

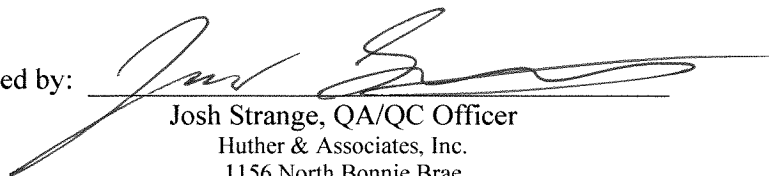
**CITY OF ROGERS**  
OUTFALL 001

Chronic Biomonitoring Report  
Permit Number NPDES AR0043397  
AFIN 04-00155

*Ceriodaphnia dubia*  
*Pimephales promelas*

April 21, 2020

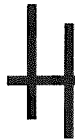
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TOXICITY TEST REPORT - CHRONIC

Client ..... City of Rogers Laboratory I.D. .... 31374
Permit No. .... NPDES AR0043397 Begin Date ..... April 21, 2020
Sample.....Outfall 001

Results: Pass Ceriodaphnia dubia survival and reproduction and Pimephales promelas survival and growth at the critical concentration (82% effluent).

SAMPLE COLLECTION

Composite effluent samples from City of Rogers were delivered by Federal Express courier to Huthur & Associates on April 21, April 23, and April 25, 2020. 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 samples were analyzed for total residual chlorine (Standard Methods, 23rd Edition, 4500-Cl D) and contained <0.01 mg/L, <0.01 mg/L, and <0.01 mg/L, respectively. Effluent and laboratory 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 1430 hours, April 21, 2020. Five concentrations were prepared (26%, 35%, 46%, 62% and 82% effluent) utilizing distilled, deionized laboratory water reconstituted to match the hardness, alkalinity and pH of the receiving stream (Osage Creek). The test was conducted in 25 mL distilled water rinsed plastic beakers containing 15 mL of solution (one organism 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.



**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: 82% 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**                      **PMSD: 8.4%**  
**NOEC: 82% Effluent**

**SUMMARY**

There were no statistically significant differences between the control and the critical low flow concentration (82% 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 AR0043397 for City of Rogers, Outfall 001 **passed** for this testing period.

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

CLIENT	City of Rogers	SAMPLE TYPE	24 Hour Composite
NPDES #	AR0043397	DATE COLLECTED	04/20/20 04/22/20 04/24/20
LAB ID #	31374	DATE RECEIVED	04/21/20 04/23/20 04/25/20
TEST TYPE	7 Day Chronic	BEGIN DATE/TIME	04/21/20 1430
TEST ORGANISM	<i>Ceriodaphnia dubia</i>	END DATE/TIME	04/28/20 1430
ORGANISM AGE	< 24-Hours	TEST TEMPERATURE (°C)	25 ± 1
ORGANISM SOURCE	In House	PHOTO PERIOD	16-hr. Light 8-hr. Dark
RECEIVING WATER	Osage Creek	LIGHT INTENSITY	50-100 ft. candl.
DILUTION WATER	Laboratory	TECHNICIAN	T. Geiger

**SURVIVAL & REPRODUCTION SUMMARY**

Control										
Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
04/22/20	A	A	A	A	A	A	A	A	A	A
04/23/20	0	0	0	0	0	0	0	0	0	0
04/24/20	0	0	0	0	0	0	0	0	0	0
04/25/20	3	2	2	5	4	3	5	4	4	3
04/26/20	10	9	10	7	6	8	6	9	9	11
04/27/20	13	11	12	12	10	11	11	13	13	14
04/28/20	26	23	25	26	23	23	24	27	26	27
x# Young 25.0					C.V. 6.53%					
x%Survival 100%					C.V. 0.00%					

26%Effluent										
Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
04/22/20	A	A	A	A	A	A	A	A	A	A
04/23/20	0	0	0	0	0	0	0	0	0	0
04/24/20	0	0	0	0	0	0	0	0	0	0
04/25/20	2	4	4	3	2	5	3	2	4	3
04/26/20	11	8	9	10	7	7	6	9	8	8
04/27/20	13	12	13	13	9	12	9	11	12	11
04/28/20	25	24	26	26	23	25	21	24	25	24
x# Young 24.3					C.V. 6.15%					
x%Survival 100%					C.V. 0.00%					

35%Effluent										
Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
04/22/20	A	A	A	A	A	A	A	A	A	A
04/23/20	0	0	0	0	0	0	0	0	0	0
04/24/20	0	0	0	0	0	0	0	0	0	0
04/25/20	3	5	4	3	2	4	3	4	3	3
04/26/20	8	8	10	7	10	11	7	9	10	10
04/27/20	11	13	14	10	12	15	10	13	13	13
04/28/20	25	26	26	22	25	28	24	26	25	26
x# Young 25.3					C.V. 6.19%					
x%Survival 100%					C.V. 0.00%					

46%Effluent										
Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
04/22/20	A	A	A	A	A	A	A	A	A	A
04/23/20	0	0	0	0	0	0	0	0	0	0
04/24/20	0	0	0	0	0	0	0	0	0	0
04/25/20	2	2	3	4	3	5	4	3	2	5
04/26/20	9	8	6	6	7	10	9	8	7	6
04/27/20	11	10	9	10	10	15	13	11	9	11
04/28/20	24	23	22	22	23	28	25	25	22	23
x# Young 23.7					C.V. 7.97%					
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

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

Rogers

Lab ID# 31374

Test Date: April 21, 2020

62%Effluent										
Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
04/22/20	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
04/23/20	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
04/24/20	0	0	0	0	0	0	0	0	0	0
	4	3	2	4	3	4	5	3	2	3
04/25/20	4	3	2	4	3	4	5	3	2	3
	9	9	8	10	6	6	9	10	7	10
04/26/20	13	12	10	14	9	10	14	13	9	13
	A	A	A	A	A	A	A	A	A	A
04/27/20	13	12	10	14	9	10	14	13	9	13
	13	12	14	13	12	13	12	13	13	12
04/28/20	26	24	24	27	21	23	26	26	22	25
	x# Young 24.4					C.V. 8.01%				
x%Survival 100%					C.V. 0.00%					

82%Effluent										
Date	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
04/22/20	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
04/23/20	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
04/24/20	A	A	A	A	A	A	A	A	A	A
	0	0	0	0	0	0	0	0	0	0
04/25/20	2	4	4	3	2	5	4	3	3	2
	2	4	4	3	2	5	4	3	3	2
04/26/20	10	6	10	11	7	9	10	8	9	9
	12	10	14	14	9	14	14	11	12	11
04/27/20	A	A	A	A	A	A	A	A	A	A
	12	10	14	14	9	14	14	11	12	11
04/28/20	13	12	14	13	12	12	12	12	13	13
	25	22	28	27	21	26	26	23	25	24
x# Young 24.7					C.V. 8.96%					
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

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

Rogers

Lab ID# 31374

Test Date: April 21, 2020

**WET CHEMISTRY MEASUREMENTS**

Date	Time	Temp	Samp. No.	pH of Solution						Analyst
				CON	26%	35%	46%	62%	82%	
04/21/20	Start	25.0	1	7.88	7.90	7.84	7.79	7.73	7.72	SD
04/22/20	24 Hr.	24.5	1	8.20	8.08	8.03	7.98	7.96	7.96	LM
04/22/20	Renew	24.7	1	8.11	8.00	7.92	7.88	7.85	7.82	LM
04/23/20	48 Hr.	24.2	1	7.86	7.85	7.90	7.92	7.95	7.96	SD
04/23/20	Renew	25.0	2	7.76	7.68	7.63	7.56	7.51	7.44	SD
04/24/20	72 Hr.	24.3	2	8.28	8.26	8.25	8.23	8.23	8.20	LM
04/24/20	Renew	24.3	2	7.82	7.76	7.73	7.64	7.57	7.51	LM
04/25/20	96 Hr.	24.4	2	8.42	8.24	8.14	8.11	8.07	8.08	LM
04/25/20	Renew	25.0	3	7.69	7.71	7.73	7.72	7.67	7.57	SD
04/26/20	120 Hr.	24.2	3	8.19	8.14	8.12	8.11	8.11	8.12	LM
04/26/20	Renew	24.2	3	7.55	7.77	7.85	7.90	7.81	7.79	LM
04/27/20	144 Hr.	24.3	3	8.00	7.93	7.90	7.89	7.82	7.75	LM
04/27/20	Renew	24.3	3	7.89	7.87	7.84	7.85	7.74	7.69	LM
04/28/20	168 Hr.	25.0	3	7.93	7.95	7.98	7.98	7.96	7.96	SD

Date	Time	Temp	Samp. No.	DO (mg/L) of Solution						Analyst
				CON	26%	35%	46%	62%	82%	
04/21/20	Start	25.0	1	7.87	7.92	7.93	7.93	7.94	8.28	SD
04/22/20	24 Hr.	24.5	1	8.14	8.16	8.17	7.73	7.51	8.10	LM
04/22/20	Renew	24.7	1	8.64	8.60	7.66	8.40	8.39	8.62	LM
04/23/20	48 Hr.	24.2	1	8.62	8.58	8.49	8.28	8.34	8.32	SD
04/23/20	Renew	25.0	2	8.00	8.44	8.65	7.82	7.76	7.73	SD
04/24/20	72 Hr.	24.3	2	7.74	7.77	8.40	8.52	8.56	8.53	LM
04/24/20	Renew	24.3	2	8.52	8.38	8.50	8.48	8.53	8.43	LM
04/25/20	96 Hr.	24.4	2	7.70	8.52	8.60	7.70	8.64	7.69	LM
04/25/20	Renew	25.0	3	8.63	8.55	8.52	8.58	8.64	8.60	SD
04/26/20	120 Hr.	24.2	3	8.00	7.91	7.92	7.98	8.20	8.49	LM
04/26/20	Renew	24.2	3	7.75	8.53	8.59	7.64	7.86	7.30	LM
04/27/20	144 Hr.	24.3	3	7.95	8.61	8.46	8.17	8.58	8.63	LM
04/27/20	Renew	24.3	3	7.83	8.41	8.17	8.41	8.52	8.58	LM
04/28/20	168 Hr.	25.0	3	7.85	8.56	8.65	7.88	8.55	8.40	SD



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

Rogers

Lab ID# 31374

Test Date: April 21, 2020

**INITIAL CHEMISTRY MEASUREMENTS @ 100% EFFLUENT**

Date	Samp. No.	pH <sup>1</sup>	DO <sup>1</sup>	Hardness mg/L CaCO <sub>3</sub> <sup>1</sup>	Alkalinity mg/L CaCO <sub>3</sub> <sup>1</sup>	Conduct. μS/cm <sup>1</sup>	Resid. Cl <sub>2</sub> mg/L <sup>1</sup>	Dechlor(mL) Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> mg/L <sup>1</sup>	Analyst
04/21/20	1	7.82	7.70	124	104	587	<0.01	N/A	SD
04/23/20	2	7.62	7.60	116	104	592	<0.01	N/A	SD
04/25/20	3	7.25	8.30	148	94	527	<0.01	N/A	SD
04/21/20	CON	7.88	7.87	120	72	417	-	-	JS

<sup>1</sup> Measurements taken in 100% solution.

*CERIODAPHNIA DUBIA* STATISTICAL ANALYSES  
 Reproduction

Summary Statistics on Transformed Data Table 1 of 2

Grp	Identification	N	Min	Max	Mean
1	Control	10	23.000	27.000	25.000
2	26% Effluent	10	21.000	26.000	24.300
3	35% Effluent	10	22.000	28.000	25.300
4	46% Effluent	10	22.000	28.000	23.700
5	62% Effluent	10	21.000	27.000	24.400
6	82% Effluent	10	21.000	28.000	24.700

Summary Statistics on Transformed Data Table 2 of 2

Grp	Identification	Variance	Sd	Sem	C.V.%
1	Control	2.667	1.633	0.516	6.53
2	26% Effluent	2.233	1.494	0.473	6.15
3	35% Effluent	2.456	1.567	0.496	6.19
4	46% Effluent	3.567	1.889	0.597	7.97
5	62% Effluent	3.822	1.955	0.618	8.01
6	82% Effluent	4.900	2.214	0.700	8.96

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	4	13	24	17	2

Calculated Chi-Square goodness of fit test statistic = 1.6487

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 = 2.00

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	15.933	3.187	0.973
Within (Error)	54	176.800	3.274	
Total	59	192.733		

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	Transformed Mean	Mean	T Stat	Sig
			Calculated In Original Units		
1	Control	25.000	25.000		
2	26% Effluent	24.300	24.300	0.865	
3	35% Effluent	25.300	25.300	-0.371	
4	46% Effluent	23.700	23.700	1.607	
5	62% Effluent	24.400	24.400	0.741	
6	82% Effluent	24.700	24.700	0.371	

Dunnett table value = 2.31 (1 Tailed Value, P=0.05, DF=40,5)

**No statistically significant difference**

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

Grp	Identification	Num of Reps	Minimum Sig	% of Control	Difference
			Diff (In Orig. Units)		from Control
1	Control	10			
2	26% Effluent	10	1.869	7.5	0.700
3	35% Effluent	10	1.869	7.5	-0.300
4	46% Effluent	10	1.869	7.5	1.300
5	62% Effluent	10	1.869	7.5	0.600
6	82% Effluent	10	1.869	7.5	0.300

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

CLIENT	City of Rogers	SAMPLE TYPE	24 Hour Composite
NPDES #	AR0043397	DATE COLLECTED	04/20/20 04/22/20 04/24/20
LAB ID #	31374	DATE RECEIVED	04/21/20 04/23/20 04/25/20
TEST TYPE	7 Day Chronic	BEGIN DATE/TIME	04/21/20 1600
TEST ORGANISM	<i>Pimephales promelas</i>	END DATE/TIME	04/28/20 1600
ORGANISM AGE	< 24-Hours	TEST TEMPERATURE (°C)	25 ± 1
ORGANISM SOURCE	In House	PHOTO PERIOD	16-hr. Light 8-hr. Dark
RECEIVING WATER	Osage Creek	LIGHT INTENSITY	50-100 ft. cndl.
DILUTION WATER	Laboratory	TECHNICIAN	J. Castillo

**SURVIVAL SUMMARY**

Conc.	04/22/20					04/23/20					04/24/20					04/25/20					04/26/20				
	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
CON	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
26%	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
35%	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
46%	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
62%	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
82%	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.	04/27/20					04/28/20					x % Survival	C.V. %
	A	B	C	D	E	A	B	C	D	E		
CON	8	8	8	8	8	8	8	8	8	8	100.0	0.00
26%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
35%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
46%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
62%	8	8	8	8	8	8	8	8	8	8	100.0	0.00
82%	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. %
CON	0.4290	0.4630	0.4540	0.4270	0.4830	0.4512	5.24
26%	0.4710	0.4770	0.4820	0.4360	0.4450	0.4622	4.42
35%	0.4490	0.4670	0.4230	0.4890	0.4820	0.4620	5.77
46%	0.4670	0.4530	0.4790	0.4900	0.4240	0.4626	5.53
62%	0.4150	0.4730	0.4860	0.4670	0.4880	0.4658	6.38
82%	0.4530	0.4210	0.4740	0.4830	0.4790	0.4620	5.56

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

Rogers

Lab ID# 31374

Test Date: April 21, 2020

**WET CHEMISTRY MEASUREMENTS**

Date	Time	Temp	Samp. No.	pH of Solution						Analyst
				CON	26%	35%	46%	62%	82%	
04/21/20	Start	25.0	1	7.88	7.90	7.84	7.79	7.73	7.72	SD
04/22/20	24 Hr.	24.8	1	8.31	8.13	8.06	8.00	7.91	7.84	LM
04/22/20	Renew	24.7	1	8.11	8.00	7.92	7.88	7.85	7.82	LM
04/23/20	48 Hr.	24.5	1	8.05	8.03	7.90	7.89	7.90	7.87	SD
04/23/20	Renew	25.0	2	7.76	7.68	7.63	7.56	7.51	7.44	SD
04/24/20	72 Hr.	24.7	2	7.73	7.74	7.76	7.69	7.84	7.73	LM
04/24/20	Renew	24.3	2	7.82	7.76	7.73	7.64	7.57	7.51	LM
04/25/20	96 Hr.	24.6	2	7.88	7.86	7.79	7.75	7.74	7.73	SD
04/25/20	Renew	25.0	3	7.69	7.71	7.73	7.72	7.67	7.57	SD
04/26/20	120 Hr.	24.5	3	7.92	7.84	7.88	7.84	7.79	7.80	LM
04/26/20	Renew	24.2	3	7.55	7.77	7.85	7.90	7.81	7.79	LM
04/27/20	144 Hr.	24.7	3	8.01	7.81	7.91	7.83	7.80	7.71	LM
04/27/20	Renew	24.3	3	7.89	7.87	7.84	7.85	7.74	7.69	LM
04/28/20	168 Hr.	25.2	3	7.66	7.55	7.53	7.46	7.45	7.54	SD

Date	Time	Temp	Samp. No.	DO (mg/L) of Solution						Analyst
				CON	26%	35%	46%	62%	82%	
04/21/20	Start	25.0	1	7.87	7.92	7.93	7.93	7.94	8.28	SD
04/22/20	24 Hr.	24.8	1	7.96	8.04	8.32	8.17	7.87	8.42	LM
04/22/20	Renew	24.7	1	8.64	8.60	7.66	8.40	8.39	8.62	LM
04/23/20	48 Hr.	24.5	1	8.16	8.19	8.07	7.85	7.69	7.66	SD
04/23/20	Renew	25.0	2	8.00	8.44	8.65	7.82	7.76	7.73	SD
04/24/20	72 Hr.	24.7	2	8.63	8.40	8.39	8.45	8.53	8.57	LM
04/24/20	Renew	24.3	2	8.52	8.38	8.50	8.48	8.53	8.43	LM
04/25/20	96 Hr.	24.6	2	8.62	7.67	8.49	8.34	8.37	8.49	SD
04/25/20	Renew	25.0	3	8.63	8.55	8.52	8.58	8.64	8.60	SD
04/26/20	120 Hr.	24.5	3	8.37	7.77	7.78	7.77	7.70	8.47	LM
04/26/20	Renew	24.2	3	7.75	8.53	8.59	7.64	7.86	7.30	LM
04/27/20	144 Hr.	24.7	3	8.60	8.58	8.41	7.43	7.27	8.55	LM
04/27/20	Renew	24.3	3	7.83	8.41	8.17	8.41	8.52	8.58	LM
04/28/20	168 Hr.	25.2	3	7.87	7.77	7.69	7.74	7.86	7.71	SD

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

Rogers

Lab ID# 31374

Test Date: April 21, 2020

**INITIAL CHEMISTRY MEASUREMENTS @ 100% EFFLUENT**

Date	Samp. No.	pH <sup>1</sup>	DO <sup>1</sup>	Hardness mg/L CaCO <sub>3</sub> <sup>1</sup>	Alkalinity mg/L CaCO <sub>3</sub> <sup>1</sup>	Conduct. μS/cm <sup>1</sup>	Resid.Cl <sub>2</sub> mg/L <sup>1</sup>	Dechlor(mL) Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> mg/L <sup>1</sup>	Analyst
04/21/20	1	7.82	7.70	124	104	587	<0.01	N/A	SD
04/23/20	2	7.62	7.60	116	104	592	<0.01	N/A	SD
04/25/20	3	7.25	8.30	148	94	527	<0.01	N/A	SD
04/21/20	CON	7.88	7.87	120	72	417	-	-	JS

<sup>1</sup> Measurements taken in 100% solution.

*PIMEPHALES PROMELAS* STATISTICAL ANALYSES  
 Growth

Summary Statistics on Transformed Data Table 1 of 2

Grp	Identification	N	Min	Max	Mean
1	Control	5	0.427	0.483	0.451
2	26% Effluent	5	0.436	0.482	0.462
3	35% Effluent	5	0.423	0.489	0.462
4	46% Effluent	5	0.424	0.490	0.463
5	62% Effluent	5	0.415	0.488	0.466
6	82% Effluent	5	0.421	0.483	0.462

Summary Statistics on Transformed Data Table 2 of 2

Grp	Identification	Variance	Sd	Sem	C.V.%
1	Control	0.001	0.024	0.011	5.24
2	26% Effluent	0.000	0.020	0.009	4.42
3	35% Effluent	0.001	0.027	0.012	5.77
4	46% Effluent	0.001	0.026	0.011	5.53
5	62% Effluent	0.001	0.030	0.013	6.38
6	82% Effluent	0.001	0.026	0.011	5.56

Shapiro - Wilk's Test For Normality

D = 0.016

W = 0.916

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 = 0.55

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	0.193
Within (Error)	24	0.016	0.001	
Total	29	0.016		

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	Mean		T Stat	Sig
			Original Units	Calculated In		
1	Control	0.451	0.451			
2	26% Effluent	0.462	0.462		-0.683	
3	35% Effluent	0.462	0.462		-0.671	
4	46% Effluent	0.463	0.463		-0.708	
5	62% Effluent	0.466	0.466		-0.907	
6	82% Effluent	0.462	0.462		-0.671	

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

**No statistically significant difference**

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

Grp	Identification	Num of Reps	Minimum Sig		Difference from Control
			Diff (In Orig. Units)	% of Control	
1	Control	5			
2	26% Effluent	5	0.038	8.4	-0.011
3	35% Effluent	5	0.038	8.4	-0.011
4	46% Effluent	5	0.038	8.4	-0.011
5	62% Effluent	5	0.038	8.4	-0.015
6	82% Effluent	5	0.038	8.4	-0.011

**APPENDIX A  
RAW DATA**

7-DAY CERIODAPHNIA DUBLA SURVIVAL & REPRODUCTION

DAILY RAW DATA TABLE

PAGE 1 OF 2

CLIENT Rogers

START DATE/TIME 4-21-20 TG 1430

OUTFALL 001

END DATE/TIME 4-28-20 TG 1430

LAB ID # 31374

Con

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
4/22	A	A	A	A	A	A	A	A	A	A	TG	1430
4/23	A	A	A	A	A	A	A	A	A	A	TG	1400
04/24	A	A	A	A	A	A	A	A	A	A	DK	1315
4/25	3	2	2	5	4	3	5	4	4	3	TG	1315
4/26	10	9	10	7	6	8	6	9	9	11	TG	1315
4/27	A	A	A	A	A	A	A	A	A	A	MH	1200
4/28	13	12	13	14	13	12	13	14	13	13	TG	1430
	26	23	25	26	23	23	24	27	26	27		

$\bar{x}$  # Young w/o Dead = 25.0 CV% = 6.53

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

$\bar{x}$  % Survival = 100 CV% = 0.00

26

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
4/22	A	A	A	A	A	A	A	A	A	A	TG	1430
4/23	A	A	X	A	A	A	A	A	A	A	TG	1400
04/24	A	A	A	A	A	A	A	A	A	A	DK	1315
4/25	2	4	4	3	2	5	3	2	4	3	TG	1315
4/26	11	8	9	10	7	7	6	9	8	8	TG	1315
4/27	A	A	A	A	A	A	A	A	A	A	MH	1200
4/28	12	12	13	13	14	13	12	13	13	13	TG	1430
	25	24	26	26	23	25	21	24	25	24		

$\bar{x}$  # Young w/o Dead = 24.3 CV% = 6.15

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

$\bar{x}$  % Survival = 100 CV% = 0.00

35

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
4/22	A	A	A	A	A	A	A	A	A	A	TG	1430
4/23	A	A	A	A	A	A	A	A	A	A	TG	1400
04/24	A	A	A	A	A	A	A	A	A	A	DK	1315
4/25	3	5	4	3	2	4	3	4	3	3	TG	1315
4/26	8	8	10	7	10	11	7	9	10	10	TG	1315
4/27	A	A	A	A	A	A	A	A	A	A	MH	1200
4/28	14	13	12	12	13	13	14	13	12	13	TG	1430
	25	26	26	22	25	28	24	26	25	26		

$\bar{x}$  # Young w/o Dead = 25.3 CV% = 6.19

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

$\bar{x}$  % Survival = 100 CV% = 0.00

46

Date	Rep1	Rep2	Rep3	Rep4	Rep5	Rep6	Rep7	Rep8	Rep9	Rep10	Analyst	Time
4/22	A	A	A	A	A	A	A	A	A	A	TG	1430
4	A	A	A	A	A	A	A	A	A	A	TG	1400
04/24	A	A	A	A	A	A	A	A	A	A	DK	1315
4/25	2	2	3	4	3	5	4	3	2	5	TG	1315
4/26	9	8	6	6	7	10	9	8	7	6	TG	1315
4/27	A	A	A	A	A	A	A	A	A	A	MH	1200
4/28	13	13	13	12	13	13	12	14	13	12	TG	1430
	24	23	22	22	23	28	25	25	22	23		

$\bar{x}$  # Young w/o Dead = 23.7 CV% = 7.97

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

$\bar{x}$  % Survival = 100 CV% = 0.00





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

CLIENT/FACILITY: Rogers DATE/TIME STARTED: 4-21-20 5c 1600  
 OUTFALL #: 001 PROJECT #: 31374 DATE/TIME ENDED: 4-28-20 5c 1600  
 ORGANISM ID#: PP0-20-111

Conc.	4-22-20 5c 1600					4-23-20 5c 1405					4-24-20 MH 0955					4-25-20 5c 825					4-26-20 5c 1000									
Initials Date/Time	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					
con	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	8	8	8	8	8
26	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	8	8	8	8	8
35	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	8	8	8	8	8
46	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	8	8	8	8	8
62	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	8	8	8	8	8
82	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	8	8	8	8	8

Conc.	4-27-20 MH 0905					4-28-20 5c 1600					Mean Survival	C.V. %
Initials Date/Time	A	B	C	D	E	A	B	C	D	E		
con	8	8	8	8	8	8	8	8	8	8	100	0.00
26	8	8	8	8	8	8	8	8	8	8	100	0.00
35	8	8	8	8	8	8	8	8	8	8	100	0.00
46	8	8	8	8	8	8	8	8	8	8	100	0.00
62	8	8	8	8	8	8	8	8	8	8	100	0.00
82	8	8	8	8	8	8	8	8	8	8	100	0.00



Client / Facility Rogers  
 Lab ID Number 31374  
 Outfall Number 001  
 Test Date 4-21-20

**INITIAL CHEMISTRY MEASUREMENTS @ 100% EFFLUENT**

Date	Samp. No.	pH	DO	Hardness mg/L CaCO <sub>3</sub> <sup>1</sup>	Alkalinity mg/L CaCO <sub>3</sub> <sup>1</sup>	Conduct. umhos/cm <sup>1</sup>	Resid. Cl <sub>2</sub> mg/L <sup>1</sup>	Dechlor(mL) Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> mg/L <sup>1</sup>	Analyst
4-21-20	1	7.82	7.70	124	104	587	<0.01	N/A	SD
4-23-20	2	7.62	7.60	116	104	592	<0.01	N/A	SD
4-25-20	3	7.25	8.30	148	94	527	<0.01	N/A	SD
4-21-20	CON	7.88	7.87	120	72	417	-	-	JS

**INITIAL CHEMISTRY MEASUREMENTS @ RECEIVING WATER**

Date	Samp. No.	pH	DO	Hardness mg/L CaCO <sub>3</sub> <sup>1</sup>	Alkalinity mg/L CaCO <sub>3</sub> <sup>1</sup>	Conduct. umhos/cm <sup>1</sup>	Resid. Cl <sub>2</sub> mg/L <sup>1</sup>	Dechlor(mL) Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> mg/L <sup>1</sup>	Analyst

Notes:

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**APPENDIX B**  
**REFERENCE TOXICANTS**

**CHRONIC REFERENCE TOXICANT TEST RESULTS**

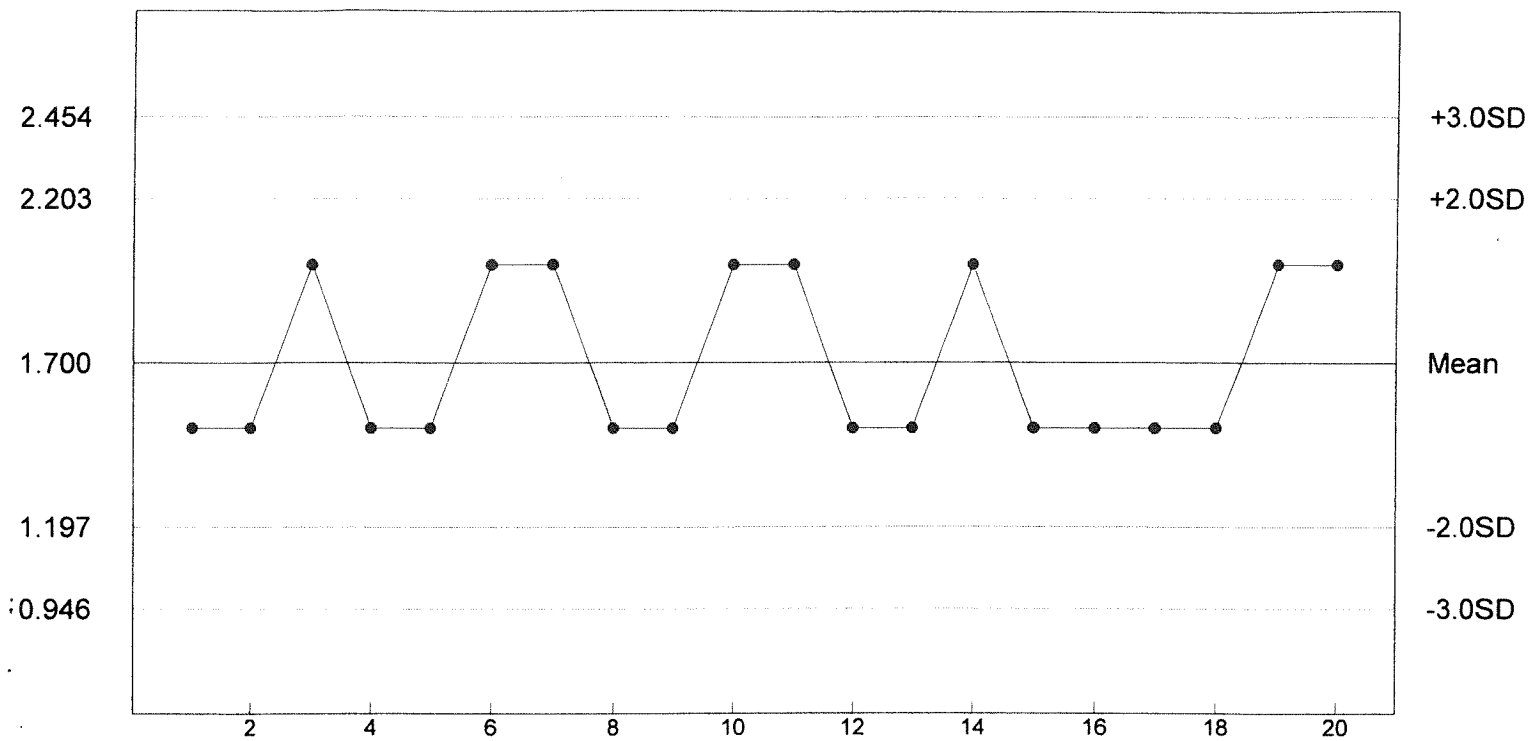
SPECIES: *Ceriodaphnia dubia*  
 CHEMICAL: Sodium Chloride  
 DURATION: 7-Days  
 TEST NUMBER: 4  
 TEST DATE: 04/01/20 - 04/08/20  
 1600 Hrs - 1600 Hrs  
 STATISTICAL METHOD: Dunnetts/Steels

CONCENTRATION (ug/L)	NUMBER EXPOSED	NUMBER DEAD
0.5	10	0
1.0	10	0
1.5	10	0
2.0	10	2
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

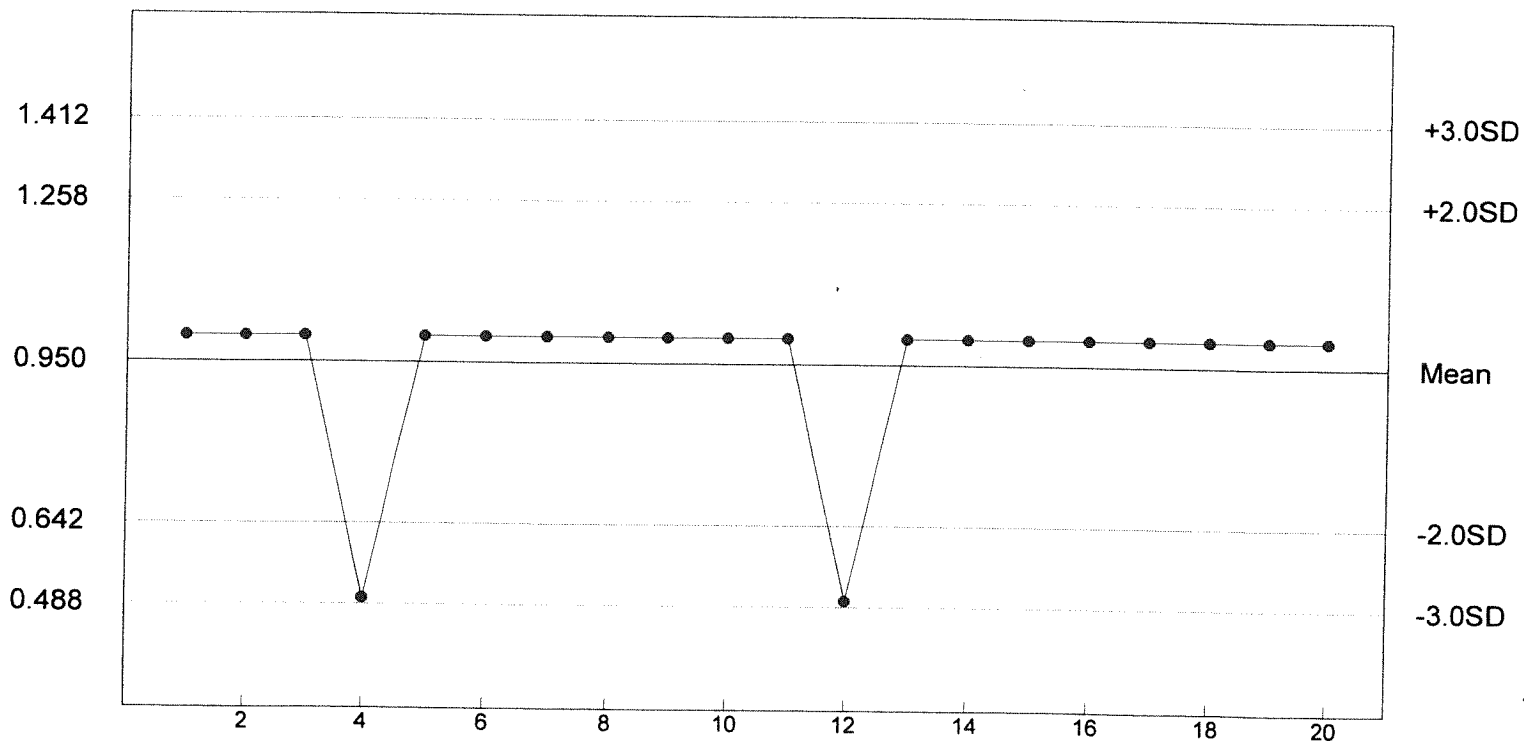
C. dubia Survival - NOEC



n= 20 Mean= 1.700 SD= 0.251 CV= 14.78% Min= 1.500 Max= 2.000

Reference Tox Sodium Chloride g/L

C. dubia Reproduction - NOEC



n= 20 Mean= 0.950 SD= 0.154 CV= 16.20% Min= 0.500 Max= 1.000

**CHRONIC REFERENCE TOXICANT TEST RESULTS**

SPECIES: *Pimephales promelas*  
 CHEMICAL: Copper Nitrate  
 DURATION: 7-Days  
 TEST NUMBER: 4  
 TEST DATE: 04/01/20 - 04/08/20  
 1440 Hrs - 1440 Hrs  
 STATISTICAL METHOD: Dunnetts/Steels

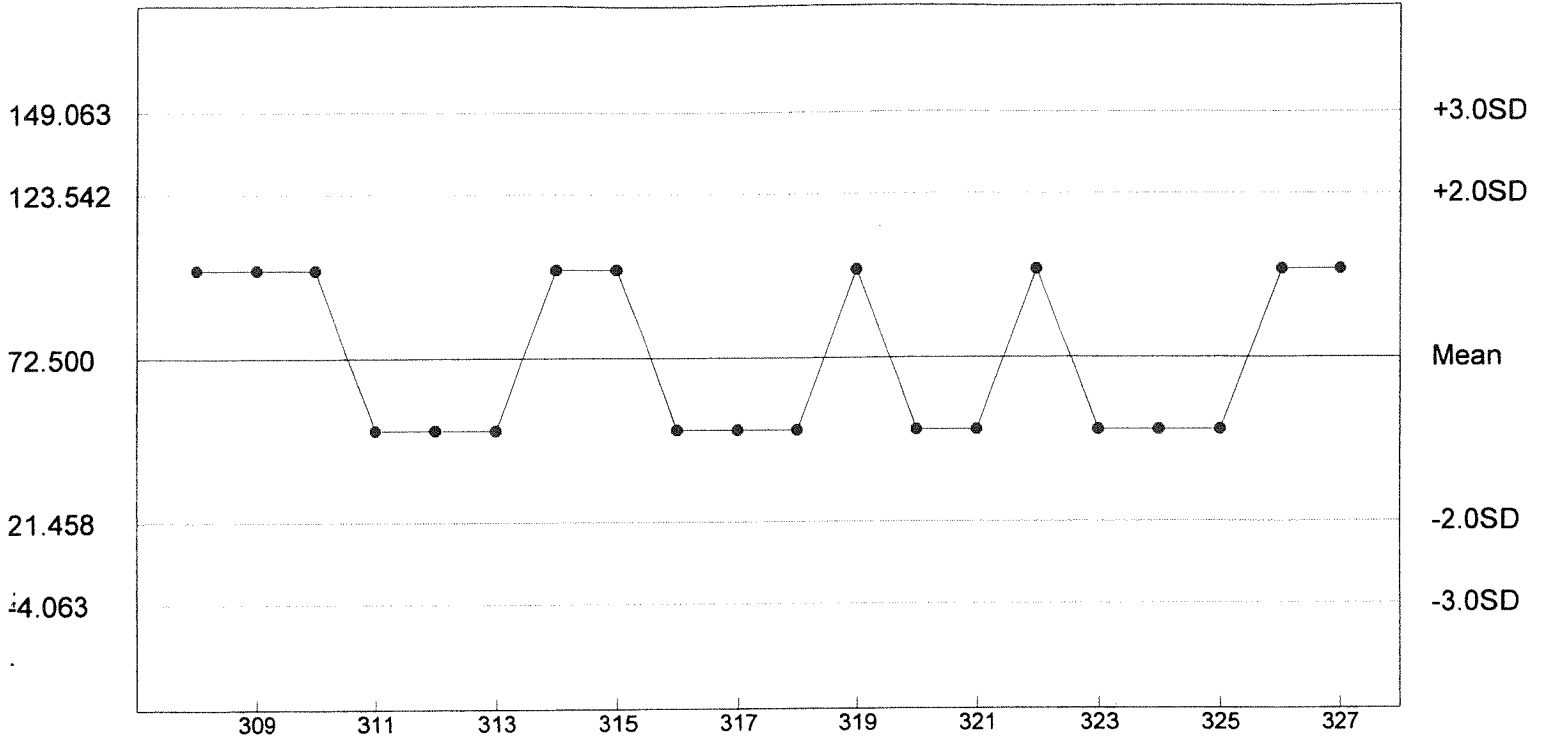
CONCENTRATION (ug/L)	NUMBER EXPOSED	NUMBER DEAD
12.5	40	0
25	40	0
50	40	0
100	40	2
200	40	19
400	40	29
800	40	40

LOEC FOR SURVIVAL	NOEC FOR SURVIVAL	LOEC FOR GROWTH	NOEC FOR GROWTH
<b>200 ug/L</b>	<b>100 ug/L</b>	<b>200 ug/L</b>	<b>100 ug/L</b>



Reference Tox Copper Nitrate ug/L

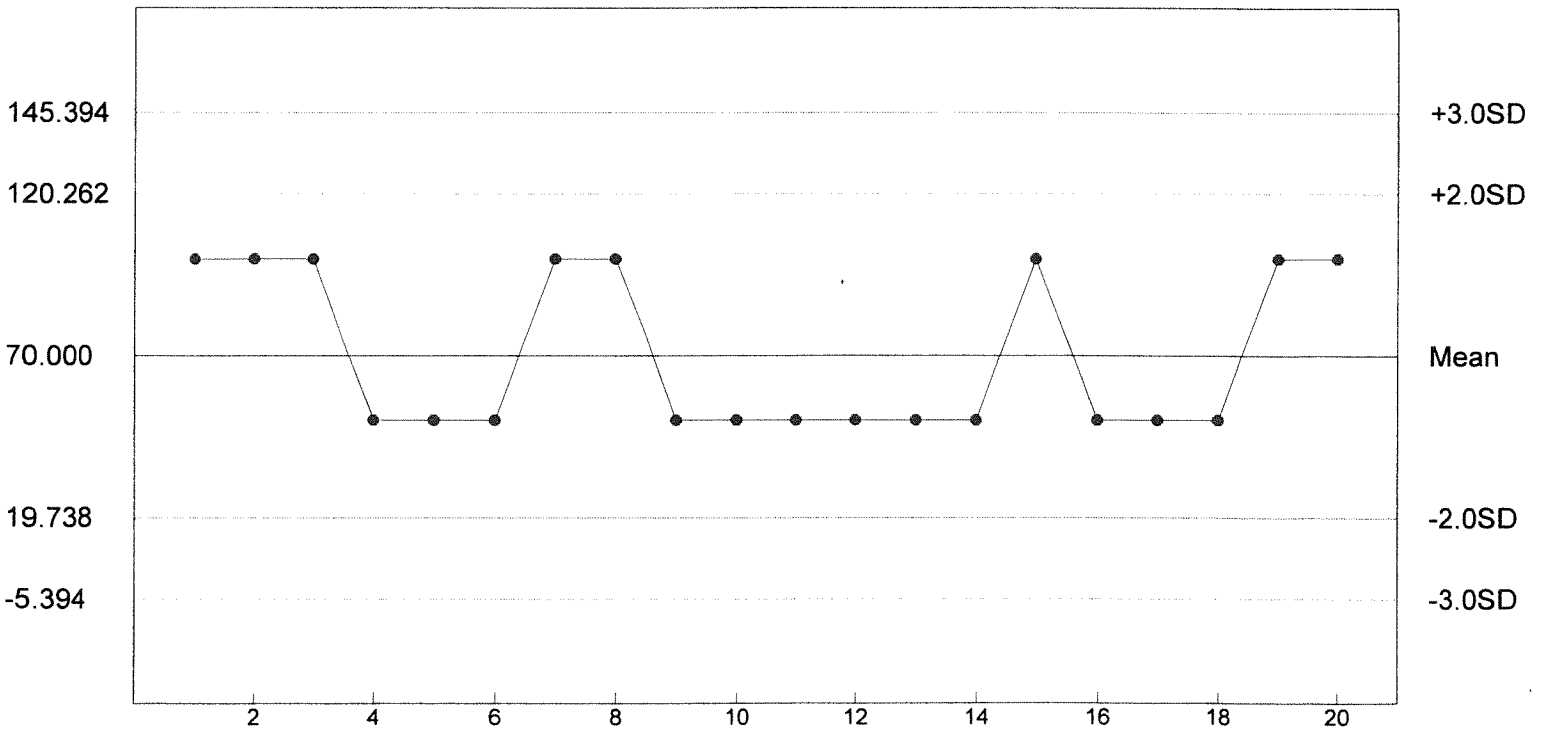
P. promelas Chronic Survival - NOEC



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

Reference Tox Copper Nitrate ug/L

P. promelas Growth - NOEC



n= 20 Mean= 70.000 SD= 25.131 CV= 35.90% Min= 50.000 Max= 100.000

**APPENDIX C**  
**CHAIN OF CUSTODY SHEETS**







**CITY OF ROGERS  
 NPDES PERMIT NO. AR0043397  
 AFIN NUMBER: 04-00155  
 BIOMONITORING REPORTING  
 TEST DATE: 04/21/20**

**I. *Ceriodaphnia dubia***

**Response**

(A) If the NOEC for survival is less than the critical dilution, enter a "1"; otherwise, enter a "0". Parameter No. TLP3B.	0
(B) Report the NOEC value for survival, Parameter No. TOP3B.	82%
(C) Report the NOEC value for reproduction, Parameter No. TPP3B.	82%
(D) If the NOEC for reproduction is less than the critical dilution, enter a "1"; otherwise, enter a "0". Parameter No. TGP3B.	0
(E) Report the higher (critical dilution or control) Coefficient of Variation, Parameter No. TQP3B.	8.96%

**II. *Pimephales promelas* (fathead minnow)**

**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) Report the NOEC value for survival, Parameter No. TOP6C.	82%
(C) Report the NOEC value for growth, Parameter No. TPP6C.	82%
(D) 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
(E) Report the highest (critical dilution or control) Coefficient of Variation, Parameter No. TQP6C.	5.56%

22414 - 10	82%
22414 - PO	82%