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ANALYTICAL REPORT

PREPARED FOR

Attn: Mr. John Davis
Malvern Water Works
506 Overman
Malvern, Arkansas 72104

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JOB DESCRIPTION

Biomonitoring

JOB NUMBER

192-2047-1

Eurofins Arkansas
8600 Kanis Rd
Little Rock AR 72204

See page two for job notes and contact information.

Eurofins Arkansas

Job Notes

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Authorization

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Biomonitoring Testing
for
Outfall 001

Prepared for:

Mr. John Davis
Malvern Water Works
506 Overman
Malvern, AR 72104

Control No. 274584-1

Malvern Water Works
ATTN: Mr. John Davis
506 Overman
Malvern, AR 72104

Re: Chronic 7-Day Renewal*Pimephales promelas* (Fathead minnow) and *Ceriodaphnia dubia* - Outfall 001
NPDES Permit No. AR0034126 AFIN 30-00040

Dear Mr. John Davis:

This report is the analytical results and supporting information for the samples submitted to Eurofins Arkansas. The following results are applicable only to the sample identified by the control number referenced above. Accurate assessment of the data requires access to the entire document. Each section of the report has been reviewed and approved by the Chief Operating Officer or qualified designee.

Testing procedures and Quality Assurance were in accordance with "Short Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms" EPA-821-R-02-013, Fourth Edition, October 2002. Test results are summarized below:

Method 1000.0 Chronic *Pimephales promelas* (Fathead minnow) Survival and Growth Test: The No Observable Effects Concentration (NOEC) for survival occurred at 16 % effluent, which is above the critical dilution of 12 %. The NOEC for growth occurred at 16 % effluent, which is above the critical dilution of 12 %. **The sample, therefore, PASSED both lethal and sub-lethal effects for the Fathead minnow test.**

Method 1002.0 Chronic *Ceriodaphnia dubia* Survival and Reproduction Test: The No Observable Effects Concentration (NOEC) for survival occurred at 16 % effluent, which is above the critical dilution of 12 %. The percent minimum significant difference (PMSD) was below the limit of 13. Following additional calculations provided in the EPA document "Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications under the National Pollutant Discharge Elimination Systems Program", the NOEC for sublethal effects was calculated to be 16 %. **The sample, therefore, PASSED both lethal and sub-lethal effects for the *Ceriodaphnia dubia* test.**

Eurofins Arkansas

John Overbey
Chief Operating Officer

PDF cc: Malvern Water Works
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jdavis@malvernar.gov

Malvern Water Works
ATTN: Mr. Carl Wheatley
cwheatley@malvernar.gov

Table of Contents

I. Control Acceptance Criteria

Pimephales promelas (Fathead minnow) Method 1000.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	97.5	PASS
Control Growth > or = 0.25 mg per Surviving minnow	0.486	PASS
Control Growth CV < or = 40%	11.4	PASS
Growth Minimum Significant Difference 12 to 30%	13.5	PASS
Critical Dilution CV < or = 40%	6.91	PASS

Ceriodaphnia dubia Method 1002.0

CRITERIA	RESULTS	PASS/FAIL
Control Survival > or = 80%	100	PASS
Control Reproduction > or = 15 per Surviving Female	29.3	PASS
Control CV < or = 40% per Surviving Female	6.64	PASS
Reproduction Minimum Significant Difference 13 to 47%	11.7	BELOW
Critical Dilution CV < or = 40%	7.47	PASS

II. Outlined Report

A. Introduction

1. Permit Number: AR0034126 AFIN 30-00040
2. Test Requirements: qtr
   Test Methods 1000.0 and 1002.0

B. Source of Effluent/Dilution Water:

1. Effluent Samples:
   a. Sampling Point:
   b. Chemical Data:

Analysis	Sample 1	Sample 2	Sample 3
Dissolved oxygen (mg/l)	8.4	8.6	8.8
pH (standard units)	6.8	7.1	7.0
Alkalinity (mg/l as CaCO3)	16	16	17
Hardness (mg/l as CaCO3)	22	21	21
Conductivity (umhos/cm)	130	130	140
Residual Chlorine (mg/l)	0.060	0.060	0.070
Ammonia as N (mg/l)	1.9	1.8	1.9

2. Dilution Water Samples:

Analysis	192-1716-A-1	192-1841-A-1
Dissolved oxygen (mg/l)	8.0	8.5
pH (standard units)	7.3	7.5
Alkalinity (mg/l as CaCO3)	31	31
Hardness (mg/l as CaCO3)	47	45
Conductivity (umhos/cm)	170	160
Residual Chlorine (mg/l)	<0.05	<0.05

C. Test Methods

1. Test methods used:
   Short Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, EPA-821-R-02-013; test Methods 1000.0 and 1002.0, Fathead Minnow Survival and Growth and *Ceriodaphnia dubia* Survival and Reproduction.
2. Endpoint: No Observable Effects Concentration (NOEC)
3. Test Conditions:

Pimephales promelas (Fathead minnow) Survival and Growth Method 1000.0

Date & Time Test Initiated:	May 23, 2023 at 1342
Date & Time Test Terminated:	May 30, 2023 at 1307
Type & Volume of Test Chamber:	500 ml disposable beaker
Volume of Sample:	250 ml
Number of Organisms per replicate:	8
Number of Replicates per dilution:	5

Ceriodaphnia dubia Survival and Reproduction Method 1002.0

Date & Time Test Initiated:	May 23, 2023 at 1320
Date & Time Test Terminated:	May 29, 2023 at 1500
Type & Volume of Test Chamber:	30 ml disposable beaker
Volume of Sample:	15 ml
Number of Organisms per replicate:	1
Number of Replicates per dilution:	10

4. Source of test organisms: In-house culture
5. Test Temperature: 25 +/- 1 degree Celsius

D. Test Organisms

1. Scientific Name
   a. Test 1000.0 *Pimephales promelas*
   b. Test 1002.0 *Ceriodaphnia dubia*

III. Data Analysis

The data was analyzed using Eurofins's Laboratory Information Management Software based on Toxstat and following EPA method criteria.

Pimephales promelas (Fathead minnow) survival data was transformed using the Arc Sine transformation. Normality and homogeneity of variance were checked using Shapiro-Wilk's. The survival data was then analyzed using Steel's Many-One Rank Test to determine the No Observable Effects Concentration (NOEC).

Fathead minnow growth data was analyzed for normality and homogeneity of variance using Shapiro-Wilk's and Bartlett's test. Dunnett's Test was used to determine the No Observable Effects Concentration (NOEC) for growth.

Ceriodaphnia dubia survival data was analyzed with Fisher's Exact Test. Reproduction data was analyzed using Kolmogorov's Test for Normality and Bartlett's test and analyzed with Steel's Many-One Rank Test to determine the No Observable Effects Concentration (NOEC) for Reproduction. Dunnett's Test was used to calculate the PMSD.

IV. Standard Reference Toxicants

The sensitivity of the offspring is determined by performing a standard reference toxicant test monthly. Sodium chloride in synthetic moderately hard water is used as prescribed in EPA-821-R-02-013.

Pimephales promelas (Fathead minnow)

A chronic reference test was performed on May 02, 2023 at 1137 to May 09, 2023 at 1125
The results were as follows: (Control No. 274557-1.)

Survival LC-50: 3105 mg/l
Growth IC-25: 2200 mg/l
Growth PMSD: 22.7

Ceriodaphnia dubia

A chronic reference test was performed on May 02, 2023 at 1010 to May 08, 2023 at 1157
The results were as follows: (Control No. 274557-2.)

Survival LC-50: 1611.6 mg/l
Reproduction IC-25: 913 mg/l
Reproduction PMSD: 17.9

V. Organism History

Pimephales promelas (Fathead minnow)

Date: May 23, 2023
Age: <24 hours
Source: In-house culture
Water: Moderately hard synthetic
Temperature: 25 deg.C

Ceriodaphnia dubia

Date: May 23, 2023
Age: <24 hours
Source: In-house culture
Water: Moderately hard synthetic
Temperature: 25 deg.C

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

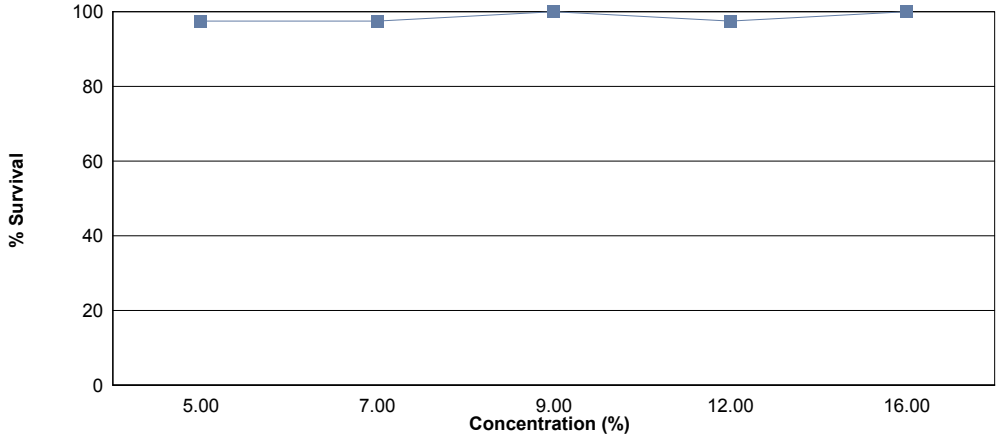
Larvae are exposed in a static renewal system for seven days to different concentrations of effluent with dilution water. Test results are based on the survival and growth (weight) of the larvae.

Effluent dilutions for this test were 5 %, 7 %, 9 %, 12 %, 16 % in accordance with the NPDES permit.

The low flow or 'critical' dilution is specified in the NPDES permit as 12 % effluent.

The test was initiated on May 23, 2023 at 1342 and continued through May 30, 2023 at 1307. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

a.) NOEC survival = 16 % effluent
b.) NOEC growth = 16 % effluent



Summary of the 7-day Fathead Minnow Survival and Growth		
Concentration	Percent Survival	Mean Growth (mg)
Control	97.5	0.474
5 %	97.5	0.449
7 %	97.5	0.428
9 %	100	0.459
12 %	97.5	0.471
16 %	100	0.488

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

Neonates are exposed in a static renewal system to different concentrations of effluent with dilution water until 60% of surviving control organisms have three broods of offspring or a maximum of eight test days.

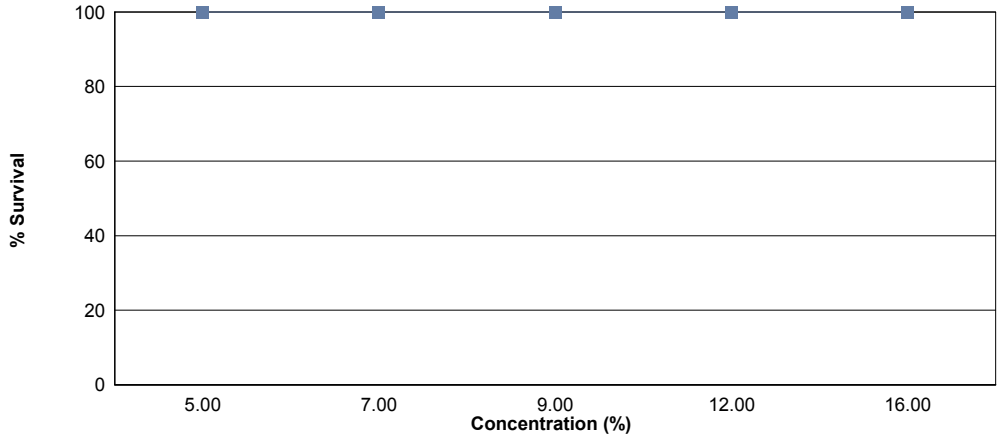
Effluent dilutions for this test were 5 %, 7 %, 9 %, 12 %, 16 % in accordance with the NPDES permit.

The low flow or 'critical' dilution is specified in the NPDES permit as 12 % effluent.

The test was initiated on May 23, 2023 at 1320 and continued through May 29, 2023 at 1500. Statistical analyses were performed on the observed data and the no observable effects concentrations (NOECs) were as follows:

a.) NOEC survival = 16 % effluent
b.) NOEC reproduction = 16 % effluent

(NOEC for sublethal effects was determined by Lower PMSD Bound Test.)



Summary of the 6-day *Ceriodaphnia dubia* Survival and Reproduction Data		
Concentration	Percent Survival	Mean Reproduction
Control	100	29.3
5 %	100	29.5
7 %	100	30.4
9 %	100	30.5
12 %	100	30.8
16 %	100	25.9

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Survival

Date and Time Test Initiated: May 23, 2023 at 1342
Date and Time Test Terminated: May 30, 2023 at 1307

Concentration	Replicate	Number of Survivors						
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	A	8	8	8	8	8	8	8
	B	8	8	8	7	7	7	7
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
5 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	7	7	7
7 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	7
	E	8	8	8	8	8	8	8
9 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
12 %	A	7	7	7	7	7	7	7
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8
16 %	A	8	8	8	8	8	8	8
	B	8	8	8	8	8	8	8
	C	8	8	8	8	8	8	8
	D	8	8	8	8	8	8	8
	E	8	8	8	8	8	8	8

Appendix A1: Test 1000.0

Pimephales promelas (Fathead Minnow) 7-Day Growth

Test Initiated: May 23, 2023 at 1342
Test Terminated: May 30, 2023 at 1307

Concentration	Replicate	Weight of pan	Weight of pan + fish	Total weight of fish (g)	Original # of fish	Mean dry weight (mg)
Control	A	.71487	.71935	0.00448	8	0.560
	B	.71717	.72088	0.00371	8	0.464
	C	.70270	.70633	0.00363	8	0.454
	D	.72777	.73107	0.00330	8	0.412
	E	.70647	.71030	0.00383	8	0.479
5 %	A	.72660	.72981	0.00321	8	0.401
	B	.68849	.69276	0.00427	8	0.534
	C	.71375	.71739	0.00364	8	0.455
	D	.71007	.71367	0.00360	8	0.450
	E	.71210	.71533	0.00323	8	0.404
7 %	A	.70292	.70652	0.00360	8	0.450
	B	.72230	.72572	0.00342	8	0.428
	C	.72594	.72927	0.00333	8	0.416
	D	.71364	.71663	0.00299	8	0.374
	E	.71599	.71975	0.00376	8	0.470
9 %	A	.70709	.71065	0.00356	8	0.445
	B	.71260	.71613	0.00353	8	0.441
	C	.72232	.72600	0.00368	8	0.460
	D	.69725	.70120	0.00395	8	0.494
	E	.71784	.72149	0.00365	8	0.456
12 %	A	.68455	.68806	0.00351	8	0.439
	B	.70009	.70422	0.00413	8	0.516
	C	.72123	.72519	0.00396	8	0.495
	D	.71161	.71522	0.00361	8	0.451
	E	.71338	.71703	0.00365	8	0.456
16 %	A	.71271	.71693	0.00422	8	0.528
	B	.70769	.71144	0.00375	8	0.469
	C	.71200	.71619	0.00419	8	0.524
	D	.69248	.69655	0.00407	8	0.509
	E	.70958	.71287	0.00329	8	0.411

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: May 23, 2023 at 1320
Date and Time Test Terminated: May 29, 2023 at 1500

Concentration: Control													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	6	5	5	5	0	4	6	5	4	6	46	10	4.60
4	9	0	0	0	3	9	0	0	0	10	31	10	3.10
5	0	10	12	10	10	0	11	9	11	0	73	10	7.30
6	14	14	15	17	12	16	12	15	15	13	143	10	14.3
7													
8													
TOTAL	29	29	32	32	25	29	29	29	30	29	293	10	29.3

Concentration: 5 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	5	4	5	4	3	4	6	5	6	6	48	10	4.80
4	9	0	0	10	0	9	9	8	0	0	45	10	4.50
5	0	11	11	0	10	0	0	0	11	11	54	10	5.40
6	14	16	15	16	14	15	16	11	15	16	148	10	14.8
7													
8													
TOTAL	28	31	31	30	27	28	31	24	32	33	295	10	29.5

Concentration: 7 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	4	4	5	6	4	5	6	0	4	6	44	10	4.40
4	0	0	0	0	0	9	0	5	0	10	24	10	2.40
5	12	10	9	9	10	0	11	10	11	0	82	10	8.20
6	16	17	15	15	14	17	15	13	16	16	154	10	15.4
7													
8													
TOTAL	32	31	29	30	28	31	32	28	31	32	304	10	30.4

Appendix A1: Test 1002.0

Ceriodaphnia dubia Survival and Reproduction

Date and Time Test Initiated: May 23, 2023 at 1320
Date and Time Test Terminated: May 29, 2023 at 1500

Concentration: 9 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	5	6	4	6	4	6	5	4	5	6	51	10	5.10
4	0	0	0	10	0	0	0	10	0	0	20	10	2.00
5	11	10	9	0	9	7	9	0	10	11	76	10	7.60
6	17	16	18	16	13	17	14	15	18	14	158	10	15.8
7													
8													
TOTAL	33	32	31	32	26	30	28	29	33	31	305	10	30.5

Concentration: 12 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	4	4	6	5	3	5	6	6	5	6	50	10	5.00
4	0	0	0	0	0	10	0	9	0	9	28	10	2.80
5	10	10	12	10	11	0	9	0	11	0	73	10	7.30
6	16	15	18	14	14	15	16	16	17	16	157	10	15.7
7													
8													
TOTAL	30	29	36	29	28	30	31	31	33	31	308	10	30.8

Concentration: 16 %													
Day	Replicate										No. of Young	No. of Adults	Young per Adult
	1	2	3	4	5	6	7	8	9	10			
1	0	0	0	0	0	0	0	0	0	0	0	10	0.00
2	0	0	0	0	0	0	0	0	0	0	0	10	0.00
3	5	6	4	0	0	5	5	0	4	5	34	10	3.40
4	0	0	0	5	6	0	0	9	0	0	20	10	2.00
5	11	10	12	10	0	8	10	10	11	6	88	10	8.80
6	16	15	16	0	11	14	15	0	14	16	117	10	11.7
7													
8													
TOTAL	32	31	32	15	17	27	30	19	29	27	259	10	25.9

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Survival

		Transformation of Data		Transform: Arc Sin(Square Root(Y))
Group	Identification	Rep	Value	Transformed
1	Control	1	1.00000	1.39310
1	Control	2	0.87500	1.20940
1	Control	3	1.00000	1.39310
1	Control	4	1.00000	1.39310
1	Control	5	1.00000	1.39310
2	5 %	1	1.00000	1.39310
2	5 %	2	1.00000	1.39310
2	5 %	3	1.00000	1.39310
2	5 %	4	1.00000	1.39310
2	5 %	5	0.87500	1.20940
3	7 %	1	1.00000	1.39310
3	7 %	2	1.00000	1.39310
3	7 %	3	1.00000	1.39310
3	7 %	4	0.87500	1.20940
3	7 %	5	1.00000	1.39310
4	9 %	1	1.00000	1.39310
4	9 %	2	1.00000	1.39310
4	9 %	3	1.00000	1.39310
4	9 %	4	1.00000	1.39310
4	9 %	5	1.00000	1.39310
5	12 %	1	0.87500	1.20940
5	12 %	2	1.00000	1.39310
5	12 %	3	1.00000	1.39310
5	12 %	4	1.00000	1.39310
5	12 %	5	1.00000	1.39310
6	16 %	1	1.00000	1.39310
6	16 %	2	1.00000	1.39310
6	16 %	3	1.00000	1.39310
6	16 %	4	1.00000	1.39310
6	16 %	5	1.00000	1.39310

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Survival

Shapiro - Wilk's Test for Normality		Transform: Arc Sin(Square Root(Y))
D = 0.108		
W = 0.5958		
Critical W = 0.9	(alpha = 0.01, N = 30)	
Critical W = 0.927	(alpha = 0.05, N = 30)	

Data FAIL normality test (alpha = 0.01).

Steel's Many-One Rank Test					Transform: Arc Sin(Square Root(Y))
Ho:Control<Treatment					
Group	Identification	Rank Sum	Critical Value	DF	Sig 0.05
1	Control				
2	5 %	27.50	16.00	5.00	
3	7 %	27.50	16.00	5.00	
4	9 %	30.00	16.00	5.00	
5	12 %	27.50	16.00	5.00	
6	16 %	30.00	16.00	5.00	

Critical values are 1 tailed (k=5)

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Growth

Shapiro - Wilk's Test for Normality		No Transformation
D = 0.04429		
W = 0.973		
Critical W = 0.9	(alpha = 0.01, N = 30)	
Critical W = 0.927	(alpha = 0.05, N = 30)	
Data PASS normality test (alpha = 0.01).		

Bartlett's Test for Homogeneity of Variance		No Transformation
Calculated B1 statistic = 4.143		
Critical B = 15.086	(alpha = 0.01, df = 5)	
Data PASS B1 homogeneity test at 0.01 level.		

Appendix A2: Statistics

Pimephales promelas (Fathead minnow) Growth

ANOVA Table				No Transformation
SOURCE	DF	SS	MS	F
Between	5	0.01139	0.002278	1.235
Within (Error)	24	0.04429	0.001845	
Total	29	0.05568		

Critical F = 3.9 (alpha = 0.01, df = 5,24)
2.62 (alpha = 0.05, df = 5,24)

Since F < Critical F FAIL TO REJECT Ho: All equal (alpha = 0.05)

Dunnett's Test - Table 1 of 2					No Transformation
Ho:Control<Treatment					
Group	Identification	Transformed Mean	Mean In Original Units	T Stat	Sig 0.05
1	Control	0.4738	0.4738		
2	5 %	0.4488	0.4488	0.9203	
3	7 %	0.4276	0.4276	1.701	
4	9 %	0.4592	0.4592	0.5374	
5	12 %	0.4714	0.4714	0.08835	
6	16 %	0.4882	0.4882	-0.5301	

Dunnett's critical value = 2.36 (1 Tailed, alpha = 0.05, df = 5,24)

Dunnett's Test - Table 2 of 2					No Transformation
Ho:Control<Treatment					
Group	Identification	Num of Reps	Min Sig Diff (In Orig. Units)	% of Control	Difference From Control
1	Control	5			
2	5 %	5	0.06411	13.5	0.025
3	7 %	5	0.06411	13.5	0.0462
4	9 %	5	0.06411	13.5	0.0146
5	12 %	5	0.06411	13.5	0.0024
6	16 %	5	0.06411	13.5	-0.0144

Appendix A2: Statistics

Ceriodaphnia dubia Survival

Fisher's Exact Test

Identification	Alive	Dead	Total Animals
Control	10	0	10
5 %	10	0	10
Total	20	0	20

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

Fisher's Exact Test

Identification	Alive	Dead	Total Animals
Control	10	0	10
7 %	10	0	10
Total	20	0	20

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

Fisher's Exact Test

Identification	Alive	Dead	Total Animals
Control	10	0	10
9 %	10	0	10
Total	20	0	20

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

Fisher's Exact Test

Identification	Alive	Dead	Total Animals
Control	10	0	10
12 %	10	0	10
Total	20	0	20

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

Appendix A2: Statistics

Ceriodaphnia dubia Survival

Fisher's Exact Test			
Identification	Alive	Dead	Total Animals
Control	10	0	10
16 %	10	0	10
Total	20	0	20

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

Summary of Fisher's Exact Test				
Group	Identification	Exposed	Dead	Sig 0.05
0	Control	10	0	
1	5 %	10	0	
2	7 %	10	0	
3	9 %	10	0	
4	12 %	10	0	
5	16 %	10	0	

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

Kolmogorov Test for Normality	No Transformation
D = 0.1197 D* = 0.9391 Critical D* = 1.035 (alpha = 0.01, N = 60) Data PASS normality test (alpha = 0.01).	

Bartlett's Test for Homogeneity of Variance	No Transformation
Calculated B1 statistic = 26.62 Critical B = 15.086 (alpha = 0.01, df = 5) Data FAIL B1 homogeneity test at 0.01 level.	

Steel's Many-One Rank Test					No Transformation
Ho:Control<Treatment					
Group	Identification	Rank Sum	Critical Value	DF	Sig 0.05
1	Control				
2	5 %	108.50	75.00	10.00	
3	7 %	119.50	75.00	10.00	
4	9 %	122.50	75.00	10.00	
5	12 %	123.00	75.00	10.00	
6	16 %	94.50	75.00	10.00	
Critical values are 1 tailed (k=5)					

Appendix A2: Statistics

Ceriodaphnia dubia Reproduction

Dunnett's Test for PMSD Calculation

ANOVA Table				No Transformation
SOURCE	DF	SS	MS	F
Between	5	164.4	32.88	3
Within (Error)	54	592	10.96	
Total	59	756.4		

Critical F = 3.38 (alpha = 0.01, df = 5,54)
2.38 (alpha = 0.05, df = 5,54)

Since F > Critical F REJECT Ho: All equal (alpha = 0.05)

Dunnett's Test - Table 1 of 2					No Transformation
Ho:Control<Treatment					
Group	Identification	Transformed Mean	Mean In Original Units	T Stat	Sig 0.05
1	Control	29.3	29.3		
2	5 %	29.5	29.5	-0.1351	
3	7 %	30.4	30.4	-0.743	
4	9 %	30.5	30.5	-0.8105	
5	12 %	30.8	30.8	-1.013	
6	16 %	25.9	25.9	2.296	

Dunnett's critical value = 2.31 (1 Tailed, alpha = 0.05, df [used] = 5,40) (Actual df = 5,54)

Dunnett's Test - Table 2 of 2					No Transformation
Ho:Control<Treatment					
Group	Identification	Num of Reps	Min Sig Diff (In Orig. Units)	% of Control	Difference From Control
1	Control	10			
2	5 %	10	3.42	11.7	-0.2
3	7 %	10	3.42	11.7	-1.1
4	9 %	10	3.42	11.7	-1.2
5	12 %	10	3.42	11.7	-1.5
6	16 %	10	3.42	11.7	3.4

Lower PMSD Bound Test for Ceriodaphnia dubia

Concentration	Reproduction	Relative Difference from Control	Pass/Fail
Control	29.3	-	
5 %	29.5	-0.683	PASS
7 %	30.4	-3.75	PASS
9 %	30.5	-4.10	PASS
12 %	30.8	-5.12	PASS
16 %	25.9	11.6	PASS

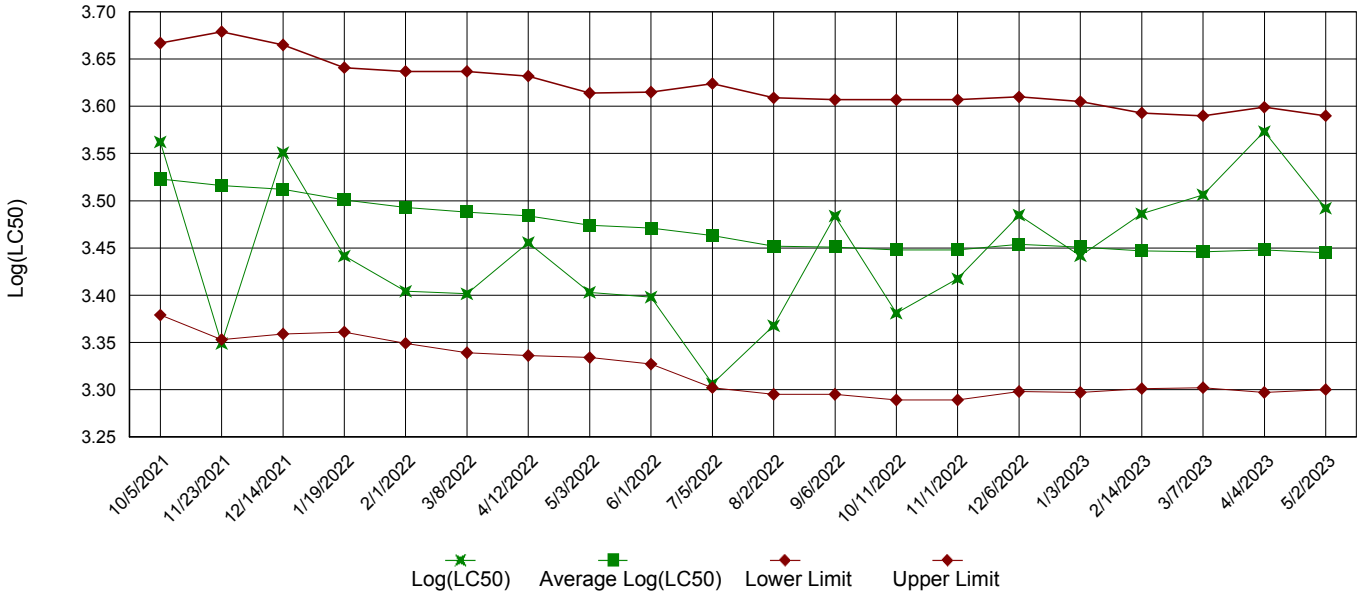
Limit = 13

NOEC = 16 %
LOEC = 16 %

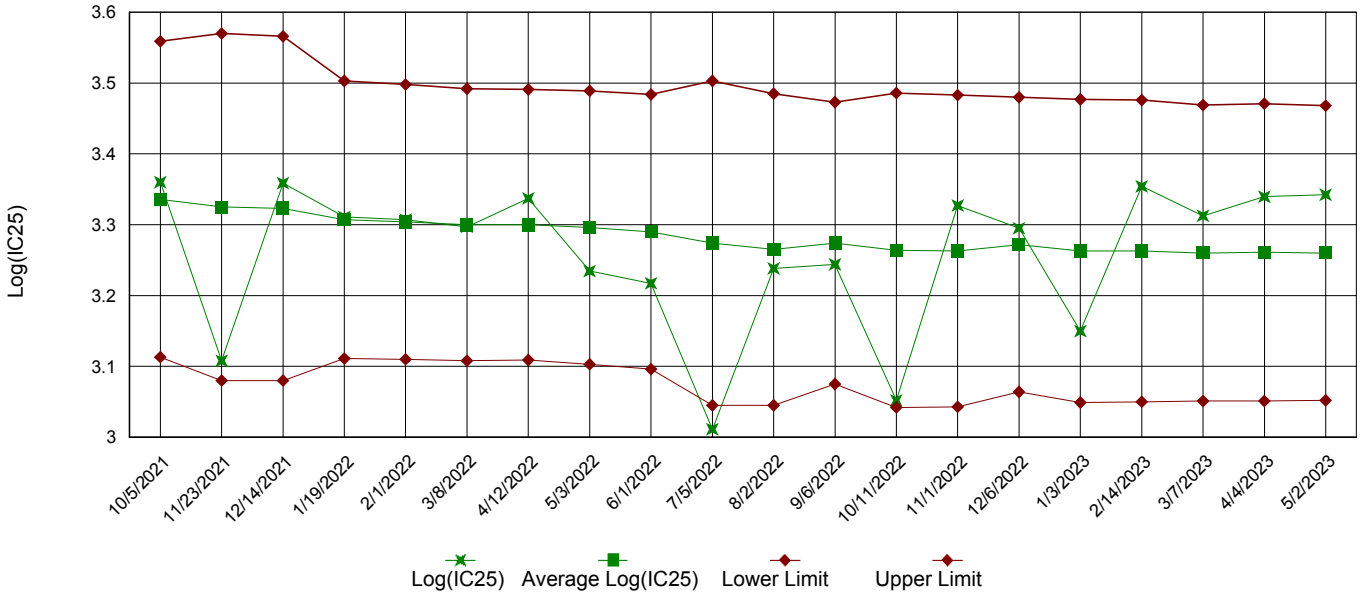
Appendix A3: Test 1000.0

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

LC50 Survival Data



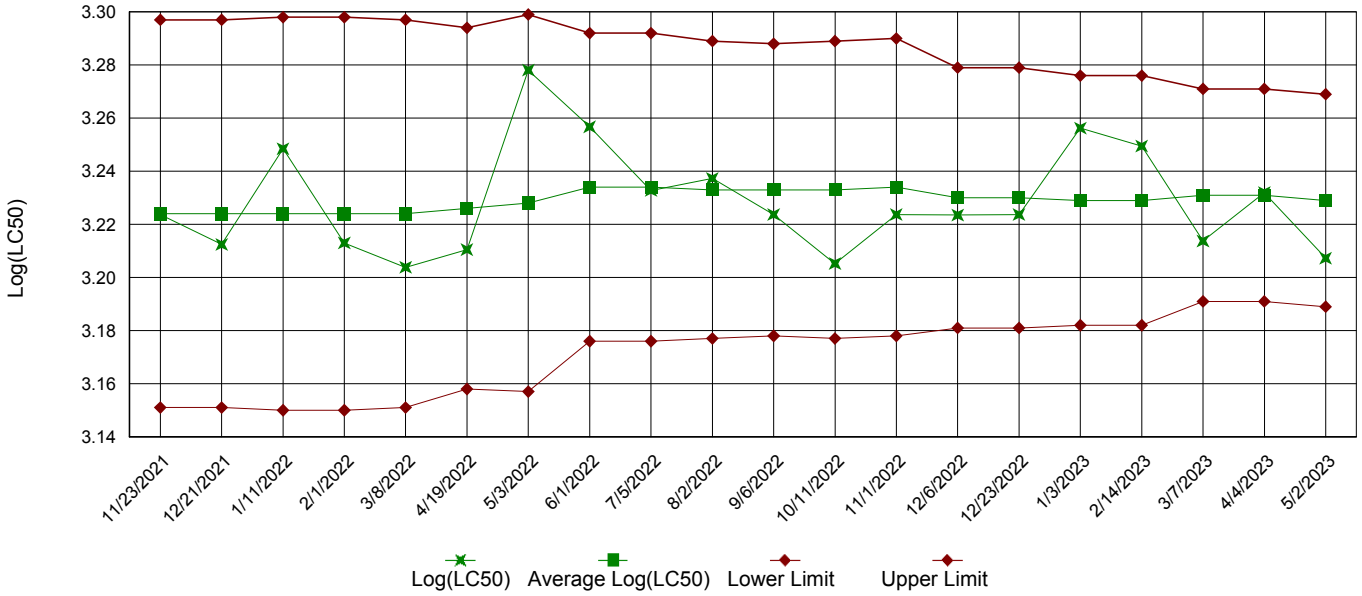
IC25 Growth Data



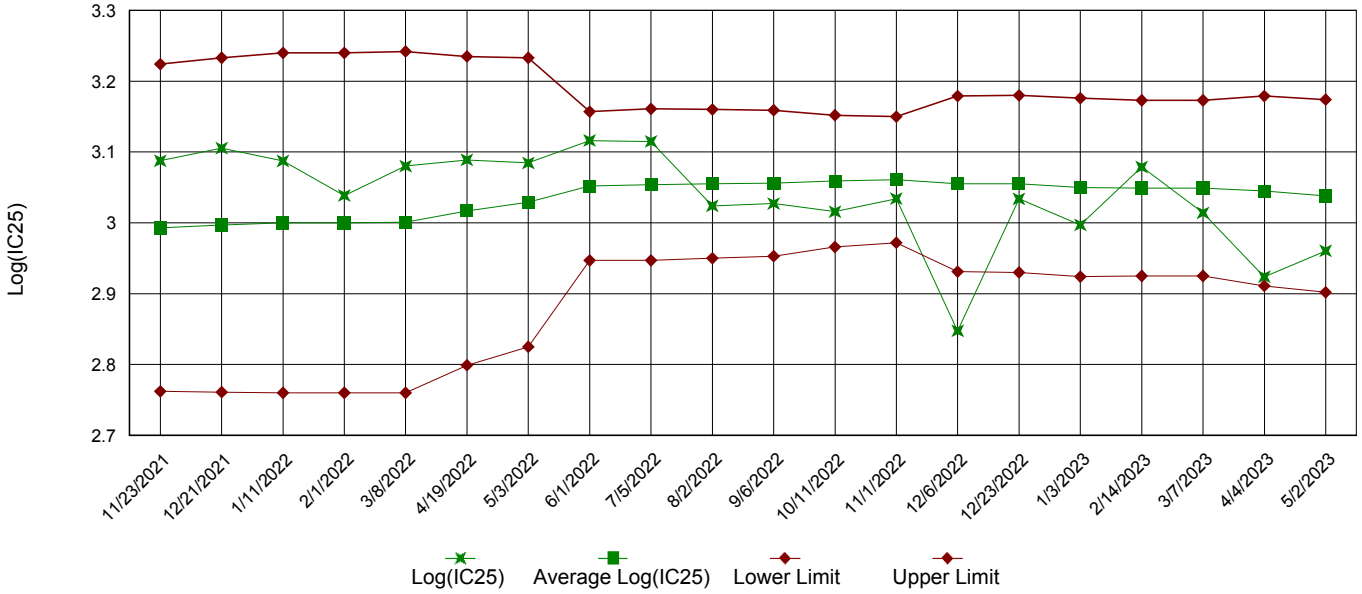
Appendix A3: Test 1002.0

Chronic Reference Toxicant, *Ceriodaphnia dubia*

LC50 Survival Data



IC25 Reproduction Data



Appendix B: Test 1000.0

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

Permittee: Malvern Water Works

NPDES No.: AR0034126 AFIN 30-00040

Date and Time Test Initiated: May 23, 2023 at 1342
Date and Time Test Terminated: May 30, 2023 at 1307
Dilution water used:

DATA TABLE FOR SURVIVAL

Effluent Conc. %	Percent Survival in replicate chambers A	B	C	D	E	Mean percent survival 24 hr	48 hr	7 days	CV%
Control	100	87.5	100	100	100	100	100	97.5	5.73
5 %	100	100	100	100	87.5	100	100	97.5	5.73
7 %	100	100	100	87.5	100	100	100	97.5	5.73
9 %	100	100	100	100	100	100	100	100	0.00
12 %	87.5	100	100	100	100	97.5	97.5	97.5	5.73
16 %	100	100	100	100	100	100	100	100	0.00

DATA TABLE FOR GROWTH

Effluent Conc. %	Average dry weight, mg replicate chambers A	B	C	D	E	Mean dry weight, mg	CV%
Control	0.560	0.464	0.454	0.412	0.479	0.474	11.4
5 %	0.401	0.534	0.455	0.450	0.404	0.449	12.0
7 %	0.450	0.428	0.416	0.374	0.470	0.428	8.52
9 %	0.445	0.441	0.460	0.494	0.456	0.459	4.56
12 %	0.439	0.516	0.495	0.451	0.456	0.471	6.91
16 %	0.528	0.469	0.524	0.509	0.411	0.488	10.0

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

Appendix B: Test 1000.0

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

1. Steel's Many-One Rank Test:

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

   a.) LOW FLOW OR CRITICAL DILUTION _____YES ___X___ NO
   b.) 1/2 LOW FLOW DILUTION _____YES _____ NO

2. Dunnett's Test:

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

   a.) LOW FLOW OR CRITICAL DILUTION _____YES ___X___ NO
   b.) 1/2 LOW FLOW DILUTION _____YES _____ NO

3. If you answered NO to 1.a) enter [0] otherwise enter [1]: 0 (TLP6C)

4. If you answered NO to 2.a) enter [0] otherwise enter [1]: 0 (TGP6C)

5. NOEC Pimephales Lethality: 16 % (TOP6C)

6. LOEC Pimephales Lethality: 16 % (TXP6C)

7. NOEC Pimephales Sublethality: 16 % (TPP6C)

8. LOEC Pimephales Sublethality: 16 % (TYP6C)

9. Coefficient of variation for Pimephales growth: 11.4 (TQP6C)

10. Sublethality for this test: 16 % (51714 or 51714S)

Appendix B: Test 1000.0

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

PERMITTEE: Malvern Water Works
NPDES NO.: AR0034126 AFIN 30-00040
CONTACT: Mr. John Davis
ANALYST: 280, 343, 357, 358

Test Initiated: DATE: May 23, 2023 TIME: 1342
Test Terminated: DATE: May 30, 2023 TIME: 1307

DILUTION Control	DAY 1	2	3	4	5	6	7
D.O. Initial	8.0	8.2	8.5	8.2	7.9	8.2	8.2
Final	6.2	6.2	5.7	6.0	7.3	5.6	6.4
pH Initial	7.3	7.5	7.5	7.5	7.6	7.6	7.6
Final	7.0	7.1	7.0	7.2	7.3	7.1	7.0

DILUTION 5 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.8	8.4	8.5	8.2	8.2	8.2	8.2
Final	6.6	6.2	6.2	6.0	6.0	6.1	6.4
pH Initial	7.4	7.5	7.5	7.5	7.5	7.6	7.6
Final	7.1	7.1	7.0	7.1	7.1	7.1	6.9

DILUTION 7 %	DAY 1	2	3	4	5	6	7
D.O. Initial	8.0	8.4	8.5	8.2	8.2	8.2	8.2
Final	6.6	5.8	6.4	5.9	6.0	5.8	6.3
pH Initial	7.4	7.5	7.5	7.5	7.5	7.6	7.6
Final	7.0	7.1	7.0	7.1	7.1	7.1	6.9

DILUTION 9 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.9	8.5	8.4	8.2	8.1	8.2	8.2
Final	6.7	6.0	5.9	5.9	5.7	5.8	6.2
pH Initial	7.4	7.4	7.6	7.5	7.6	7.6	7.6
Final	7.0	7.1	7.0	7.0	7.0	7.0	6.9

DILUTION 12 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.9	8.2	8.3	8.2	8.3	7.9	8.1
Final	6.3	5.9	5.9	5.9	5.7	5.7	6.1
pH Initial	7.4	7.4	7.6	7.5	7.6	7.6	7.6
Final	7.0	7.1	7.0	7.1	7.0	7.0	6.8

DILUTION 16 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.9	8.5	8.5	8.1	8.4	8.1	8.0
Final	6.2	5.9	6.2	5.8	5.7	5.6	6.2
pH Initial	7.4	7.4	7.5	7.4	7.5	7.5	7.5
Final	7.0	7.0	7.0	7.0	7.0	6.9	6.8

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
16	22	130	0.060	MWW Bio #1 23-MAY-23
16	21	130	0.060	MWW Bio #2 24-MAY-23
17	21	140	0.070	MWW Bio #3 26-MAY-23

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
31	47	170	<0.05	192-1716-A-1
31	45	160	<0.05	192-1841-A-1

Appendix B: Test 1002.0

SUMMARY REPORTING FORMS
CHRONIC BIOMONITORING
Ceriodaphnia dubia
SURVIVAL AND REPRODUCTION

Permittee: Malvern Water Works

NPDES No.: AR0034126 AFIN 30-00040

Date and Time Test Initiated: May 23, 2023 at 1320
Date and Time Test Terminated: May 29, 2023 at 1500
Dilution water used:

PERCENT SURVIVAL

Time of Reading	Control	Percent Effluent 5 %	7 %	9 %	12 %	16 %
24 hour	100	100	100	100	100	100
48 hour	100	100	100	100	100	100
6 day	100	100	100	100	100	100

NUMBER OF YOUNG PRODUCED PER FEMALE @ 6 DAYS

Replicates	Control	Percent Effluent 5 %	7 %	9 %	12 %	16 %
A	29	28	32	33	30	32
B	29	31	31	32	29	31
C	32	31	29	31	36	32
D	32	30	30	32	29	15
E	25	27	28	26	28	17
F	29	28	31	30	30	27
G	29	31	32	28	31	30
H	29	24	28	29	31	19
I	30	32	31	33	33	29
J	29	33	32	31	31	27
Mean per Adult	29.3	29.5	30.4	30.5	30.8	25.9
Mean per Surviving Adult	29.3	29.5	30.4	30.5	30.8	25.9
CV %	6.64	9.21	5.19	7.45	7.47	24.9

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

Appendix B: Test 1002.0

SUMMARY REPORTING FORMS
CHRONIC BIOMONITORING
Ceriodaphnia dubia
SURVIVAL AND REPRODUCTION

1. Fisher's Exact Test:

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

   a.) LOW FLOW OR CRITICAL DILUTION _____YES ___X___ NO
   b.) 1/2 LOW FLOW DILUTION _____YES _____ NO

2. Steel's Many-One Rank Test:

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

   a.) LOW FLOW OR CRITICAL DILUTION _____YES ___X___ NO
   b.) 1/2 LOW FLOW DILUTION _____YES _____ NO

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

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

5. NOEC Ceriodaphnia Lethality: 16 % (TOP3B)

6. LOEC Ceriodaphnia Lethality: 16 % (TXP3B)

7. NOEC Ceriodaphnia Sublethality: 16 % (TPP3B)

8. LOEC Ceriodaphnia Sublethality: 16 % (TYP3B)

9. Coefficient of variation for Ceriodaphnia Reproduction: 7.47 (TQP3B)

10. Sublethality for this test: 16 % (51710 or 51710Q)

Appendix B: Test 1002.0

CHRONIC TOXICITY SUMMARY FORM
Ceriodaphnia dubia
CHEMICAL PARAMETERS CHART

PERMITTEE: Malvern Water Works
NPDES NO.: AR0034126 AFIN 30-00040
CONTACT: Mr. John Davis
ANALYST: 280, 343, 357, 358

Test Initiated: DATE: May 23, 2023 TIME: 1320
Test Terminated: DATE: May 29, 2023 TIME: 1500

DILUTION Control	DAY 1	2	3	4	5	6	7
D.O. Initial	8.0	8.2	8.5	8.2	7.9	8.2	8.2
Final	8.3	8.4	8.1	8.1	8.0	7.9	--
pH Initial	7.3	7.5	7.5	7.5	7.6	7.6	7.6
Final	7.8	7.9	7.8	7.8	7.8	7.7	--

DILUTION 5 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.8	8.4	8.5	8.2	8.2	8.2	8.2
Final	8.2	8.3	8.1	8.0	8.0	7.9	--
pH Initial	7.4	7.5	7.5	7.5	7.5	7.6	7.6
Final	7.7	7.8	7.8	7.8	7.8	7.7	--

DILUTION 7 %	DAY 1	2	3	4	5	6	7
D.O. Initial	8.0	8.4	8.5	8.2	8.2	8.2	8.2
Final	8.3	8.2	8.1	8.2	8.0	7.9	--
pH Initial	7.4	7.5	7.5	7.5	7.5	7.6	7.6
Final	7.7	7.8	7.7	7.8	7.8	7.7	--

DILUTION 9 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.9	8.5	8.4	8.2	8.1	8.2	8.2
Final	8.2	8.3	8.2	8.0	8.0	7.9	--
pH Initial	7.4	7.4	7.6	7.5	7.6	7.6	7.6
Final	7.7	7.8	7.8	7.7	7.7	7.7	--

DILUTION 12 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.9	8.2	8.3	8.2	8.3	7.9	8.1
Final	8.2	8.2	8.3	8.1	8.0	7.8	--
pH Initial	7.4	7.4	7.6	7.5	7.6	7.6	7.6
Final	7.7	7.8	7.7	7.8	7.7	7.7	--

DILUTION 16 %	DAY 1	2	3	4	5	6	7
D.O. Initial	7.9	8.5	8.5	8.1	8.4	8.1	8.0
Final	8.3	8.3	8.2	8.1	8.0	7.9	--
pH Initial	7.4	7.4	7.5	7.4	7.5	7.5	7.5
Final	7.7	7.8	7.7	7.8	7.8	7.7	--

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
16	22	130	0.060	MWW Bio #1 23-MAY-23
16	21	130	0.060	MWW Bio #2 24-MAY-23
17	21	140	0.070	MWW Bio #3 26-MAY-23

Alkalinity	Hardness	Conductivity	Chlorine	Sample ID
31	47	170	<0.05	192-1716-A-1
31	45	160	<0.05	192-1841-A-1

AMERICAN INTERPLEX CORPORATION
LABORATORIES

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

192-2047 COC

Client: Malvern Waste Water
Project Reference:
Project Manager:
Sampled By: Devon Baugh

PO No.

MATRIX: W A T E R | S O I L

NO OF BOTTLES

ANALYSES REQUESTED: Biomonitoring | P, NO3+NO2-N

AIC CONTROL NO: LIMS 2744584
AIC PROPOSAL NO: TALS 2047
Carrier:
Received on ice? Yes (No) Temp. 1.7 °C

AIC No.	Sample Identification	Date/Time Collected	GRAB	COMP	WATER	SOIL		BOTTLES	Biomonitoring	P, NO3+NO2-N	Remarks
1	Mww Bio #1	5.23.23 8:05Am		X				1	X		
	Mww P, NO3+NO2-N	5.23.23 10:30Am	X					1		X	TALS: 2049
		Container Type									Field pH calibration
		Preservative									on _____ @ _____
											Buffer:

G = Glass P = Plastic V = VOA vials H = HCl to pH2 T = Sodium Thiosulfate
NO = none S = Sulfuric acid pH2 N = Nitric acid pH2 B = NaOH to pH12 Z = Zinc acetate A=$(NH_4)_2SO_4$, NH_4OH

Turnaround Time Requested: (Please circle)
NORMAL or EXPEDITED IN ____ DAYS
Expedited results requested by: _____
Who should AIC contact with questions:
Phone:_____ Fax:
Report Attention to:
Report Address to:

Email Address:

Relinquished By:	Date/Time	Received By:	Date/Time
Devon Baugh	5.23.23 11:35Am		
Relinquished By:	Date/Time	Received in Lab By:	Date/Time
			9-23-23 1137

Comments:

9/2014 FORM 0060

6/2/2023

AMERICAN INTERPLEX CORPORATION
LABORATORIES

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

Client: Malvern Waste Water
Project Reference:
Project Manager:
Sampled By: Devan Baugh
PO No.
MATRIX

AIC No	Sample Identification	Date/Time Collected	GRAB	COMP	WATER	SOIL	NO OF BOTTLES	Biomonitoring
2	MWW Bio #2	5-24-23 9:15Am		X			1	X
		Container Type						
		Preservative						

AIC CONTROL NO: 274584 / 2047
AIC PROPOSAL NO:
Carrier:
Received on ice? Yes No Temp. 1.9 °C
Remarks
Field pH calibration on _____ @ _____
Buffer:

G = Glass P = Plastic V = VOA vials H = HCl to pH2 T = Sodium Thiosulfate
NO = none S = Sulfuric acid pH2 N = Nitric acid pH2 B = NaOH to pH12 Z = Zinc acetate A=$(NH_4)_2SO_4$, NH_4OH

Turnaround Time Requested: (Please circle)
NORMAL or EXPEDITED IN ____ DAYS
Expedited results requested by: _____
Who should AIC contact with questions: _____
Contact Phone: _____
Report Attention to: _____
Email Address:

Relinquished By:	Date/Time	Received By:	Date/Time
Devan Baugh	5-24-23 10:35Am		
Relinquished By:	Date/Time	Received in Lab By: DANNY BIZOU	Date/Time 5-24-23 1035

Comments.

FORM 0060

AMERICAN INTERPLEX CORPORATION
LABORATORIES

CHAIN OF CUSTODY / ANALYSIS REQUEST FORM

PAGE OF

Client: Malvern Waste Water
Project Reference:
Project Manager:
Sampled By: Devan Baugh

PO No.

MATRIX: W A T E R | S O I L

NO OF BOTTLES

ANALYSES REQUESTED: Biomonitoring

AIC CONTROL NO:
AIC PROPOSAL NO:
Carrier:
Received on ice? Yes No Temp. 0.7°C °C

AIC No	Sample Identification	Date/Time Collected	GRAB	COMP	WATER	SOIL		NO OF BOTTLES	Biomonitoring	Remarks
	B.O #3	5/24/23 7:15AM		X				1	X	
		Container Type								Field pH calibration
		Preservative								on _____ @ _____
										Buffer:

G = Glass P = Plastic V = VOA vials H = HCl to pH2 T = Sodium Thiosulfate
NO = none S = Sulfuric acid pH2 N = Nitric acid pH2 B = NaOH to pH12 Z = Zinc acetate A=$(NH_4)_2SO_4$, NH_4OH

Turnaround Time Requested: (Please circle)
NORMAL or EXPEDITED IN ____ DAYS
Expedited results requested by: _____
Who should AIC contact with questions: _____
Contact Phone: _____
Report Attention to: _____
_____
_____
Email Address:

Relinquished By:	Date/Time	Received By:	Date/Time
Devan Baugh	5-26-23 8:57AM		
Relinquished By:	Date/Time	Received in Lab By:	Date/Time
		Cage Hayes	5-26-23 0857

Comments.

FORM 0060