

## Bio-Analytical Laboratories' Executive Summary

**Permittee:** Nashville Public Works  
Nashville, AR 71852

**Project #:** X9028

**Outfall:** 001 (treated municipal wastewater)

**Permit #:** AR0021776/ AFIN #31-00036

**Contact:** Southern Petroleum Laboratories  
4720 Viking Drive, Ste A  
Bossier City, LA 71111

**Test Dates:** November 28 – December 5, 2023

**Test Type:** Chronic Static Renewal Survival and Reproduction Test using  
*Ceriodaphnia dubia* (EPA Method 1002.0)  
Chronic Static Renewal Survival and Growth Test using  
*Pimephales promelas* (EPA Method 1000.0)

### Results:

#### For *Ceriodaphnia dubia*:

1. If the NOEC for survival is less than the critical dilution (78.0%), enter a "1"; otherwise, enter a "0" for Parameter TLP3B - 0 -**Pass**
2. If the NOEC for reproduction is less than the critical dilution, enter a "1"; otherwise, enter a "0" for Parameter TGP3B - 0 - **Pass**
3. Report the NOEC value for survival, Parameter TOP3B - 78.0%.
4. Report the NOEC value for reproduction, Parameter TPP3B - 78.0%.
5. Report the largest % coefficient of variation between the control and the critical dilution, Parameter TQP3B – 23.65%
6. Chronic WET Limit, DMR CODE 51710, - 78.0%
7. PMSD Reproduction-31.65%(13.0 – 47.0%) moderate precision, acceptable for passing test.

#### For *Pimephales promelas*:

1. If the NOEC for survival is less than the critical dilution (78.0%), enter a "1"; otherwise, enter a "0" for Parameter TLP6C - 0 -**Pass**
2. If the NOEC for growth is less than the critical dilution, enter a "1"; otherwise, enter a "0" for Parameter TGP6C - 0 -**Pass**
3. Report the NOEC value for survival, Parameter TOP6C - 78.0%.
4. Report the NOEC value for growth, Parameter TPP6C - 78.0%.
5. Report the largest % coefficient of variation between the control and the critical dilution, Parameter TQP6C –18.01%.
6. Chronic WET Limit, DMR CODE 51714, - 78.0%
7. PMSD Biomass-29.96% (12.0 – 30.0%)-low precision, See Results and Discussion.

This report contains a total of 54 pages, including this page. The results contained within pertain only to the samples listed on the chain of custody documents in Appendix A. The information meets the standards set forth by ADEQ. The chemical data in this report is for monitoring purposes only and should not be reported on discharge monitoring reports.



## **Bio-Analytical Laboratories**

3240 Spurgin Road  
Post Office Box 527  
Doyline, LA 71023

(318) 745-2772  
1-800-259-1246  
Fax: (318) 745-2773

**THE RESULTS OF TWO CHRONIC  
DEFINITIVE TOXICITY TESTS  
FOR OUTFALL 001**

**AT**

**NASHVILLE PUBLIC WORKS  
Nashville, Arkansas**

**NPDES #AR0021776  
AFIN #31-00036**

**EPA Methods 1000.0 and 1002.0**

**Project X9028**

**Test Dates: November 28 – December 5, 2023**

**Report Date: December 8, 2023**

**Prepared for:**

Southern Petroleum Laboratories  
4720 Viking Drive, Ste A  
Bossier City, LA 71111

**Prepared by:**

Ginger Briggs  
Bio-Analytical Laboratories  
P.O. Box 527  
Doyline, LA 71023  
ADEQ #88-0630

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## 1.0 Introduction

Bio-Analytical Laboratories (BAL), Doyline, Louisiana conducted two chronic definitive toxicity tests for Outfall 001 at Nashville Public Works, Nashville, Arkansas. The test organisms used were the cladoceran, *Ceriodaphnia dubia*, and the fathead minnow, *Pimephales promelas*. The purpose of this study is to determine if appropriately dilute effluent samples adversely affect the survival, reproduction and/or growth of the test organisms. Toxicity is defined as a statistically significant difference at the 95 percent confidence level between the survival, reproduction and/or growth of the test organism in the critical dilution (the effluent concentration representative of the proportion of effluent in the receiving water during critical low flow or critical mixing conditions) compared to the survival, reproduction and/or growth of the test organism in the control. The test endpoint is the No-Observed-Effect-Concentration (NOEC), the highest effluent concentration that is not significantly different from the control.

## 2.0 Methods and Materials

### 2.1 Test Methods

All methods followed were according to the latest edition of “Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms” (EPA-821-R-02-013), “Standard Methods for The Examination of Water and Wastewater, 22<sup>nd</sup> Edition” (APHA 2012), and BAL’s standard operating procedure.

### 2.2 Test Organisms

The *Ceriodaphnia dubia* test organisms were cultured in-house at test temperature and dilution water hardness and were less than 24 hours old at test initiation. The neonates were released within the same 8-hour period. The fathead minnows were also raised in-house at test temperature and dilution water hardness and were less than 24 hours old at test initiation. Monthly chronic reference toxicant tests were conducted in order to document organism sensitivity and demonstration of capability.

### 2.3 Dilution Water

Moderately hard reconstituted water, made per method guidelines, was used as the dilution water and the control for the toxicity tests. Conductivity (SM 2510 B) and pH (SM 4500-H+ B) measurements, in umhos/cm and standard units, respectively, were checked on each lot of water prior to use to confirm water hardness.

## 2.4 Test Concentrations

The test concentrations used in the chronic toxicity tests (for a 3.5 MGD design flow) were 78.0, 59.0, 44.0, 33.0 and 25.0 percent effluent and a reconstituted water control. The critical dilution was 78.0 percent effluent. The *Ceriodaphnia* test was conducted using 10 replicates of one animal each for a total of 10 animals per concentration. The fathead minnow test was conducted using five replicates of eight animals each for a total of 40 animals per concentration.

## 2.5 Sample Collection

Three 24-hour composite samples of Outfall 001 were collected by Nashville Public Works personnel on November 27, 29 and December 1, 2023, at 0651, 0418 and 0437 hours, respectively. Upon collection and completion of each composite, the samples were packed in ice and delivered to the laboratory the day of collection by Southern Petroleum Laboratories (SPL) personnel. The sample temperature upon arrival of each sample was -0.3, 0.5 and 1.0<sup>o</sup> Celsius, respectively.

## 2.6 Sample Preparation

Upon arrival, the samples were logged in, given an identification number, and refrigerated unless needed. Prior to use, the samples were warmed to 25±1<sup>o</sup> Celsius. Total residual chlorine levels were measured in milligrams/Liter (mg/L) using a test strip and recorded if present. Total ammonia levels were measured in mg/L using a test strip. Dissolved oxygen (SM 4500-0 G) and pH (SM 4500-H+ B) measurements, in mg/L and standard units, respectively, were measured on the control and each concentration at test initiation, at test renewal and at test termination. Conductivity (SM 2510 B) measurements, in umhos/cm, were also taken at test initiation and at each renewal. Alkalinity (SM 2320 B) and hardness (SM 2340 C) levels were measured in mg/L as CaCO<sub>3</sub> on the control and the undiluted effluent samples.

## 2.7 Monitoring of the Tests

The cladoceran test was run in a Precision<sup>R</sup> dual-programmable, illuminated incubator at a temperature of 25±1<sup>o</sup> Celsius. The fathead minnow test was run in a circulating waterbath, using a Remcor<sup>R</sup> heated liquid circulator to keep a constant temperature of 25±1<sup>o</sup> Celsius. Data-loggers were used to monitor diurnal test temperature. Test temperatures were recorded at the beginning of the day, after test renewal and at the end of the day. Light cycles and intensities were recorded twice a month.

## 2.8 Data Analysis

*Ceriodaphnia dubia* survival data was analyzed using Fisher's Exact Test, an equality test comparing concentration data to control data. Reproduction data was analyzed using Steel's Many-One Rank Test, a nonparametric test comparing concentration data to control data. Fathead minnow survival and growth data was analyzed using Dunnett's Test, a parametric test. Other endpoints were obtained by approved EPA methods of analysis.

## 3.0 Results and Discussion

The results of the *Ceriodaphnia dubia* test can be found in Table 1. Ninety percent survival occurred in the control and in the 78.0 percent critical dilution after seven days of exposure. The average number of neonates per female after three broods in the control and in the critical dilution was 19.3 and 21.0, respectively. The No-Observed-Effect-Concentration (NOEC) for survival and reproduction in this test was 78.0 percent effluent (p=.05).

**Table 1: Results of the Chronic Definitive *Ceriodaphnia dubia* Test**

Percent Effluent	Percent Survival	Sig.*	Mean # Neonates-Surviving	Mean # Neonates -Total	Sig.*
Control	90.0		21.0	19.3	
25.0	100.0		20.5	20.5	
33.0	100.0		18.1	18.1	
44.0	80.0		21.5	172	
59.0	100.0		19.8	19.8	
78.0	100.0		21.0	21.0	

\*significant when compared to the control (p=.05). Test validity based on mean number of neonates per surviving female. NOEC value based on total mean number of neonates.

The fathead minnow test results can be found in Table 2. After seven days of exposure, 82.5 percent survival occurred in the control and 85.0 percent survival occurred in the 78.0 percent critical dilution. The average weight gained in the control and in the 78.0 percent critical dilution was 0.553 and 0.513 milligram (mg), respectively. The NOEC for survival and growth (biomass) was 78.0 percent effluent (p=.05).

**Table 2: Results of the Chronic Definitive Fathead Minnow Test**

Percent Effluent	Percent Survival	Sig.*	Mean Dry Weight (mg)	Sig.*
Control	82.5		0.553	
25.0	70.0		0.413	
33.0	70.0		0.443	
44.0	95.0		0.720	
59.0	90.0		0.620	
78.0	85.0		0.513	

\*significant when compared to the control (p=.05).

The most recent reference toxicant tests showed the test organisms to be within the respective sensitivity range. The graphs of the results of the chronic reference toxicant tests can be found in Appendix D- Quality Assurance Charts.

#### 4.0 Conclusions

The three composite samples of Outfall 001 collected on November 27, 29 and December 1, 2023, from Nashville Public Works, Nashville, Arkansas, were not found to be lethally toxic to the *Ceriodaphnia dubia* test organisms nor the *Pimephales promelas* test organisms in the 78.0 percent critical dilution after seven days of exposure (p=.05). Sublethal effects (i.e., lack of growth or reproduction) were not noted in the 78.0 percent critical dilution in either test (p=.05).

## 5.0 References

- EPA, 2002. Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms. Fourth Edition. EPA-821-R-02-013, Office of Water.
- EPA, 2000. Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination System. EPA-833-R-00-003, Office of Wastewater Management.
- EPA, 2000. Method Guidance and Recommendations for Whole Effluent (WET) Testing. EPA-821-B-00-04, Office of Water
- APHA, 2012. Standard Methods for The Examination of Water and Wastewater. 22<sup>nd</sup> Edition.



**APPENDIX A**  
**CHAIN-OF-CUSTODY DOCUMENTS**

X 9028 / C259113

color; clear  
 odor; None  
 Temp; -0.3  
 H; 29  
 Tech; PM

Bio-Analytical Laboratories  
 3240 Spurgeon Rd  
 Doyline, LA 71023  
 (318)7452772

Date 11-27-23 Time 1100

Sampler Printed Name KEVIN FUNDARBURG

Sampler Signature CLIENT

Sampler Affiliation NASH

Please analyze the following samples:

Sample Identification	Client Code	Date	Time	Testing Required- FCSC
Bio Monitoring	NASH	11-26-23	0951	Chronic ceriodaphnia dubia
		11-27-23	0651	Chronic pimephales promelas

Requires RUSH status (24 hr Turn Around Time)  Requires E-mail as soon as completed  
 (skeeter@ana-lab.com)

If you have any questions about the testing, contact Skeeter Ludewig at (903)984-0551.

Date	Time	Relinquished		Received	
11-27-23	1637	Printed Name <u>Mark Hensch</u>	Affiliation <u>SPL</u>	Printed Name <u>ERIN G. BRIGGS</u>	Affiliation <u>BAC</u>
		Signature <u>[Signature]</u>		Signature <u>[Signature]</u>	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	

X9028/C25957

Bio-Analytical Laboratories  
 3240 Spurgeon Rd  
 Doyline, LA 71023  
 (318)7452772

Odor: None  
 Color: Clear  
 Temperature upon arrival: 0.5°C  
 Thermometer #: #29  
 Tech: PM  
 Date: 11/29/23

Date 11.29.23 Time 1045  
 Sampler Printed Name KEVIN FUNDALBURY  
 Sampler Signature CLIENT  
 Sampler Affiliation NASH

Please analyze the following samples:

Sample Identification	Client Code	Date	Time	Testing Required- FCSC
Bio Monitoring	NASH	11-28-23	0750	Chronic ceriodaphnia dubia
		11-29-23	0418	Chronic pimephales promelas
		H 11-29-23		

Requires RUSH status (24 hr Turn Around Time)  Requires E-mail as soon as completed  
 (skeeter@ana-lab.com)

If you have any questions about the testing, contact Skeeter Ludewig at (903)984-0551.

Date	Time	Relinquished		Received	
11-29-23	1100	Printed Name MARK HIRSCH	Affiliation SPL	Printed Name Skeeter Ludewig	Affiliation AEC
		Signature Mark Hirsch		Signature	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	

X9028 / C2664

1.0<sup>cc</sup> #29  
 clear  
 no odor

SPW 12/01/23

Bio-Analytical Laboratories  
 3240 Spurgeon Rd  
 Doyline, LA 71023  
 (318)7452772

Date 12.1.23 Time 0427

Sampler Printed Name KEVIN FUNDARBOUN

Sampler Signature CLIENT

Sampler Affiliation NASH

Please analyze the following samples:

Sample Identification	Client Code	Date	Time	Testing Required- FCSC
Bio Monitoring	NASH	11.30.23	0835	Chronic ceriodaphnia dubia
		12.1.23	0437	Chronic pimephales promelas

Requires RUSH status (24 hr Turn Around Time)  Requires E-mail as soon as completed  
 (skeeter@ana-lab.com)

If you have any questions about the testing, contact Skeeter Ludewig at (903)984-0551.

Date	Time	Relinquished		Received	
12.1.23	13510	Printed Name <u>Mark Hinsat</u>	Affiliation <u>SPW</u>	Printed Name <u>Kevin F. Fundarboun</u>	Affiliation <u>ANA</u>
		Signature <u>[Signature]</u>		Signature <u>[Signature]</u>	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	
		Printed Name	Affiliation	Printed Name	Affiliation
		Signature		Signature	

**APPENDIX B  
RAW DATA SHEETS**

BIO-ANALYTICAL LABORATORIES CERIODAPHNIA DUBIA SURVIVAL AND REPRODUCTION TEST

Project# X9028 Date start: 11/28/23 Date end: 12/15/23

Client/Contact: NASH/Nashville Public Works

Address: 426 North Main, Nashville, AR 71852

NPDES#: AR0021776/ AFIN 31-00036

Sample Description: 001 Dilution Water: MH RECONSTITUTED

Adults isolated: Date 11/28/23 Time: 0645

Neonates collected: Date 11/28/23 Time: 1245 Board: 205

Dissolved Oxygen Meter#: 2

pH Meter#: 3 Conductivity Meter#: 9

ORP Meter#: \_\_\_\_\_ Salinity Meter#: \_\_\_\_\_

Effluent Initial D.O. (mg/L & %)/Tech	Aerate?/Minutes /Final D.O. (mg/L & %)/Tech	Receiving Water Initial D.O. (mg/L & %)/Tech	Aerate?/Minutes /Final D.O. (mg/L & %)/Tech
0. <u>9.2</u> / <u>111.6</u> % / <u>3</u> / <u>1mv</u>	<u>Y</u> / <u>112</u> / <u>99.2</u> % / <u>1mv</u>	0. _____	0. _____
1. <u>10.4</u> / <u>126.4</u> % / <u>PM</u>	1. <u>Y</u> / <u>6</u> / <u>8.5</u> / <u>100.5</u> % / <u>PM</u>	1. _____	1. _____
2. <u>10.6</u> / <u>133.5</u> % / <u>PM</u>	2. <u>Y</u> / <u>6</u> / <u>8.7</u> / <u>100.7</u> % / <u>PM</u>	2. _____	2. _____
3. <u>10.4</u> / <u>125.5</u> % / <u>PM</u>	3. <u>Y</u> / <u>6</u> / <u>8.5</u> / <u>100.2</u> % / <u>PM</u>	3. _____	3. _____
4. <u>9.2</u> / <u>108.6</u> % / <u>1mv</u>	4. <u>Y</u> / <u>110</u> / <u>83</u> / <u>95.3</u> % / <u>1mv</u>	4. _____	4. _____
5. <u>9.5</u> / <u>115.3</u> % / <u>1mv</u>	5. <u>Y</u> / <u>112</u> / <u>86</u> / <u>98.3</u> % / <u>1mv</u>	5. _____	5. _____
6. <u>10.6</u> / <u>134.2</u> % / <u>PM</u>	6. <u>Y</u> / <u>6</u> / <u>8.6</u> / <u>100.5</u> % / <u>PM</u>	6. _____	6. _____
7. _____	7. _____	7. _____	7. _____

Total Residual Chlorine (mg/L)/Tech

Dechlorinated? Amount?/Tech

Ammonia (NH3) (mg/L)/Tech

BAL Sample # Date in use

1. 40.5 / 1mv  
 2. 40.5 / PM  
 3. 40.5 / 1mv

1. NO / 1mv  
 2. NO / PM  
 3. NO / 1mv

1. 40.5 / 1mv  
 2. 40.5 / PM  
 3. 40.5 / 20.5 / 1mv

1. C25943 11/28/23  
 2. C25948 11/30/23  
 3. C25964 12/2/23

PM 11/30/23

Comments:



Project# X9028 Client Nashville

Sample ID 001

Test started: Date 11/28/2013 Time 12:10

Test ended: Date 12/5/2013 Time 15:57

Date/Tech: Day 0 11/28/2013 1 11/29/2013 2 11/30/2013 3 12/1/2013 4 12/2/2013 5 12/3/2013 6 12/4/2013 7 12/5/2013 8

Time: Day 0 12:10 1 15:10 2 13:30 3 14:45 4 11:32 5 11:53 6 17:00 7 15:57 8

Temp. (°C): Day 0 24.5 1 24.0 2 24.3 3 24.0 4 24.3 5 23.2 6 24.1 7 23.6 8

Conc	Day	1	2	3	4	5	6	7	8	9	10	Number of Live	
0	1	0										10	
	2	0										10	
	3	0										10	
	4	112	111	112	114	112	113	112	113	112	114	10	
	5	0			X							10	
	6	214	215	214		217	215	214	216	215	216	10	
	7	3115	3112	3114		3115	3112	3115	3114	3114	3111	9	
	8												
25.0	1	0										10	
	2	0										10	
	3	0										10	
	4	114	112	114	112	111	112	111	112	114	115	10	
	5	0										10	
	6	0	214	215	214	213	216	214	214	215	215	10	
	7	213	3115	3112	3113	3113	3116	3114	3113	3115	3115	3112	10
	8												
33.0	1	0										10	
	2	0										10	
	3	0										10	
	4	0	0	112	111	115	112	0	112	111	112	10	
	5	0										10	
	6	113	115	214	213	216	214	114	215	213	216	10	
	7	218	214	3111	3112	3113	3114	219	3116	3116	3110	10	
	8												
44.0	1	0										10	
	2	0										10	
	3	0										10	
	4	113	112	114	115	114	115	112	0	0	0	10	
	5	0										10	
	6	216	215	X	2110	216	217	215	113	114	X	10	
	7	3116	3114		3113	3112	3115	3115	218	217		8	
	8												
59.0	1	0										10	
	2	0										10	
	3	0										10	
	4	112	115	112	113	112	114	115	112	0	112	10	
	5	0										10	
	6	215	216	214	216	215	217	0	0	214	214	10	
	7	3112	3113	3114	3115	3114	3113	2112	2110	3113	3114	10	
	8												
78.0	1	0										10	
	2	0										10	
	3	0										10	
	4	111	115	112	114	115	112	114	115	112	114	10	
	5	0										10	
	6	216	213	216	216	0	214	215	0	0	216	10	
	7	3116	3114	3115	3112	217	3116	3115	2118	2110	3115	10	
	8												

Key: X=dead adult, X<sup>n</sup>=adult had n neonates before death, M=male.

B/N = Brood count/#neonates



BIO-ANALYTICAL LABORATORIES CHRONIC WATER QUALITY DATA (CHR CHEM Rev. 4.0)

Project# X9028 Client Nashville

Organism C. dub.ig

Date	Day 0 11/28/23 5562	Day 1 11/29/23	Day 2 11/30/23	Day 3 12/01/23	Day 4 12/2/23	Day 5 12/3/23	Day 6 12/4/23	Day 7 12/5/23	Day 8
Concentration:	0 MH								
Temperature (°C)		24.4	23.9	24.1	23.4	22.4	24.0	23.2	
pH	24.1	21.9	21.6	24.7	22.4	21.6	19.7		
DO (mg/L)	7.8	7.6	7.6	7.5	7.9	7.7	7.9		
Cond (umhos/cm)	298	315	324	330	323	334	350		
Concentration:	25.0%								
Temperature (°C)		24.4	23.4	23.9	23.4	22.5	24.1	23.2	
pH	23.1	22.3	23.4	24.7	20.9	19.5	20.4		
DO (mg/L)	7.7	8.1	7.3	7.3	7.5	7.1	7.4	7.2	
Cond (umhos/cm)	7.1	8.5	8.4	7.6	7.1	7.4	8.9	8.3	
Concentration:	33.0%								
Temperature (°C)		24.4	23.4	24.1	23.4	22.5	24.3	23.2	
pH	23.3	22.5	23.6	24.6	21.4	20.2	20.6		
DO (mg/L)	7.9	8.2	7.7	7.4	7.8	7.8	8.0	8.0	
Cond (umhos/cm)	7.8	8.6	8.3	7.9	7.9	7.8	8.2	8.2	
Prerenewal Tech Initials/Time		1510 PM	1330	1445	1134 MV	1154 MV	1705 PM	1557 MV	
Postrenewal Tech Initials/Time	1046 MV	1145 PM	1150 PM	1030	1000 MV	1048 MV	1055 PM		

Control Alkalinity (mg/L as CaCO<sub>3</sub>)

Control Hardness (mg/L as CaCO<sub>3</sub>)

ID# 5562 Result 4.0 Date Tested 11/30/23 ID# 5562 Result 88.0 Date Tested 11/30/23  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_ ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_ ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

Sample Alkalinity (mg/L as CaCO<sub>3</sub>)

Sample Hardness (mg/L as CaCO<sub>3</sub>)

ID# 25943 Result 164 Date Tested 11/30/23 ID# 25943 Result 256 Date Tested 11/30/23  
 ID# 25957 Result 160 Date Tested 11/30/23 ID# 25957 Result 249 Date Tested 11/30/23  
 ID# 25964 Result 160 Date Tested 12/7/23 ID# 25964 Result 220 Date Tested 12/7/23



BIO-ANALYTICAL LABORATORIES CHRONIC WATER QUALITY DATA (CHR CHEM Rev. 4.0)

Project# X9028 Client Nashville

Organism C. dubia

Date	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8
Concentration: <u>44.0%</u>									
Temperature (°C)		<u>24.4</u>	<u>23.7</u>	<u>24.1</u>	<u>23.5</u>	<u>22.7</u>	<u>24.3</u>	<u>23.1</u>	
	<u>23.5</u>	<u>22.8</u>	<u>23.6</u>	<u>24.7</u>	<u>21.5</u>	<u>20.7</u>	<u>22.9</u>		
pH		<u>8.2</u>	<u>8.0</u>	<u>7.6</u>	<u>7.2</u>	<u>7.1</u>	<u>8.0</u>	<u>7.0</u>	
	<u>7.0</u>	<u>7.9</u>	<u>7.7</u>	<u>7.7</u>	<u>7.8</u>	<u>7.8</u>	<u>8.0</u>		
DO (mg/l)		<u>8.6</u>	<u>7.9</u>	<u>7.5</u>	<u>7.1</u>	<u>7.2</u>	<u>8.9</u>	<u>7.8</u>	
	<u>8.3</u>	<u>8.5</u>	<u>8.4</u>	<u>7.9</u>	<u>7.9</u>	<u>7.5</u>	<u>8.2</u>		
Cond (umhos/cm)		<u>412</u>	<u>425</u>	<u>431</u>	<u>416</u>	<u>419</u>	<u>421</u>		
	<u>379</u>								
Concentration: <u>59.0%</u>									
Temperature (°C)		<u>24.7</u>	<u>24.1</u>	<u>23.9</u>	<u>23.2</u>	<u>22.5</u>	<u>24.4</u>	<u>23.1</u>	
	<u>23.7</u>	<u>23.1</u>	<u>23.6</u>	<u>24.7</u>	<u>21.7</u>	<u>20.7</u>	<u>22.3</u>		
pH		<u>8.3</u>	<u>7.9</u>	<u>7.4</u>	<u>7.1</u>	<u>7.1</u>	<u>8.2</u>	<u>7.1</u>	
	<u>7.0</u>	<u>7.9</u>	<u>7.7</u>	<u>7.7</u>	<u>7.8</u>	<u>7.8</u>	<u>8.0</u>		
DO (mg/l)		<u>8.2</u>	<u>7.9</u>	<u>7.6</u>	<u>7.5</u>	<u>7.6</u>	<u>9.1</u>	<u>7.3</u>	
	<u>7.6</u>	<u>8.5</u>	<u>8.4</u>	<u>8.2</u>	<u>8.1</u>	<u>7.5</u>	<u>8.2</u>		
Cond (umhos/cm)		<u>444</u>	<u>459</u>	<u>471</u>	<u>444</u>	<u>453</u>	<u>454</u>		
	<u>414</u>								
Concentration: <u>78.0%</u>									
Temperature (°C)		<u>24.6</u>	<u>23.9</u>	<u>23.7</u>	<u>23.3</u>	<u>22.5</u>	<u>24.4</u>	<u>23.2</u>	
	<u>23.8</u>	<u>23.6</u>	<u>23.9</u>	<u>24.8</u>	<u>21.9</u>	<u>21.2</u>	<u>22.6</u>		
pH		<u>8.3</u>	<u>7.9</u>	<u>7.6</u>	<u>7.2</u>	<u>7.1</u>	<u>8.3</u>	<u>7.2</u>	
	<u>7.1</u>	<u>8.0</u>	<u>7.9</u>	<u>7.8</u>	<u>7.8</u>	<u>7.9</u>	<u>8.0</u>		
DO (mg/l)		<u>8.2</u>	<u>7.9</u>	<u>7.5</u>	<u>7.4</u>	<u>7.6</u>	<u>9.1</u>	<u>7.6</u>	
	<u>7.8</u>	<u>8.4</u>	<u>7.9</u>	<u>8.0</u>	<u>7.9</u>	<u>7.3</u>	<u>8.2</u>		
Cond (umhos/cm)		<u>487</u>	<u>502</u>	<u>515</u>	<u>482</u>	<u>488</u>	<u>450</u>		
	<u>471</u>								
Prerenewal Tech Initials/Time		<u>1510 PM</u>	<u>EDU 1330</u>	<u>EDU 1445</u>	<u>1132 MV</u>	<u>1154 MV</u>	<u>1715 PM</u>	<u>1557 MV</u>	
Postrenewal Tech Initials/Time	<u>1040 MV</u>	<u>1145 PM</u>	<u>1150 PM</u>	<u>EDU 1030</u>	<u>1000 MV</u>	<u>1048 MV</u>	<u>1055 PM</u>		

Control Alkalinity (mg/L as CaCO<sub>3</sub>)

Control Hardness (mg/L as CaCO<sub>3</sub>)

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

Sample Alkalinity (mg/L as CaCO<sub>3</sub>)

Sample Hardness (mg/L as CaCO<sub>3</sub>)

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

**CETIS Test Data Worksheet**

Report Date: 27 Nov-23 10:17 (p 1 of 2)  
 Test Code/ID: 61A912C9 / 16-3847-0345

**Ceriodaphnia 7-d Survival and Reproduction Test**

**Bio-Analytical Laboratories**

Start Date: 28 Nov-23 15:03 Species: Ceriodaphnia dubia Sample Code: 56025932  
 End Date: 05 Dec-23 17:05 Protocol: EPA/821/R-02-013 (2002) Sample Source: AR0021776  
 Sample Date: 27 Nov-23 09:57 Material: POTW Effluent Sample Station: 001

Conc-%	Code	Rep	Pos	# Exposed	1d Surv	2d Surv	3d Surv	4d Surv	5d Surv	6d Surv	7d Surv	8d Surv	2d Neo	3d Neo	4d Neo	5d Neo	6d Neo	7d Neo	8d Neo	Male	Notes	
59		10	1																			
59		4	2																			
78		8	3																			
25		3	4																			
59		1	5																			
25		7	6																			
44		6	7																			
0	D	2	8																			
33		4	9																			
78		9	10																			
0	D	7	11																			
25		10	12																			
78		5	13																			
25		1	14																			
44		7	15																			
59		6	16																			
25		2	17																			
33		6	18																			
25		4	19																			
25		5	20																			
59		5	21																			
59		9	22																			
0	D	9	23																			
59		3	24																			
33		5	25																			
78		2	26																			
0	D	6	27																			
0	D	10	28																			
44		1	29																			
33		3	30																			
25		6	31																			
59		8	32																			
33		1	33																			
78		1	34																			
33		9	35																			
0	D	5	36																			
0	D	8	37																			
78		6	38																			
78		7	39																			
78		10	40																			
44		2	41																			

**CETIS Test Data Worksheet**

Report Date: 27 Nov-23 10:17 (p 2 of 2)  
 Test Code/ID: 61A912C9 / 16-3847-0345

Conc-%	Code	Rep	Pos	# Exposed	1d Surv	2d Surv	3d Surv	4d Surv	5d Surv	6d Surv	7d Surv	8d Surv	2d Neo	3d Neo	4d Neo	5d Neo	6d Neo	7d Neo	8d Neo	Male	Notes	
44		3	42																			
44		9	43																			
33		8	44																			
33		10	45																			
25		9	46																			
0	D	3	47																			
0	D	1	48																			
44		4	49																			
33		2	50																			
44		8	51																			
0	D	4	52																			
44		10	53																			
25		8	54																			
59		7	55																			
44		5	56																			
78		4	57																			
33		7	58																			
78		3	59																			
59		2	60																			

Analyst: *EPV* 11/27/23  
 QA: *EB* 12/18/23

BIO-ANALYTICAL LABORATORIES  
GERIODAPHNIA DUBIA CHRONIC TEST  
PARENTAL BLOCK ASSIGNMENT RANDOM NUMBER TEMPLATE #3  
FOR 6 CONCENTRATION TEST

MV  
11/28/23

Set #1

2, 4, 1, 6, 3, 5 Parent# 3c

Set #2

3, 5, 1, 2, 4, 6 Parent# ST

Set #3

3, 6, 2, 1, 5, 4 Parent# LA

Set #4

2, 4, 6, 1, 5, 3 Parent# UH

Set #5

6, 2, 5, 3, 1, 4 Parent# UD

Set #6

1, 3, 6, 5, 2, 4 Parent# UA

Set #7

1, 2, 5, 6, 3, 4 Parent# 3A

Set #8

1, 6, 5, 3, 2, 4 Parent# 1C

Set #9

1, 6, 2, 4, 5, 3 Parent# 3I

Set #10

2, 1, 5, 3, 6, 4 Parent# 4B

BIO-ANALYTICAL LABORATORIES  
PIMEPHALES PROMELAS SURVIVAL AND GROWTH DATA SHEET

Project# X9028 Date started: 11/28/23 Date ended 12/5/23

Client/Contact: NASH/Nashville Public Works

Address: 426 North Main, Nashville, AR 71852

NPDES# AR0021776/AFIN 31-00036

Sample Description: 001 Dilution Water: MH Reconstituted  
Test organism age: 24 hrs Vendor/ID# BAL 112823

Day	Feeding Times		
	AM	NOON	PM
0			
1	<u>EW/0915/0.10ml</u>	<u>PM/1135/0.10ml</u>	<u>PM/1420/0.2ml</u>
2	<u>PM/0940/0.10ml</u>	<u>EW/1245/0.10ml</u>	<u>PM/1420/0.10ml</u>
3	<u>EW/0915/0.10ml</u>	<u>EW/1245/0.10ml</u>	<u>MV/1720/0.10ml</u>
4	<u>MV/1005/0.20ml</u>	<u>EW/1245/0.10ml</u>	<u>EW/1600/0.10ml</u>
5	<u>MV/1000/0.20ml</u>		<u>MV/1305/0.20ml</u>
6	<u>PM/1090/0.10ml</u>	<u>PM/1105/0.10ml</u>	<u>MV/1237/0.20ml</u> <u>MV/1826/0.10ml</u>

Dissolved Oxygen Meter #: 2  
pH Meter#: 3 Conductivity Meter#: 9  
ORP Meter#: --- Salinity Meter #: ---

Effluent Initial DO (mg/L & %)/Tech	Aerate?/Minutes /Final DO (mg/L & %)/Tech	Receiving Water Initial DO (mg/L & %)/Tech	Aerate?/Minutes /Final DO (mg/L & %)/Tech
0. <u>9.2/116.3%/mv</u>	<u>Y/12/99.2%/mv</u>	0. _____	0. _____
1. <u>10.4/126.4%/PM</u>	1. <u>Y/6/85.5/100.3%/PM</u>	1. _____	1. _____
2. <u>10.6/133.5%/PM</u>	2. <u>Y/6/8.7/99.7%/PM</u>	2. _____	2. _____
3. <u>10.4/128.5%/SPW</u>	3. <u>Y/6/8.5/100.2%/SPW</u>	3. _____	3. _____
4. <u>9.2/102.6%/mv</u>	4. <u>Y/10/95.3%/mv</u>	4. _____	4. _____
5. <u>9.5/115.3%/mv</u>	5. <u>Y/12/98.3%/mv</u>	5. _____	5. _____
6. <u>10.6/134.2%/PM</u>	6. <u>Y/6/8.4/100.3%/PM</u>	6. _____	6. _____

Total Residual Chlorine (mg/L)/Tech	Dechlorinated? Amount?/Tech	Ammonia (NH3) (mg/L)/Tech	BAL Sample # Date in use
1. <u>20.5/mv</u>	1. <u>NO/mv</u>	1. <u>20.5/mv</u>	1. <u>C25943</u>
2. <u>20.5/PM</u>	2. <u>NO/PM</u>	2. <u>20.5/PM</u>	2. <u>C25957</u>
3. <u>20.5/mv</u>	3. <u>NO/mv</u>	3. <u>20.5/mv</u>	3. <u>C25764</u>

11/28/23

Comments:

9/12/23



BIO-ANALYTICAL LABORATORIES 7-DAY CHRONIC MINNOW SURVIVAL DATA- EPA 1000, OECD 210

Project# X9028 Test started: Date 11/28/23 Time 1345

Client Nashville Sample ID 001 Test ended: Date 12/13/23 Time

Date/Tech: Day 0 11/28/23 1 11/29/23 2 11/30/23 3 12/01/23 4 12/02/23 5 12/03/23 6 12/04/23 7 12/05/23

Time: Day 0 1345 1 1430 2 1245 3 1113 4 1030 5 1137 6 1305 7 1340

Temp (°C) Day 0 25.5 1 25.2 2 24.8 3 25.4 4 23.0 5 22.8 6 22.0 7 22.6

Conc. <sup>g/l</sup>	Rep.	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
0 MH	1	8	8	8	8	8	8	8	8
	2	8	8	8	8	8	7	7	7
	3	8	8	8	8	8	8	7	7
	4	8	8	8	8	8	8	8	7
	5	8	8	8	8	8	7	5	4
25.0	1	8	8	8	8	8	8	8	6
	2	8	8	8	8	8	8	6	6
	3	8	8	8	8	8	8	7	7
	4	8	8	8	8	8	8	4	3
	5	8	8	8	8	8	8	1	6
33.0	1	8	8	7	7	7	7	7	5
	2	8	8	8	8	8	8	5	4
	3	8	8	8	8	8	5	4	4
	4	8	8	8	8	8	8	8	8
	5	8	8	8	8	8	8	7	7
44.0	1	8	8	8	8	8	8	8	8
	2	8	8	8	8	8	8	8	8
	3	8	8	8	8	8	8	8	8
	4	8	8	8	8	8	8	8	7
	5	8	8	8	8	8	8	7	7
59.0	1	8	8	8	8	8	8	8	8
	2	8	8	8	8	8	8	8	8
	3	8	8	8	8	8	7	7	6
	4	8	8	8	8	8	8	8	7
	5	8	8	8	8	8	8	8	7
78.0	1	8	8	8	8	8	8	8	8
	2	8	8	8	8	8	8	8	8
	3	8	8	8	8	8	7	7	6
	4	8	8	8	8	8	8	8	6
	5	8	8	8	8	8	8	8	8

OPN 12/4/23

Project#/Client: X9028 Nashville Temp Start (°C): 90.1 Tech: PM Date: 12/13 Time: 1340  
 Temp End (°C): 11.0 Tech: EB Date: 12/13 Time: 0745

Conc. %	Replicate/ Pan number	Wt. of pan(g) Date <u>11/30/23</u> weighed: Tech: <u>MV</u>	Wt. of pan + larvae(g) Date <u>12/1/23</u> weighed: Tech: <u>EB</u>	Total wt. of larvae (g)	Original # of larvae at test initiation	Mean Dry wt. of larvae (mg)	Mean Dry wt. - surviving larvae (mg) Control Only*
0	1	31	1.0888	1.0929 <sup>EB</sup>	12/1/23		
	2	32	1.0678	1.0720			
	3	33	1.0376	1.0427			
	4	34	1.0122	1.0672			
	5	35	1.0587	1.0628			
25	1	36	1.0697	1.0731			
	2	37	1.0643	1.0682			
	3	38	1.0786	1.0826			
	4	39	1.0796	1.0810			
	5	40	1.0913	1.0951			
33	1	41	1.0697	1.0733			
	2	42	1.0842	1.0866			
	3	43	1.0688	1.0709			
	4	44	1.0696	1.0745			
	5	45	1.0670	1.0717			
44	1	46	1.0223	1.0892			
	2	47	1.0701	1.0754			
	3	48	1.0506	1.0572			
	4	49	1.0837	1.0884			
	5	50	1.0849	1.0902			
59	1	51	1.0676	1.0721			
	2	52	1.0599	1.0647			
	3	53	1.0440	1.0498			
	4	54	1.0770	1.0821			
	5	55	1.0478	1.0524			
78	1	56	1.0577	1.0630			
	2	57	1.0571	1.0613			
	3	58	1.0664	1.0699			
	4	59	1.0565	1.0605			
	5	60	1.0627	1.0662			

\* Test acceptance of control weight based on surviving larvae at end of test.

Calculated by: LETIS Calculations checked by: EB 12/1/23



BIO-ANALYTICAL LABORATORIES CHRONIC WATER QUALITY DATA (CHR CHEM Rev.4.0)

Project# X9028 Client Nashville Organism P. promelas

Date	Day 0 11/28/23 5558	Day 1 11/29/23	Day 2 11/30/23	Day 3 12/01/23 5563	Day 4 12/01/23	Day 5 12/01/23	Day 6 12/01/23	Day 7 12/01/23	Day 8
Concentration:	0 M H								
Temperature (°C)	23.3	24.9	24.0	23.9	23.1	22.5	21.7	22.8	
pH	7.7	6.9	7.0	7.4	6.5	7.0	6.7	6.7	
DO (mg/l)	7.6	6.7	6.9	7.2	6.8	7.8	7.0	7.0	
Cond (umhos/cm)	305	8.4	8.4	7.9	7.6	8.5	8.4		
Concentration:	25.0%								
Temperature (°C)	23.9	25.0	23.8	24.5	22.6	21.7	21.7	22.6	
pH	7.9	7.2	7.3	7.2	6.8	6.5	7.0	6.9	
DO (mg/l)	7.5	6.4	6.5	7.4	6.2	7.2	7.2	6.6	
Cond (umhos/cm)	363	8.3	8.5	7.9	7.6	8.3	8.4		
Concentration:	33.0%								
Temperature (°C)	24.1	24.9	24.1	24.8	22.6	21.8	21.7	22.9	
pH	8.1	7.3	7.3	7.3	6.9	6.7	7.1	7.2	
DO (mg/l)	7.5	6.4	6.7	7.4	7.0	7.3	6.9	6.5	
Cond (umhos/cm)	374	8.3	8.5	8.2	8.4	8.2	8.4		
Prerenewal Tech Initials/Time		1435 PM	1245 PM	EDV 1113	1030 MV	1137 MV	1305 PM	1345 PM	
Postrenewal Tech Initials/Time	1050 MV	1130 PM	1150 PM	800 2030	0753 MV	1044 MV	1100 PM		

Control Alkalinity (mg/L as CaCO<sub>3</sub>)

Control Hardness (mg/L as CaCO<sub>3</sub>)

ID# 5558 Result 60 Date Tested 11/30/23 ID# 5558 Result 108 Date Tested 11/30/23  
 ID# 5563 Result 50.0 Date Tested 12/1/23 ID# 5563 Result 92.0 Date Tested 12/1/23  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_ ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

Sample Alkalinity (mg/L as CaCO<sub>3</sub>)

Sample Hardness (mg/L as CaCO<sub>3</sub>)

ID# 25943 Result 164 Date Tested 11/30/23 ID# 25943 Result 756 Date Tested 11/30/23  
 ID# 25955 Result 160 Date Tested 11/30/23 ID# 25957 Result 248 Date Tested 11/30/23  
 ID# 25964 Result 160 Date Tested 12/1/23 ID# 25964 Result 200 Date Tested 12/1/23



BIO-ANALYTICAL LABORATORIES CHRONIC WATER QUALITY DATA (CHR CHEM Rev.4.0)

Project# X9028 Client Nashville

Organism P. Promelas

Date	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8
Concentration: <u>44.0%</u>									
Temperature (°C)		<u>24.8</u>	<u>23.9</u>	<u>25.1</u>	<u>22.4</u>	<u>21.7</u>	<u>21.6</u>	<u>22.8</u>	
	<u>24.3</u>	<u>24.2</u>	<u>23.9</u>	<u>24.9</u>	<u>23.3</u>	<u>22.7</u>	<u>22.5</u>		
pH		<u>7.5</u>	<u>7.3</u>	<u>7.2</u>	<u>7.1</u>	<u>7.1</u>	<u>7.2</u>	<u>7.2</u>	
	<u>8.2</u>	<u>7.9</u>	<u>7.7</u>	<u>7.9</u>	<u>7.8</u>	<u>7.8</u>	<u>8.0</u>		
DO (mg/l)		<u>6.4</u>	<u>7.0</u>	<u>7.6</u>	<u>5.9</u>	<u>7.7</u>	<u>6.6</u>	<u>6.2</u>	
	<u>7.4</u>	<u>8.3</u>	<u>8.4</u>	<u>8.3</u>	<u>7.6</u>	<u>8.2</u>	<u>8.25</u>		
Cond (umhos/cm)		<u>399</u>	<u>455</u>	<u>427</u>	<u>436</u>	<u>415</u>	<u>427</u>	<u>423</u>	
	<u>399</u>	<u>455</u>	<u>427</u>	<u>436</u>	<u>415</u>	<u>427</u>	<u>423</u>		
Concentration: <u>59.0%</u>									
Temperature (°C)		<u>24.8</u>	<u>24.1</u>	<u>25.4</u>	<u>22.4</u>	<u>21.8</u>	<u>21.6</u>	<u>22.8</u>	
	<u>24.3</u>	<u>24.2</u>	<u>24.0</u>	<u>24.9</u>	<u>23.2</u>	<u>22.7</u>	<u>22.6</u>		
pH		<u>7.6</u>	<u>7.3</u>	<u>7.2</u>	<u>7.2</u>	<u>7.1</u>	<u>7.3</u>	<u>7.3</u>	
	<u>8.2</u>	<u>7.9</u>	<u>7.8</u>	<u>8.0</u>	<u>7.8</u>	<u>7.8</u>	<u>7.9</u>		
DO (mg/l)		<u>6.8</u>	<u>7.1</u>	<u>7.1</u>	<u>5.8</u>	<u>6.9</u>	<u>6.8</u>	<u>6.4</u>	
	<u>7.6</u>	<u>8.3</u>	<u>8.4</u>	<u>8.2</u>	<u>7.1</u>	<u>8.3</u>	<u>8.2</u>		
Cond (umhos/cm)		<u>429</u>	<u>457</u>	<u>462</u>	<u>477</u>	<u>446</u>	<u>458</u>	<u>455</u>	
	<u>429</u>	<u>457</u>	<u>462</u>	<u>477</u>	<u>446</u>	<u>458</u>	<u>455</u>		
Concentration: <u>78.0%</u>									
Temperature (°C)		<u>24.7</u>	<u>24.1</u>	<u>24.7</u>	<u>22.6</u>	<u>21.7</u>	<u>21.5</u>	<u>22.8</u>	
	<u>24.4</u>	<u>24.5</u>	<u>24.3</u>	<u>25.1</u>	<u>22.9</u>	<u>22.7</u>	<u>22.8</u>		
pH		<u>7.8</u>	<u>7.3</u>	<u>7.4</u>	<u>7.3</u>	<u>7.2</u>	<u>7.4</u>	<u>7.5</u>	
	<u>8.2</u>	<u>8.0</u>	<u>7.8</u>	<u>7.9</u>	<u>7.8</u>	<u>7.9</u>	<u>8.0</u>		
DO (mg/l)		<u>6.7</u>	<u>6.9</u>	<u>7.4</u>	<u>5.8</u>	<u>6.5</u>	<u>6.6</u>	<u>6.3</u>	
	<u>7.6</u>	<u>8.3</u>	<u>8.4</u>	<u>7.9</u>	<u>7.9</u>	<u>8.4</u>	<u>9.0</u>		
Cond (umhos/cm)		<u>473</u>	<u>504</u>	<u>507</u>	<u>517</u>	<u>488</u>	<u>502</u>	<u>502</u>	
	<u>473</u>	<u>504</u>	<u>507</u>	<u>517</u>	<u>488</u>	<u>502</u>	<u>502</u>		
Prerenewal Tech Initials/Time		<u>1435</u>	<u>EM</u>	<u>EM</u>	<u>1030</u>	<u>1137</u>	<u>1305</u>	<u>1340</u>	
		<u>PM</u>	<u>1245</u>	<u>1113</u>	<u>MU</u>	<u>MU</u>	<u>PM</u>	<u>PM</u>	
Postrenewal Tech Initials/Time	<u>1050</u>	<u>1130</u>	<u>1150</u>	<u>EM</u>	<u>0753</u>	<u>1644</u>	<u>1100</u>		
	<u>MU</u>	<u>PM</u>	<u>PM</u>	<u>1030</u>	<u>MU</u>	<u>MU</u>	<u>PM</u>		

Control Alkalinity (mg/L as CaCO<sub>3</sub>)

Control Hardness (mg/L as CaCO<sub>3</sub>)

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

Sample Alkalinity (mg/L as CaCO<sub>3</sub>)

Sample Hardness (mg/L as CaCO<sub>3</sub>)

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_  
 ID# \_\_\_\_\_ Result \_\_\_\_\_ Date Tested \_\_\_\_\_

**CETIS Test Data Worksheet**

Report Date: 27 Nov-23 10:17 (p 1 of 1)  
 Test Code/ID: 3B0583DB / 09-9021-7179

**Fathead Minnow 7-d Larval Survival and Growth Test** Bio-Analytical Laboratories

Start Date: 28 Nov-23 15:00 Species: Pimephales promelas Sample Code: 606DCAD3  
 End Date: 05 Dec-23 15:50 Protocol: EPA/821/R-02-013 (2002) Sample Source: AR0021776  
 Sample Date: 27 Nov-23 09:00 Material: POTW Effluent Sample Station: 001

Conc-%	Code	Rep	Pos	# Exposed	1d Survival	2d Survival	3d Survival	4d Survival	5d Survival	6d Survival	7d Survival	Weight-mg Total	Weight-mg Tare	Pan Count	Notes
59		1	1												
• 33		4	2												
• 59		4	3												
25		3	4												
• 44		3	5												
• 78		5	6												
• 78		4	7												
• 25		4	8												
• 44		5	9												
• 44		2	10												
• 59		5	11												
• 59		3	12												
• 78		2	13												
• 59		2	14												
• 33		3	15												
• 33		2	16												
• 78		3	17												
• 33		1	18												
• 0	D	3	19												
• 25		1	20												
• 25		5	21												
• 0	D	4	22												
• 0	D	2	23												
• 25		2	24												
• 44		1	25												
• 78		1	26												
• 0	D	5	27												
• 0	D	1	28												
• 44		4	29												
33		5	30												

**APPENDIX C**  
**STATISTICAL ANALYSES**

# CETIS Analytical Report

Report Date: 07 Dec-23 13:22 (p 1 of 2)  
Test Code/ID: 61A912C9 / 16-3847-0345

**Ceriodaphnia 7-d Survival and Reproduction Test** Bio-Analytical Laboratories

Analysis ID: 02-8627-4694	Endpoint: 7d Survival Rate	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 13:21	Analysis: STP 2xK Contingency Tables	Status Level: 1
Edit Date: 07 Dec-23 13:10	MD5 Hash: B93B372A644C8A5EA663044E6D0505C8	Editor ID: 008-522-314-5
Batch ID: 01-0354-5901	Test Type: Reproduction-Survival (2-8d)	Analyst: Lab Tech
Start Date: 28 Nov-23 12:10	Protocol: EPA/821/R-02-013 (2002)	Diluent: Reconstituted Water
Ending Date: 05 Dec-23 15:57	Species: Ceriodaphnia dubia	Brine:
Test Length: 7d 4h	Taxon: Branchiopoda	Source: In-House Culture <span style="float: right;">Age: &lt;24</span>
Sample ID: 14-4299-4482	Code: X9028	Project: WET Quarterly Compliance Test (4Q)
Sample Date: 27 Nov-23 11:00	Material: POTW Effluent	Source: AR0021776 (AR0021776)
Receipt Date: 27 Nov-23 16:37	CAS (PC):	Station: 001
Sample Age: 25h (-0.3 °C)	Client: Nashville Public Works	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	Tox Units
Untransformed	C > T	78	>78	---	1.3

**Fisher Exact/Bonferroni-Holm Test**

Control	vs	Conc-%	Test Stat	P-Type	P-Value	Decision(α:5%)
Dilution Water		25	1.0000	Exact	1.0000	Non-Significant Effect
		33	1.0000	Exact	1.0000	Non-Significant Effect
		44	0.5000	Exact	1.0000	Non-Significant Effect
		59	1.0000	Exact	1.0000	Non-Significant Effect
		78	1.0000	Exact	1.0000	Non-Significant Effect

**7d Survival Rate Frequencies**

Conc-%	Code	NR	R	NR + R	Prop NR	Prop R	%Effect
0	D	9	1	10	0.9000	0.1000	0.00%
25		10	0	10	1.0000	0.0000	-11.11%
33		10	0	10	1.0000	0.0000	-11.11%
44		8	2	10	0.8000	0.2000	11.11%
59		10	0	10	1.0000	0.0000	-11.11%
78		10	0	10	1.0000	0.0000	-11.11%

**7d Survival Rate Summary**

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	10	0.9000	0.6738	1.0000	1.0000	0.0000	1.0000	0.1000	35.14%	0.00%
25		10	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-11.11%
33		10	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-11.11%
44		10	0.8000	0.4984	1.0000	1.0000	0.0000	1.0000	0.1333	52.70%	11.11%
59		10	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-11.11%
78		10	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-11.11%

**7d Survival Rate Detail**

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	D	1.0000	1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
25		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
33		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
44		1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000
59		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
78		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000

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12/18/23

# CETIS Analytical Report

Report Date: 07 Dec-23 13:22 (p 2 of 2)  
 Test Code/ID: 61A912C9 / 16-3847-0345

## Ceriodaphnia 7-d Survival and Reproduction Test

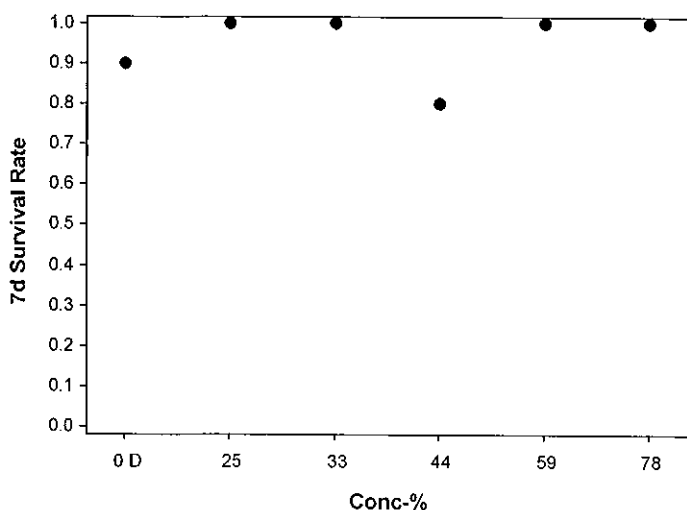
Bio-Analytical Laboratories

Analysis ID: 02-8627-4694	Endpoint: 7d Survival Rate	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 13:21	Analysis: STP 2xK Contingency Tables	Status Level: 1
Edit Date: 07 Dec-23 13:10	MD5 Hash: B93B372A644C8A5EA663044E6D0505C8	Editor ID: 008-522-314-5

### 7d Survival Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	D	1/1	1/1	1/1	0/1	1/1	1/1	1/1	1/1	1/1	1/1
25		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
33		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
44		1/1	1/1	0/1	1/1	1/1	1/1	1/1	1/1	1/1	0/1
59		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
78		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1

### Graphics



*EWB*  
*12/8/23*

**CETIS Analytical Report**

Report Date: 07 Dec-23 13:23 (p 1 of 2)  
Test Code/ID: 61A912C9 / 16-3847-0345

Ceriodaphnia 7-d Survival and Reproduction Test **SURVIVING** Bio-Analytical Laboratories

Analysis ID: 11-8707-2448 Endpoint: Reproduction CETIS Version: CETIS v2.1.5  
Analyzed: 07 Dec-23 13:22 Analysis: Nonparametric-Multiple Comparison Status Level: 1  
Edit Date: 07 Dec-23 13:10 MD5 Hash: 3FBDC94D6EFA5CE9C2EFEAB4D09CB59 Editor ID: 008-522-314-5

Batch ID: 01-0354-5901 Test Type: Reproduction-Survival (2-8d) Analyst: Lab Tech  
Start Date: 28 Nov-23 12:10 Protocol: EPA/821/R-02-013 (2002) Diluent: Reconstituted Water  
Ending Date: 05 Dec-23 15:57 Species: Ceriodaphnia dubia Brine:  
Test Length: 7d 4h Taxon: Branchiopoda Source: In-House Culture Age: <24

Sample ID: 14-4299-4482 Code: X9028 Project: WET Quarterly Compliance Test (4Q)  
Sample Date: 27 Nov-23 11:00 Material: POTW Effluent Source: AR0021776 (AR0021776)  
Receipt Date: 27 Nov-23 16:37 CAS (PC): Station: 001  
Sample Age: 25h (-0.3 °C) Client: Nashville Public Works

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	Tox Units	MSDu	RMSD
Untransformed	C > T	78	>78	---	1.3	4.695	22.35%

**Wilcoxon/Bonferroni Adj Test**

Control	vs	Conc-%	df	Test Stat	Critical	Ties	P-Type	P-Value	Decision(α:5%)
Dilution Water		25	17	94.5	---	4	Exact	1.0000	Non-Significant Effect
		33	17	76.5	---	4	Exact	0.1338	Non-Significant Effect
		44	15	83.5	---	2	Exact	1.0000	Non-Significant Effect
		59	17	90.5	---	3	Exact	1.0000	Non-Significant Effect
		78	17	116	---	2	Exact	1.0000	Non-Significant Effect

**ANOVA Table**

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	72.0526	14.4105	5	0.7962	0.5575	Non-Significant Effect
Error	923	18.098	51			
Total	995.053		56			

**ANOVA Assumptions Tests**

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test	19.57	15.09	0.0015	Unequal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9327	0.9434	0.0035	Non-Normal Distribution

**Reproduction Summary**

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	9	21	19.67	22.33	21	18	24	0.5774	8.25%	0.00%
25		10	20.5	19.06	21.94	20.5	18	24	0.6368	9.82%	2.38%
33		10	18.1	15.19	21.01	18.5	11	24	1.286	22.47%	13.81%
44		8	21.5	15.5	27.5	23	11	31	2.535	33.36%	-2.38%
59		10	19.8	17.06	22.54	20	12	24	1.209	19.31%	5.71%
78		10	21	17.45	24.55	22.5	12	27	1.571	23.65%	0.00%

**Reproduction Detail**

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	D	21	18	20	24	20	21	23	21	21	
25		19	18	22	19	20	22	18	21	24	22
33		11	19	17	16	24	20	13	23	20	18
44		25	21	31	24	27	22	11	11		
59		19	24	20	24	21	24	17	12	17	20
78		23	22	23	22	12	22	24	23	12	27

*EWB*  
*12/8/23*

**CETIS Analytical Report**

Report Date: 07 Dec-23 13:35 (p 1 of 2)  
Test Code/ID: 61A912C9 / 16-3847-0345

**Ceriodaphnia 7-d Survival and Reproduction Test**

Bio-Analytical Laboratories

Analysis ID: 08-3359-2869	Endpoint: Reproduction	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 13:25	Analysis: Nonparametric-Control vs Treatments	Status Level: 1
Edit Date: 07 Dec-23 13:10	MD5 Hash: 06D08648A03663EA2937B224394386CB	Editor ID: 008-522-314-5
Batch ID: 01-0354-5901	Test Type: Reproduction-Survival (2-8d)	Analyst: Lab Tech
Start Date: 28 Nov-23 12:10	Protocol: EPA/821/R-02-013 (2002)	Diluent: Reconstituted Water
Ending Date: 05 Dec-23 15:57	Species: Ceriodaphnia dubia	Brine:
Test Length: 7d 4h	Taxon: Branchiopoda	Source: In-House Culture Age: <24
Sample ID: 14-4299-4482	Code: X9028	Project: WET Quarterly Compliance Test (4Q)
Sample Date: 27 Nov-23 11:00	Material: POTW Effluent	Source: AR0021776 (AR0021776)
Receipt Date: 27 Nov-23 16:37	CAS (PC):	Station: 001
Sample Age: 25h (-0.3 °C)	Client: Nashville Public Works	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	Tox Units	MSDu	PMSD
Untransformed	C > T	78	>78	---	1.3	6.109	31.65%

**Steel Many-One Rank Sum Test**

Control	vs	Conc-%	df	Test Stat	Critical	Ties	P-Type	P-Value	Decision(α:5%)
Dilution Water		25	18	104.5	75	4	CDF	0.8218	Non-Significant Effect
		33	18	86.5	75	4	CDF	0.2473	Non-Significant Effect
		44	18	110.5	75	2	CDF	0.9287	Non-Significant Effect
		59	18	100.5	75	3	CDF	0.7129	Non-Significant Effect
		78	18	126	75	2	CDF	0.9980	Non-Significant Effect

**ANOVA Table**

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	104.283	20.8567	5	0.5858	0.7107	Non-Significant Effect
Error	1922.7	35.6056	54			
Total	2026.98		59			

**ANOVA Assumptions Tests**

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test	26.6	15.09	6.8E-05	Unequal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9066	0.9459	0.0002	Non-Normal Distribution

**Reproduction Summary**

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	10	19.3	15.28	23.32	21	4	24	1.777	29.11%	0.00%
25		10	20.5	19.06	21.94	20.5	18	24	0.6368	9.82%	-6.22%
33		10	18.1	15.19	21.01	18.5	11	24	1.286	22.47%	6.22%
44		10	17.2	9.293	25.11	21.5	0	31	3.495	64.26%	10.88%
59		10	19.8	17.06	22.54	20	12	24	1.209	19.31%	-2.59%
78		10	21	17.45	24.55	22.5	12	27	1.571	23.65%	-8.81%

**Reproduction Detail**

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	D	21	18	20	4	24	20	21	23	21	21
25		19	18	22	19	20	22	18	21	24	22
33		11	19	17	16	24	20	13	23	20	18
44		25	21	0	31	24	27	22	11	11	0
59		19	24	20	24	21	24	17	12	17	20
78		23	22	23	22	12	22	24	23	12	27

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# CETIS Analytical Report

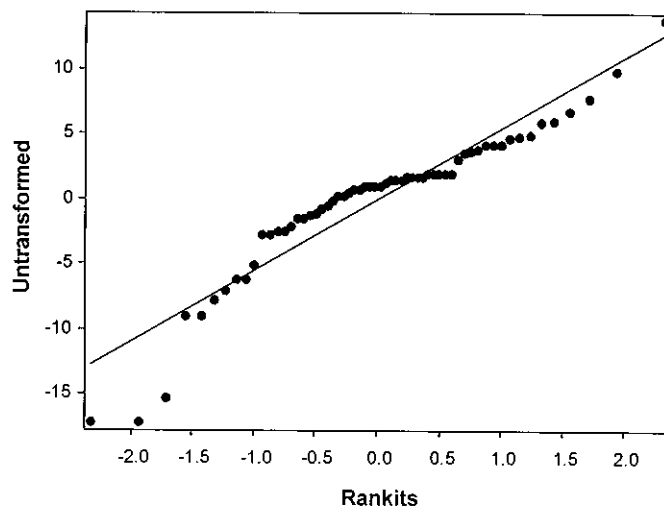
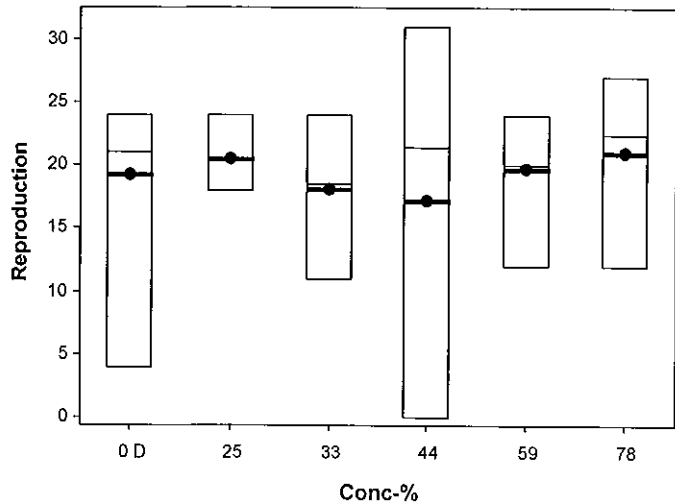
Report Date: 07 Dec-23 13:35 (p 2 of 2)  
Test Code/ID: 61A912C9 / 16-3847-0345

## Ceriodaphnia 7-d Survival and Reproduction Test

Bio-Analytical Laboratories

Analysis ID: 08-3359-2869	Endpoint: Reproduction	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 13:25	Analysis: Nonparametric-Control vs Treatments	Status Level: 1
Edit Date: 07 Dec-23 13:10	MD5 Hash: 06D08648A03663EA2937B224394386CB	Editor ID: 008-522-314-5

### Graphics



*ELB*  
*12/8/23*



# CETIS Analytical Report

Report Date: 07 Dec-23 13:36 (p 1 of 2)  
Test Code/ID: 61A912C9 / 16-3847-0345

## Ceriodaphnia 7-d Survival and Reproduction Test

Bio-Analytical Laboratories

Analysis ID: 08-0458-5684	Endpoint: Reproduction	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 13:35	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Edit Date: 07 Dec-23 13:10	MD5 Hash: 06D08648A03663EA2937B224394386CB	Editor ID: 008-522-314-5
Batch ID: 01-0354-5901	Test Type: Reproduction-Survival (2-8d)	Analyst: Lab Tech
Start Date: 28 Nov-23 12:10	Protocol: EPA/821/R-02-013 (2002)	Diluent: Reconstituted Water
Ending Date: 05 Dec-23 15:57	Species: Ceriodaphnia dubia	Brine:
Test Length: 7d 4h	Taxon: Branchiopoda	Source: In-House Culture      Age: <24
Sample ID: 14-4299-4482	Code: X9028	Project: WET Quarterly Compliance Test (4Q)
Sample Date: 27 Nov-23 11:00	Material: POTW Effluent	Source: AR0021776 (AR0021776)
Receipt Date: 27 Nov-23 16:37	CAS (PC):	Station: 001
Sample Age: 25h (-0.3 °C)	Client: Nashville Public Works	

### Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	2067266	1000	Yes	Two-Point Interpolation

### Point Estimates

Level	%	95% LCL	95% UCL	Tox Units	95% LCL	95% UCL
IC15	>78	---	---	<1.3	---	---
IC20	>78	---	---	<1.3	---	---
IC25	>78	---	---	<1.3	---	---
IC40	>78	---	---	<1.3	---	---
IC50	>78	---	---	<1.3	---	---

### Reproduction Summary

Conc-%	Code	Count	Calculated Variate						Isotonic Variate	
			Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect
0	D	10	19.3	21	4	24	29.11%	0.00%	19.9	0.00%
25		10	20.5	20.5	18	24	9.82%	-6.22%	19.9	0.00%
33		10	18.1	18.5	11	24	22.47%	6.22%	19.02	4.40%
44		10	17.2	21.5	0	31	64.26%	10.88%	19.02	4.40%
59		10	19.8	20	12	24	19.31%	-2.59%	19.02	4.40%
78		10	21	22.5	12	27	23.65%	-8.81%	19.02	4.40%

### Reproduction Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	D	21	18	20	4	24	20	21	23	21	21
25		19	18	22	19	20	22	18	21	24	22
33		11	19	17	16	24	20	13	23	20	18
44		25	21	0	31	24	27	22	11	11	0
59		19	24	20	24	21	24	17	12	17	20
78		23	22	23	22	12	22	24	23	12	27

*EWB*  
*12/18/23*

# CETIS Analytical Report

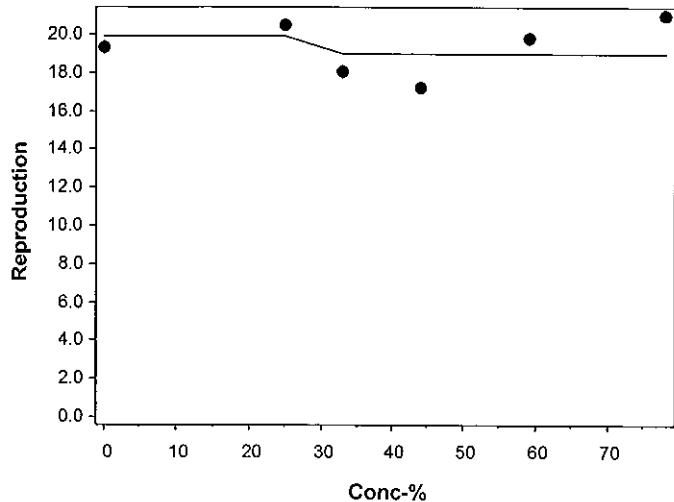
Report Date: 07 Dec-23 13:36 (p 2 of 2)  
Test Code/ID: 61A912C9 / 16-3847-0345

## Ceriodaphnia 7-d Survival and Reproduction Test

Bio-Analytical Laboratories

Analysis ID: 08-0458-5684	Endpoint: Reproduction	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 13:35	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Edit Date: 07 Dec-23 13:10	MD5 Hash: 06D08648A03663EA2937B224394386CB	Editor ID: 008-522-314-5

### Graphics



*EUB*  
*12/8/23*

# CETIS Analytical Report

Report Date: 07 Dec-23 13:59 (p 1 of 2)  
Test Code/ID: 3B0583DB / 09-9021-7179

## Fathead Minnow 7-d Larval Survival and Growth Test

Bio-Analytical Laboratories

Analysis ID: 15-4066-2484	Endpoint: 7d Survival Rate	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 13:58	Analysis: Parametric-Control vs Treatments	Status Level: 1
Edit Date: 07 Dec-23 13:53	MD5 Hash: 3990F848C9814977C9FD583026DA20F6	Editor ID: 008-522-314-5
Batch ID: 15-8045-4251	Test Type: Growth-Survival (7d)	Analyst: Lab Tech
Start Date: 28 Nov-23 13:45	Protocol: EPA/821/R-02-013 (2002)	Diluent: Reconstituted Water
Ending Date: 05 Dec-23 13:40	Species: Pimephales promelas	Brine:
Test Length: 7d	Taxon: Actinopterygii	Source: In-House Culture Age: <24
Sample ID: 16-1780-8083	Code: X9028	Project: WET Monthly Compliance Test (NOV)
Sample Date: 27 Nov-23 11:00	Material: POTW Effluent	Source: AR0021776 (AR0021776)
Receipt Date: 27 Nov-23 16:37	CAS (PC):	Station: 001
Sample Age: 27h (-0.3 °C)	Client: Nashville Public Works	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	Tox Units	MSDu	PMSD
Angular (Corrected)	C > T	78	>78	---	1.3	0.2442	29.60%

### Dunnnett Multiple Comparison Test

Control	vs	Conc-%	df	Test Stat	Critical	MSD	P-Type	P-Value	Decision(α:5%)
Dilution Water		25	8	1.276	2.362	0.2948	CDF	0.3039	Non-Significant Effect
		33	8	1.156	2.362	0.2948	CDF	0.3523	Non-Significant Effect
		44	8	-1.268	2.362	0.2948	CDF	0.9923	Non-Significant Effect
		59	8	-0.7137	2.362	0.2948	CDF	0.9643	Non-Significant Effect
		78	8	-0.1938	2.362	0.2948	CDF	0.8842	Non-Significant Effect

### Test Acceptability Criteria

Attribute	Test Stat	TAC Limits		Overlap	Decision
		Lower	Upper		
Control Resp	0.825	0.8	>>	Yes	Passes Criteria

### ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.398119	0.0796238	5	2.044	0.1084	Non-Significant Effect
Error	0.934978	0.0389574	24			
Total	1.3331		29			

### ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test	3.865	15.09	0.5690	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9726	0.9031	0.6130	Normal Distribution

### 7d Survival Rate Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	5	0.8250	0.5896	1.0000	0.8750	0.5000	1.0000	0.0848	22.98%	0.00%
25		5	0.7000	0.4646	0.9354	0.7500	0.3750	0.8750	0.0848	27.08%	15.15%
33		5	0.7000	0.4181	0.9819	0.6250	0.5000	1.0000	0.1016	32.44%	15.15%
44		5	0.9500	0.8650	1.0000	1.0000	0.8750	1.0000	0.0306	7.21%	-15.15%
59		5	0.9000	0.7701	1.0000	0.8750	0.7500	1.0000	0.0468	11.62%	-9.09%
78		5	0.8500	0.6800	1.0000	0.7500	0.7500	1.0000	0.0612	16.11%	-3.03%

### Angular (Corrected) Transformed Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	5	1.1610	0.8823	1.4400	1.2090	0.7854	1.3930	0.1005	19.35%	0.00%
25		5	1.0020	0.7485	1.2560	1.0470	0.6591	1.2090	0.0913	20.38%	13.72%
33		5	1.0170	0.6788	1.3550	0.9117	0.7854	1.3930	0.1218	26.79%	12.43%
44		5	1.3200	1.1950	1.4450	1.3930	1.2090	1.3930	0.0450	7.62%	-13.63%
59		5	1.2500	1.0690	1.4320	1.2090	1.0470	1.3930	0.0653	11.68%	-7.67%
78		5	1.1860	0.9503	1.4210	1.0470	1.0470	1.3930	0.0847	15.98%	-2.08%

*EBB*  
*12/8/23*

# CETIS Analytical Report

Report Date: 07 Dec-23 13:59 (p 2 of 2)  
 Test Code/ID: 3B0583DB / 09-9021-7179

## Fathead Minnow 7-d Larval Survival and Growth Test

Bio-Analytical Laboratories

Analysis ID: 15-4066-2484      Endpoint: 7d Survival Rate      CETIS Version: CETIS v2.1.5  
 Analyzed: 07 Dec-23 13:58      Analysis: Parametric-Control vs Treatments      Status Level: 1  
 Edit Date: 07 Dec-23 13:53      MD5 Hash: 3990F848C9814977C9FD583026DA20F6      Editor ID: 008-522-314-5

### 7d Survival Rate Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	D	1.0000	0.8750	0.8750	0.8750	0.5000
25		0.7500	0.7500	0.8750	0.3750	0.7500
33		0.6250	0.5000	0.5000	1.0000	0.8750
44		1.0000	1.0000	1.0000	0.8750	0.8750
59		1.0000	1.0000	0.7500	0.8750	0.8750
78		1.0000	0.7500	0.7500	0.7500	1.0000

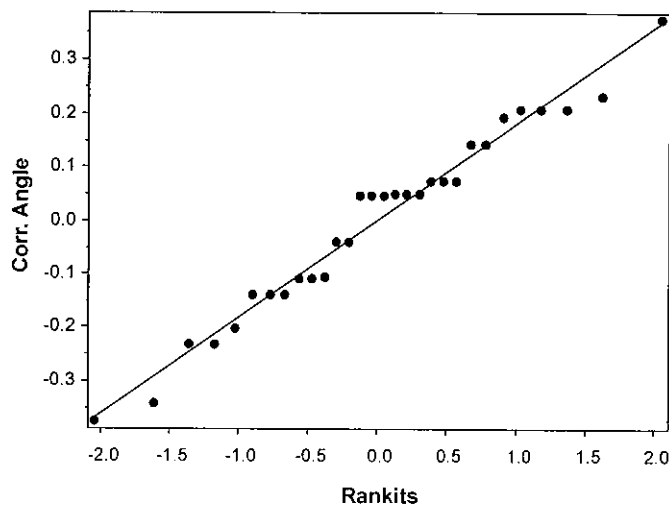
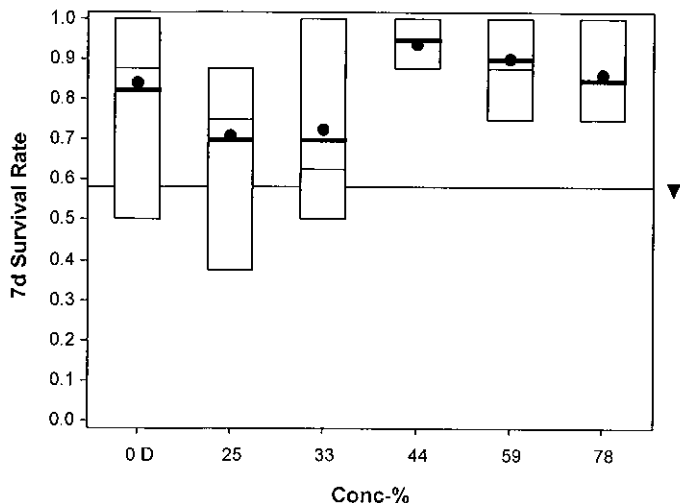
### Angular (Corrected) Transformed Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	D	1.3930	1.2090	1.2090	1.2090	0.7854
25		1.0470	1.0470	1.2090	0.6591	1.0470
33		0.9117	0.7854	0.7854	1.3930	1.2090
44		1.3930	1.3930	1.3930	1.2090	1.2090
59		1.3930	1.3930	1.0470	1.2090	1.2090
78		1.3930	1.0470	1.0470	1.0470	1.3930

### 7d Survival Rate Binomials

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	D	8/8	7/8	7/8	7/8	4/8
25		6/8	6/8	7/8	3/8	6/8
33		5/8	4/8	4/8	8/8	7/8
44		8/8	8/8	8/8	7/8	7/8
59		8/8	8/8	6/8	7/8	7/8
78		8/8	6/8	6/8	6/8	8/8

### Graphics



*EUB*  
 12/8/23

# CETIS Analytical Report

Report Date: 07 Dec-23 14:07 (p 1 of 2)  
Test Code/ID: 3B0583DB / 09-9021-7179

## Fathead Minnow 7-d Larval Survival and Growth Test

Bio-Analytical Laboratories

Analysis ID: 13-3230-6725	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 14:04	Analysis: Parametric-Control vs Treatments	Status Level: 1
Edit Date: 07 Dec-23 13:53	MD5 Hash: 2CE56282793EE411CB258337B5F03C5C	Editor ID: 008-522-314-5
Batch ID: 15-8045-4251	Test Type: Growth-Survival (7d)	Analyst: Lab Tech
Start Date: 28 Nov-23 13:45	Protocol: EPA/821/R-02-013 (2002)	Diluent: Reconstituted Water
Ending Date: 05 Dec-23 13:40	Species: Pimephales promelas	Brine:
Test Length: 7d	Taxon: Actinopterygii	Source: In-House Culture Age: <24
Sample ID: 16-1780-8083	Code: X9028	Project: WET Monthly Compliance Test (NOV)
Sample Date: 27 Nov-23 11:00	Material: POTW Effluent	Source: AR0021776 (AR0021776)
Receipt Date: 27 Nov-23 16:37	CAS (PC):	Station: 001
Sample Age: 27h (-0.3 °C)	Client: Nashville Public Works	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	Tox Units	MSDu	PMSD
Untransformed	C > T	78	>78	---	1.3	0.1655	29.96%

### Dunnnett Multiple Comparison Test

Control	vs	Conc-%	df	Test Stat	Critical	MSD	P-Type	P-Value	Decision(α:5%)
Dilution Water		25	8	1.998	2.362	0.1655	CDF	0.0998	Non-Significant Effect
		33	8	1.569	2.362	0.1655	CDF	0.2022	Non-Significant Effect
		44	8	-2.39	2.362	0.1655	CDF	0.9998	Non-Significant Effect
		59	8	-0.9631	2.362	0.1655	CDF	0.9815	Non-Significant Effect
		78	8	0.5707	2.362	0.1655	CDF	0.6157	Non-Significant Effect

### Test Acceptability Criteria

Attribute	Test Stat	TAC Limits			Decision
		Lower	Upper	Overlap	
Control Resp	0.5525	0.25	>>	Yes	Passes Criteria
PMSD	0.2996	0.12	0.3	Yes	Passes Criteria

### ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.327041	0.0654082	5	5.326	0.0020	Significant Effect
Error	0.294744	0.012281	24			
Total	0.621785		29			

### ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test	5.81	15.09	0.3251	Equal Variances
Distribution	Shapiro-Wilk W Normality Test	0.9725	0.9031	0.6093	Normal Distribution

### Mean Dry Biomass-mg Summary

Conc-%	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	5	0.5525	0.4854	0.6196	0.525	0.5125	0.6375	0.02417	9.78%	0.00%
25		5	0.4125	0.2439	0.5811	0.475	0.175	0.5	0.06072	32.92%	25.34%
33		5	0.4425	0.2436	0.6414	0.45	0.2625	0.6125	0.07165	36.21%	19.91%
44		5	0.72	0.5737	0.8663	0.6625	0.5875	0.8625	0.05268	16.36%	-30.32%
59		5	0.62	0.5389	0.7011	0.6	0.5625	0.725	0.02921	10.53%	-12.22%
78		5	0.5125	0.3979	0.6271	0.5	0.4375	0.6625	0.04127	18.01%	7.24%

### Mean Dry Biomass-mg Detail

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	D	0.5125	0.525	0.6375	0.575	0.5125
25		0.425	0.4875	0.5	0.175	0.475
33		0.45	0.3	0.2625	0.6125	0.5875
44		0.8625	0.6625	0.825	0.5875	0.6625
59		0.5625	0.6	0.725	0.6375	0.575
78		0.6625	0.525	0.4375	0.5	0.4375

*EUB*  
*12/8/23*

# CETIS Analytical Report

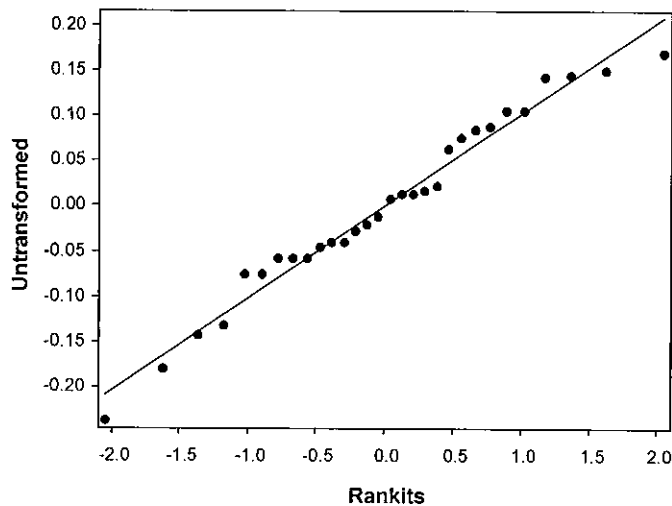
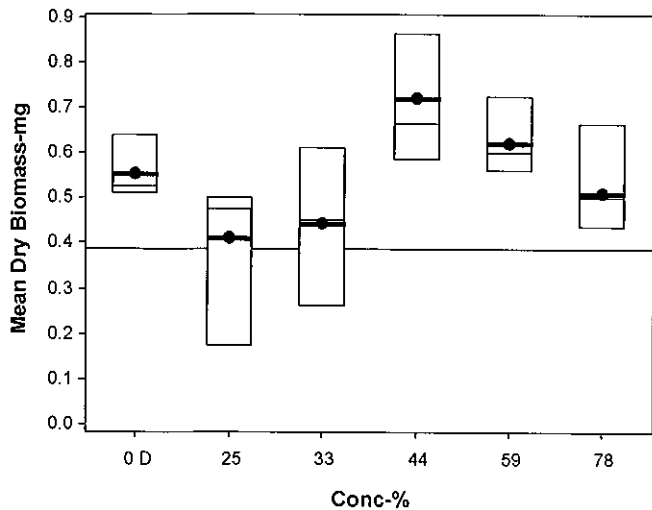
Report Date: 07 Dec-23 14:07 (p 2 of 2)  
Test Code/ID: 3B0583DB / 09-9021-7179

## Fathead Minnow 7-d Larval Survival and Growth Test

Bio-Analytical Laboratories

Analysis ID: 13-3230-6725	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 14:04	Analysis: Parametric-Control vs Treatments	Status Level: 1
Edit Date: 07 Dec-23 13:53	MD5 Hash: 2CE56282793EE411CB258337B5F03C5C	Editor ID: 008-522-314-5

### Graphics



*EB*  
*12/8/23*

**CETIS Analytical Report**

Report Date: 07 Dec-23 14:09 (p 1 of 2)  
 Test Code/ID: 3B0583DB / 09-9021-7179

**Fathead Minnow 7-d Larval Survival and Growth Test**

Bio-Analytical Laboratories

Analysis ID: 02-7055-9223	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 14:08	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Edit Date: 07 Dec-23 13:53	MD5 Hash: 2CE56282793EE411CB258337B5F03C5C	Editor ID: 008-522-314-5
Batch ID: 15-8045-4251	Test Type: Growth-Survival (7d)	Analyst: Lab Tech
Start Date: 28 Nov-23 13:45	Protocol: EPA/821/R-02-013 (2002)	Diluent: Reconstituted Water
Ending Date: 05 Dec-23 13:40	Species: Pimephales promelas	Brine:
Test Length: 7d	Taxon: Actinopterygii	Source: In-House Culture Age: <24
Sample ID: 16-1780-8083	Code: X9028	Project: WET Monthly Compliance Test (NOV)
Sample Date: 27 Nov-23 11:00	Material: POTW Effluent	Source: AR0021776 (AR0021776)
Receipt Date: 27 Nov-23 16:37	CAS (PC):	Station: 001
Sample Age: 27h (-0.3 °C)	Client: Nashville Public Works	

**Linear Interpolation Options**

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	1941671	1000	Yes	Two-Point Interpolation

**Test Acceptability Criteria**

**TAC Limits**

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	0.5525	0.25	>>	Yes	Passes Criteria

**Point Estimates**

Level	%	95% LCL	95% UCL	Tox Units	95% LCL	95% UCL
IC15	>78	---	---	<1.3	---	---
IC20	>78	---	---	<1.3	---	---
IC25	>78	---	---	<1.3	---	---
IC40	>78	---	---	<1.3	---	---
IC50	>78	---	---	<1.3	---	---

**Mean Dry Biomass-mg Summary**

**Calculated Variate**

**Isotonic Variate**

Conc-%	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect
0	D	5	0.5525	0.525	0.5125	0.6375	9.78%	0.00%	0.5525	0.00%
25		5	0.4125	0.475	0.175	0.5	32.92%	25.34%	0.5487	0.68%
33		5	0.4425	0.45	0.2625	0.6125	36.21%	19.91%	0.5487	0.68%
44		5	0.72	0.6625	0.5875	0.8625	16.36%	-30.32%	0.5487	0.68%
59		5	0.62	0.6	0.5625	0.725	10.53%	-12.22%	0.5487	0.68%
78		5	0.5125	0.5	0.4375	0.6625	18.01%	7.24%	0.5125	7.24%

**Mean Dry Biomass-mg Detail**

Conc-%	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5
0	D	0.5125	0.525	0.6375	0.575	0.5125
25		0.425	0.4875	0.5	0.175	0.475
33		0.45	0.3	0.2625	0.6125	0.5875
44		0.8625	0.6625	0.825	0.5875	0.6625
59		0.5625	0.6	0.725	0.6375	0.575
78		0.6625	0.525	0.4375	0.5	0.4375

*EUB*  
*12/18/23*

# CETIS Analytical Report

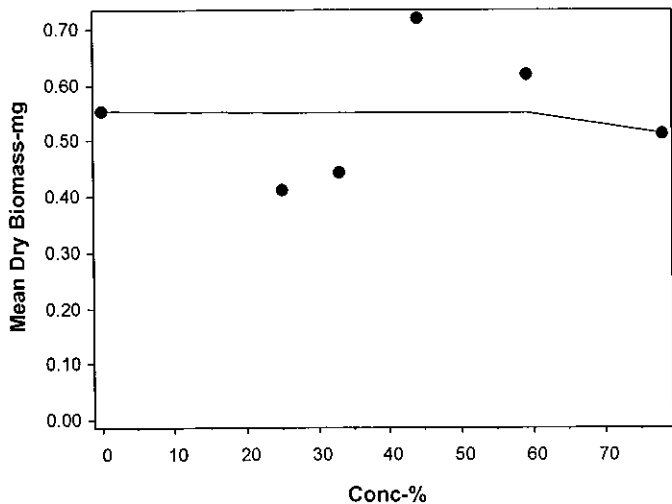
Report Date: 07 Dec-23 14:09 (p 2 of 2)  
Test Code/ID: 3B0583DB / 09-9021-7179

## Fathead Minnow 7-d Larval Survival and Growth Test

Bio-Analytical Laboratories

Analysis ID: 02-7055-9223	Endpoint: Mean Dry Biomass-mg	CETIS Version: CETIS v2.1.5
Analyzed: 07 Dec-23 14:08	Analysis: Linear Interpolation (ICPIN)	Status Level: 1
Edit Date: 07 Dec-23 13:53	MD5 Hash: 2CE56282793EE411CB258337B5F03C5C	Editor ID: 008-522-314-5

### Graphics

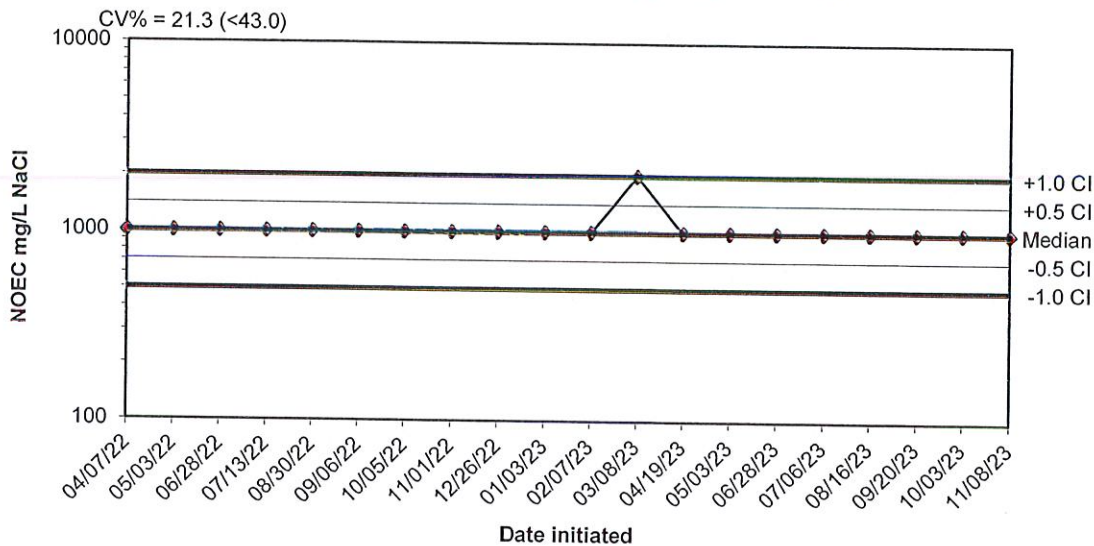


*EVB*  
*12/8/23*



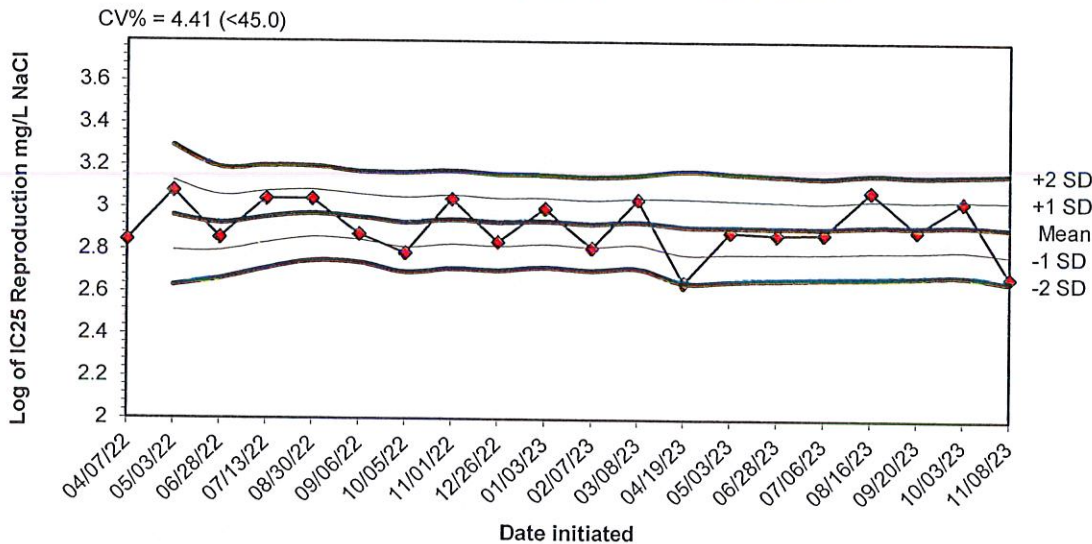
**APPENDIX D**  
**QUALITY ASSURANCE CHARTS**

**CHRONIC REFERENCE TOXICANT TEST RESULTS FOR  
 CERIODAPHNIA DUBIA IN MODERATELY HARD WATER**



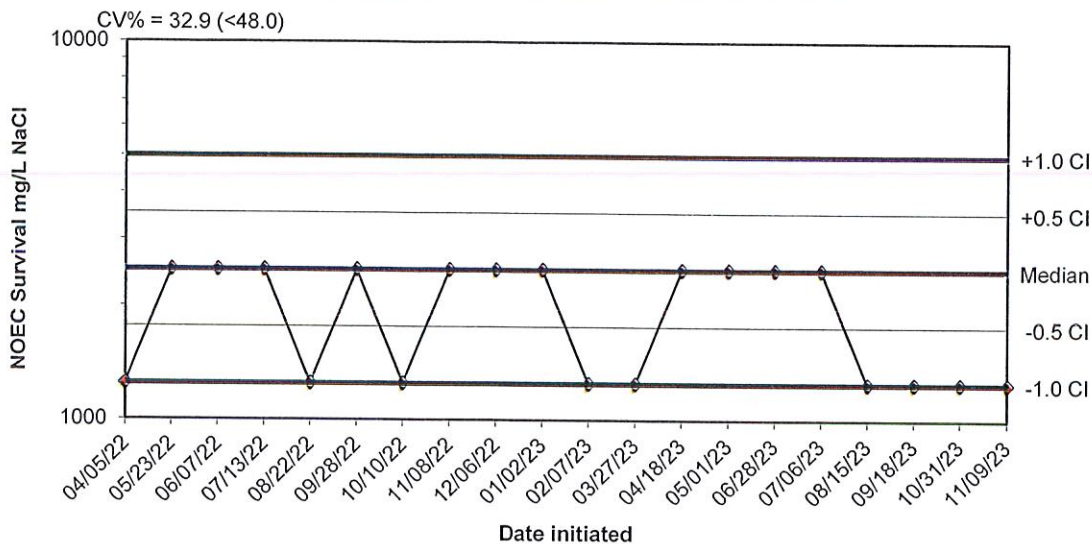
Dates	Values	Median	-0.5 CI	-1.0 CI	+0.5 CI	+1.0 CI
04/07/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
05/03/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
06/28/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
07/13/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
08/30/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
09/06/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
10/05/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
11/01/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
12/26/22	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
01/03/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
02/07/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
03/08/23	2000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
04/19/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
05/03/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
06/28/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
07/06/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
08/16/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
09/20/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
10/03/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000
11/08/23	1000.0000	1000.0000	707.1068	500.0000	1414.2136	2000.0000

**CHRONIC REFERENCE TOXICANT TEST RESULTS FOR  
 CERIODAPHNIA DUBIA IN MODERATELY HARD WATER**



Dates	Values	Mean	-1 SD	-2 SD	+1 SD	+2 SD
04/07/22	2.8451					
05/03/22	3.0792	2.9621	2.7966	2.6311	3.1277	3.2932
06/28/22	2.8573	2.9272	2.7954	2.6637	3.0590	3.1907
07/13/22	3.0414	2.9558	2.8340	2.7122	3.0775	3.1993
08/30/22	3.0414	2.9729	2.8607	2.7485	3.0851	3.1973
09/06/22	2.8751	2.9566	2.8486	2.7405	3.0646	3.1726
10/05/22	2.7853	2.9321	2.8142	2.6962	3.0501	3.1680
11/01/22	3.0414	2.9458	2.8299	2.7141	3.0616	3.1774
12/26/22	2.8388	2.9339	2.8198	2.7058	3.0480	3.1620
01/03/23	3.0000	2.9405	2.8309	2.7214	3.0501	3.1596
02/07/23	2.8129	2.9289	2.8181	2.7073	3.0397	3.1506
03/08/23	3.0414	2.9383	2.8277	2.7172	3.0488	3.1594
04/19/23	2.6501	2.9161	2.7835	2.6509	3.0487	3.1814
05/03/23	2.8890	2.9142	2.7865	2.6589	3.0418	3.1694
06/28/23	2.8774	2.9117	2.7884	2.6650	3.0351	3.1584
07/06/23	2.8824	2.9099	2.7905	2.6711	3.0293	3.1487
08/16/23	3.0839	2.9201	2.7971	2.6740	3.0432	3.1662
09/20/23	2.8981	2.9189	2.7994	2.6799	3.0384	3.1579
10/03/23	3.0350	2.9250	2.8059	2.6867	3.0442	3.1633
11/08/23	2.6784	2.9127	2.7843	2.6559	3.0411	3.1695

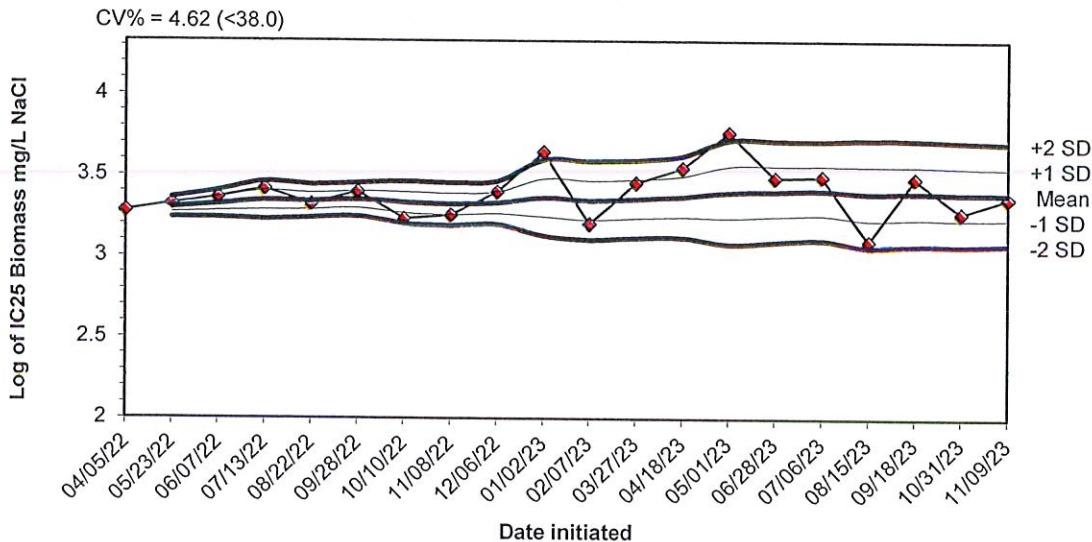
### CHRONIC REFERENCE TOXICANT TEST RESULTS FOR PIMEPHALES PROMELAS IN MODERATELY HARD WATER



Dates	Values	Median	-0.5 CI	-1.0 CI	+0.5 CI	+1.0 CI
04/05/22	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
05/23/22	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
06/07/22	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
07/13/22	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
08/22/22	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
09/28/22	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
10/10/22	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
11/08/22	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
12/06/22	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
01/02/23	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
02/07/23	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
03/27/23	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
04/18/23	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
05/01/23	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
06/28/23	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
07/06/23	2500.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
08/15/23	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
09/18/23	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
10/31/23	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000
11/09/23	1250.0000	2500.0000	1767.7670	1250.0000	3535.5339	5000.0000



**CHRONIC REFERENCE TOXICANT TEST RESULTS FOR PIMEPHALES  
 PROMELAS IN MODERATELY HARD WATER**



Dates	Values	Mean	-1 SD	-2 SD	+1 SD	+2 SD
04/05/22	3.2788					
05/23/22	3.3222	3.3005	3.2698	3.2390	3.3312	3.3620
06/07/22	3.3617	3.3209	3.2794	3.2379	3.3624	3.4039
07/13/22	3.4150	3.3444	3.2864	3.2285	3.4024	3.4604
08/22/22	3.3222	3.3400	3.2888	3.2376	3.3912	3.4423
09/28/22	3.3979	3.3496	3.2981	3.2466	3.4012	3.4527
10/10/22	3.2304	3.3326	3.2675	3.2023	3.3977	3.4629
11/08/22	3.2553	3.3229	3.2567	3.1905	3.3892	3.4554
12/06/22	3.3979	3.3313	3.2645	3.1977	3.3981	3.4649
01/02/23	3.6435	3.3625	3.2454	3.1283	3.4796	3.5967
02/07/23	3.2041	3.3481	3.2272	3.1063	3.4690	3.5899
03/27/23	3.4550	3.3570	3.2377	3.1183	3.4763	3.5957
04/18/23	3.5469	3.3716	3.2458	3.1200	3.4974	3.6232
05/01/23	3.7654	3.3997	3.2395	3.0792	3.5600	3.7203
06/28/23	3.4878	3.4056	3.2495	3.0934	3.5617	3.7178
07/06/23	3.4949	3.4112	3.2587	3.1063	3.5636	3.7161
08/15/23	3.0973	3.3927	3.2266	3.0605	3.5588	3.7249
09/18/23	3.4823	3.3977	3.2352	3.0727	3.5602	3.7227
10/31/23	3.2704	3.3910	3.2304	3.0698	3.5516	3.7122
11/09/23	3.3632	3.3896	3.2332	3.0767	3.5461	3.7025

**APPENDIX E  
AGENCY FORMS**



**SUMMARY REPORTING FORMS  
 CHRONIC BIOMONITORING  
Ceriodaphnia dubia Survival and Reproduction**

Permittee: Nashville Public Works

NPDES No.: AR0021776/ AFIN 31-00036

	Time	Date	Time	Date
Composite 1 Collected From	0951	11/26/23	To 0651	11/27/23
Composite 2 Collected From	0750	11/28/23	To 0418	11/29/23
Composite 3 Collected From	0835	11/30/23	To 0437	12/01/23
Test initiated:	1210	am/pm	11/28/23	Date
Test terminated:	1557	am/pm	12/05/23	Date
Dilution water used:	Receiving	X	Reconstituted	

**PERCENT SURVIVAL**

Time of Reading	Percent Effluent					
	0	25.0	33.0	44.0	59.0	78.0
24h	100.0	100.0	100.0	100.0	100.0	100.0
48h	100.0	100.0	100.0	100.0	100.0	100.0
End of test	90.0	100.0	100.0	80.0	100.0	100.0

**NUMBER OF YOUNG PRODUCED PER FEMALE @ END OF TEST**

Rep	0	25.0	33.0	44.0	59.0	78.0
A	21	19	11	25	19	23
B	18	18	19	21	24	22
C	20	22	17	D	20	23
D	D4	19	16	31	24	22
E	24	20	24	24	21	12
F	20	22	20	27	24	22
G	21	18	13	22	17	24
H	23	21	23	11	12	23
I	21	24	20	11	17	12
J	21	22	18	D	20	27
Surv. Mean	21.0	20.5	18.1	21.5	19.8	21.0
Total Mean	19.3	20.5	18.1	17.2	19.8	21.0
CV%*	8.25	9.82	22.47	33.36	19.31	23.65

\*coefficient of variation = standard deviation x 100/mean. D=dead adult  
 PMSD =31.65%

Ceriodaphnia dubia  
Survival and Reproduction (continued)

**1. Fisher's Exact Test:**

Is the mean survival at the end of the test significantly different ( $p=.05$ ) than the control survival for the % effluent corresponding to (lethality):

- |   |     |   |    |
|---|-----|---|----|
| a) LOW FLOW OR CRITICAL DILUTION (78.0%): | YES | X | NO |
| b) ½ LOW FLOW DILUTION (NA%):             | YES |   | NO |

**2. Dunnett's Procedure or Steel's Many-One Rank Test as appropriate:**

Is the mean number of young produced per female significantly different ( $p=.05$ ) than the control's number of young per female for the % effluent corresponding to (significant non-lethal effects):

- |   |     |   |    |
|---|-----|---|----|
| a) LOW FLOW OR CRITICAL DILUTION (78.0%): | YES | X | NO |
| b) ½ LOW FLOW DILUTION (NA%):             | YES |   | NO |

3. If you answered NO to 1. a) and 2. a) enter (0) otherwise enter (1): 0

4. If you answered NO to 1. b) and 2. b) enter (0) otherwise enter (1):

5. Enter response to item 3 on DMR Form, parameter #TEP3B.

6. Enter response to item 4 on DMR Form, parameter #TFP3B.

7. Enter percent effluent corresponding to each NOEC below:

- |                       |                 |
|-----------------------|-----------------|
| a) NOEC survival:     | 78.0 % effluent |
| b) NOEC reproduction: | 78.0 % effluent |

Biomonitoring Form  
Chronic Toxicity Summary Form for Ceriodaphnia dubia  
Chemical Parameters Chart

Permittee: Nashville Public Works  
NPDES#: AR0021776/ AFIN 31-00036  
Contact: Larry Dunaway  
Analysts: Water, Valle, Miller

Sample #1 Collected: 11/27/2023 Time: 651  
Sample #2 Collected: 11/29/2023 Time: 418  
Sample #3 Collected: 12/1/2023 Time: 437  
Test Begin: 11/28/2023 Time: 1210  
Test End: 12/5/2023 Time: 1557

Dilution:	0%							44.0%						
Day:	1	2	3	4	5	6	7	1	2	3	4	5	6	7
T (°C)	24.0	24.3	24.0	24.8	25.2	24.1	23.6	24.0	24.3	24.0	24.8	25.2	24.1	23.6
DO Initial	7.9	8.0	7.9	8.0	8.0	8.6	8.6	8.6	7.9	7.5	8.1	8.2	8.9	8.8
DO Final	8.9	9.1	7.7	8.2	8.0	9.2	8.6	8.5	8.4	7.9	7.9	8.5	8.2	8.2
pH Initial	6.9	7.0	7.3	7.1	6.8	7.4	6.5	8.2	8.0	7.6	8.2	8.1	8.0	8.0
pH Final	7.6	7.6	7.5	7.9	7.7	7.9		7.9	7.7	7.7	7.8	7.8	8.0	
Conductivity	315.0	324.0	30.0	323.0	334.0	350.0		412.0	425.0	431.0	416.0	419.0	421.0	
Alkalinity	64.0													
Hardness	88.0													
Chlorine	<0.5													
Dilution:	25.0%							59.0%						
Day:	1	2	3	4	5	6	7	1	2	3	4	5	6	7
T (°C)	24.0	24.3	24.0	24.8	25.2	24.1	23.6	24.0	24.3	24.0	24.8	25.2	24.1	23.6
DO Initial	8.5	8.4	7.6	7.1	8.4	8.9	8.3	8.2	7.9	7.6	8.5	8.6	9.1	8.3
DO Final	8.1	8.5	7.4	8.1	8.2	9.0	8.1	8.5	8.4	8.2	8.1	8.5	8.2	8.2
pH Initial	8.1	7.3	7.3	7.5	7.1	7.4	7.2	8.3	7.9	7.4	8.1	8.1	8.2	8.1
pH Final	7.8	7.6	7.2	7.8	7.7	7.9		7.9	7.7	7.7	7.8	7.8	8.0	
Conductivity	366.0	379.0	384.0	376.0	373.0	395.0		444.0	459.0	471.0	444.0	453.0	454.0	
Alkalinity														
Hardness														
Chlorine														
Dilution:	33.0%							78.0%						
Day:	1	2	3	4	5	6	7	1	2	3	4	5	6	7
T (°C)	24.0	24.3	24.0	24.8	25.2	24.1	23.6	24.0	24.3	24.0	24.8	25.2	24.1	23.6
DO Initial	8.6	8.3	7.9	7.9	8.8	8.2	8.2	8.2	7.9	7.5	8.4	8.6	9.1	8.6
DO Final	8.7	8.5	7.7	8.1	8.8	8.9	8.0	8.4	7.9	8.0	7.9	8.3	8.2	8.6
pH Initial	8.2	7.7	7.2	8.1	8.0	8.0	8.0	8.3	7.9	7.6	8.2	8.1	8.3	8.2
pH Final	7.9	7.7	7.4	7.8	7.8	8.0		8.0	7.9	7.8	7.8	7.9	8.0	
Conductivity	382.0	400.0	405.0	394.0	392.0	411.0		487.0	502.0	515.0	482.0	488.0	501.0	
Alkalinity								164.0	160.0	160.0				
Hardness								256.0	248.0	220.0				
Chlorine								<0.5	<0.5	<0.5				

Comments:

**SUMMARY REPORTING FORMS CHRONIC BIOMONITORING  
 FATHEAD MINNOW LARVAE GROWTH AND SURVIVAL  
 (*Pimephales promelas*)**

Permittee: Nashville Public Works NPDES No.: AR0021776/ AFIN 31-00036

	Time	Date		Time	Date
Composite 1 Collected from:	0951	11/26/23	To	0651	11/27/23
Composite 2 Collected from:	0750	11/28/23	To	0418	11/29/23
Composite 3 Collected from:	0835	11/30/23	To	0437	12/01/23
Test initiated:	1345	am/pm		11/28/23	date
Test terminated:	1340	am/pm		12/05/23	date
Dilution water used:		Receiving	X	Reconstituted	

**DATA TABLE FOR SURVIVAL**

Effluent Conc. %	Percent Survival in Replicate Chambers					Mean Percent Survival			CV%*
	A	B	C	D	E	24h	48h	7 days	
<b>0</b>	100.0	87.5	87.5	87.5	500.0	100.0	100.0	82.5	19.35
<b>25.0</b>	75.0	75.0	87.3	37.5	75.0	100.0	100.0	70.0	20.38
<b>33.0</b>	62.5	50.0	50.0	100.0	87.5	100.0	97.5	70.0	26.79
<b>44.0</b>	400.0	100.0	100.0	87.5	87.5	100.0	100.0	95.0	7.62
<b>59.0</b>	100.0	100.0	75.0	87.5	87.5	100.0	100.0	90.0	11.68
<b>78.0</b>	100.0	75.0	75.0	75.0	100.0	100.0	100.0	85.0	15.98

**DATA TABLE FOR GROWTH**

Effluent Conc. %	Average Dry Weight in milligrams in replicate chambers					Mean Dry Weight mg	CV*
	A	B	C	D	E		
<b>0</b>	0.513	0.525	0.68	0.575	0.513	0.553	9.78
<b>25.0</b>	0.425	0.488	0.500	0.175	0.475	0.413	32.92
<b>33.0</b>	0.450	0.300	0.253	0.613	0.588	0.443	36.21
<b>44.0</b>	0.863	0.663	0.825	0.588	0.663	0.720	16.36
<b>59.0</b>	0.563	0.600	0.725	0.638	0.575	0.620	10.53
<b>78.0</b>	0.663	0.525	0.438	0.500	0.438	0.513	18.01

\*coefficient of variation = standard deviation x 100/mean.

PMSD: 29.96%

FATHEAD MINNOW LARVAE GROWTH AND SURVIVAL (cont)  
(Pimephales promelas)

1. Dunnett's Procedure or Steels Many-One Rank Test as appropriate:

Is the mean survival at 7 days significantly different ( $p=.05$ ) than the control survival for the % effluent corresponding to:

a) LOW FLOW OR CRITICAL DILUTION (78.0 %)	YES	X	NO
b) ½ LOW FLOW DILUTION ( NA %)	YES		NO

2. Dunnett's Procedure (or appropriate test):

Is the mean dry weight (growth) at 7 days significantly different ( $p=.05$ ) than the control's dry weight for the % effluent corresponding to (significant non-lethal effects):

a) LOW FLOW OR CRITICAL DILUTION (78.0 %)	YES	X	NO
b) ½ LOW FLOW DILUTION ( NA %)	YES		NO

3. If you answered NO to 1. a) and 2. a) enter (0) otherwise enter (1): 0

4. If you answered NO to 1. b) and 2. b) enter (0) otherwise enter (1):

5. Enter response to item 3 on DMR Form, parameter #TEP6C.

6. Enter response to item 4 on DMR Form, parameter #TFP6C.

7. Enter percent effluent corresponding to each NOEC below:

a.) NOEC survival	78.0% effluent.
b.) NOEC growth	78.0% effluent.

Biomonitoring Form  
Chronic Toxicity Summary Form for Pimephales promelas  
Chemical Parameters Chart

Permittee: Nashville Public Works  
NPDES#: AR0021776/AFIN 31-00036  
Contact: Larry Dunaway  
Analyst: Ware, Valle, Miller

Sample #1 Collected: 11/27/2023 Time: 651  
Sample #2 Collected: 11/29/2023 Time: 418  
Sample #3 Collected: 12/1/2023 Time: 437  
Test Begin: 11/28/2023 Time: 1345  
Test End: 12/5/2023 Time: 1340

Dilution: 0%							Dilution: 44.0%							Dilution: 59.0%							Dilution: 78.0%										
Day:	1	2	3	4	5	6	7	Day:	1	2	3	4	5	6	7	Day:	1	2	3	4	5	6	7	Day:	1	2	3	4	5	6	7
T (°C)	25.2	24.8	25.4	23.0	22.8	22.8	22.6	T (°C)	25.2	24.8	25.4	23.0	22.8	22.8	22.6	T (°C)	25.2	24.8	25.4	23.0	22.8	22.8	22.6	T (°C)	25.2	24.8	25.4	23.0	22.8	22.8	22.6
DO Initial	6.7	6.7	7.2	6.8	7.8	7.0	7.0	DO Initial	6.4	7.0	7.6	5.9	7.9	6.0	6.2	DO Initial	6.3	8.4	8.3	7.6	7.2	7.1	7.2	DO Initial	6.7	6.9	7.4	5.8	6.9	6.6	6.3
DO Final	8.4	8.4	7.9	7.6	8.5	8.4	8.4	DO Final	8.3	8.4	8.3	7.2	7.1	8.5	8.5	DO Final	8.3	8.4	7.9	7.9	8.4	9.0	9.0	DO Final	8.3	8.4	7.9	7.9	8.4	9.0	9.0
pH Initial	6.9	7.0	7.4	6.5	7.0	6.7	6.7	pH Initial	7.5	7.3	7.2	7.1	7.1	7.2	7.2	pH Initial	7.8	7.3	7.4	7.3	7.2	7.4	7.5	pH Initial	7.3	7.2	7.3	6.9	6.7	7.1	7.2
pH Final	7.3	6.9	7.8	7.6	6.7	7.9	7.9	pH Final	7.9	7.7	7.9	7.8	7.8	8.0	8.0	pH Final	7.8	7.3	7.4	7.3	7.2	7.4	7.5	pH Final	7.8	7.6	7.9	7.8	7.9	8.0	8.0
Conductivity	324.0	328.0	320.0	318.0	324.0	324.0	324.0	Conductivity	455.0	427.0	436.0	415.0	427.0	423.0	423.0	Conductivity	504.0	507.0	517.0	488.0	502.0	502.0	502.0	Conductivity	395.0	402.0	414.0	393.0	398.0	398.0	398.0
Alkalinity	60.0		56.0					Alkalinity								Alkalinity	164.0	160.0	160.0					Alkalinity							
Hardness	108.0		92.0					Hardness								Hardness	256.0	248.0	220.0					Hardness							
Chlorine	<0.5		<0.5					Chlorine								Chlorine	<0.5	<0.5	<0.5					Chlorine							

Comments:



**APPENDIX F**  
**REPORT QUALITY ASSURANCE FORM**



## Bio-Analytical Laboratories

3240 Spurgin Road  
Post Office Box 527  
Doyline, LA 71023

(318) 745-2772  
1-800-259-1246  
Fax: (318) 745-2773

### REPORT QUALITY ASSURANCE FORM

Client: City of Nashville AR

Project#: X9028

Chain of Custody Documents Checked by: EBB 12/8/23  
Technician/Date

Raw Data Documents Checked by: EBB 12/8/23  
Technician/Date

Statistical Analysis Package Checked by: EBB 12/8/23  
Quality Manager/Date

Quality Control Data Checked by: EBB 12/8/23  
Quality Manager/Date

Report Checked by: EBB 12/11/23  
Quality Manager/Date

I certify that this document was prepared under my direction or supervision in accordance with a system designed to assure that qualified personnel properly gather and evaluate the information submitted. The information contained in this document, to the best of my knowledge, is true, accurate and complete.

Paul Brugg, BS  
Quality Manager

12/11/23  
Date

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