCO ₃ ¹	Conduct. umhos/cm ¹	Resid. Cl ₂ mg/L ¹	Dechlor(mL) Na ₂ S ₂ O ₃ mg/L ¹	Analyst
12/02/14	1	7.61	8.61	80	18	410	<0.01	N/A	TN
12/04/14	2	7.61	8.53	84	20	441	<0.01	N/A	TN
12/06/14	3	7.62	8.48	88	22	444	<0.01	N/A	TN

INITIAL CHEMISTRY MEASUREMENTS @ RECEIVING WATER

Date	Samp. No.	Ph	DO	Hardness mg/L CaCO ₃ ¹	Alkalinity mg/L CaCO ₃ ¹	Conduct. umhos/cm ¹	Resid. Cl ₂ mg/L ¹	Dechlor(mL) Na ₂ S ₂ O ₃ mg/L ¹	Analyst
12/02/14	RS1	7.40	8.88	108	58	520	<0.01	N/A	TN
12/04/14	RS2	7.39	8.81	112	56	555	<0.01	N/A	TN
12/06/14	RS3	7.34	8.87	116	58	550	<0.01	N/A	TN

¹ Measurements taken in 100% solution.

Huther and Associates, Inc.
 Begin Date: December 02, 2014
 Lab I.D.# 23040

PIMEPHALES PROMELAS STATISTICAL ANALYSES
 Growth

Summary Statistics on Transformed Data Table 1 of 2

Grp	Identification	N	Min	Max	Mean
1	Control	5	0.426	0.493	0.460
2	32% Effluent	5	0.420	0.501	0.474
3	42% Effluent	5	0.426	0.504	0.468
4	56% Effluent	5	0.427	0.504	0.473
5	75% Effluent	5	0.444	0.504	0.479
6	100% Effluent	5	0.421	0.505	0.478

Summary Statistics on Transformed Data Table 2 of 2

Grp	Identification	Variance	Sd	Sem	C.V.%
1	Control	0.001	0.026	0.012	5.67
2	32% Effluent	0.001	0.032	0.014	6.73
3	42% Effluent	0.001	0.033	0.015	7.03
4	56% Effluent	0.001	0.031	0.014	6.65
5	75% Effluent	0.001	0.024	0.011	5.06
6	100% Effluent	0.001	0.036	0.016	7.44

Shapiro - Wilk's Test For Normality

D = 0.023

W = 0.907

Critical W (P = 0.05) (n = 30) = 0.927

Critical W (P = 0.01) (n = 30) = 0.900

Data Pass normality test at P=0.01 level. Continue analysis.

Bartlett's Test For Homogeneity of Variance

Calculated B1 statistic = 0.75

Table Chi-square value = 15.09 (alpha = 0.01, DF = 5)

Table Chi-square value = 11.07 (alpha = 0.05, DF = 5)

Data Pass B1 homogeneity test at 0.01 level. Continue analysis.

ANOVA Table

SOURCE	DF	SS	MS	F
Between	5	0.001	0.000	0.261
Within (Error)	24	0.023	0.001	
Total	29	0.024		

Critical F value = 2.62 (0.05,5,24)

Since F < Critical F Fail to Reject Ho: All equal

Dunnett's Test - Table 1 of 2 Ho:Control < Treatment

Grp	Identification	Transformed	Mean	T Stat	Sig
		Mean	Calculated In Original Units		
1	Control	0.460	0.460		
2	32% Effluent	0.474	0.474	-0.723	
3	42% Effluent	0.468	0.468	-0.423	
4	56% Effluent	0.473	0.473	-0.651	
5	75% Effluent	0.479	0.479	-0.960	
6	100% Effluent	0.478	0.478	-0.940	

Dunnett table value = 2.36 (1 Tailed Value, P=0.05, DF=24,5)

No statistically significant difference

Dunnett's Test - Table 1 of 2 Ho:Control < Treatment

Grp	Identification	Num of Reps	Minimum Sig Diff (In Orig. Units)	% of Control	Difference
					from Control
1	Control	5			
2	32% Effluent	5	0.046	9.9	-0.014
3	42% Effluent	5	0.046	9.9	-0.008
4	56% Effluent	5	0.046	9.9	-0.013
5	75% Effluent	5	0.046	9.9	-0.019
6	100% Effluent	5	0.046	9.9	-0.018

**APPENDIX A
RAW DATA**

7-DAY CERIODAPHNIA DUBIA SURVIVAL & REPRODUCTION
 DAILY RAW DATA TABLE
 PAGE 1 OF 2

CLIENT EEG Clarksville
 OUTFALL 001
 LAB ID # 23040

START DATE/TIME 12-2-14 ZG 1400
 END DATE/TIME 12-9-14 NL 1400

Pcon

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
12/3	A	A	A	A	A	A	A	A	A	A	ZG	1400
12/4	A	A	A	A	A	A	A	A	A	A	ZG	1450
12/5	A	A	A	A	A	A	A	A	A	A	TG	1105
12/6	3	2	2	2	3	4	2	3	4	2	NL	1445
12/7	A	A	A	A	A	A	A	A	A	A	NL	1000
12/8	6	6	6	7	6	6	7	8	7	6	ZG	1200
12/9	11	13	12	12	12	13	12	12	13	12	NL	1400

\bar{x} # Young w/o Dead = 21.4 CV% = 6.68

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = 100.0 CV% = 0.00

32

Tcon

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
12/3	A	A	A	A	A	A	A	A	A	A	ZG	1400
12/4	A	A	A	A	A	A	A	A	A	A	ZG	1450
12/5	A	A	A	A	A	A	A	A	A	A	TG	1105
12/6	4	3	3	4	2	4	2	3	4	2	NL	1445
12/7	A	A	A	A	A	A	A	A	A	A	NL	1000
12/8	8	9	8	7	10	7	6	8	6	9	ZG	1200
12/9	12	11	12	12	13	14	12	14	13	12	NL	1400

\bar{x} # Young w/o Dead = 23.4 CV% = 6.43

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = 100.0 CV% = 0.00

42

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
12/3	A	A	A	A	A	A	A	A	A	A	ZG	1400
12/4	A	A	A	A	A	A	A	A	A	A	ZG	1450
12/5	A	A	A	A	A	A	A	A	A	A	TG	1105
12/6	3	5	3	4	2	4	2	4	3	2	NL	1445
12/7	A	A	A	A	A	A	A	A	A	A	NL	1000
12/8	8	10	9	6	7	10	11	9	8	8	ZG	1200
12/9	12	13	13	12	14	13	12	11	12	11	NL	1400

\bar{x} # Young w/o Dead = 24.1 CV% = 9.06

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = 100.0 CV% = 0.00

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
12/3	A	A	A	A	A	A	A	A	A	A	ZG	1400
12/4	A	A	A	A	A	A	A	A	A	A	ZG	1450
12/5	A	A	A	A	A	A	A	A	A	A	TG	1105
12/6	2	2	3	4	2	4	3	2	4	3	NL	1445
12/7	A	A	A	A	A	A	A	A	A	7	NL	1000
12/8	9	10	8	6	9	6	10	7	7	8	ZG	1200
12/9	13	14	12	13	13	12	12	11	12	13	NL	1400

\bar{x} # Young w/o Dead = 23.3 CV% = 7.02

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = 100.0 CV% = 0.00

7-DAY CERIODAPHNIA DUBIA SURVIVAL & REPRODUCTION
DAILY RAW DATA TABLE
PAGE 2 OF 2

CLIENT EEG Clarksville
OUTFALL 001
LAB ID # 23040

START DATE/TIME 12-2-14 2G 1400
END DATE/TIME 12-9-14 NL 1400

56

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
12/3	A	A	A	A	A	A	A	A	A	A	ZG	1400
12/4	A	A	A	A	A	A	A	A	A	A	ZG	1450
12/5	A	A	A	A	A	A	A	A	A	A	TG	1105
12/6	2	4	3	2	3	4	2	2	2	4	NL	1445
12/7	A	A	A	A	A	A	A	A	A	A	NL	1000
12/8	10	7	6	8	9	6	7	10	9	9	ZG	1200
12/9	13	13	12	14	12	14	13	12	11		NL	1400
	25	24	21	24	24	22	23	25	23	24		

\bar{x} # Young w/o Dead = 23.5 CV% = 5.40

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = 100.0 CV% = 0.00

75

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
12/3	A	A	A	A	A	A	A	A	A	A	ZG	1400
12/4	A	A	A	A	A	A	A	A	A	A	ZG	1450
12/5	A	A	A	A	A	A	A	A	A	A	TG	1105
12/6	2	4	4	3	2	3	2	2	3	2	NL	1445
12/7	A	A	A	A	A	A	A	A	A	A	NL	1000
12/8	10	10	6	7	9	6	7	6	10	9	ZG	1200
12/9	13	12	11	13	13	11	13	14	13	12	NL	1400
	25	26	21	23	24	20	22	22	26	23		

\bar{x} # Young w/o Dead = 23.2 CV% = 8.81

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = 100.0 CV% = 0.00

100

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
12/3	A	A	A	A	A	A	A	A	A	A	ZG	1400
12/4	A	A	A	A	A	A	A	A	A	A	ZG	1450
12/5	A	A	A	A	A	A	A	A	A	A	TG	1105
12/6	2	4	3	3	2	2	2	3	4	2	NL	1445
12/7	A	A	A	A	A	A	A	A	A	A	NL	1000
12/8	10	8	9	6	10	8	7	10	9	10	ZG	1200
12/9	12	12	13	12	12	12	13	12	11	12	NL	1400
	24	24	25	21	24	22	22	25	24	24		

\bar{x} # Young w/o Dead = 23.5 CV% = 5.76

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = 100.0 CV% = 0.00

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time

\bar{x} # Young w/o Dead = CV% =

\bar{x} # Young w/Dead = CV% =

\bar{x} % Survival = CV% =

**7-DAY CHRONIC TOXICITY TEST
PIMEPHALES PROMELAS (fathead minnow) SURVIVAL**

CLIENT/FACILITY

EEG Clarksville

DATE/TIME STARTED

12-2-14 MH 1510

OUTFALL #

001

PROJECT #

23040

DATE/TIME ENDED

12-9-14 MH 1510

ORGANISM ID#

PP0-14-335

Conc.	A					B					C					D					E				
	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E
Pcon	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
Tcon	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
32	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
42	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
56	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
75	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
100	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
Initials Date/Time	12-3-14 MH 1510					12-4-14 MH 0855					12-5-14 TG 0820					12-6-14 MH 0820					12-7-14 MH 0900				

Conc.	A					B					Mean Survival	C.V. %
	A	B	C	D	E	A	B	C	D	E		
Pcon	8	8	8	8	8	8	8	8	8	8	100.0	0.00
Tcon	8	8	8	8	8	8	8	8	8	8	100.0	0.00
32	8	8	8	8	8	8	8	8	8	8	100.0	0.00
42	8	8	8	8	8	8	8	8	8	8	100.0	0.00
56	8	8	8	8	8	8	8	8	8	8	100.0	0.00
75	8	8	8	8	8	8	8	8	8	8	100.0	0.00
100	8	8	8	8	8	8	8	8	8	8	100.0	0.00
Initials Date/Time	12-8-14 TN 0820					12-9-14 MH 1510						

Huther and Associates, Inc.

environmental toxicologists, biologists, and consultants

Client / Facility EEG Marksville
 Lab ID Number 23040
 Outfall Number 001
 Test Date 12-2-14

INITIAL CHEMISTRY MEASUREMENTS @ 100% EFFLUENT

Date	Samp. No.	pH	DO	Hardness mg/L CaCO ₃ ¹	Alkalinity mg/L CaCO ₃ ¹	Conduct. umhos/cm ¹	Resid. Cl ₂ mg/L ¹	Dechlor(mL) Na ₂ S ₂ O ₃ mg/L ¹	Analyst
12/2	1	7.61	8.61	80	18	410	20.01	Na	TG
12/4	2	7.61	8.53	84	20	441	5	5	5
12/6	3	7.62	8.48	88	22	444	5	5	5

INITIAL CHEMISTRY MEASUREMENTS @ RECEIVING WATER

Date	Samp. No.	pH	DO	Hardness mg/L CaCO ₃ ¹	Alkalinity mg/L CaCO ₃ ¹	Conduct. umhos/cm ¹	Resid. Cl ₂ mg/L ¹	Dechlor(mL) Na ₂ S ₂ O ₃ mg/L ¹	Analyst
12/2	RS1	7.40	8.88	108	58	520	20.01	Na	TG
12/4	RS2	7.39	8.81	112	56	555	5	5	5
12/6	RS3	7.34	8.87	116	58	550	5	5	5

Notes:

**APPENDIX B
REFERENCE TOXICANTS**

CHRONIC REFERENCE TOXICANT TEST RESULTS

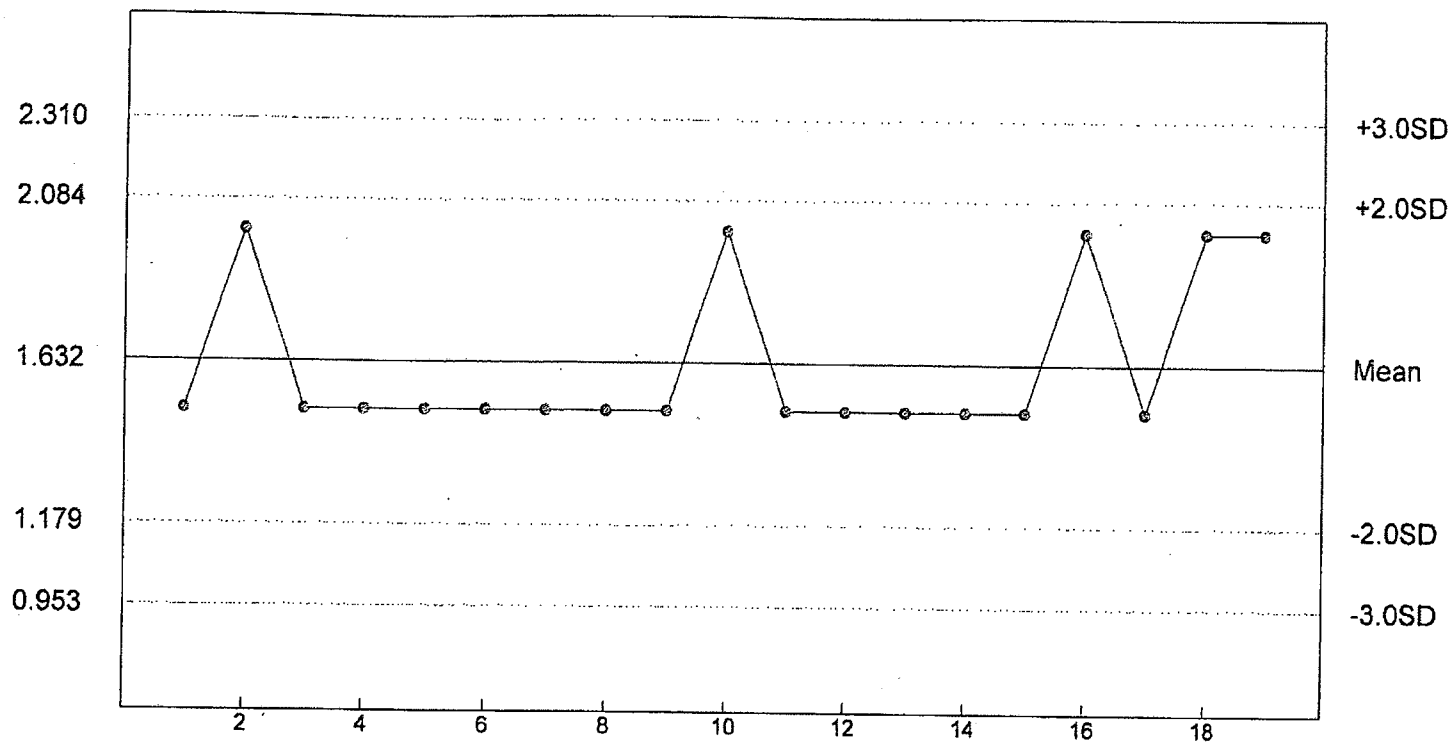
SPECIES: *Ceriodaphnia dubia*
 CHEMICAL: Sodium Chloride
 DURATION: 7-Days
 TEST NUMBER: 12
 TEST DATE/TIME: 12/01/14 - 12/08/14
 1100 Hrs - 1100 Hrs
 STATISTICAL METHOD: Fishers, Dunnetts/Steels

CONCENTRATION (g/L)	NUMBER EXPOSED	NUMBER DEAD
0.5	10	0
1.0	10	0
1.5	10	0
2.0	10	1
2.5	10	10
3.0	10	10
4.0	10	10

LOEC FOR SURVIVAL	NOEC FOR SURVIVAL	LOEC FOR REPRODUCTION	NOEC FOR REPRODUCTION
2.5 g/L	2.0 g/L	1.5 g/L	1.0 g/L

Reference Tox Sodium Chloride g/L

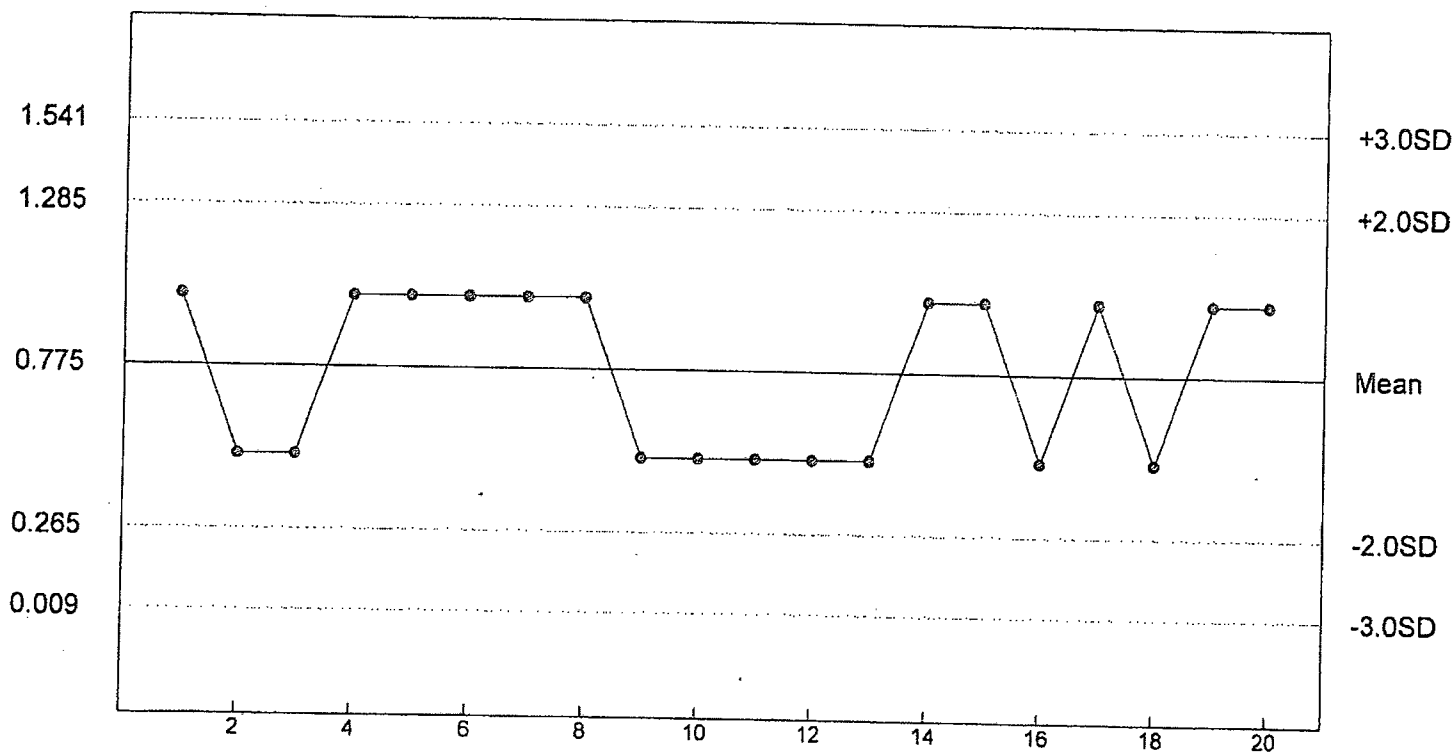
C. dubia Survival - NOEC



n= 19 Mean= 1.632 SD= 0.226 CV= 13.86% Min= 1.500 Max= 2.000

Reference Tox Sodium Chloride g/L

C. dubia Reproduction - NOEC



n= 20 Mean= 0.775 SD= 0.255 CV= 32.93% Min= 0.500 Max= 1.000

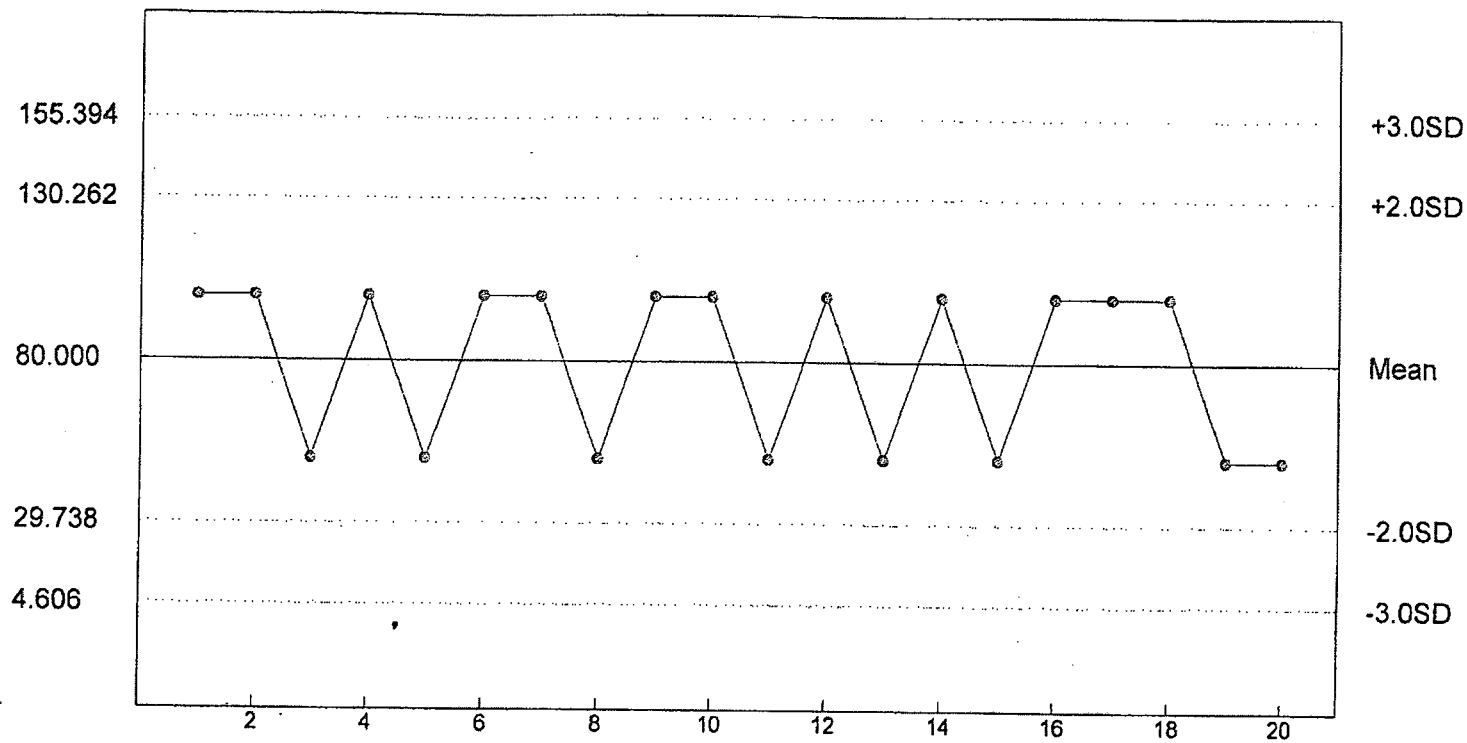
CHRONIC REFERENCE TOXICANT TEST RESULTS

SPECIES: *Pimephales promelas*
 CHEMICAL: Copper Nitrate
 DURATION: 7-Days
 TEST NUMBER: 12
 TEST DATE/TIME: 12/01/14 - 12/08/14
 1300 Hrs - 1300 Hrs
 STATISTICAL METHOD: Dunnetts/Steels

CONCENTRATION (ug/L)	NUMBER EXPOSED	NUMBER DEAD
12.5	40	0
25	40	0
50	40	0
100	40	17
200	40	26
400	40	40
800	40	40

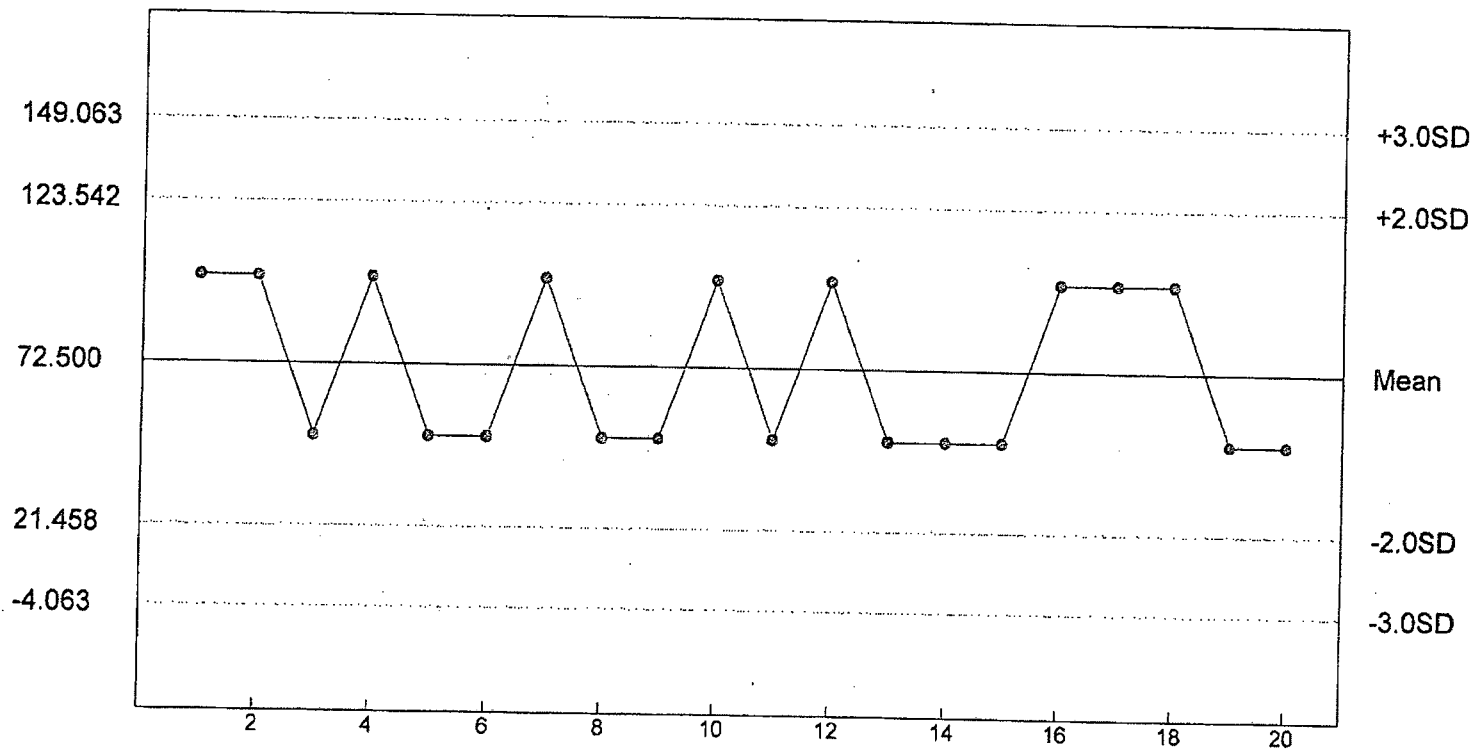
LOEC FOR SURVIVAL	NOEC FOR SURVIVAL	LOEC FOR GROWTH	NOEC FOR GROWTH
100 ug/L	50 ug/L	100 ug/L	50 ug/L

Reference Tox Copper Nitrate ug/L
P. promelas Chronic Survival - NOEC



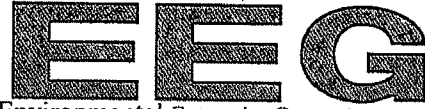
n= 20 Mean= 80.000 SD= 25.131 CV= 31.41% Min= 50.000 Max= 100.000

Reference Tox Copper Nitrate ug/L
P. promelas Growth - NOEC



n= 20 Mean= 72.500 SD= 25.521 CV= 35.20% Min= 50.000 Max= 100.000

**APPENDIX C
CHAIN OF CUSTODY SHEETS**

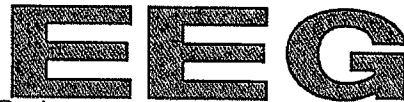


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L444-049203

Environmental Enterprise Group, Inc.
220 North Knoxville, Suite 200
Russellville, Arkansas 72801
(479) 968-6767 Fax (479) 968-1956

Company Name:										Phone #:										Requested Analysis												Laboratory Control Number	Remarks (Please note special detection limits below.)	
Clarksville Light and Water										(479) 754-7929										7-Day Chronic Bio-Monitoring														
Address:										Fax #:																								
P.O. Box 1807 Clarksville, AR 72830										(479) 754-8181																								
Project Name or Number:										Purchase Order #:																								
Bio-Monitoring										Bio-Monitoring																								
Sampling Personnel Signature(s): <i>Alan Bratton</i> <i>Porshe Russell</i>										Printed: Alan Bratton Porshe Russell																								
Sample I.D.	Date	Time	Comp.	Grab	Cont. Type		# of Containers	Method Preserved							Sample Matrix					7-Day Chronic Bio-Monitoring														
					Plast.	Glass		H2SO4	HNO3	NAOH	HCL	Ice	None	Water	Soil	Air	Sludge	Other																
Outfall 001	11/20/14	0636	X		X		1												X							1214005								
	12-1-14	0720																																
Relinquished by: <i>Alan Bratton</i>										Date: 12-1-14					Time: 1300					Received By: <i>Alan Bratton</i>					Date: 12/1/14					Time: 1337				
Received by: <i>Alan Bratton</i>										Date: 12-1-14					Time: 1300					Relinquished By: <i>Alan Bratton</i>					Date: 12/1/14					Time: 1400				
Relinquished by: <i>Alan Bratton</i>										Date: 12-1-14					Time: 1337					Received by Laboratory: <i>Matt Warner</i>					Date: 12-2-14					Time: 1010				
Comments: 0.5°C										UPS																								

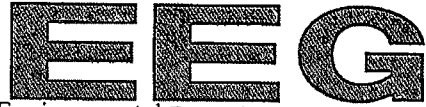


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 Russellville, Arkansas 72801
 (479) 968-6767 Fax (479) 968-1956

Company Name:		Phone #:		Requested Analysis												Laboratory Control Number	Remarks (Please note special detection limits below.)								
Clarksville Light and Water		(479) 754-7929																							
Address:		Fax #:														7-Day Chronic Bio-Monitoring									
P.O. Box 1807 Clarksville, AR 72830		(479) 754-8181																							
Project Name or Number:		Purchase Order #:																							
Bio-Monitoring																									
Sampling Personnel Signature(s):								Printed :																	
<i>Genny Underbrugh</i>								Gary Yarbrough																	
Sample I.D.	Date	Time	Comp.	Grab	Cont. Type		# of Containers	Method Preserved					Sample Matrix					7-Day Chronic Bio-Monitoring							
					Plast.	Glass		H2SO4	HNO3	NaOH	HCL	Ice	None	Water	Soil	Air	Sludge				Other				
Receiving Water	12-1-14	7:30		X	X		1						X		X				X				1214004		
Relinquished by:		Date:		Time:		Received By:		Date:		Time:		Received by Laboratory:		Date:		Time:									
<i>Genny Underbrugh</i>		12-1-14		1300		<i>Genny Underbrugh</i>		12/1/14		1337		<i>Matthew</i>		12/1/14		1600									
Received by:		Date:		Time:		Relinquished By:		Date:		Time:		Received by Laboratory:		Date:		Time:									
<i>sc 31</i>		12-1-14		1300		<i>Genny Underbrugh</i>		12/1/14		1600		<i>Matthew</i>		12-2-14		1010									
Relinquished by:		Date:		Time:		Received By:		Date:		Time:		Received by Laboratory:		Date:		Time:									
<i>sc 31</i>		12-1-14		1337		<i>Matthew</i>		12-2-14		1010		<i>Matthew</i>		12-2-14		1010									
Comments:																									
UPS																									

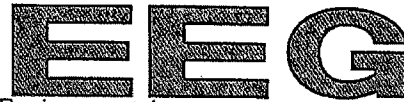


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Russellville, Arkansas 72801
(479) 968-6767 Fax (479) 968-1956

Company Name: Clarksville Light and Water											Phone #: (479) 754-7929				Requested Analysis								Laboratory Control Number	Remarks (Please note special detection limits below.)						
Address: P.O. Box 1807 Clarksville, AR 72830											Fax #: (479) 754-8181				7-Day Chronic Bio-Monitoring															
Project Name or Number: Bio-Monitoring											Purchase Order #:																			
Sampling Personnel Signature(s): <i>Porsha Russell</i>											Printed: <i>Porsha Russell</i>																			
Sample I.D.	Date	Time	Comp.	Grab	Cont. Type		# of Containers	Method Preserved						Sample Matrix																
					Plast.	Glass		H ₂ SO ₄	HNO ₃	NAOH	HCL	Ice	None	Water	Soil	Air	Sludge	Other												
Outfall 001	12-2-14 12-3-14	0740 0745	X		X		1						X	X						X		12/4005								
Relinquished by: <i>Porsha Russell</i>											Date: 12-2-14			Time: 1045		Received By: <i>Stacy Perren</i>											Date: 12/3/14		Time: 1135	
Received by: <i>slz</i>											Date: 12-3-14			Time: 1045		Relinquished By: <i>Stacy Perren</i>											Date: 12/3/14		Time: 1600	
Relinquished by: <i>slz</i>											Date: 12-3-14			Time: 1135		Received by Laboratory: <i>Matt Horner</i>											Date: 12-4-14		Time: 0935	
Comments: <i>0.9°C</i>																														

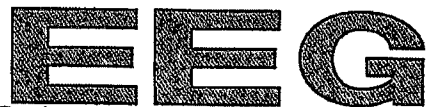


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Russellville, Arkansas 72801
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Company Name:		Phone #:		Requested Analysis										Laboratory Control Number	Remarks (Please note special detection limits below.)									
Clarksville Light and Water		(479) 754-7929		7-Day Chronic Bio-Monitoring																				
Address:		Fax #:																						
P.O. Box 1807 Clarksville, AR 72830		(479) 754-8181																						
Project Name or Number:		Purchase Order #:																						
Bio-Monitoring		Printed:																						
Sampling Personnel Signature(s):		Gary Yarbrough																						
Sample I.D.	Date	Time	Comp.	Grab	Cont Type		# of Containers	Method Preserved				Sample Matrix				7-Day Chronic Bio-Monitoring								
					Plast.	Glass		H2SO4	HNO3	NAOH	HCL	Ice	None	Water	Soil			Air	Sludge	Other				
Receiving Water	12-3-14	8:30		X	X		1				X		X					X					1214004	
Relinquished by:		Date:		Time:		Received By:		Date:		Time:		Relinquished By:		Date:		Time:		Received by Laboratory:		Date:		Time:		
Gary Yarbrough		12-3-14		1044		Stacy Kren		12/3/14		1135		Stacy Kren		12/3/14		1600		Matt Horner		12-4-14		0935		
Received by:		Date:		Time:		Relinquished By:		Date:		Time:		Received by Laboratory:		Date:		Time:		Received by Laboratory:		Date:		Time:		
[Signature]		12-3-14		1049		Stacy Kren		12/3/14		1600		Matt Horner		12-4-14		0935		Matt Horner		12-4-14		0935		
Relinquished by:		Date:		Time:		Received By:		Date:		Time:		Relinquished By:		Date:		Time:		Received by Laboratory:		Date:		Time:		
[Signature]		12-3-14		1135		Matt Horner		12-4-14		0935		Matt Horner		12-4-14		0935		Matt Horner		12-4-14		0935		
Comments:																								
UPS																								



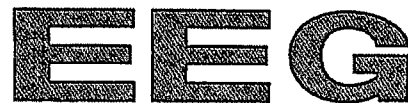
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220 North Knoxville, Suite 200
Russellville, Arkansas 72801
(479) 968-6767 Fax (479) 968-1956

L444-049203

Company Name:							Phone #:							Requested Analysis								Laboratory Control Number	Remarks (Please note special detection limits below.)
Clarksville Light and Water							(479) 754-7929							7-Day Chronic Bio-Monitoring									
Address:							Fax #:																
P.O. Box 1807 Clarksville, AR 72830							(479) 754-8181																
Project Name or Number:							Purchase Order #:																
Bio-Monitoring																							
Sampling Personnel Signature(s): <i>Porsha Russell</i>							Printed: <i>Porsha Russell</i>																
Sample I.D.	Date	Time	Comp.	Grab	Cont. Type		# of Containers	Method Preserved					Sample Matrix					7-Day Chronic Bio-Monitoring	Laboratory Control Number	Remarks			
					Plast.	Glass		H2SO4	HNO3	NAOH	HCL	Ice	None	Water	Soil	Air	Sludge				Other		
Outfall 001	12-4-14 12-5-14	0740 0730	X		X		1						X		X				X	1214005			
Relinquished by: <i>Porsha Russell</i>							Date: 12-5-14		Time: 0900		Received By: <i>Stacy Kren</i>					Date: 12/5/14		Time: 0945					
Received by: <i>Neil Z</i>							Date: 12-5-14		Time: 0900		Relinquished By: <i>Stacy Kren</i>					Date: 12/5/14		Time: 1600					
Relinquished by: <i>Neil Z</i>							Date: 12-5-14		Time: 0945		Received by Laboratory: <i>Matt Turner</i>					Date: 12-6X-14		Time: 1205					
Comments: <i>1.90C</i> <i>UPS</i>																							

DMH 12-6-14



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Russellville, Arkansas 72801
(479) 968-6767 Fax (479) 968-1956

Company Name:					Phone #:					Requested Analysis											
Clarksville Light and Water					(479) 754-7929					Laboratory Control Number 1274004 Remarks (Please note special detection limits below.)											
Address:					Fax #:																
P.O. Box 1807 Clarksville, AR 72830					(479) 754-8181																
Project Name or Number:					Purchase Order #:																
Bio-Monitoring																					
Sampling Personnel Signature(s): <i>Gary Yarbrough</i>							Printed : GARY YARBROUGH							7-Day Chronic Bio-Monitoring							
Sample I.D.	Date	Time	Comp.	Grab	Cont. Type		# of Containers	Method Preserved					Sample Matrix								
					Plast.	Glass		H2SO4	HNO3	NAOH	HCL	Ice	None	Water	Soil	Air	Sludge	Other			
Receiving Water	12-5-14	0945	X	X			1									X					
Relinquished by: <i>Gary Yarbrough</i>							Date: 12-5-14		Time: 0900		Received By: <i>Stacy Kren</i>					Date: 12/5/14		Time: 0945			
Received by: <i>Ed Zel</i>							Date: 12-5-14		Time: 0900		Relinquished By: <i>Stacy Kren</i>					Date: 12/5/14		Time: 1600			
Relinquished by: <i>Ed Zel</i>							Date: 12-5-14		Time: 0945		Received by Laboratory: <i>Matt Warner</i>					Date: 12-6-14		Time: 1205			
Comments: UPS																					

ENVIRONMENTAL ENTERPRISE GROUP
CITY OF CLARKSVILLE WWTP - OUTFALL 001
NPDES PERMIT NO. AR0022187
AFIN NO. 36-00038
BIOMONITORING REPORTING
TEST DATE: 12/02/14

II. *Ceriodaphnia dubia*

	Response
A. If the No Observed Effect Concentration (NOEC) for survival is less than the critical dilution, enter a "1"; otherwise, enter a "0". Parameter No. TLP3B.	0
B. If the No Observed Effect Concentration (NOEC) for reproduction is less than the critical dilution, enter a "1"; otherwise, enter a "0". Parameter No. TGP3B.	0
C. Report the NOEC value for survival, Parameter No. TOP3B.	100%
D. Report the NOEC value for reproduction, Parameter No. TPP3B.	100%
E. Report the highest (critical dilution or control) Coefficient of Variation, Parameter No. TQP3B.	6.43%

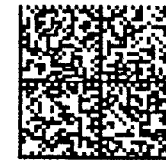
I. *Pimephales promelas*

	Response
A. If the No Observed Effect Concentration (NOEC) for survival is less than the critical dilution, enter a "1"; otherwise, enter a "0". Parameter No. TLP6C.	0
B. If the No Observed Effect Concentration (NOEC) for growth is less than the critical dilution, enter a "1"; otherwise, enter a "0". Parameter No. TGP6C.	0
C. Report the NOEC value for survival, Parameter No. TOP6C.	100%
D. Report the NOEC value for growth, Parameter No. TPP6C.	100%
E. Report the highest (critical dilution or control) Coefficient of Variation, Parameter No. TQP6C.	7.44%

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0001370120 DEC. 29. 2014

Clarksville Light & Water
400 West Main · P.O. Box 1807
Clarksville, AR 72830
Phone (479) 754-3148

Arkansas Department of Environmental Quality
5301 Northshore Drive
N. Little Rock, AR 72118

RETURN RECEIPT
REQUESTED

