

Arkansas Analytical, Inc.

Toxicity Test Results
City of DeQueen
NPDES PERMIT NUMBER: AR0021733
Second Quarter 2012
AFIN # 67-00023

Fathead Minnow, *Pimephales promelas*, Larval Survival and Growth Test
Test 1000.0

Ceriodaphnia dubia, Survival and Reproduction Test
Test 1002.0

Prepared for: **Mr. Mike Sims**
City of DeQueen
P.O. Box 730
DeQueen, Arkansas 71832

Prepared by: **Arkansas Analytical, Inc.**
11701 I-30, Bldg 1, Suite 115
Little Rock, Arkansas 72209
Lab Number K1206003

Monday, June 25, 2012

Introduction

This report contains test results for toxicity testing for the City of DeQueen. The NPDES permit number is AR0021733. The facility is located 1/8 mile south from intersection of Coulter Ave. and south of 9th Street on Philip Cox Blvd, in Section 36, Township 8 South, Range 32 West in Sevier County, Arkansas. The discharge is to receiving waters named: an unnamed ditch around pond to Bear Creek to Little River to Red River in Segment 1C of the Red River Basin.

The permit requires chronic biomonitoring testing quarterly for both *Ceriodaphnia dubia* and *Pimephales promelas*. The test results in this report represent the testing for the second quarter of 2012.

Plant Operations

To be provided by permittee.

Source of Effluent and Dilution Water

Effluent samples were collected as follows:

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-5-12, 1000	6-6-12, 1000
Sample #2:	6-6-12, 1000	6-7-12, 1000
Sample #3:	6-10-12, 1000	6-11-12, 1000

The samples were composites collected at the final discharge from City of DeQueen Wastewater Plant outfall.

The following information was collected upon immediate receipt of the samples at the laboratory:

Sample Receiving Information:	Date, Time Sample(s) Received	Temperature Upon Receipt (°C)
Sample #1:	6-7-12, 1145	1
Sample #2:	6-8-12, 0920	3
Sample #3:	6-12-12, 1100	3

Chain of custody documentation is located in Appendix A.

The permit designates the receiving water to be used as dilution water for the toxicity tests. Due to its earlier characterization as toxic, synthetic dilution water was substituted.

The dilution water used in the toxicity tests was moderately hard synthetic. It was prepared using Elga Maxima ultra pure water according to EPA specifications. Each batch was analyzed for pH, hardness, total alkalinity, and conductivity. Results are provided in Appendix B.

Dilution Series

Five dilutions in addition to a control (0% effluent) were used in the toxicity tests. The dilutions, which were made with synthetic water, were 32%, 42%, 56%, 75%, and 100%. The low-flow effluent concentration (**critical dilution**) was defined as **100% effluent**.

Test Methods

EPA Method 1000.0, Fathead Minnow, *Pimephales promelas*, Larval Survival and Growth Test, was used in this bioassay. Larvae are exposed in a static renewal system for seven days and the results are based on the survival and growth (increase in weight) of the larvae. The alternate method suggested in the method (11.3.4.5) for combating pathogen interference, was run in place of the original fathead minnow test. The test chambers were 30 ml plastic cups with 20 ml of test solution. Each chamber contained 2 organisms. The total number of fish was 40 per test solution. The fish were then combined to perform growth analysis. The test temperature was 25 degrees Centigrade. Raw data and statistics are provided in Appendix C.

EPA Method 1002.0, Cladoceran, *Ceriodaphnia dubia*, Survival and Reproduction Test, was also used. Neonates are exposed in a static renewal system until at least 60% of the control organisms have produced a third brood. Results are based on the survival and reproduction of the organisms. One neonate was placed in each of ten replicate chambers using a randomizing template. Test chambers were 30 ml plastic cups filled with 15 ml of test solution. The test temperature was 25 degrees Centigrade. Raw data and statistics are provided in Appendix D.

Test Organisms

The organisms used in Test 1000.0 were < 24 hour old Fathead Minnows, *Pimephales promelas*, which were purchased from Aquatox; a copy of the organism history is provided in Appendix E.

The organisms used in Test 1002.0 were < 24 hour old *Ceriodaphnia dubia* neonates, (all born within the same eight hours), obtained from an in-house culture. An organism history is provided in Appendix E.

Quality Assurance

Test Acceptability

TEST ACCEPTANCE CRITERIA for *Ceriodaphnia dubia*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	90%	X	
Average of 15 or more young per surviving female	15.6	X	
At least 60% of surviving females should have produced 3 broods	88.9%	X	
The percent coefficient of variation between replicates must be 40% or less for the young of surviving females	16.1	X	

TEST ACCEPTANCE CRITERIA for *Pimephales promelas*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	100%	X	
The percent coefficient of variation between replicates must be 40% or less for survival	0.00	X	
Minimum of 0.25 mg average dry weight of surviving controls	0.381	X	
The percent coefficient of variation between replicates must be 40% or less for growth	12.4	X	

Reference Toxicant

The reference toxicant used was Potassium Chloride prepared in-house. The tests were performed using moderately hard synthetic as dilution water. The results of the reference toxicant were:

REFERENCE TOXICANT

<i>Ceriodaphnia dubia</i> 5/8-15/12		<i>Pimephales promelas</i> 5/8-15/12	
NOEC Survival:	250 ppm KCl	NOEC Survival:	500 ppm KCl
LOEC Survival:	500 ppm KCl	LOEC Survival:	1000 ppm KCl
NOEC Reproduction:	250 ppm KCl	NOEC Growth:	500 ppm KCl
LOEC Reproduction:	500 ppm KCl	LOEC Growth:	1000 ppm KCl

Quality Assurance charts are provided in Appendix F.

Summary of Results
City of DeQueen

<i>Ceriodaphnia dubia</i>		<i>Pimephales promelas</i>	
NOEC / LOEC Survival	100% / NA	NOEC / LOEC survival	100% / NA
NOEC / LOEC Reproduction	100% / NA	NOEC / LOEC growth	100% / NA
Mean number of neonates (critical dilution)	14.3	%CV survival (critical dilution)	0.00
%CV Reproduction (critical dilution)	19.7	Mean dry weight (critical dilution) in milligrams	0.423
		%CV growth (critical dilution)	13.1
PMSD Reproduction	44.1	PMSD Growth	18.8

Conclusion

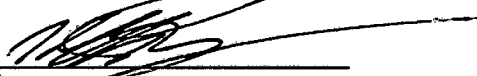
Chronic static renewal larval survival and growth test using fathead minnow, *Pimephales promelas*, (Method 1000.0)

The permit issued to the City of DeQueen, AR0021733, specifies that the **critical dilution is 100% effluent**. The effluent samples **did not** exhibit lethal and sublethal effects at the critical dilution, and, as such, **passed** both portions of the test.

Chronic static renewal survival and reproduction test using *Ceriodaphnia dubia*, (Method 1002.0)

The permit issued to the City of DeQueen, AR0021733, specifies that the **critical dilution is 100% effluent**. The effluent samples **did not** exhibit lethal effects or sublethal effects at the critical dilution, and, as such, **passed** both portions of the test.

Biomonitoring Analysts:


Kenneth Pigue


Melissa Bird

**SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
FATHEAD MINNOW LARVAE GROWTH AND SURVIVAL
*PIMEPHALES PROMELAS***

PERMITTEE: City of DeQueen

NPDES #: AR0021733

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-5-12, 1000	6-6-12, 1000
Sample #2:	6-6-12, 1000	6-7-12, 1000
Sample #3:	6-10-12, 1000	6-11-12, 1000

Test initiated (date, time): 6-7-12, 1510 Test terminated (date, time): 6-14-12, 0855

Dilution water used: Moderately Hard Synthetic

DATA TABLE FOR FATHEAD MINNOW SURVIVAL

Effluent Conc %	Percent Survival in Replicate Chambers					CV %	Mean Percent Survival		
	A	B	C	D	E		24 hours	48 hours	7 days
0%	100	100	100	100	100	0.00	100	100	100
32%	100	100	100	100	100		100	100	100
42%	100	100	100	100	100		100	100	100
56%	100	100	100	87.5	100		100	100	97.5
75%	100	100	100	100	100		100	100	100
100%	100	100	100	100	100	0.00	100	100	100

DATA TABLE FOR GROWTH OF FATHEAD MINNOWS

Effluent Conc %	A	B	C	D	E	CV%	Mean Dry Weight	
							Mean Dry Weight	CV%
0%	0.355	0.310	0.419	0.418	0.403	12.40	0.381	
32%	0.322	0.410	0.430	0.442	0.401		0.401	
42%	0.369	0.410	0.381	0.349	0.431		0.388	
56%	0.315	0.345	0.353	0.418	0.387		0.364	
75%	0.270	0.385	0.376	0.412	0.436		0.376	
100%	0.328	0.430	0.441	0.445	0.471	13.10	0.423	

Average Dry Weight in milligrams in replicate chambers
Coefficient of Variation = standard deviation / mean * 100

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
FATHEAD MINNOW LARVAE GROWTH AND SURVIVAL

Pimephales promelas

1. Dunnett's procedure or Steel's Many-One Rank Test as appropriate:

Is the mean survival at 7 days significantly different ($p=0.05$) than the control survival for:

a) LOW FLOW OR CRITICAL DILUTION, (100%) YES _____ NO X _____

2. Dunnett's Procedure

Is the mean dry weight (growth) at 7 days significantly different ($p=0.05$) than the control's dry weight (growth) for:

a) LOW FLOW OR CRITICAL DILUTION, (100%) YES _____ NO X _____

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

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

5. Enter percentage corresponding to each parameter below:

a) NOEC survival (parameter TOP6C)= _____ 100 _____ % effluent

b) NOEC growth (parameter TPP6C)= _____ 100 _____ % effluent

c) Coefficient of variation (parameter TQP6C)= _____ 13.1 _____ %

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
Ceriodaphnia dubia SURVIVAL AND REPRODUCTION

PERMITTEE: City of DeQueen

NPDES #: AR0021733

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-5-12, 1000	6-6-12, 1000
Sample #2:	6-6-12, 1000	6-7-12, 1000
Sample #3:	6-10-12, 1000	6-11-12, 1000

Test initiated (date, time): 6-7-12, 1500 Test terminated (date, time): 6-14-12, 0835

Dilution water used: Moderately Hard Synthetic

Ceriodaphnia dubia SURVIVAL AND REPRODUCTION
 NUMBER OF YOUNG PRODUCED PER FEMALE @ TEST TERMINATION
 PERCENT EFFLUENT

Replicate	0%	32%	42%	56%	75%	100%
A	19	19	26	25	19	14
B	18	25	19	14	16	20
C	12	25	13	14	21	11
D	16	x0	22	15	13	x0
E	x0	14	12	12	8	12
F	16	15	12	15	12	15
G	13	16	13	19	20	12
H	18	17	15	25	3	17
I	13	8	16	4	19	13
J	15	22	10	13	15	15
Mean	14.0	16.1	15.8	15.6	14.6	12.9
Mean/surviving female	15.6	17.9	15.8	15.6	14.6	14.3
CV%*	16.1					19.7

X= Dead Adult; M= Male (Not considered in statistics)

*Coefficient of Variation = standard deviation/ mean * 100; CV% calculation based on young per surviving female

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
Ceriodaphnia dubia SURVIVAL AND REPRODUCTION

PERMITTEE: City of DeQueen

NPDES #: AR0021733

PERCENT SURVIVAL

PERCENT EFFLUENT	0%	32%	42%	56%	75%	100%
Time of Reading: 24 HOURS	100	90	100	100	100	90
48 HOURS	100	90	100	100	100	90
Test termination	90	90	100	100	100	90

1. Fisher's Exact Test:

Is the mean survival at test termination significantly different ($p=0.05$) than the control survival for:

a) LOW FLOW OR CRITICAL DILUTION, (100%): YES _____ NO X _____

2. Dunnett's Procedure or Steel's Many One Rank Test:

Is the mean number of young produced per female significantly different ($p=0.05$) than the controls number of young per female for:

a) LOW FLOW OR CRITICAL DILUTION, (100%): YES _____ NO X _____

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

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

5. Enter percentage corresponding to each parameter below:

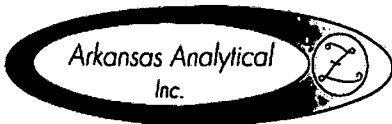
a) NOEC survival (parameter TOP3B)= 100 % effluent

b) NOEC reproduction (parameter TPP3B)= 100 % effluent

c) Coefficient of variation (parameter TQP3B)= 19.7 %

APPENDIX A

Chain of Custody Forms



11701 Interstate 30, Bldg. 1, Ste. 115
 Little Rock, AR 72209
 PHONE: 501-455-3233
 FAX: 501-455-6118

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		BILLING		Project Description		Turnaround Time		Preservation Codes:							
City of DeQueen Wastewater Plant 514 South 9th DeQueen, AR 71832		City of DeQueen Wastewater Plan P.O. Box 730 DeQueen, AR 71832		Chronic Toxicity		24 Hour 48 Hour 72 Hour Routine		1. Cool, 4 Degrees Centigrade 2. Sulfuric Acid (H ₂ SO ₄), pH < 2 3. Nitric Acid (HNO ₃), pH < 1			4. Thiosulfate for Dechlorination 5. Hydrochloric Acid(HCl) 6. Sodium Hydroxide (NaOH), pH > 12				
Attn: Mike Sims				Telephone: 870-642-5231 Fax: 870-642-3117 Email: msims@cityofdequeen.com		Preservative Code: 1 Bottle Type: P		TEST PARAMETERS						Bottle Type Code G = Glass; P = Plastic V = Septum; A = Amber	
Sampler(s) Signature				Sampler(s) Printed				Chronic Biomonitoring							Arkansas Analytical Work Order Number: K/206- 003A
Field Number	SAMPLE COLLECTION		Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION								
	Date/s	Time/s					DQ Bio								
	6-5/6-12	10:00/10:00		X		Water									
1. Relinquished by: (Signature) 		Date/Time 6-6-12 11:45am		2. Received by: (Signature) UPS		SAMPLE CONDITION UPON RECEIPT IN LAB						REMARKS / SAMPLE COMMENTS			
3. Relinquished by: (Signature) UPS		Date/Time 6/7/12 1132		4. Received by lab: (Signature) Sydney James		1. CUSTODY SEALS: <input checked="" type="checkbox"/> Yes ___ No 2. CONTAINERS CORRECT: <input checked="" type="checkbox"/> Yes ___ No 3. COC/LABELS AGREE: <input checked="" type="checkbox"/> Yes ___ No 4. PRESERVATION CONFIRMED: <input checked="" type="checkbox"/> Yes ___ No 5. RECEIVED ON ICE: <input checked="" type="checkbox"/> Yes ___ No 6. TEMPERATURE ON RECEIPT: 1°C						P.O. Number: 66776			
Revision 1												FOR COMPLETION BY LAB ONLY			



11701 Interstate 30, Bldg. 1, Ste. 115
 Little Rock, AR 72209
 PHONE: 501-455-3233
 FAX: 501-455-6118

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		BILLING		Project Description		Turnaround Time		Preservation Codes:													
City of DeQueen Wastewater Plant 514 South 9th DeQueen, AR 71832		City of DeQueen Wastewater Plant P.O. Box 730 DeQueen, AR 71832		Chronic Toxicity		24 Hour 48 Hour 72 Hour Routine		1. Cool, 4 Degrees Centigrade 2. Sulfuric Acid (H ₂ SO ₄), pH < 2 3. Nitric Acid (HNO ₃), pH < 2				4. Thiosulfate for Dechlorination 5. Hydrochloric Acid(HCl) 6. Sodium Hydroxide (NaOH), pH > 12									
Attn: Mike Sims				Reporting Information Telephone: 870-642-5231 Fax: 870-642-3117 Email: msims@cityofdequeen.com		Preservative Code: Bottle Type:		TEST PARAMETERS								Bottle Type Code G = Glass; P = Plastic V = Septum; A = Amber					
Sampler(s) Signature				Sampler(s) Printed <i>MS</i>								Chronic Biomonitoring		Arkansas Analytical Work Order Number: <i>K1206003</i> <i>B</i>							
Field Number		SAMPLE COLLECTION Date/s Time/s		Grab Comp		Number of Bottles Sample Matrix		SAMPLE IDENTIFICATION/ DESCRIPTION													
		<i>6-6/7-12 10:00/10:00</i>		<i>X</i>		<i>Water</i>		<i>DQ Bio</i>								<i>X</i>					
1. Relinquished by: (Signature)		Date/Time		2. Received by: (Signature)		SAMPLE CONDITION UPON RECEIPT IN LAB								REMARKS / SAMPLE COMMENTS							
		<i>6-7-12 11:45am</i>		<i>UPS</i>		1. CUSTODY SEALS: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No 2. CONTAINERS CORRECT: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No 3. COC/LABELS AGREE: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No 4. PRESERVATION CONFIRMED: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No 5. RECEIVED ON ICE: <input type="checkbox"/> Yes <input checked="" type="checkbox"/> No 6. TEMPERATURE ON RECEIPT: <i>13</i>								P.O. Number: <i>66776</i>							
3. Relinquished by: (Signature)		Date/Time		4. Received by lab: (Signature)		FOR COMPLETION BY LAB ONLY															
<i>UPS</i>		<i>6/8/12 0920</i>		<i>Amanda Forbush</i>																	



11701 Interstate 30, Bldg. 1, Ste. 115
 Little Rock, AR 72209
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 FAX: 501-455-6118

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		BILLING		Project Description		Turnaround Time		Preservation Codes:							
City of DeQueen Wastewater Plant		City of DeQueen Wastewater Plant		Chronic Toxicity		24 Hour		1. Cool, 4 Degrees Centigrade			4. Thiosulfate for Dechlorination				
514 South 9th		P.O. Box 730				48 Hour		2. Sulfuric Acid (H ₂ SO ₄), pH < 2			5. Hydrochloric Acid(HCl)				
DeQueen, AR 71832		DeQueen, AR 71832		Reporting Information		72 Hour		3. Nitric Acid (HNO ₃), pH < 2			6. Sodium Hydroxide (NaOH), pH > 12				
Attn: Mike Sims				Telephone: 870-642-5231		Routine		TEST PARAMETERS						Bottle Type Code	
				Fax: 870-642-3117		Preservative Code:		1							G = Glass; P = Plastic
				Email: msims@cityofdequeen.com		Bottle Type:		P							V = Septum; A = Amber
Sampler(s) Signature				Sampler(s) Printed				Chronic Biomonitoring							Arkansas Analytical Work Order Number: K1206-003C
Field Number	SAMPLE COLLECTION		Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION								
	Date/s	Time/s													
	6-10-11-12	10:30/10:45		X		Water	DQ Bio								
1. Relinquished by: (Signature)		Date/Time		2. Received by: (Signature)		SAMPLE CONDITION UPON RECEIPT IN LAB						REMARKS / SAMPLE COMMENTS			
		6-11-12 11:45am		UPS		1. CUSTODY SEALS: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No						P.O. Number: 66776			
						2. CONTAINERS CORRECT: <input type="checkbox"/> Yes <input type="checkbox"/> No									
						3. COC/LABELS AGREE: <input type="checkbox"/> Yes <input type="checkbox"/> No									
3. Relinquished by: (Signature)		Date/Time		4. Received by lab: (Signature)		4. PRESERVATION CONFIRMED: <input type="checkbox"/> Yes <input type="checkbox"/> No									
UPS		6/12/12, 1100		Sydney James		5. RECEIVED ON ICE: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No									
Revision 1						6. TEMPERATURE ON RECEIPT: 30C									
						FOR COMPLETION BY LAB ONLY									

APPENDIX B

Effluent and Dilution Water Data

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING							Fathead Minnow		
Lab # / Sample ID <i>K1206003</i>				Test Start (Date/Time) <i>6/17/12</i>					
Client: <i>DeQueen</i>				Test End (Date/Time) <i>6/19/12</i>					
		Day of Test							
		1	2	3	4	5	6	7	notes/remarks
Control	MHS551	<i>6/17</i>	<i>6/18</i>	<i>6/19</i>	<i>6/10</i>	<i>6/11</i>	<i>6/12</i>	<i>6/13</i>	
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.5</i>	<i>8.4</i>	<i>8.3</i>	<i>8.3</i>	<i>8.5</i>	<i>8.5</i>	
	FINAL	<i>7.7</i>	<i>8.0</i>	<i>8.0</i>	<i>8.1</i>	<i>8.0</i>	<i>8.3</i>	<i>8.3</i>	
pH (s.u.)	INITIAL	<i>8.2</i>	<i>8.2</i>	<i>8.0</i>	<i>7.8</i>	<i>7.9</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.8</i>	<i>8.0</i>	<i>8.0</i>	<i>8.0</i>	<i>7.5</i>	<i>7.7</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.0</i>	<i>22.2</i>	<i>22.7</i>	<i>22.5</i>	<i>23.4</i>	<i>22.5</i>	<i>22.0</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
ALKALINITY (mg/L)		<i>58</i>							
HARDNESS (mg/L)		<i>32</i>							
CONDUCTIVITY (umhos/cm)		<i>301</i>							
CHLORINE (mg/L)		<i><0.05</i>							
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.3</i>	<i>8.4</i>	<i>8.3</i>	<i>8.71</i>	<i>8.9</i>	<i>8.7</i>	
	FINAL	<i>7.9</i>	<i>8.0</i>	<i>8.0</i>	<i>8.1</i>	<i>8.0</i>	<i>8.0</i>	<i>8.1</i>	
pH (s.u.)	INITIAL	<i>7.9</i>	<i>7.7</i>	<i>8.0</i>	<i>7.4</i>	<i>7.8</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.8</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	<i>7.6</i>	<i>7.6</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.0</i>	<i>23.3</i>	<i>22.7</i>	<i>22.7</i>	<i>24.0</i>	<i>21.0</i>	<i>21.1</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.5</i>	<i>8.4</i>	<i>8.3</i>	<i>8.71</i>	<i>9.1</i>	<i>8.8</i>	
	FINAL	<i>7.6</i>	<i>8.1</i>	<i>7.9</i>	<i>8.0</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	
pH (mg/L)	INITIAL	<i>7.9</i>	<i>7.8</i>	<i>7.9</i>	<i>7.5</i>	<i>7.7</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.8</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	<i>7.7</i>	<i>7.7</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.0</i>	<i>23.3</i>	<i>22.9</i>	<i>22.8</i>	<i>24.1</i>	<i>21.4</i>	<i>24.4</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.7</i>	<i>8.5</i>	<i>8.4</i>	<i>9.11</i>	<i>9.2</i>	<i>8.9</i>	
	FINAL	<i>7.6</i>	<i>8.2</i>	<i>8.0</i>	<i>8.0</i>	<i>8.0</i>	<i>7.9</i>	<i>8.1</i>	
pH (s.u.)	INITIAL	<i>7.9</i>	<i>7.8</i>	<i>7.9</i>	<i>7.4</i>	<i>7.62</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.5</i>	<i>8.0</i>	<i>7.9</i>	<i>7.9</i>	<i>7.7</i>	<i>7.7</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.1</i>	<i>23.6</i>	<i>23.0</i>	<i>22.9</i>	<i>24.2</i>	<i>23.3</i>	<i>24.8</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.7</i>	<i>8.5</i>	<i>8.4</i>	<i>9.0</i>	<i>9.3</i>	<i>9.1</i>	
	FINAL	<i>7.6</i>	<i>8.2</i>	<i>8.1</i>	<i>8.0</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	
pH (s.u.)	INITIAL	<i>7.8</i>	<i>7.7</i>	<i>7.8</i>	<i>7.3</i>	<i>7.61</i>	<i>7.7</i>	<i>7.5</i>	
	FINAL	<i>7.6</i>	<i>7.9</i>	<i>7.9</i>	<i>7.9</i>	<i>7.7</i>	<i>7.6</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.1</i>	<i>23.8</i>	<i>23.1</i>	<i>23.0</i>	<i>24.3</i>	<i>23.6</i>	<i>25.9</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.7</i>	<i>8.5</i>	<i>8.6</i>	<i>9.62</i>	<i>8.8</i>	<i>8.8</i>	
	FINAL	<i>7.5</i>	<i>8.2</i>	<i>8.1</i>	<i>8.0</i>	<i>7.9</i>	<i>8.0</i>	<i>8.1</i>	
pH (s.u.)	INITIAL	<i>7.8</i>	<i>7.6</i>	<i>7.7</i>	<i>7.2</i>	<i>7.45</i>	<i>7.4</i>	<i>7.3</i>	
	FINAL	<i>7.6</i>	<i>7.8</i>	<i>7.9</i>	<i>7.8</i>	<i>7.7</i>	<i>7.6</i>	<i>7.8</i>	
temp (C)	INITIAL	<i>22.2</i>	<i>23.9</i>	<i>23.2</i>	<i>23.1</i>	<i>24.7</i>	<i>23.3</i>	<i>27.4</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:	100%	<i>A</i>	<i>A</i>	<i>A</i>	<i>B</i>	<i>B</i>	<i>C</i>	<i>C</i>	
ALKALINITY (mg/L)		<i>30</i>			<i>24</i>		<i>18</i>		
HARDNESS (mg/L)		<i>52</i>			<i>34</i>		<i>38</i>		
CONDUCTIVITY (umhos/cm)		<i>697</i>			<i>727</i>		<i>644</i>		
CHLORINE (mg/L)		<i><0.05</i>			<i><0.05</i>		<i><0.05</i>		

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING		Cerodaphnia Dubia							
Lab # / Sample ID		K1206003		Test Start (Date/Time)		6/7/12			
Client:		Dequon		Test End (Date/Time)		6/14/12			
		Day of Test							
		1	2	3	4	5	6	7	notes/remarks
Control	MHS551	6/10	6/8	6/9	6/10	6/11	6/12	6/13	
D.O. (mg/L)	INITIAL	8.4	8.5	8.4	8.3	8.3	8.5	8.5	
	FINAL	8.3	8.4	8.4	8.7	8.6	8.6		
pH (s.u.)	INITIAL	8.2	8.2	8.0	7.8	7.9	7.7	7.6	
	FINAL	8.1	8.0	8.3	8.3	6.9	7.3		
temp (C)	INITIAL	22.9	22.2	22.7	22.5	23.4	22.5	22.0	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
ALKALINITY (mg/L)		58							
HARDNESS (mg/L)		82							
CONDUCTIVITY (umhos/cm)		301							
CHLORINE (mg/L)		0.05							
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.3	8.4	8.3	8.71	8.1	8.7	
	FINAL	8.3	8.3	8.4	8.4	8.4	8.5		
pH (s.u.)	INITIAL	7.9	7.7	8.0	7.6	7.8	7.7	7.6	
	FINAL	8.1	8.0	8.3	8.3	7.0	7.4		
temp (C)	INITIAL	22.0	23.3	22.7	22.7	24.0	21.0	23.9	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.5	8.4	8.3	8.71	9.1	8.8	
	FINAL	8.3	8.4	8.4	8.4	8.4	8.5		
pH (mg/L)	INITIAL	7.9	7.8	7.9	7.5	7.7	7.7	7.6	
	FINAL	8.1	7.9	8.2	8.2	7.2	7.6		
temp (C)	INITIAL	22.0	23.3	22.9	22.8	24.1	21.4	24.4	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.7	8.5	8.4	9.11	9.2	8.9	
	FINAL	8.3	8.4	8.3	8.3	8.4	8.5		
pH (s.u.)	INITIAL	7.9	7.8	7.9	7.4	7.62	7.7	7.6	
	FINAL	8.0	7.9	8.2	8.2	7.4	7.7		
temp (C)	INITIAL	22.1	23.6	23.0	22.9	24.2	23.3	24.8	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.7	8.5	8.4	9.0	8.8	9.1	
	FINAL	8.3	8.5	8.3	8.3	8.3	8.5		
pH (s.u.)	INITIAL	7.8	7.7	7.8	7.3	7.61	7.7	7.5	
	FINAL	8.0	7.9	8.1	8.1	7.4	7.8		
temp (C)	INITIAL	22.1	23.8	23.1	23.0	24.3	23.6	25.9	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.7	8.5	8.6	9.11	8.8	8.8	
	FINAL	8.3	8.5	8.3	8.4	8.4	8.5		
pH (s.u.)	INITIAL	7.8	7.7	7.7	7.2	7.45	7.4	7.3	
	FINAL	8.0	7.9	8.0	8.1	7.5	7.8		
temp (C)	INITIAL	22.2	23.9	23.2	23.1	24.7	23.3	27.4	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC: 100%		A	A	A	B	B	C	C	
ALKALINITY (mg/L)		30			24		18		
HARDNESS (mg/L)		52			34		38		
CONDUCTIVITY (umhos/cm)		697			727		644		
CHLORINE (mg/L)		0.05			0.05		0.05		

APPENDIX C

Fathead minnow raw data and statistics

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID K1206003 TEST START DATE 6/17/12 TIME 1570
 CLIENT Duke TEST END DATE 6/14/12 TIME 0855

AGE AND SOURCE OF MINNOWS

Summary Page

CONC:	REP #	start	DAY (NUMBER SURVIVING)						7%	MEAN %	CV
			1	2	3	4	5	6			
0	A	8	8	8	8	8	8	8	100	100	0.00
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	100		
	D	8	8	8	8	8	8	8	100		
	E	8	8	8	8	8	8	8	100		
32	A	8	8	8	8	8	8	8	100	100	0
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	100		
	D	8	8	8	8	8	8	8	100		
	E	8	8	8	8	8	8	8	100		
42	A	8	8	8	8	8	8	8	100	100	
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	100		
	D	8	8	8	8	8	8	8	100		
	E	8	8	8	8	8	8	8	100		
56	A	8	8	8	8	8	8	8	100	97.5	
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	100		
	D	8	8	8	8	8	8	8	100		
	E	8	8	8	8	8	8	8	100		
72	A	8	8	8	8	8	8	8	100	100	
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	100		
	D	8	8	8	8	8	8	8	100		
	E	8	8	8	8	8	8	8	100		
160	A	8	8	8	8	8	8	8	100	100	0.00
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	100		
	D	8	8	8	8	8	8	8	100		
	E	8	8	8	8	8	8	8	100		
ANALYST											
DATE:											
TIME:											

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		DATE		TIME				
CLIENT		TEST END DATE		DATE		TIME				
AGE AND SOURCE OF MINNOWS										
DAY (NUMBER SURVIVING)										
REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
CONC: 0	A	2	2	2	2	2	2	2		
	B	1	1	2	2	1	1	1		
	C	1	1	2	2	1	1	1		
	D	1	1	2	2	1	1	1		
	E									
CONC: 32	A	2	2	2	2	2	2	2		
	B	1	1	2	2	1	1	1		
	C	1	1	2	2	1	1	1		
	D	1	1	2	2	1	1	1		
	E									
CONC: 42	A	2	2	2	2	2	2	2		
	B	1	1	2	2	1	1	1		
	C	1	1	2	2	1	1	1		
	D	1	1	2	2	1	1	1		
	E									
CONC: 56	A	2	2	2	2	2	2	2		
	B	1	1	2	2	1	1	1		
	C	1	1	2	2	1	1	1		
	D	1	1	2	2	1	1	1		
	E									
CONC: 75	A	2	2	2	2	2	2	2		
	B	1	1	2	2	1	1	1		
	C	1	1	2	2	1	1	1		
	D	1	1	2	2	1	1	1		
	E									
CONC: 100	A	2	2	2	2	2	2	2		
	B	1	1	2	2	1	1	1		
	C	1	1	2	2	1	1	1		
	D	1	1	2	2	1	1	1		
	E									
ANALYST	KP	KP	mb	mb	ILP	KP	KP	KP		
DATE:	6/7/12	6/8/12	6-9-12	6-10-12	6/11/12	6/12/12	6/13/12	6/14/12		
TIME:	1510	1336	0900	1030	1330	1300	1100	0855		

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		6/7/12		TIME		1510			
CLIENT		Dequon		TEST END DATE		TIME					
AGE AND SOURCE OF MINNOWS											
B											
DAY (NUMBER SURVIVING)											
SURVIVAL											
CONC: 0	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
CONC: 32	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
CONC: 42	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
CONC: 56	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
CONC: 75	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
CONC: 100	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
ANALYST	KP										
DATE:	6/7/12										
TIME:	1510										

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		TIME						
CLIENT		TEST END DATE		TIME						
AGE AND SOURCE OF MINNOWS										
DAY (NUMBER SURVIVING)								SURVIVAL		
REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
CONC: 0	A	2	2	2	2	2	2			
	B	1	2	2	2	2	2			
	C	1	2	2	2	2	2			
	D	1	2	2	2	2	2			
	E									
CONC: 32	A	2	2	2	2	2	2			
	B	1	2	2	2	2	2			
	C	1	2	2	2	2	2			
	D	1	2	2	2	2	2			
	E									
CONC: 42	A	2	2	2	2	2	2			
	B	1	2	2	2	2	2			
	C	1	2	2	2	2	2			
	D	1	2	2	2	2	2			
	E									
CONC: 56	A	2	2	2	2	2	2			
	B	1	2	2	2	2	2			
	C	1	2	2	2	2	2			
	D	1	2	2	2	2	2			
	E									
CONC: 75	A	2	2	2	2	2	2			
	B	1	2	2	2	2	2			
	C	1	2	2	2	2	2			
	D	1	2	2	2	2	2			
	E									
CONC: 100	A	2	2	2	2	2	2			
	B	1	2	2	2	2	2			
	C	1	2	2	2	2	2			
	D	1	2	2	2	2	2			
	E									
ANALYST	KP									
DATE:	6/7/12									
TIME:	1510									

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE	6/7/12		TIME	1510					
CLIENT		Dequinn		TEST END DATE		TIME					
		AGE AND SOURCE OF MINNOWS									
		DAY (NUMBER SURVIVING)									
		SURVIVAL									
CONC: 0	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	2	2	1	1	1	1			
	C	1	2	2	1	1	1	1			
	D	1	2	2	1	1	1	1			
	E										
CONC: 32	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	2	2	1	1	1	1			
	C	1	2	2	1	1	1	1			
	D	1	2	2	1	1	1	1			
	E										
CONC: 42	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	2	2	1	1	1	1			
	C	1	2	2	1	1	1	1			
	D	1	2	2	1	1	1	1			
	E										
CONC: 56	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	2	2	1	1	1	1			
	C	1	2	2	1	1	1	1			
	D	1	2	1	1	1	1	1			
	E										
CONC: 75	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	2	2	1	1	1	1			
	C	1	2	2	1	1	1	1			
	D	1	2	2	1	1	1	1			
	E										
CONC: 100	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
	A	2	2	2	2	2	2	2			
	B	1	2	2	1	1	1	1			
	C	1	2	2	1	1	1	1			
	D	1	2	2	1	1	1	1			
	E										
ANALYST:	KP										
DATE:	6/7/12										
TIME:	1510										

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID TEST START DATE 6/7/12 TIME 1510
 CLIENT Dequon TEST END DATE TIME
 AGE AND SOURCE OF MINNOWS
 DAY (NUMBER SURVIVING)

CONC:	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
02	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
32	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
42	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
56	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
75	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
100	A	2	2	2	2	2	2	2			
	B	1	1	2	2	1	1	1			
	C	1	1	2	2	1	1	1			
	D	1	1	2	2	1	1	1			
	E										
ANALYST	KP										
DATE:	6/7/12										
TIME:	1510										

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

Pimephales promelas

FATHEAD MINNOW

TEST 1000.0

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB # / #s:		K1206003				TEST DATES (BEGIN / END):		6/7-14/12	
CLIENT:		City of DeQueen				WEIGHING DATE / TIME:		6-15-12, 1400	
ANALYSTS:		KP				DRYING TEMP (DEGREES C):		60	
SAMPLE ID:						DRYING TIME (HOURS):		24	
	REP #	FINAL DRY WEIGHT TIN+LARVAE (g)	INITIAL WEIGHT TIN (g)	TOTAL DRY WEIGHT OF LARVAE (g)	NUMBER OF LARVAE	DRY WEIGHT OF LARVAE (mg)			
CONTROL	A	0.97700	0.97416	0.00284	8	0.355	AVG DRY WEIGHT (mg)	0.381	
	B	0.97542	0.97294	0.00248	8	0.310			
	C	1.01428	1.01093	0.00335	8	0.419	CV	12.41	
	D	0.98611	0.98277	0.00334	8	0.418			
	E	0.96878	0.96556	0.00322	8	0.403			
CONC:	A	0.97254	0.96996	0.00258	8	0.322	AVG DRY WEIGHT (mg)	0.401	
	B	0.97711	0.97383	0.00328	8	0.410			
	32%	C	0.98327	0.97983	0.00344	8	0.430	CV	
	D	1.00341	0.99987	0.00354	8	0.442			
	E	1.02314	1.01993	0.00321	8	0.401			
CONC:	A	1.00698	1.00403	0.00295	8	0.369	AVG DRY WEIGHT (mg)	0.388	
	B	0.99814	0.99486	0.00328	8	0.410			
	42%	C	1.00491	1.00186	0.00305	8	0.381	CV	
	D	1.00834	1.00555	0.00279	8	0.349			
	E	0.97488	0.97143	0.00345	8	0.431			
CONC:	A	0.99367	0.99115	0.00252	8	0.315	AVG DRY WEIGHT (mg)	0.364	
	B	1.02483	1.02207	0.00276	8	0.345			
	56%	C	0.98755	0.98473	0.00282	8	0.353	CV	
	D	0.99380	0.99046	0.00334	8	0.418			
	E	0.98627	0.98317	0.00310	8	0.387			
CONC:	A	0.98040	0.97824	0.00216	8	0.270	AVG DRY WEIGHT (mg)	0.376	
	B	1.00010	0.99702	0.00308	8	0.385			
	75%	C	1.00548	1.00247	0.00301	8	0.376	CV	
	D	1.00674	1.00344	0.00330	8	0.412			
	E	1.01380	1.01031	0.00349	8	0.436			
CONC:	A	0.99026	0.98764	0.00262	8	0.328	AVG DRY WEIGHT (mg)	0.423	
	B	0.98426	0.98082	0.00344	8	0.430			
	100%	C	0.96535	0.96182	0.00353	8	0.441	CV	13.12
	D	0.97411	0.97055	0.00356	8	0.445			
	E	0.98395	0.98018	0.00377	8	0.471			

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.027

W = 0.416

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

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

Data FAIL normality test. Try another transformation.

Warning - The first three homogeneity tests are sensitive to non-normal data and should not be performed.

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

Hartley's test for homogeneity of variance
Bartlett's test for homogeneity of variance

These two tests can not be performed because at least one group has zero variance.

Data FAIL to meet homogeneity of variance assumption.
Additional transformations are useless.

TITLE: AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
FILE: Z:\TOXSTAT\MONTE\FHSURV.
TRANSFORM: ARC SINE(SQUARE ROOT(Y)) NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	1.0000	1.3931
1	CONTROL	2	1.0000	1.3931
1	CONTROL	3	1.0000	1.3931
1	CONTROL	4	1.0000	1.3931
1	CONTROL	5	1.0000	1.3931
2	32 % EFFLUENT	1	1.0000	1.3931
2	32 % EFFLUENT	2	1.0000	1.3931
2	32 % EFFLUENT	3	1.0000	1.3931
2	32 % EFFLUENT	4	1.0000	1.3931
2	32 % EFFLUENT	5	1.0000	1.3931

3	42 %	EFFLUENT	1	1.0000	1.3931
3	42 %	EFFLUENT	2	1.0000	1.3931
3	42 %	EFFLUENT	3	1.0000	1.3931
3	42 %	EFFLUENT	4	1.0000	1.3931
3	42 %	EFFLUENT	5	1.0000	1.3931
4	56 %	EFFLUENT	1	1.0000	1.3931
4	56 %	EFFLUENT	2	1.0000	1.3931
4	56 %	EFFLUENT	3	1.0000	1.3931
4	56 %	EFFLUENT	4	0.8750	1.2094
4	56 %	EFFLUENT	5	1.0000	1.3931
5	75 %	EFFLUENT	1	1.0000	1.3931
5	75 %	EFFLUENT	2	1.0000	1.3931
5	75 %	EFFLUENT	3	1.0000	1.3931
5	75 %	EFFLUENT	4	1.0000	1.3931
5	75 %	EFFLUENT	5	1.0000	1.3931
6	100 %	EFFLUENT	1	1.0000	1.3931
6	100 %	EFFLUENT	2	1.0000	1.3931
6	100 %	EFFLUENT	3	1.0000	1.3931
6	100 %	EFFLUENT	4	1.0000	1.3931
6	100 %	EFFLUENT	5	1.0000	1.3931

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.006	0.001	1.000
Within (Error)	24	0.027	0.001	
Total	29	0.033		

Critical F value = 2.62 (0.05,5,24)
 Since F < Critical F FAIL TO REJECT Ho: All equal

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	CONTROL	1.393	1.000		
2	32 % EFFLUENT	1.393	1.000	0.000	
3	42 % EFFLUENT	1.393	1.000	0.000	
4	56 % EFFLUENT	1.356	0.975	1.732	
5	75 % EFFLUENT	1.393	1.000	0.000	
6	100 % EFFLUENT	1.393	1.000	0.000	

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

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12

File: Z:\TOXSTAT\MONTE\FHSURV.

Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST - TABLE 2 OF 2

Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	CONTROL	5			
2	32 % EFFLUENT	5	0.020	2.0	0.000
3	42 % EFFLUENT	5	0.020	2.0	0.000
4	56 % EFFLUENT	5	0.020	2.0	0.025
5	75 % EFFLUENT	5	0.020	2.0	0.000
6	100 % EFFLUENT	5	0.020	2.0	0.000

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12

File: Z:\TOXSTAT\MONTE\FHSURV.

Transform: ARC SINE(SQUARE ROOT(Y))

STEEL'S MANY-ONE RANK TEST

Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	1.393				
2	32 % EFFLUENT	1.393	27.50	16.00	5.00	
3	42 % EFFLUENT	1.393	27.50	16.00	5.00	
4	56 % EFFLUENT	1.356	25.00	16.00	5.00	
5	75 % EFFLUENT	1.393	27.50	16.00	5.00	
6	100 % EFFLUENT	1.393	27.50	16.00	5.00	

Critical values use $k = 5$, are 1 tailed, and $\alpha = 0.05$

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.061

W = 0.910

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

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

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

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 2.06

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

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

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

TITLE: AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
FILE: Z:\TOXSTAT\MONTE\FHGR.
TRANSFORM: ARC SINE(SQUARE ROOT(Y)) NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.3550	0.6383
1	CONTROL	2	0.3100	0.5905
1	CONTROL	3	0.4190	0.7040
1	CONTROL	4	0.4180	0.7030
1	CONTROL	5	0.4030	0.6878
2	32 % EFFLUENT	1	0.3220	0.6034
2	32 % EFFLUENT	2	0.4100	0.6949
2	32 % EFFLUENT	3	0.4300	0.7152
2	32 % EFFLUENT	4	0.4420	0.7273
2	32 % EFFLUENT	5	0.4010	0.6857
3	42 % EFFLUENT	1	0.3690	0.6529
3	42 % EFFLUENT	2	0.4100	0.6949
3	42 % EFFLUENT	3	0.3810	0.6652
3	42 % EFFLUENT	4	0.3490	0.6320
3	42 % EFFLUENT	5	0.4310	0.7162
4	56 % EFFLUENT	1	0.3150	0.5959

4	56 %	EFFLUENT	2	0.3450	0.6278
4	56 %	EFFLUENT	3	0.3530	0.6362
4	56 %	EFFLUENT	4	0.4180	0.7030
4	56 %	EFFLUENT	5	0.3870	0.6714
5	75 %	EFFLUENT	1	0.2700	0.5464
5	75 %	EFFLUENT	2	0.3850	0.6694
5	75 %	EFFLUENT	3	0.3760	0.6601
5	75 %	EFFLUENT	4	0.4120	0.6969
5	75 %	EFFLUENT	5	0.4360	0.7212
6	100 %	EFFLUENT	1	0.3280	0.6098
6	100 %	EFFLUENT	2	0.4300	0.7152
6	100 %	EFFLUENT	3	0.4410	0.7263
6	100 %	EFFLUENT	4	0.4450	0.7303
6	100 %	EFFLUENT	5	0.4710	0.7564

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.011	0.002	0.898
Within (Error)	24	0.061	0.003	
Total	29	0.073		

Critical F value = 2.62 (0.05,5,24)
 Since F < Critical F FAIL TO REJECT Ho: All equal

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	CONTROL	0.665	0.381		
2	32 % EFFLUENT	0.685	0.401	-0.643	
3	42 % EFFLUENT	0.672	0.388	-0.235	
4	56 % EFFLUENT	0.647	0.364	0.558	
5	75 % EFFLUENT	0.659	0.376	0.185	
6	100 % EFFLUENT	0.708	0.423	-1.339	

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

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	CONTROL	5			
2	32 % EFFLUENT	5	0.072	18.8	-0.020
3	42 % EFFLUENT	5	0.072	18.8	-0.007
4	56 % EFFLUENT	5	0.072	18.8	0.017
5	75 % EFFLUENT	5	0.072	18.8	0.005
6	100 % EFFLUENT	5	0.072	18.8	-0.042

APPENDIX D

***Ceriodaphnia dubia* Raw Data and Statistics**

Cerodaphnia dubia

SURVIVAL AND REPRODUCTION TEST

Discharger: Dequane Lab Number/s: K120603
 Location: K120603
 Date Sample Collected:

Analyst: KP
 Test Start - Date/Time: 6/7/12, 15:00
 Test Stop - Date/Time: 6/14/12, 08:30

Conc 1		Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
%	Day	A	B	C	D	E	F	G	H	I	J				
0	1	0	0	0	0	0	0	0	0	0	0	0	10	0	KP
	2	0	0	0	0	0	0	0	0	0	0	0	10	0	mb
	3	0	0	1	0	0	0	1	0	2	3	7	9	0.3	KP
	4	4	4	1	0	3	1	2	4	1	2	29	9	3.2	KP
	5	8	9	1	0	5	2	8	1	2	4	43	9	4.8	KP
	6	7	5	9	9	8	9	3	6	9	1	65	9	7.2	KP
	7														
	8														
	Total	19	18	12	10	10	16	13	18	13	15	140		$\bar{x} = 5.6$	

Conc 4		Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
%	Day	A	B	C	D	E	F	G	H	I	J				
56	1	0	0	0	0	0	0	0	0	0	0	0	10	0	
	2	0	0	0	0	0	0	0	0	0	0	0	10	0	
	3	2	0	0	0	0	0	0	0	0	0	2	10	0.2	
	4	6	5	5	7	5	6	5	7	3	2	51	10	5.1	
	5	6	4	0	7	5	9	3	8	1	6	49	10	4.9	
	6	11	5	9	1	2	0	11	10	0	5	54	10	5.4	
	7														
	8														
	Total	25	14	14	15	12	15	19	25	4	13	156			

Conc 2		Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
%	Day	A	B	C	D	E	F	G	H	I	J				
32	1	0	0	0	0	0	0	0	0	0	0	0	9	0	
	2	0	0	0	0	0	0	0	0	0	0	0	9	0	
	3	2	2	3	0	0	0	2	2	3	3	14	9	1.6	
	4	5	4	7	0	9	5	5	2	3	6	46	9	5.1	
	5	10	3	3	0	5	8	0	2	1	6	38	9	4.2	
	6	2	16	12	0	0	9	11	11	2	7	63	9	7.0	
	7														
	8														
	Total	19	25	25	0	14	15	16	17	8	12	161		$CV = 16.1$	

Conc 5		Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
%	Day	A	B	C	D	E	F	G	H	I	J				
75	1	0	0	0	0	0	0	0	0	0	0	0	10	0	
	2	0	0	0	0	0	0	0	0	0	0	0	10	0	
	3	4	7	1	0	0	1	1	0	3	0	11	10	1.1	
	4	6	3	6	5	6	1	5	3	4	5	38	10	3.8	
	5	5	8	7	4	2	10	1	0	4	3	44	10	4.4	
	6	10	4	7	4	0	0	13	0	8	7	53	10	5.3	
	7														
	8														
	Total	19	16	21	13	8	12	20	3	19	15	146			

Conc 3		Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
%	Day	A	B	C	D	E	F	G	H	I	J				
42	1	0	0	0	0	0	0	0	0	0	0	0	10	0	
	2	0	0	0	0	0	0	0	0	0	0	0	10	0	
	3	0	0	1	3	0	0	0	0	1	0	5	10	0.5	
	4	9	7	3	6	5	7	3	6	5	3	49	10	4.9	
	5	13	10	3	5	6	1	3	9	8	3	61	10	6.1	
	6	4	2	6	8	1	9	7	0	2	4	43	10	4.3	
	7														
	8														
	Total	26	19	13	22	12	12	13	15	16	10	158			

Conc 6		Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
%	Day	A	B	C	D	E	F	G	H	I	J				
100	1	0	0	0	0	0	0	0	0	0	0	0	9	0	
	2	0	0	0	0	0	0	0	0	0	0	0	9	0	
	3	0	0	0	0	0	0	0	0	0	1	1	9	0.1	
	4	4	6	0	0	0	0	0	2	4	2	29	9	3.2	
	5	7	3	2	0	5	9	2	10	0	4	45	9	5.0	
	6	3	12	5	0	7	1	7	2	11	6	54	9	6.0	
	7														
	8														
	Total	14	20	11	0	12	15	12	17	13	5	129			

X= DEAD; Y= MALE

Revision 1
11/30/10

$\bar{x} = 14.3$
 $CV = 19.7$

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

Shapiro - Wilk's test for normality

***** Shapiro - Wilk's Test is aborted *****

This test can not be performed because total number of replicates
is greater than 50.

Total number of replicates = 60

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 2.18

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

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

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	9	1	10
32%	9	1	10
TOTAL	18	2	20

CRITICAL FISHER'S VALUE (10,10,9) (p=0.05) IS 4. b VALUE IS 9.
 Since b is greater than 4 there is no significant difference
 between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	DEAD	ALIVE	TOTAL ANIMALS
CONTROL	1	9	10
42%	0	10	10
TOTAL	1	19	20

CRITICAL FISHER'S VALUE (10,10,1) (p=0.05) IS LESS THAN 0. b VALUE IS 0.
 NO SIGNIFICANT DIFFERENCE

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	DEAD	ALIVE	TOTAL ANIMALS
CONTROL	1	9	10
56%	0	10	10

TOTAL 1 19 20

CRITICAL FISHER'S VALUE (10,10,1) (p=0.05) IS LESS THAN 0. b VALUE IS 0.
NO SIGNIFICANT DIFFERENCE

FISHER'S EXACT TEST

NUMBER OF

IDENTIFICATION	DEAD	ALIVE	TOTAL ANIMALS
CONTROL	1	9	10
75%	0	10	10
TOTAL	1	19	20

CRITICAL FISHER'S VALUE (10,10,1) (p=0.05) IS LESS THAN 0. b VALUE IS 0.
NO SIGNIFICANT DIFFERENCE

FISHER'S EXACT TEST

NUMBER OF

IDENTIFICATION	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	9	1	10
100%	9	1	10
TOTAL	18	2	20

CRITICAL FISHER'S VALUE (10,10,9) (p=0.05) IS 4. b VALUE IS 9.

Since b is greater than 4 there is no significant difference between CONTROL and TREATMENT at the 0.05 level.

SUMMARY OF FISHER'S EXACT TESTS

GROUP	IDENTIFICATION	NUMBER EXPOSED	NUMBER DEAD	SIG (P=.05)
	CONTROL	10	1	

1	32%	10	1
2	42%	10	0
3	56%	10	0
4	75%	10	0
5	100%	10	1

TITLE: AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
FILE: Z:\TOXSTAT\MONTE\CD.
TRANSFORM: NO TRANSFORMATION NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	19.0000	19.0000
1	CONTROL	2	18.0000	18.0000
1	CONTROL	3	12.0000	12.0000
1	CONTROL	4	16.0000	16.0000
1	CONTROL	5	0.0000	0.0000
1	CONTROL	6	16.0000	16.0000
1	CONTROL	7	13.0000	13.0000
1	CONTROL	8	18.0000	18.0000
1	CONTROL	9	13.0000	13.0000
1	CONTROL	10	15.0000	15.0000
2	32 % EFFLUENT	1	19.0000	19.0000
2	32 % EFFLUENT	2	25.0000	25.0000
2	32 % EFFLUENT	3	25.0000	25.0000
2	32 % EFFLUENT	4	0.0000	0.0000
2	32 % EFFLUENT	5	14.0000	14.0000
2	32 % EFFLUENT	6	15.0000	15.0000
2	32 % EFFLUENT	7	16.0000	16.0000
2	32 % EFFLUENT	8	17.0000	17.0000
2	32 % EFFLUENT	9	8.0000	8.0000
2	32 % EFFLUENT	10	22.0000	22.0000
3	42 % EFFLUENT	1	26.0000	26.0000
3	42 % EFFLUENT	2	19.0000	19.0000
3	42 % EFFLUENT	3	13.0000	13.0000
3	42 % EFFLUENT	4	22.0000	22.0000
3	42 % EFFLUENT	5	12.0000	12.0000
3	42 % EFFLUENT	6	12.0000	12.0000
3	42 % EFFLUENT	7	13.0000	13.0000
3	42 % EFFLUENT	8	15.0000	15.0000
3	42 % EFFLUENT	9	16.0000	16.0000
3	42 % EFFLUENT	10	10.0000	10.0000
4	56 % EFFLUENT	1	25.0000	25.0000
4	56 % EFFLUENT	2	14.0000	14.0000
4	56 % EFFLUENT	3	14.0000	14.0000
4	56 % EFFLUENT	4	15.0000	15.0000
4	56 % EFFLUENT	5	12.0000	12.0000
4	56 % EFFLUENT	6	15.0000	15.0000
4	56 % EFFLUENT	7	19.0000	19.0000
4	56 % EFFLUENT	8	25.0000	25.0000
4	56 % EFFLUENT	9	4.0000	4.0000
4	56 % EFFLUENT	10	13.0000	13.0000
5	75 % EFFLUENT	1	19.0000	19.0000

5	75 %	EFFLUENT	2	16.0000	16.0000
5	75 %	EFFLUENT	3	21.0000	21.0000
5	75 %	EFFLUENT	4	13.0000	13.0000
5	75 %	EFFLUENT	5	8.0000	8.0000
5	75 %	EFFLUENT	6	12.0000	12.0000
5	75 %	EFFLUENT	7	20.0000	20.0000
5	75 %	EFFLUENT	8	3.0000	3.0000
5	75 %	EFFLUENT	9	19.0000	19.0000
5	75 %	EFFLUENT	10	15.0000	15.0000
6	100 %	EFFLUENT	1	14.0000	14.0000
6	100 %	EFFLUENT	2	20.0000	20.0000
6	100 %	EFFLUENT	3	11.0000	11.0000
6	100 %	EFFLUENT	4	0.0000	0.0000
6	100 %	EFFLUENT	5	12.0000	12.0000
6	100 %	EFFLUENT	6	15.0000	15.0000
6	100 %	EFFLUENT	7	12.0000	12.0000
6	100 %	EFFLUENT	8	17.0000	17.0000
6	100 %	EFFLUENT	9	13.0000	13.0000
6	100 %	EFFLUENT	10	15.0000	15.0000

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
 File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	76.133	15.227	0.426
Within (Error)	54	1928.200	35.707	
Total	59	2004.333		

Critical F value = 2.45 (0.05, 5, 40)
 Since F < Critical F FAIL TO REJECT Ho: All equal

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
 File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

DUNNETT'S TEST - TABLE 1 OF 2 Ho: Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	CONTROL	14.000	14.000		
2	32 % EFFLUENT	16.100	16.100	-0.786	
3	42 % EFFLUENT	15.800	15.800	-0.674	
4	56 % EFFLUENT	15.600	15.600	-0.599	
5	75 % EFFLUENT	14.600	14.600	-0.225	
6	100 % EFFLUENT	12.900	12.900	0.412	

Dunnnett table value = 2.31 (1 Tailed Value, P=0.05, df=40,5)

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
 File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

DUNNETT'S TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	CONTROL	10			
2	32 % EFFLUENT	10	6.173	44.1	-2.100
3	42 % EFFLUENT	10	6.173	44.1	-1.800
4	56 % EFFLUENT	10	6.173	44.1	-1.600
5	75 % EFFLUENT	10	6.173	44.1	-0.600
6	100 % EFFLUENT	10	6.173	44.1	1.100

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
 File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

STEEL'S MANY-ONE RANK TEST - Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	14.000				
2	32 % EFFLUENT	16.100	117.50	75.00	10.00	
3	42 % EFFLUENT	15.800	105.00	75.00	10.00	
4	56 % EFFLUENT	15.600	107.00	75.00	10.00	
5	75 % EFFLUENT	14.600	111.00	75.00	10.00	
6	100 % EFFLUENT	12.900	92.50	75.00	10.00	

Critical values use k = 5, are 1 tailed, and alpha = 0.05

APPENDIX E

Organism History

AQUATOX, INC.

416 TWIN POINTS ROAD
HOT SPRINGS, ARKANSAS 71913
501-520-0560

TEST ORGANISM HISTORY

DATE SHIPPED 6/7/12 CLIENT AR Analytical

Purchase Order #: _____ Ken

SPECIES: Pimephales promelas

Quantity Shipped: 300

Age: hatched 6/6/12 1500 EST

Brood Stock Source: Anderson Farms, AR

Culture Water: Groundwater

Hardness (Mg/l CaCO₃): 160

Dissolved Oxygen (Mg/l): 8.2

Temperature (°C): 25.1°C

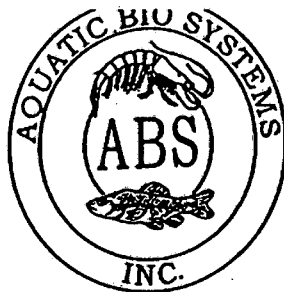
Feeding: Artemia

Comments: _____

Shipped Via: Federal Express UPS Overnight Shuttle

Packaged By: _____

1300 Blue Spruce Drive, Suite C
Fort Collins, Colorado 80524



Toll Free: 800/331-5916
Tel: 970/484-5091 Fax: 970/484-2514

ORGANISM HISTORY

DATE: 6/22/09

SPECIES: Ceriodaphnia dubia

AGE: Variable

LIFE STAGE: Adult


HATCH DATE: Variable

BEGAN FEEDING: Immediately

FOOD: YTC, Selenastrum sp.

Water Chemistry Record:	Current	Range
TEMPERATURE:	<u>25°C</u>	<u>20-25°C</u>
SALINITY/CONDUCTIVITY:	<u>--</u>	<u>--</u>
TOTAL HARDNESS (as CaCO ₃):	<u>142 mg/l</u>	<u>86-124 mg/l</u>
TOTAL ALKALINITY (as CaCO ₃):	<u>100 mg/l</u>	<u>65-130 mg/l</u>
pH:	<u>7.92</u>	<u>7.56-8.35</u>

Comments:

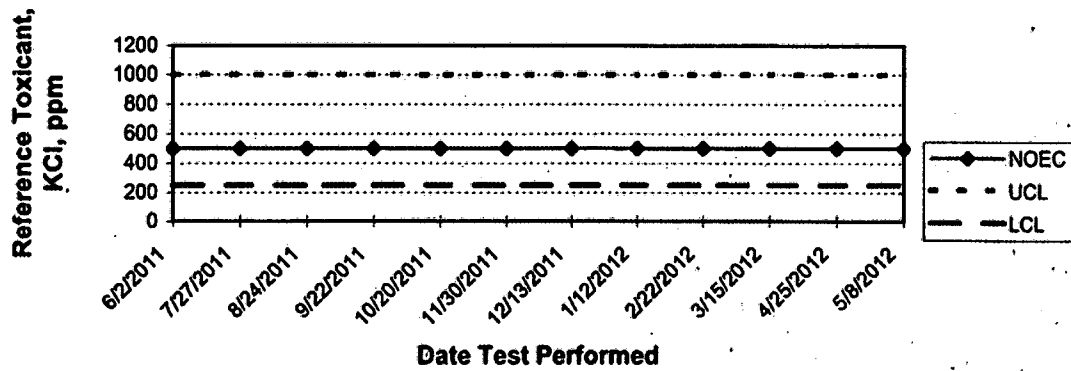


Facility Supervisor

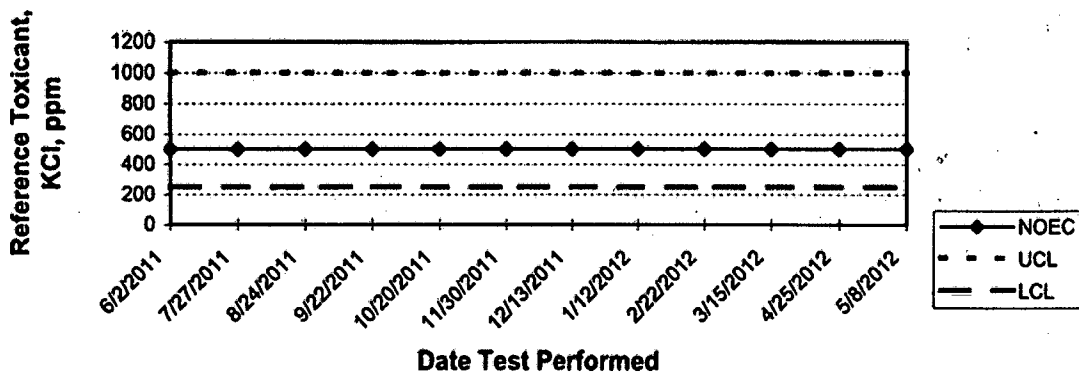
APPENDIX F

Quality Assurance Charts

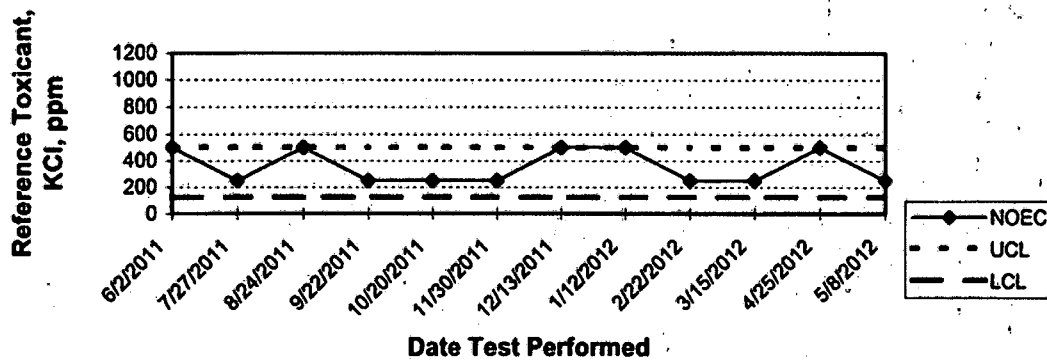
ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW SURVIVAL
QUALITY ASSURANCE



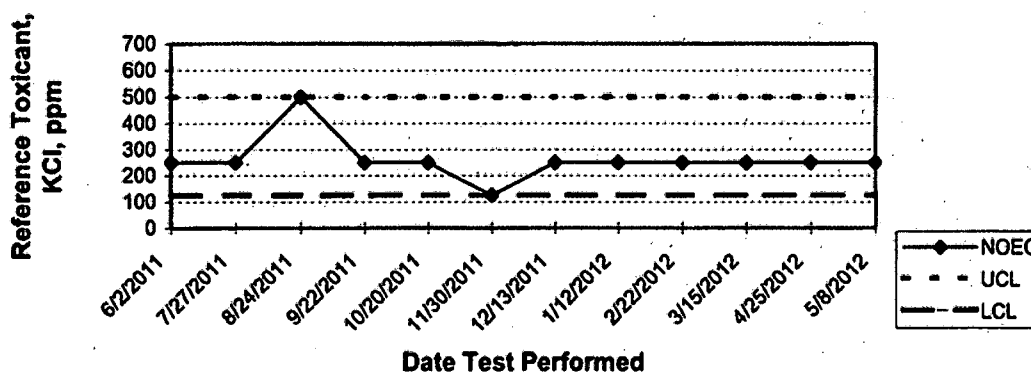
ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW GROWTH
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.
CERIODAPHNIA DUBIA SURVIVAL
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.
CERIODAPHNIA DUBIA REPRODUCTION
QUALITY ASSURANCE



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

July 5, 2012

WATER DIVISION
NPDES Enforcement Section

RE: City of De Queen Wastewater Plant, NPDES Permit AR0021733
AFIN 67-00023, 2nd Quarter WET report & DMR.

Michael Sims
City of De Queen
Wastewater Superintendent
PO Box 730
De Queen AR 71832

PERMITTEE NAME/ADDRESS (Include Facility Name/Location if Different)

NAME: DE QUEEN, CITY OF
 ADDRESS: P.O. BOX 730
 DE QUEEN, AR 71832

FACILITY: DEQUEEN, CITY OF
 LOCATION: 670 SOUTH 9TH STREET
 DEQUEEN, AR 71832

ATTN: MICHAEL SIMS, MANAGER

NATIONAL POLLUTANT DISCHARGE ELIMINATION SYSTEM (NPDES)

DISCHARGE MONITORING REPORT (DMR)

Arkansas Analytical
 11701 I-30 Bldg. 1 Ste. 115
 Little Rock Ar 72209

Form Approved
 OMB No. 2040-0004

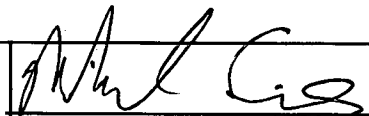
AR0021733	TX1-Q
PERMIT NUMBER	DISCHARGE NUMBER
MONITORING PERIOD	
MM/DD/YYYY	MM/DD/YYYY
4/1/2012	6/30/2012

DMR Mailing ZIP CODE: 71832
 MAJOR \$

TX1-QUARTERLY-W.E.T. REPORTS
 External Outfall

No Discharge

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		VALUE	VALUE	UNITS	VALUE	VALUE	VALUE	UNITS			
NOEC Lethal Static Renewal 7 Day Chronic Pimephales promelas	SAMPLE MEASUREMENT	*****	*****	*****	*****	100	*****		0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%		Quarterly	COMP24
NOEC Sub-Lethal Static Renewal 7 Day Chronic Ceriodaphnia dubia	SAMPLE MEASUREMENT	*****	*****	*****	*****	100	*****		0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%		Quarterly	COMP24
NOEC Sub-Lethal Static Renewal 7 Day Chronic Pimephales promelas	SAMPLE MEASUREMENT	*****	*****	*****	*****	100	*****		0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%		Quarterly	COMP24
Coef Of Var Statre 7Day Chronic Ceriodaphnia	SAMPLE MEASUREMENT	*****	*****	*****	*****	19.7	*****		0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%		Quarterly	COMP24
Coef Of Var Statre 7Day Chronic Pimephales	SAMPLE MEASUREMENT	*****	*****	*****	*****	13.1	*****		0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%		Quarterly	COMP24

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER Michael Sims TYPED OR PRINTED	I certify under penalty of law that this document and all attachments were prepared under my direction or supervision in accordance with a system designed to assure that qualified personnel properly gather and evaluate the information submitted. Based on my inquiry of the person or persons who manage the system, or those persons directly responsible for gathering the information, the information submitted is, to the best of my knowledge and belief, true, accurate, and complete. I am aware that there are significant penalties for submitting false information, including the possibility of fine and imprisonment for knowing violations.	 SIGNATURE OF PRINCIPAL EXECUTIVE OFFICER OR AUTHORIZED AGENT	TELEPHONE	DATE
			870-642-5231 AREA Code NUMBER	07/06/2012 MM/DD/YYYY

COMMENTS AND EXPLANATION OF ANY VIOLATIONS (Reference all attachments here)

(PASS=0/FAIL=1) IF THE NOEC VALUE IS LESS THAN THE CRITICAL DILUTION, REPORT "1"; OTHERWISE, REPORT "0". SEE PART III, CONDITION #9.

67-00023

Arkansas Analytical
11701 I-30 Bldg 1, Ste 115
Little Rock AR 72209

PERMITTEE NAME/ADDRESS (Include Facility Name/Location if Different)
NAME: DE QUEEN, CITY OF
ADDRESS: P.O. BOX 730
 DE QUEEN, AR 71832
FACILITY: DEQUEEN, CITY OF
LOCATION: 670 SOUTH 9TH STREET
 DEQUEEN, AR 71832
ATTN: MICHAEL SIMS, MANAGER

NATIONAL POLLUTANT DISCHARGE ELIMINATION SYSTEM (NPDES)

DISCHARGE MONITORING REPORT (DMR)

AR0021733	TX1-Q
PERMIT NUMBER	DISCHARGE NUMBER
MONITORING PERIOD	
MM/DD/YYYY	MM/DD/YYYY
4/1/2012	6/30/2012

Form Approved
OMB No. 2040-0004

DMR Mailing ZIP CODE: 71832
 MAJOR \$

TX1-QUARTERLY-W.E.T. REPORTS
 External Outfall

No Discharge

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		VALUE	VALUE	UNITS	VALUE	VALUE	VALUE	UNITS			
Whole effluent toxicity 22414 1 0 Effluent Gross	SAMPLE MEASUREMENT	*****	*****	*****	100	*****	*****			1/90	Comp 24
	PERMIT REQUIREMENT	*****	*****	*****	100 DLYAVMIN	*****	*****	%		Quarterly	COMP24
Whole effluent toxicity 22414 P 0 See Comments	SAMPLE MEASUREMENT	*****	*****	*****	100	*****	*****			1/90	Comp 24
	PERMIT REQUIREMENT	*****	*****	*****	100 7 DA MIN	*****	*****	%		Quarterly	COMP24
Pass/Fail Static Renewal 7 Day Chronic Ceriodaphnia	SAMPLE MEASUREMENT	*****	*****	*****		0	*****			1/90	Comp 24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
Pass/Fail Statre 7Day Chronic Pimephales Promelas	SAMPLE MEASUREMENT	*****	*****	*****		0	*****			1/90	Comp 24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
Low Flow Pass/Fail Survival Test Static Renewal 7 Day Chronic Ceriodaphnia dubia	SAMPLE MEASUREMENT	*****	*****	*****		0	*****			1/90	Comp 24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
Low Flow Pass/Fail Survival Test Static Renewal 7 Day Chronic Pimephales promelas	SAMPLE MEASUREMENT	*****	*****	*****		0	*****			1/90	Comp 24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
NOEC Lethal Static Renewal 7 Day Chronic Ceriodaphnia dubia	SAMPLE MEASUREMENT	*****	*****	*****		100	*****			1/90	Comp 24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%		Quarterly	COMP24

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			870-642-5231	07/06/2012	
			AREA Code	NUMBER	MM/DD/YYYY

COMMENTS AND EXPLANATION OF ANY VIOLATIONS (Reference all attachments here)
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67-00023

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 ADDRESS: P.O. BOX 730
 DE QUEEN, AR 71832

FACILITY: DEQUEEN, CITY OF
 LOCATION: 670 SOUTH 9TH STREET
 DEQUEEN, AR 71832
 ATTN: MICHAEL SIMS, MANAGER

NATIONAL POLLUTANT DISCHARGE ELIMINATION SYSTEM (NPDES)

DISCHARGE MONITORING REPORT (DMR)

Arkansas Analytical
 11701 I-30 Bldg 1, Ste 115
 Little Rock Ar 72209

Form Approved
 OMB No. 2040-0004

AR0021733	TX1-Q
PERMIT NUMBER	DISCHARGE NUMBER
MONITORING PERIOD	
MM/DD/YYYY	MM/DD/YYYY
4/1/2012	6/30/2012

DMR Mailing ZIP CODE: 71832
 MAJOR \$

TX1-QUARTERLY-W.E.T. REPORTS
 External Outfall

No Discharge

PARAMETER	SAMPLE MEASUREMENT	QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE
		VALUE	VALUE	UNITS	VALUE	VALUE	VALUE	UNITS			
Whole effluent toxicity	SAMPLE MEASUREMENT	*****	*****	*****	100	*****	*****		0	1/90	Comp 24
22414 10 Effluent Gross	PERMIT REQUIREMENT	*****	*****	*****	100 DLYAVMIN	*****	*****	%		Quarterly	COMP24
Whole effluent toxicity	SAMPLE MEASUREMENT	*****	*****	*****	100	*****	*****		0	1/90	Comp 24
22414 P 0 See Comments	PERMIT REQUIREMENT	*****	*****	*****	100 7 DA MIN	*****	*****	%		Quarterly	COMP24
Pass/Fail Static Renewal 7 Day Chronic Ceriodaphnia	SAMPLE MEASUREMENT	*****	*****	*****		0	*****		0	1/90	Comp 24
TGP3B 10 Effluent Gross	PERMIT REQUIREMENT	*****	*****	*****		Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
Pass/Fail Statre 7Day Chronic Pimephales Promelas	SAMPLE MEASUREMENT	*****	*****	*****		0	*****		0	1/90	Comp 24
TGP6C 10 Effluent Gross	PERMIT REQUIREMENT	*****	*****	*****		Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
Low Flow Pass/Fail Survival Test Static Renewal 7 Day Chronic Ceriodaphnia dubia	SAMPLE MEASUREMENT	*****	*****	*****		0	*****		0	1/90	Comp 24
TLP3B 10 Effluent Gross	PERMIT REQUIREMENT	*****	*****	*****		Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
Low Flow Pass/Fail Survival Test Static Renewal 7 Day Chronic Pimephales promelas	SAMPLE MEASUREMENT	*****	*****	*****		0	*****		0	1/90	Comp 24
TLP6C 10 Effluent Gross	PERMIT REQUIREMENT	*****	*****	*****		Req. Mon. 7 DA AVG	*****	pass=0/fail=1		Quarterly	COMP24
NOEC Lethal Static Renewal 7 Day Chronic Ceriodaphnia dubia	SAMPLE MEASUREMENT	*****	*****	*****		100	*****		0	1/90	Comp 24
TOP3B 10 Effluent Gross	PERMIT REQUIREMENT	*****	*****	*****		Req. Mon. 7 DA AVG	*****	%		Quarterly	COMP24

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			870-642-5231 AREA Code NUMBER	07/06/2012 MM/DD/YYYY

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67-00023

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 DEQUEEN, AR 71832

ATTN: MICHAEL SIMS, MANAGER

NATIONAL POLLUTANT DISCHARGE ELIMINATION SYSTEM (NPDES)

DISCHARGE MONITORING REPORT (DMR)

Arkansas Analytical
 11701 I-30 Bldg. 1 Ste. 115
 Little Rock Ar 72209

Form Approved
 OMB No. 2040-0004


AR0021733	TX1-Q
PERMIT NUMBER	DISCHARGE NUMBER
MONITORING PERIOD	
MM/DD/YYYY	MM/DD/YYYY
4/1/2012	6/30/2012

DMR Mailing ZIP CODE: 71832
 MAJOR \$

TX1-QUARTERLY-W.E.T. REPORTS
 External Outfall

No Discharge

PARAMETER		QUANTITY OR LOADING			QUALITY OR CONCENTRATION				NO. EX	FREQUENCY OF ANALYSIS	SAMPLE TYPE	
		VALUE	VALUE	UNITS	VALUE	VALUE	VALUE	UNITS				
NOEC Lethal Static Renewal 7 Day Chronic Pimephales promelas	SAMPLE MEASUREMENT	*****	*****	*****	*****	100	*****			0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%			Quarterly	COMP24
NOEC Sub-Lethal Static Renewal 7 Day Chronic Ceriodaphnia dubia	SAMPLE MEASUREMENT	*****	*****	*****	*****	100	*****			0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%			Quarterly	COMP24
NOEC Sub-Lethal Static Renewal 7 Day Chronic Pimephales promelas	SAMPLE MEASUREMENT	*****	*****	*****	*****	100	*****			0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%			Quarterly	COMP24
Coef Of Var Statre 7Day Chronic Ceriodaphnia	SAMPLE MEASUREMENT	*****	*****	*****	*****	19.7	*****			0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%			Quarterly	COMP24
Coef Of Var Statre 7Day Chronic Pimephales	SAMPLE MEASUREMENT	*****	*****	*****	*****	13.1	*****			0	1/90	Comp24
	PERMIT REQUIREMENT	*****	*****	*****	*****	Req. Mon. 7 DA AVG	*****	%			Quarterly	COMP24

NAME/TITLE PRINCIPAL EXECUTIVE OFFICER	I certify under penalty of law that this document and all attachments were prepared under my direction or supervision in accordance with a system designed to assure that qualified personnel properly gather and evaluate the information submitted. Based on my inquiry of the person or persons who manage the system, or those persons directly responsible for gathering the information, the information submitted is, to the best of my knowledge and belief, true, accurate, and complete. I am aware that there are significant penalties for submitting false information, including the possibility of fine and imprisonment for knowing violations.		TELEPHONE		DATE
TYPED OR PRINTED			SIGNATURE OF PRINCIPAL EXECUTIVE OFFICER OR AUTHORIZED AGENT	870-642-5231	07/06/2012
			AREA Code	NUMBER	MM/DD/YYYY

COMMENTS AND EXPLANATION OF ANY VIOLATIONS (Reference all attachments here)

(PASS=0/FAIL=1) IF THE NOEC VALUE IS LESS THAN THE CRITICAL DILUTION, REPORT "1"; OTHERWISE, REPORT "0". SEE PART III, CONDITION #9.

67-00023

Arkansas Analytical, Inc.

Toxicity Test Results
City of DeQueen
NPDES PERMIT NUMBER: AR0021733
Second Quarter 2012
AFIN # 67-00023

Fathead Minnow, *Pimephales promelas*, Larval Survival and Growth Test
Test 1000.0

Ceriodaphnia dubia, Survival and Reproduction Test
Test 1002.0

Prepared for: **Mr. Mike Sims**
City of DeQueen
P.O. Box 730
DeQueen, Arkansas 71832

Prepared by: Arkansas Analytical, Inc.
11701 I-30, Bldg 1, Suite 115
Little Rock, Arkansas 72209
Lab Number K1206003

Monday, June 25, 2012

Introduction

This report contains test results for toxicity testing for the City of DeQueen. The NPDES permit number is AR0021733. The facility is located 1/8 mile south from intersection of Coulter Ave. and south of 9th Street on Philip Cox Blvd, in Section 36, Township 8 South, Range 32 West in Sevier County, Arkansas. The discharge is to receiving waters named: an unnamed ditch around pond to Bear Creek to Little River to Red River in Segment 1C of the Red River Basin.

The permit requires chronic biomonitoring testing quarterly for both *Ceriodaphnia dubia* and *Pimephales promelas*. The test results in this report represent the testing for the second quarter of 2012.

Plant Operations

To be provided by permittee.

Source of Effluent and Dilution Water

Effluent samples were collected as follows:

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-5-12, 1000	6-6-12, 1000
Sample #2:	6-6-12, 1000	6-7-12, 1000
Sample #3:	6-10-12, 1000	6-11-12, 1000

The samples were composites collected at the final discharge from City of DeQueen Wastewater Plant outfall.

The following information was collected upon immediate receipt of the samples at the laboratory:

Sample Receiving Information:	Date, Time Sample(s) Received	Temperature Upon Receipt (°C)
Sample #1:	6-7-12, 1145	1
Sample #2:	6-8-12, 0920	3
Sample #3:	6-12-12, 1100	3

Chain of custody documentation is located in Appendix A.

The permit designates the receiving water to be used as dilution water for the toxicity tests. Due to its earlier characterization as toxic, synthetic dilution water was substituted.

The dilution water used in the toxicity tests was moderately hard synthetic. It was prepared using Elga Maxima ultra pure water according to EPA specifications. Each batch was analyzed for pH, hardness, total alkalinity, and conductivity. Results are provided in Appendix B.

Dilution Series

Five dilutions in addition to a control (0% effluent) were used in the toxicity tests. The dilutions, which were made with synthetic water, were 32%, 42%, 56%, 75%, and 100%. The low-flow effluent concentration (**critical dilution**) was defined as **100% effluent**.

Test Methods

EPA Method 1000.0, Fathead Minnow, *Pimephales promelas*, Larval Survival and Growth Test, was used in this bioassay. Larvae are exposed in a static renewal system for seven days and the results are based on the survival and growth (increase in weight) of the larvae. The alternate method suggested in the method (11.3.4.5) for combating pathogen interference, was run in place of the original fathead minnow test. The test chambers were 30 ml plastic cups with 20 ml of test solution. Each chamber contained 2 organisms. The total number of fish was 40 per test solution. The fish were then combined to perform growth analysis. The test temperature was 25 degrees Centigrade. Raw data and statistics are provided in Appendix C.

EPA Method 1002.0, Cladoceran, *Ceriodaphnia dubia*, Survival and Reproduction Test, was also used. Neonates are exposed in a static renewal system until at least 60% of the control organisms have produced a third brood. Results are based on the survival and reproduction of the organisms. One neonate was placed in each of ten replicate chambers using a randomizing template. Test chambers were 30 ml plastic cups filled with 15 ml of test solution. The test temperature was 25 degrees Centigrade. Raw data and statistics are provided in Appendix D.

Test Organisms

The organisms used in Test 1000.0 were < 24 hour old Fathead Minnows, *Pimephales promelas*, which were purchased from Aquatox; a copy of the organism history is provided in Appendix E.

The organisms used in Test 1002.0 were < 24 hour old *Ceriodaphnia dubia* neonates, (all born within the same eight hours), obtained from an in-house culture. An organism history is provided in Appendix E.

Quality Assurance

Test Acceptability

TEST ACCEPTANCE CRITERIA for *Ceriodaphnia dubia*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	90%	X	
Average of 15 or more young per surviving female	15.6	X	
At least 60% of surviving females should have produced 3 broods	88.9%	X	
The percent coefficient of variation between replicates must be 40% or less for the young of surviving females	16.1	X	

TEST ACCEPTANCE CRITERIA for *Pimephales promelas*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	100%	X	
The percent coefficient of variation between replicates must be 40% or less for survival	0.00	X	
Minimum of 0.25 mg average dry weight of surviving controls	0.381	X	
The percent coefficient of variation between replicates must be 40% or less for growth	12.4	X	

Reference Toxicant

The reference toxicant used was Potassium Chloride prepared in-house. The tests were performed using moderately hard synthetic as dilution water. The results of the reference toxicant were:

REFERENCE TOXICANT

<i>Ceriodaphnia dubia</i> 5/8-15/12		<i>Pimephales promelas</i> 5/8-15/12	
NOEC Survival:	250 ppm KCl	NOEC Survival:	500 ppm KCl
LOEC Survival:	500 ppm KCl	LOEC Survival:	1000 ppm KCl
NOEC Reproduction:	250 ppm KCl	NOEC Growth:	500 ppm KCl
LOEC Reproduction:	500 ppm KCl	LOEC Growth:	1000 ppm KCl

Quality Assurance charts are provided in Appendix F.

Summary of Results
City of DeQueen

<i>Ceriodaphnia dubia</i>		<i>Pimephales promelas</i>	
NOEC / LOEC Survival	100% / NA	NOEC / LOEC survival	100% / NA
NOEC / LOEC Reproduction	100% / NA	NOEC / LOEC growth	100% / NA
Mean number of neonates (critical dilution)	14.3	%CV survival (critical dilution)	0.00
%CV Reproduction (critical dilution)	19.7	Mean dry weight (critical dilution) in milligrams	0.423
		%CV growth (critical dilution)	13.1
PMSD Reproduction	44.1	PMSD Growth	18.8

Conclusion

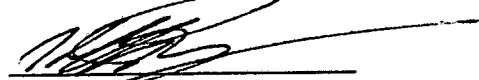
Chronic static renewal larval survival and growth test using fathead minnow, *Pimephales promelas*, (Method 1000.0)

The permit issued to the City of DeQueen, AR0021733, specifies that the **critical dilution is 100% effluent**. The effluent samples **did not** exhibit lethal and sublethal effects at the critical dilution, and, as such, **passed** both portions of the test.

Chronic static renewal survival and reproduction test using *Ceriodaphnia dubia*, (Method 1002.0)

The permit issued to the City of DeQueen, AR0021733, specifies that the **critical dilution is 100% effluent**. The effluent samples **did not** exhibit lethal effects or sublethal effects at the critical dilution, and, as such, **passed** both portions of the test.

Biomonitoring Analysts:


Kenneth Pigue


Melissa Bird

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
 FATHEAD MINNOW LARVAE GROWTH AND SURVIVAL
PIMEPHALES PROMELAS

PERMITTEE: City of DeQueen

NPDES #: AR0021733

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-5-12, 1000	6-6-12, 1000
Sample #2:	6-6-12, 1000	6-7-12, 1000
Sample #3:	6-10-12, 1000	6-11-12, 1000

Test initiated (date, time): 6-7-12, 1510 Test terminated (date, time): 6-14-12, 0855

Dilution water used: Moderately Hard Synthetic

DATA TABLE FOR FATHEAD MINNOW SURVIVAL

Effluent Conc %	Percent Survival in Replicate Chambers						Mean Percent Survival			CV %
	A	B	C	D	E		24 hours	48 hours	7 days	
0%	100	100	100	100	100		100	100	100	0.00
32%	100	100	100	100	100		100	100	100	
42%	100	100	100	100	100		100	100	100	
56%	100	100	100	87.5	100		100	100	97.5	
75%	100	100	100	100	100		100	100	100	
100%	100	100	100	100	100		100	100	100	0.00

DATA TABLE FOR GROWTH OF FATHEAD MINNOWS

Effluent Conc %	A	B	C	D	E		Mean Dry Weight	CV%
0%	0.355	0.310	0.419	0.418	0.403		0.381	12.40
32%	0.322	0.410	0.430	0.442	0.401		0.401	
42%	0.369	0.410	0.381	0.349	0.431		0.388	
56%	0.315	0.345	0.353	0.418	0.387		0.364	
75%	0.270	0.385	0.376	0.412	0.436		0.376	
100%	0.328	0.430	0.441	0.445	0.471		0.423	13.10

Average Dry Weight in milligrams in replicate chambers
 Coefficient of Variation = standard deviation / mean * 100

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
FATHEAD MINNOW LARVAE GROWTH AND SURVIVAL

Pimephales promelas

1. Dunnett's procedure or Steel's Many-One Rank Test as appropriate:
Is the mean survival at 7 days significantly different ($p=0.05$) than the control survival for:
a) LOW FLOW OR CRITICAL DILUTION, (100%) YES _____ NO X

2. Dunnett's Procedure
Is the mean dry weight (growth) at 7 days significantly different ($p=0.05$) than the control's dry weight (growth) for:
a) LOW FLOW OR CRITICAL DILUTION, (100%) YES _____ NO X

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

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

5. Enter percentage corresponding to each parameter below:
a) NOEC survival (parameter TOP6C)= 100 % effluent
b) NOEC growth (parameter TPP6C)= 100 % effluent
c) Coefficient of variation (parameter TQP6C)= 13.1 %

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
Ceriodaphnia dubia SURVIVAL AND REPRODUCTION

PERMITTEE: City of DeQueen

NPDES #: AR0021733

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-5-12, 1000	6-6-12, 1000
Sample #2:	6-6-12, 1000	6-7-12, 1000
Sample #3:	6-10-12, 1000	6-11-12, 1000

Test initiated (date, time): 6-7-12, 1500 Test terminated (date, time): 6-14-12, 0835

Dilution water used: Moderately Hard Synthetic

Ceriodaphnia dubia SURVIVAL AND REPRODUCTION
 NUMBER OF YOUNG PRODUCED PER FEMALE @ TEST TERMINATION
 PERCENT EFFLUENT

Replicate	0%	32%	42%	56%	75%	100%
A	19	19	26	25	19	14
B	18	25	19	14	16	20
C	12	25	13	14	21	11
D	16	x0	22	15	13	x0
E	x0	14	12	12	8	12
F	16	15	12	15	12	15
G	13	16	13	19	20	12
H	18	17	15	25	3	17
I	13	8	16	4	19	13
J	15	22	10	13	15	15
Mean	14.0	16.1	15.8	15.6	14.6	12.9
Mean/surviving female	15.6	17.9	15.8	15.6	14.6	14.3
CV%*	16.1					19.7

X= Dead Adult; M= Male (Not considered in statistics)

*Coefficient of Variation = standard deviation/ mean * 100; CV% calculation based on young per surviving female

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
Ceriodaphnia dubia SURVIVAL AND REPRODUCTION

PERMITTEE: City of DeQueen

NPDES #: AR0021733

PERCENT SURVIVAL

PERCENT EFFLUENT	0%	32%	42%	56%	75%	100%
Time of Reading: 24 HOURS	100	90	100	100	100	90
48 HOURS	100	90	100	100	100	90
Test termination	90	90	100	100	100	90

1. Fisher's Exact Test:

Is the mean survival at test termination significantly different ($p=0.05$) than the control survival for:

a) LOW FLOW OR CRITICAL DILUTION, (100%): YES _____ NO X _____

2. Dunnett's Procedure or Steel's Many One Rank Test:

Is the mean number of young produced per female significantly different ($p=0.05$) than the controls number of young per female for:

a) LOW FLOW OR CRITICAL DILUTION, (100%): YES _____ NO X _____

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

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

5. Enter percentage corresponding to each parameter below:

a) NOEC survival (parameter TOP3B)= 100 % effluent

b) NOEC reproduction (parameter TPP3B)= 100 % effluent

c) Coefficient of variation (parameter TQP3B)= 19.7 %

APPENDIX A

Chain of Custody Forms



11701 Interstate 30, Bldg. 1, Ste. 115
 Little Rock, AR 72209
 PHONE: 501-455-3233
 FAX: 501-455-6118

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		BILLING		Project Description		Turnaround Time		Preservation Codes:											
City of DeQueen Wastewater Plant 514 South 9th DeQueen, AR 71832		City of DeQueen Wastewater Plan P.O. Box 730 DeQueen, AR 71832		Chronic Toxicity		24 Hour 48 Hour 72 Hour Routine		1. Cool, 4 Degrees Centigrade 2. Sulfuric Acid (H ₂ SO ₄), pH < 2 3. Nitric Acid (HNO ₃), pH < 2				4. Thiosulfate for Dechlorination 5. Hydrochloric Acid(HCl) 6. Sodium Hydroxide (NaOH), pH > 12							
Attn: Mike Sims				Reporting Information Telephone: 870-642-5231 Fax: 870-642-3117 Email: msims@cityofdequeen.com		Preservative Code: Bottle Type:		TEST PARAMETERS								Bottle Type Code G = Glass; P = Plastic V = Septum; A = Amber			
Sampler(s) Signature				Sampler(s) Printed				Chronic Biomonitoring										Arkansas Analytical Work Order Number: K/206-003A	
Field Number	SAMPLE COLLECTION		Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION		Chronic Biomonitoring										
	Date/s	Time/s																	
	6-5/6-12	10:20/10:40		X		Water	DQ Bio	X											
1. Relinquished by: (Signature) 		Date/Time 6-6-12 11:45am		2. Received by: (Signature) UPS		SAMPLE CONDITION UPON RECEIPT IN LAB				REMARKS / SAMPLE COMMENTS									
3. Relinquished by: (Signature) UPS		Date/Time 6/7/12 1132		4. Received by lab: (Signature) Sydney James		1. CUSTODY SEALS: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No 2. CONTAINERS CORRECT: <input type="checkbox"/> Yes <input type="checkbox"/> No 3. COC/LABELS AGREE: <input type="checkbox"/> Yes <input type="checkbox"/> No 4. PRESERVATION CONFIRMED: <input type="checkbox"/> Yes <input type="checkbox"/> No 5. RECEIVED ON ICE: <input type="checkbox"/> Yes <input type="checkbox"/> No 6. TEMPERATURE ON RECEIPT: 1°C				P.O. Number: 66776									
Revision 1 12/1/10												FOR COMPLETION BY LAB ONLY							



11701 Interstate 30, Bldg. 1, Ste. 115
 Little Rock, AR 72209
 PHONE: 501-455-3233
 FAX: 501-455-6118

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		BILLING		Project Description		Turnaround Time		Preservation Codes:							
City of DeQueen Wastewater Plant		City of DeQueen Wastewater Plant		Chronic Toxicity		24 Hour		1. Cool, 4 Degrees Centigrade			4. Thiosulfate for Dechlorination				
514 South 9th		P.O. Box 730				48 Hour		2. Sulfuric Acid (H ₂ SO ₄), pH < 2			5. Hydrochloric Acid(HCl)				
DeQueen, AR 71832		DeQueen, AR 71832		Reporting Information		72 Hour		3. Nitric Acid (HNO ₃), pH < 2			6. Sodium Hydroxide (NaOH), pH > 12				
Attn: Mike Sims				Telephone: 870-642-5231		Routine		TEST PARAMETERS						Bottle Type Code	
				Fax: 870-642-3117		Preservative Code:		1							G = Glass; P = Plastic
				Email: msims@cityofdequeen.com		Bottle Type:		P							V = Septum; A = Amber
Sampler(s) Signature				Sampler(s) Printed <i>MS</i>				Chronic Biomonitoring							Arkansas Analytical Work Order Number: K1206003 B
Field Number	SAMPLE COLLECTION Date/s Time/s		Grab	Comp	Number of Bottles	Sample Matrix	IDENTIFICATION/ DESCRIPTION								
	6-6/7-12 10:00/10:00			X		Water	DQ Bio						X		
1. Relinquished by: (Signature)		Date/Time		2. Received by: (Signature)		SAMPLE CONDITION UPON RECEIPT IN LAB				REMARKS / SAMPLE COMMENTS					
		6-7-12 11:45am		UPS		1. CUSTODY SEALS: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No				P.O. Number: 66776					
						2. CONTAINERS CORRECT: <input type="checkbox"/> Yes <input type="checkbox"/> No									
						3. COC/LABELS AGREE: <input type="checkbox"/> Yes <input type="checkbox"/> No									
3. Relinquished by: (Signature)		Date/Time		4. Received by lab: (Signature)		4. PRESERVATION CONFIRMED: <input type="checkbox"/> Yes <input type="checkbox"/> No									
UPS		6/8/12 0920		Amanda Forbush		5. RECEIVED ON ICE: <input type="checkbox"/> Yes <input checked="" type="checkbox"/> No									
						6. TEMPERATURE ON RECEIPT: 13°									
Revision 1						FOR COMPLETION BY LAB ONLY									



11701 Interstate 30, Bldg. 1, Ste. 115
 Little Rock, AR 72209
 PHONE: 501-455-3233
 FAX: 501-455-6118

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		BILLING		Project Description		Turnaround Time		Preservation Codes:								
City of DeQueen Wastewater Plant		City of DeQueen Wastewater Plant		Chronic Toxicity		24 Hour		1. Cool, 4 Degrees Centigrade			4. Thiosulfate for Dechlorination					
514 South 9th		P.O. Box 730				48 Hour		2. Sulfuric Acid (H ₂ SO ₄), pH < 2			5. Hydrochloric Acid(HCl)					
DeQueen, AR 71832		DeQueen, AR 71832		Reporting Information		72 Hour		3. Nitric Acid (HNO ₃), pH < 2			6. Sodium Hydroxide (NaOH), pH > 12					
Attn: Mike Sims				Telephone: 870-642-5231		Routine		TEST PARAMETERS						Bottle Type Code		
				Fax: 870-642-3117		Preservative Code:		1								G = Glass; P = Plastic
				Email: msims@cityofdequeen.com		Bottle Type:		P								V = Septum; A = Amber
Sampler(s) Signature				Sampler(s) Printed				Chronic Biomonitoring							Arkansas Analytical Work Order Number: K1206-003C	
Field Number	SAMPLE COLLECTION		Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION									
	Date/s	Time/s														
	6-10-11-12	10:00/10:00		X		Water	DQ Bio									
1. Relinquished by: (Signature)		Date/Time		2. Received by: (Signature)		SAMPLE CONDITION UPON RECEIPT IN LAB				REMARKS / SAMPLE COMMENTS						
		6-11-12		UPS		1. CUSTODY SEALS: <input checked="" type="checkbox"/> Yes ___ No				P.O. Number: 66776						
		11:45am				2. CONTAINERS CORRECT: <input type="checkbox"/> Yes ___ No										
						3. COC/LABELS AGREE: <input type="checkbox"/> Yes ___ No										
3. Relinquished by: (Signature)		Date/Time		4. Received by lab: (Signature)		4. PRESERVATION CONFIRMED: <input type="checkbox"/> Yes ___ No										
UPS		6/12/12		Sydney James		5. RECEIVED ON ICE: <input checked="" type="checkbox"/> Yes ___ No										
Revision		1100				6. TEMPERATURE ON RECEIPT: 30°C										
												FOR COMPLETION BY LAB ONLY				

APPENDIX B

Effluent and Dilution Water Data

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING							Fathead Minnow		
Lab # / Sample ID <i>K1206003</i>				Test Start (Date/Time) <i>6/17/12</i>					
Client: <i>DeRuxon</i>				Test End (Date/Time) <i>6/19/12</i>					
Day of Test									
		1	2	3	4	5	6	7	notes/remarks
Control	MHS551	<i>6/17</i>	<i>6/18</i>	<i>6/19</i>	<i>6/10</i>	<i>6/11</i>	<i>6/12</i>	<i>6/13</i>	
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.5</i>	<i>8.4</i>	<i>8.3</i>	<i>8.3</i>	<i>8.5</i>	<i>8.5</i>	
	FINAL	<i>7.7</i>	<i>8.0</i>	<i>8.0</i>	<i>8.1</i>	<i>8.0</i>	<i>8.3</i>	<i>8.3</i>	
pH (s.u.)	INITIAL	<i>8.2</i>	<i>8.2</i>	<i>8.0</i>	<i>7.8</i>	<i>7.9</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.8</i>	<i>8.0</i>	<i>8.0</i>	<i>8.0</i>	<i>7.5</i>	<i>7.7</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.0</i>	<i>22.2</i>	<i>22.7</i>	<i>22.5</i>	<i>23.4</i>	<i>22.5</i>	<i>22.0</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
ALKALINITY (mg/L)		<i>58</i>							
HARDNESS (mg/L)		<i>32</i>							
CONDUCTIVITY (umhos/cm)		<i>301</i>							
CHLORINE (mg/L)		<i>0.05</i>							
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.3</i>	<i>8.4</i>	<i>8.3</i>	<i>8.71</i>	<i>8.9</i>	<i>8.7</i>	
	FINAL	<i>7.8</i>	<i>8.0</i>	<i>8.0</i>	<i>8.1</i>	<i>8.0</i>	<i>8.0</i>	<i>8.1</i>	
pH (s.u.)	INITIAL	<i>7.9</i>	<i>7.7</i>	<i>8.0</i>	<i>7.4</i>	<i>7.8</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.8</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	<i>7.6</i>	<i>7.6</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.0</i>	<i>23.3</i>	<i>22.7</i>	<i>22.7</i>	<i>24.0</i>	<i>21.0</i>	<i>21.9</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.5</i>	<i>8.4</i>	<i>8.3</i>	<i>8.71</i>	<i>9.1</i>	<i>8.8</i>	
	FINAL	<i>7.6</i>	<i>8.1</i>	<i>7.9</i>	<i>8.0</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	
pH (mg/L)	INITIAL	<i>7.9</i>	<i>7.8</i>	<i>7.9</i>	<i>7.5</i>	<i>7.7</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.8</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	<i>7.7</i>	<i>7.7</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.0</i>	<i>23.3</i>	<i>22.9</i>	<i>22.8</i>	<i>24.1</i>	<i>21.4</i>	<i>24.4</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.7</i>	<i>8.5</i>	<i>8.4</i>	<i>9.11</i>	<i>9.2</i>	<i>8.9</i>	
	FINAL	<i>7.6</i>	<i>8.2</i>	<i>8.0</i>	<i>8.0</i>	<i>8.0</i>	<i>7.9</i>	<i>8.1</i>	
pH (s.u.)	INITIAL	<i>7.9</i>	<i>7.8</i>	<i>7.9</i>	<i>7.4</i>	<i>7.62</i>	<i>7.7</i>	<i>7.6</i>	
	FINAL	<i>7.9</i>	<i>8.0</i>	<i>7.9</i>	<i>7.9</i>	<i>7.7</i>	<i>7.7</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.1</i>	<i>23.6</i>	<i>23.0</i>	<i>22.9</i>	<i>24.2</i>	<i>23.3</i>	<i>24.8</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:							<i>8.5</i>		
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.7</i>	<i>8.5</i>	<i>8.4</i>	<i>9.0</i>	<i>9.3</i>	<i>9.1</i>	
	FINAL	<i>7.6</i>	<i>8.2</i>	<i>8.1</i>	<i>8.0</i>	<i>7.9</i>	<i>7.9</i>	<i>8.0</i>	
pH (s.u.)	INITIAL	<i>7.8</i>	<i>7.7</i>	<i>7.8</i>	<i>7.3</i>	<i>7.61</i>	<i>7.7</i>	<i>7.5</i>	
	FINAL	<i>7.6</i>	<i>7.9</i>	<i>7.9</i>	<i>7.9</i>	<i>7.7</i>	<i>7.6</i>	<i>7.9</i>	
temp (C)	INITIAL	<i>22.1</i>	<i>23.8</i>	<i>23.1</i>	<i>23.0</i>	<i>24.3</i>	<i>23.6</i>	<i>25.9</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:									
D.O. (mg/L)	INITIAL	<i>8.4</i>	<i>8.7</i>	<i>8.5</i>	<i>8.0</i>	<i>9.62</i>	<i>8.8</i>	<i>8.8</i>	
	FINAL	<i>7.5</i>	<i>8.2</i>	<i>8.1</i>	<i>8.0</i>	<i>7.9</i>	<i>8.0</i>	<i>8.1</i>	
pH (s.u.)	INITIAL	<i>7.8</i>	<i>7.6</i>	<i>7.7</i>	<i>7.2</i>	<i>7.45</i>	<i>7.4</i>	<i>7.3</i>	
	FINAL	<i>7.6</i>	<i>7.8</i>	<i>7.9</i>	<i>7.8</i>	<i>7.7</i>	<i>7.6</i>	<i>7.8</i>	
temp (C)	INITIAL	<i>22.2</i>	<i>23.9</i>	<i>23.2</i>	<i>23.1</i>	<i>24.7</i>	<i>23.3</i>	<i>27.4</i>	
	FINAL	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	<i>25.0</i>	
CONC:		100%	A	A	A	B	B	C	C
ALKALINITY (mg/L)		<i>30</i>			<i>29</i>		<i>18</i>		
HARDNESS (mg/L)		<i>52</i>			<i>34</i>		<i>38</i>		
CONDUCTIVITY (umhos/cm)		<i>697</i>			<i>727</i>		<i>644</i>		
CHLORINE (mg/L)		<i>0.05</i>			<i>0.05</i>		<i>0.05</i>		

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING		Cerodaphnia Dubia							
Lab # / Sample ID K1206003		Test Start (Date/Time) 6/7/12							
Client: Dequan		Test End (Date/Time) 6/14/12							
		Day of Test							
		1	2	3	4	5	6	7	notes/remarks
Control	MHS551	6/7	6/8	6/9	6/10	6/11	6/12	6/13	
D.O. (mg/L)	INITIAL	8.4	8.5	8.4	8.3	8.3	8.5	8.5	
	FINAL	8.3	8.4	8.4	8.7	8.6	8.6		
pH (s.u.)	INITIAL	8.2	8.2	8.0	7.8	7.9	7.7	7.6	
	FINAL	8.1	8.0	8.3	8.3	6.9	7.3		
temp (C)	INITIAL	22.9	22.2	22.7	22.5	23.4	22.5	22.0	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
ALKALINITY (mg/L)		58							
HARDNESS (mg/L)		82							
CONDUCTIVITY (umhos/cm)		301							
CHLORINE (mg/L)		0.05							
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.3	8.4	8.3	8.71	8.1	8.7	
	FINAL	8.5	8.3	8.4	8.4	8.4	8.5		
pH (s.u.)	INITIAL	7.9	7.7	8.0	7.6	7.8	7.7	7.6	
	FINAL	8.1	8.0	8.3	8.3	7.0	7.4		
temp (C)	INITIAL	22.0	23.3	22.7	22.7	24.0	24.0	23.9	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.5	8.4	8.3	8.71	9.1	8.8	
	FINAL	8.3	8.4	8.4	8.4	8.4	8.5		
pH (mg/L)	INITIAL	7.9	7.8	7.9	7.5	7.7	7.7	7.6	
	FINAL	8.1	7.9	8.2	8.2	7.2	7.6		
temp (C)	INITIAL	22.0	23.3	22.9	22.8	24.1	21.4	24.4	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.7	8.5	8.4	9.11	9.2	8.9	
	FINAL	8.5	8.4	8.3	8.3	8.4	8.5		
pH (s.u.)	INITIAL	7.9	7.8	7.9	7.4	7.62	7.7	7.6	
	FINAL	8.0	7.9	8.2	8.2	7.4	7.7		
temp (C)	INITIAL	22.1	23.6	23.0	22.9	24.2	23.3	24.8	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.7	8.5	8.4	9.0	8.8	9.1	
	FINAL	8.3	8.5	8.3	8.3	8.3	8.5		
pH (s.u.)	INITIAL	7.8	7.7	7.8	7.3	7.61	7.7	7.5	
	FINAL	8.0	7.9	8.1	8.1	7.4	7.8		
temp (C)	INITIAL	22.1	23.8	23.1	23.0	24.3	23.6	25.9	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC:									
D.O. (mg/L)	INITIAL	8.4	8.7	8.5	8.6	9.11	8.8	8.8	
	FINAL	8.3	8.5	8.3	8.4	8.4	8.5		
pH (s.u.)	INITIAL	7.8	7.7	7.7	7.2	7.45	7.4	7.3	
	FINAL	8.0	7.9	8.0	8.1	7.5	7.8		
temp (C)	INITIAL	22.2	23.9	23.2	23.1	24.7	23.3	27.4	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0		
CONC: 100%		A	A	A	B	B	C	C	
ALKALINITY (mg/L)		30			24		18		
HARDNESS (mg/L)		52			34		38		
CONDUCTIVITY (umhos/cm)		697			727		644		
CHLORINE (mg/L)		0.05			0.05		0.05		

APPENDIX C

Fathead minnow raw data and statistics

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID K1206003 TEST START DATE 6/9/12 TIME 1570
 CLIENT Delaware TEST END DATE 6/14/12 TIME 0855

Summary Page

		DAY (NUMBER SURVIVING)							SURVIVAL			
CONC:	REP #	start	1	2	3	4	5	6	7%	MEAN %	CV	
0	A	8	8	8	8	8	8	8	100	100	0.00	
	B	8	8	8	8	8	8	8	100			
	C	8	8	8	8	8	8	8	100			
	D	8	8	8	8	8	8	8	100			
	E	8	8	8	8	8	8	8	100			
3.2	A	8	8	8	8	8	8	8	100	100	0	
	B	8	8	8	8	8	8	8	100			
	C	8	8	8	8	8	8	8	100			
	D	8	8	8	8	8	8	8	100			
	E	8	8	8	8	8	8	8	100			
4.2	A	8	8	8	8	8	8	8	100	100		
	B	8	8	8	8	8	8	8	100			
	C	8	8	8	8	8	8	8	100			
	D	8	8	8	8	8	8	8	100			
	E	8	8	8	8	8	8	8	100			
16	A	8	8	8	8	8	8	8	100	97.5		
	B	8	8	8	8	8	8	8	100			
	C	8	8	8	8	8	8	8	100			
	D	8	8	8	8	8	8	8	100			
	E	8	8	8	8	8	8	8	100			
75	A	8	8	8	8	8	8	8	100	100		
	B	8	8	8	8	8	8	8	100			
	C	8	8	8	8	8	8	8	100			
	D	8	8	8	8	8	8	8	100			
	E	8	8	8	8	8	8	8	100			
160	A	8	8	8	8	8	8	8	100	100	0.00	
	B	8	8	8	8	8	8	8	100			
	C	8	8	8	8	8	8	8	100			
	D	8	8	8	8	8	8	8	100			
	E	8	8	8	8	8	8	8	100			
ANALYST												
DATE:												
TIME:												

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		TIME						
CLIENT		TEST END DATE		TIME						
AGE AND SOURCE OF MINNOWS										
D A Y (NUMBER SURVIVING)										
SURVIVAL										
REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
CONC: 0	A	2	2	2	2	2	2	2		
	B	1	2	2	2	1	1	1		
	C	1	2	2	2	1	1	1		
	D	1	2	2	2	1	1	1		
	E									
CONC: 32	A	2	2	2	2	2	2	2		
	B	1	2	2	2	1	1	1		
	C	1	2	2	2	1	1	1		
	D	1	2	2	2	1	1	1		
	E									
CONC: 42	A	2	2	2	2	2	2	2		
	B	1	2	2	2	1	1	1		
	C	1	2	2	2	1	1	1		
	D	1	2	2	2	1	1	1		
	E									
CONC: 56	A	2	2	2	2	2	2	2		
	B	1	2	2	2	1	1	1		
	C	1	2	2	2	1	1	1		
	D	1	2	2	2	1	1	1		
	E									
CONC: 75	A	2	2	2	2	2	2	2		
	B	1	2	2	2	1	1	1		
	C	1	2	2	2	1	1	1		
	D	1	2	2	2	1	1	1		
	E									
CONC: 100	A	2	2	2	2	2	2	2		
	B	1	2	2	2	1	1	1		
	C	1	2	2	2	1	1	1		
	D	1	2	2	2	1	1	1		
	E									
ANALYST	KP	KP	mb	mb	KP	KP	KP	KP		
DATE:	6/7/12	6/8/12	6-9-12	6-10-12	6/11/12	6/12/12	6/13/12	6/14/12		
TIME:	1510	1536	0900	1030	1330	1300	1100	0855		

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		6/7/12		TIME		1510	
CLIENT		Dequere		TEST END DATE		TIME			
AGE AND SOURCE OF MINNOWS									
B									
DAY (NUMBER SURVIVING)									
SURVIVAL									
REP #	start	1	2	3	4	5	6	7%	MEAN % CV
CONC: 0	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E	2	2	2	2	2	2	2	
CONC: 32	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E	2	2	2	2	2	2	2	
CONC: 42	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E	2	2	2	2	2	2	2	
CONC: 56	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E	2	2	2	2	2	2	2	
CONC: 75	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E	2	2	2	2	2	2	2	
CONC: 100	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E	2	2	2	2	2	2	2	
ANALYST	KP								
DATE:	6/7/12								
TIME:	1510								

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		6/7/12		TIME		1510			
CLIENT		Dequinn		TEST END DATE		TIME					
AGE AND SOURCE OF MINNOWS											
DAY (NUMBER SURVIVING)											
SURVIVAL											
	REP #	start	1	2	3	4	5	6	7%	MEAN % CV	
CONC: 0	A	2	2	2	2	2	2	2			
	B	1	2	2	2	2	2	2			
	C	1	2	2	2	2	2	2			
	D	1	2	2	2	2	2	2			
	E	1	2	2	2	2	2	2			
	REP #	start	1	2	3	4	5	6	7%	MEAN % CV	
CONC: 32	A	2	2	2	2	2	2	2			
	B	1	2	2	2	2	2	2			
	C	1	2	2	2	2	2	2			
	D	1	2	2	2	2	2	2			
	E	1	2	2	2	2	2	2			
	REP #	start	1	2	3	4	5	6	7%	MEAN % CV	
CONC: 42	A	2	2	2	2	2	2	2			
	B	1	2	2	2	2	2	2			
	C	1	2	2	2	2	2	2			
	D	1	2	2	2	2	2	2			
	E	1	2	2	2	2	2	2			
	REP #	start	1	2	3	4	5	6	7%	MEAN % CV	
CONC: 56	A	2	2	2	2	2	2	2			
	B	1	2	2	2	2	2	2			
	C	1	2	2	2	2	2	2			
	D	1	2	2	2	2	2	2			
	E	1	2	2	2	2	2	2			
	REP #	start	1	2	3	4	5	6	7%	MEAN % CV	
CONC: 75	A	2	2	2	2	2	2	2			
	B	1	2	2	2	2	2	2			
	C	1	2	2	2	2	2	2			
	D	1	2	2	2	2	2	2			
	E	1	2	2	2	2	2	2			
	REP #	start	1	2	3	4	5	6	7%	MEAN % CV	
CONC: 100	A	2	2	2	2	2	2	2			
	B	1	2	2	2	2	2	2			
	C	1	2	2	2	2	2	2			
	D	1	2	2	2	2	2	2			
	E	1	2	2	2	2	2	2			
ANALYST	KP										
DATE:	6/7/12										
TIME:	1510										

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		6/7/12		TIME		1510	
CLIENT		TEST END DATE				TIME			
		AGE AND SOURCE OF MINNOWS							
		DAY (NUMBER SURVIVING)						SURVIVAL	
REP #	start	1	2	3	4	5	6	7%	MEAN % CV
CONC: 0	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E								
CONC: 32	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E								
CONC: 12	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E								
CONC: 56	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	1	1	1	1	1	1	
	E								
CONC: 75	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E								
CONC: 100	A	2	2	2	2	2	2	2	
	B	2	2	2	2	2	2	2	
	C	2	2	2	2	2	2	2	
	D	2	2	2	2	2	2	2	
	E								
ANALYST	KP								
DATE:	6/7/12								
TIME:	1510								

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID		TEST START DATE		TIME						
CLIENT		TEST END DATE		TIME						
AGE AND SOURCE OF MINNOWS										
DAY (NUMBER SURVIVING)										
SURVIVAL										
REP #	start	1	2	3	4	5	6	7%	MEAN %	CV
CONC: 0	A	2	2	2	2	2	2	2		
	B			2	2					
	C			2	2					
	D			2	2					
	E									
CONC: 32	A	2	2	2	2	2	2	2		
	B			2	2					
	C			2	2					
	D			2	2					
	E									
CONC: 42	A	2	2	2	2	2	2	2		
	B			2	2					
	C			2	2					
	D			2	2					
	E									
CONC: 56	A	2	2	2	2	2	2	2		
	B			2	2					
	C			2	2					
	D			2	2					
	E									
CONC: 75	A	2	2	2	2	2	2	2		
	B			2	2					
	C			2	2					
	D			2	2					
	E									
CONC: 100	A	2	2	2	2	2	2	2		
	B			2	2					
	C			2	2					
	D			2	2					
	E									
ANALYST	KP									
DATE:	6/7/12									
TIME:	1510									

CV = PERCENT COEFFICIENT OF VARIATION: STANDARD DEVIATION/MEAN * 100

Pimephales promelas

FATHEAD MINNOW

TEST 1000.0

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB # / #s:		K1206003			TEST DATES (BEGIN / END):		6/7-14/12	
CLIENT:		City of DeQueen			WEIGHING DATE / TIME:		6-15-12, 1400	
ANALYSTS:		KP			DRYING TEMP (DEGREES C):		60	
SAMPLE ID:					DRYING TIME (HOURS):		24	
	REP #	FINAL DRY WEIGHT TIN+LARVAE (g)	INITIAL WEIGHT TIN (g)	TOTAL DRY WEIGHT OF LARVAE (g)	NUMBER OF LARVAE	DRY WEIGHT OF LARVAE (mg)		
CONTROL	A	0.97700	0.97416	0.00284	8	0.355	AVG DRY	
	B	0.97542	0.97294	0.00248	8	0.310	WEIGHT (mg)	
	C	1.01428	1.01093	0.00335	8	0.419	0.381	
	D	0.98611	0.98277	0.00334	8	0.418	CV	
	E	0.96878	0.96556	0.00322	8	0.403	12.41	
CONC:	A	0.97254	0.96996	0.00258	8	0.322	AVG DRY	
	B	0.97711	0.97383	0.00328	8	0.410	WEIGHT (mg)	
	32% C	0.98327	0.97983	0.00344	8	0.430	0.401	
	D	1.00341	0.99987	0.00354	8	0.442	CV	
	E	1.02314	1.01993	0.00321	8	0.401		
CONC:	A	1.00698	1.00403	0.00295	8	0.369	AVG DRY	
	B	0.99814	0.99486	0.00328	8	0.410	WEIGHT (mg)	
	42% C	1.00491	1.00186	0.00305	8	0.381	0.388	
	D	1.00834	1.00555	0.00279	8	0.349	CV	
	E	0.97488	0.97143	0.00345	8	0.431		
CONC:	A	0.99367	0.99115	0.00252	8	0.315	AVG DRY	
	B	1.02483	1.02207	0.00276	8	0.345	WEIGHT (mg)	
	56% C	0.98755	0.98473	0.00282	8	0.353	0.364	
	D	0.99380	0.99046	0.00334	8	0.418	CV	
	E	0.98627	0.98317	0.00310	8	0.387		
CONC:	A	0.98040	0.97824	0.00216	8	0.270	AVG DRY	
	B	1.00010	0.99702	0.00308	8	0.385	WEIGHT (mg)	
	75% C	1.00548	1.00247	0.00301	8	0.376	0.376	
	D	1.00674	1.00344	0.00330	8	0.412	CV	
	E	1.01380	1.01031	0.00349	8	0.436		
CONC:	A	0.99026	0.98764	0.00262	8	0.328	AVG DRY	
	B	0.98426	0.98082	0.00344	8	0.430	WEIGHT (mg)	
	100% C	0.96535	0.96182	0.00353	8	0.441	0.423	
	D	0.97411	0.97055	0.00356	8	0.445	CV	
	E	0.98395	0.98018	0.00377	8	0.471	13.12	

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.027

W = 0.416

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

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

Data FAIL normality test. Try another transformation.

Warning - The first three homogeneity tests are sensitive to non-normal data and should not be performed.

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

Hartley's test for homogeneity of variance

Bartlett's test for homogeneity of variance

These two tests can not be performed because at least one group has zero variance.

Data FAIL to meet homogeneity of variance assumption.
Additional transformations are useless.

TITLE: AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
FILE: Z:\TOXSTAT\MONTE\FHSURV.
TRANSFORM: ARC SINE(SQUARE ROOT(Y)) NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	1.0000	1.3931
1	CONTROL	2	1.0000	1.3931
1	CONTROL	3	1.0000	1.3931
1	CONTROL	4	1.0000	1.3931
1	CONTROL	5	1.0000	1.3931
2	32 % EFFLUENT	1	1.0000	1.3931
2	32 % EFFLUENT	2	1.0000	1.3931
2	32 % EFFLUENT	3	1.0000	1.3931
2	32 % EFFLUENT	4	1.0000	1.3931
2	32 % EFFLUENT	5	1.0000	1.3931

3	42 %	EFFLUENT	1	1.0000	1.3931
3	42 %	EFFLUENT	2	1.0000	1.3931
3	42 %	EFFLUENT	3	1.0000	1.3931
3	42 %	EFFLUENT	4	1.0000	1.3931
3	42 %	EFFLUENT	5	1.0000	1.3931
4	56 %	EFFLUENT	1	1.0000	1.3931
4	56 %	EFFLUENT	2	1.0000	1.3931
4	56 %	EFFLUENT	3	1.0000	1.3931
4	56 %	EFFLUENT	4	0.8750	1.2094
4	56 %	EFFLUENT	5	1.0000	1.3931
5	75 %	EFFLUENT	1	1.0000	1.3931
5	75 %	EFFLUENT	2	1.0000	1.3931
5	75 %	EFFLUENT	3	1.0000	1.3931
5	75 %	EFFLUENT	4	1.0000	1.3931
5	75 %	EFFLUENT	5	1.0000	1.3931
6	100 %	EFFLUENT	1	1.0000	1.3931
6	100 %	EFFLUENT	2	1.0000	1.3931
6	100 %	EFFLUENT	3	1.0000	1.3931
6	100 %	EFFLUENT	4	1.0000	1.3931
6	100 %	EFFLUENT	5	1.0000	1.3931

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.006	0.001	1.000
Within (Error)	24	0.027	0.001	
Total	29	0.033		

Critical F value = 2.62 (0.05,5,24)
 Since F < Critical F FAIL TO REJECT Ho: All equal

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHSURV. Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST - TABLE 1 OF 2 Ho: Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	CONTROL	1.393	1.000		
2	32 % EFFLUENT	1.393	1.000	0.000	
3	42 % EFFLUENT	1.393	1.000	0.000	
4	56 % EFFLUENT	1.356	0.975	1.732	
5	75 % EFFLUENT	1.393	1.000	0.000	
6	100 % EFFLUENT	1.393	1.000	0.000	

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

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12

File: Z:\TOXSTAT\MONTE\FHSURV.

Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST

TABLE 2 OF 2

Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	CONTROL	5			
2	32 % EFFLUENT	5	0.020	2.0	0.000
3	42 % EFFLUENT	5	0.020	2.0	0.000
4	56 % EFFLUENT	5	0.020	2.0	0.025
5	75 % EFFLUENT	5	0.020	2.0	0.000
6	100 % EFFLUENT	5	0.020	2.0	0.000

AA# K1206003, FATHEAD MINNOW, CHRONIC, 6-7-12

File: Z:\TOXSTAT\MONTE\FHSURV.

Transform: ARC SINE(SQUARE ROOT(Y))

STEEL'S MANY-ONE RANK TEST

Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	1.393				
2	32 % EFFLUENT	1.393	27.50	16.00	5.00	
3	42 % EFFLUENT	1.393	27.50	16.00	5.00	
4	56 % EFFLUENT	1.356	25.00	16.00	5.00	
5	75 % EFFLUENT	1.393	27.50	16.00	5.00	
6	100 % EFFLUENT	1.393	27.50	16.00	5.00	

Critical values use k = 5, are 1 tailed, and alpha = 0.05

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.061

W = 0.910

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

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

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

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 2.06

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

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

TITLE: AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
FILE: Z:\TOXSTAT\MONTE\FHGR.
TRANSFORM: ARC SINE(SQUARE ROOT(Y)) NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.3550	0.6383
1	CONTROL	2	0.3100	0.5905
1	CONTROL	3	0.4190	0.7040
1	CONTROL	4	0.4180	0.7030
1	CONTROL	5	0.4030	0.6878
2	32 % EFFLUENT	1	0.3220	0.6034
2	32 % EFFLUENT	2	0.4100	0.6949
2	32 % EFFLUENT	3	0.4300	0.7152
2	32 % EFFLUENT	4	0.4420	0.7273
2	32 % EFFLUENT	5	0.4010	0.6857
3	42 % EFFLUENT	1	0.3690	0.6529
3	42 % EFFLUENT	2	0.4100	0.6949
3	42 % EFFLUENT	3	0.3810	0.6652
3	42 % EFFLUENT	4	0.3490	0.6320
3	42 % EFFLUENT	5	0.4310	0.7162
4	56 % EFFLUENT	1	0.3150	0.5959

4	56 %	EFFLUENT	2	0.3450	0.6278
4	56 %	EFFLUENT	3	0.3530	0.6362
4	56 %	EFFLUENT	4	0.4180	0.7030
4	56 %	EFFLUENT	5	0.3870	0.6714
5	75 %	EFFLUENT	1	0.2700	0.5464
5	75 %	EFFLUENT	2	0.3850	0.6694
5	75 %	EFFLUENT	3	0.3760	0.6601
5	75 %	EFFLUENT	4	0.4120	0.6969
5	75 %	EFFLUENT	5	0.4360	0.7212
6	100 %	EFFLUENT	1	0.3280	0.6098
6	100 %	EFFLUENT	2	0.4300	0.7152
6	100 %	EFFLUENT	3	0.4410	0.7263
6	100 %	EFFLUENT	4	0.4450	0.7303
6	100 %	EFFLUENT	5	0.4710	0.7564

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.011	0.002	0.898
Within (Error)	24	0.061	0.003	
Total	29	0.073		

Critical F value = 2.62 (0.05,5,24)
 Since $F < \text{Critical } F$ FAIL TO REJECT H_0 : All equal

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST - TABLE 1 OF 2 H_0 : Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	CONTROL	0.665	0.381		
2	32 % EFFLUENT	0.685	0.401	-0.643	
3	42 % EFFLUENT	0.672	0.388	-0.235	
4	56 % EFFLUENT	0.647	0.364	0.558	
5	75 % EFFLUENT	0.659	0.376	0.185	
6	100 % EFFLUENT	0.708	0.423	-1.339	

Dunnnett table value = 2.36 (1 Tailed Value, $P=0.05$, $df=24,5$)

AA# K1206003 FATHEAD MINNOW GROWTH CHRONIC, 6-7-12
 File: Z:\TOXSTAT\MONTE\FHGR. Transform: ARC SINE(SQUARE ROOT(Y))

DUNNETT'S TEST - TABLE 2 OF 2 H_0 : Control < Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	CONTROL	5			
2	32 % EFFLUENT	5	0.072	18.8	-0.020
3	42 % EFFLUENT	5	0.072	18.8	-0.007
4	56 % EFFLUENT	5	0.072	18.8	0.017
5	75 % EFFLUENT	5	0.072	18.8	0.005
6	100 % EFFLUENT	5	0.072	18.8	-0.042

APPENDIX D

Ceriodaphnia dubia Raw Data and Statistics

Cerodaphnia dubia

SURVIVAL AND REPRODUCTION TEST

Discharger: Regener Lab Number/s: K120203
 Location: K120203
 Date Sample Collected:

Analyst: KP
 Test Start - Date/Time: 6/17/12, 1500
 Test Stop - Date/Time: 6/19/12, 0835

Conc 1	% Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst	
		A	B	C	D	E	F	G	H	I	J					
0	1	0	0	0	0	0	0	0	0	0	0	0	10	0	0	KP
	2	0	0	0	0	0	0	0	0	0	0	0	0	10	0	mb
	3	0	0	1	0	0	0	1	0	2	3	7	9	0.5	0.5	KP
	4	4	4	1	0	-	3	1	2	4	1	29	9	7.8	7.8	KP
	5	8	9	1	7	-	5	2	8	1	2	43	9	6.8	6.8	KP
	6	7	5	9	9	-	8	9	3	6	9	16	9	7.2	7.2	KP
	7															
	8															
Total		19	18	12	16	x0	16	13	18	13	15	140				

Conc 4	% Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst	
		A	B	C	D	E	F	G	H	I	J					
56	1	0	0	0	0	0	0	0	0	0	0	0	10	0		
	2	0	0	0	0	0	0	0	0	0	0	0	10	0		
	3	2	0	0	0	0	0	0	0	0	0	2	10	0.2		
	4	6	9	5	7	5	6	5	7	3	2	51	10	5.1		
	5	6	4	0	7	9	3	8	1	6	49	10	4.9			
	6	11	5	9	1	2	0	11	10	0	54	10	5.4			
	7															
	8															
Total		25	14	14	15	12	15	19	25	4	13	156				

Conc 2	% Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst	
		A	B	C	D	E	F	G	H	I	J					
32	1	0	0	0	0	0	0	0	0	0	0	0	9	0		
	2	0	0	0	-	0	0	0	0	0	0	0	9	0		
	3	2	2	3	-	0	0	0	0	0	0	14	9	1.6		
	4	5	4	7	-	9	5	5	2	3	6	46	9	6.1		
	5	10	3	3	-	5	8	0	7	1	6	38	9	4.2		
	6	2	16	17	-	0	2	11	11	2	7	63	9	7.0		
	7															
	8															
Total		19	25	25	x0	14	15	16	17	8	12	161				

Conc 5	% Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst	
		A	B	C	D	E	F	G	H	I	J					
75	1	0	0	0	0	0	0	0	0	0	0	0	10	0		
	2	0	0	0	0	0	0	0	0	0	0	0	10	0		
	3	4	7	1	0	0	1	1	0	3	0	11	10	1.1		
	4	6	3	6	5	6	1	5	3	4	5	38	10	3.8		
	5	5	8	7	4	2	10	1	0	4	3	44	10	4.4		
	6	10	4	7	4	0	8	3	0	8	7	53	10	5.3		
	7															
	8															
Total		19	16	21	13	8	12	20	3	19	15	146				

Conc 3	% Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst	
		A	B	C	D	E	F	G	H	I	J					
42	1	0	0	0	0	0	0	0	0	0	0	0	10	0		
	2	0	0	0	0	0	0	0	0	0	0	0	10	0		
	3	0	0	1	3	0	0	0	0	1	0	5	10	0.5		
	4	9	7	3	6	5	7	3	6	5	3	49	10	4.9		
	5	13	10	3	5	6	7	3	9	8	3	61	10	6.1		
	6	4	7	6	8	1	9	7	0	2	4	43	10	4.3		
	7															
	8															
Total		26	19	13	22	12	12	13	15	16	10	158				

Conc 6	% Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst	
		A	B	C	D	E	F	G	H	I	J					
100	1	0	0	0	0	0	0	0	0	0	0	0	9	0		
	2	0	0	0	-	0	0	0	0	0	0	0	9	0		
	3	0	0	0	-	0	0	0	0	0	1	1	9	0.1		
	4	4	6	0	-	0	5	3	5	2	4	29	9	3.2		
	5	7	3	2	-	5	9	2	10	0	9	45	9	5.0		
	6	3	12	5	-	7	1	7	2	11	6	54	9	6.0		
	7															
	8															
Total		14	20	11	x0	12	15	12	17	13	15	129				

X = DEAD; Y = MALE

F = 14.3
 CV = 19.7

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

Shapiro - Wilk's test for normality

***** Shapiro - Wilk's Test is aborted *****

This test can not be performed because total number of replicates
is greater than 50.

Total number of replicates = 60

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 2.18

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

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

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	9	1	10
32%	9	1	10
TOTAL	18	2	20

CRITICAL FISHER'S VALUE (10,10,9) ($p=0.05$) IS 4. b VALUE IS 9.
 Since b is greater than 4 there is no significant difference
 between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	DEAD	ALIVE	TOTAL ANIMALS
CONTROL	1	9	10
42%	0	10	10
TOTAL	1	19	20

CRITICAL FISHER'S VALUE (10,10,1) ($p=0.05$) IS LESS THAN 0. b VALUE IS 0.
 NO SIGNIFICANT DIFFERENCE

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	DEAD	ALIVE	TOTAL ANIMALS
CONTROL	1	9	10
56%	0	10	10

TOTAL 1 19 20

CRITICAL FISHER'S VALUE (10,10,1) (p=0.05) IS LESS THAN 0. b VALUE IS 0.
NO SIGNIFICANT DIFFERENCE

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	DEAD	ALIVE	TOTAL ANIMALS
CONTROL	1	9	10
75%	0	10	10
TOTAL	1	19	20

CRITICAL FISHER'S VALUE (10,10,1) (p=0.05) IS LESS THAN 0. b VALUE IS 0.
NO SIGNIFICANT DIFFERENCE

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	9	1	10
100%	9	1	10
TOTAL	18	2	20

CRITICAL FISHER'S VALUE (10,10,9) (p=0.05) IS 4. b VALUE IS 9.
Since b is greater than 4 there is no significant difference
between CONTROL and TREATMENT at the 0.05 level.

SUMMARY OF FISHER'S EXACT TESTS

GROUP	IDENTIFICATION	NUMBER EXPOSED	NUMBER DEAD	SIG (P=.05)
	CONTROL	10	1	

1	32%	10	1
2	42%	10	0
3	56%	10	0
4	75%	10	0
5	100%	10	1

TITLE: AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
FILE: Z:\TOXSTAT\MONTE\CD.
TRANSFORM: NO TRANSFORMATION NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	19.0000	19.0000
1	CONTROL	2	18.0000	18.0000
1	CONTROL	3	12.0000	12.0000
1	CONTROL	4	16.0000	16.0000
1	CONTROL	5	0.0000	0.0000
1	CONTROL	6	16.0000	16.0000
1	CONTROL	7	13.0000	13.0000
1	CONTROL	8	18.0000	18.0000
1	CONTROL	9	13.0000	13.0000
1	CONTROL	10	15.0000	15.0000
2	32 % EFFLUENT	1	19.0000	19.0000
2	32 % EFFLUENT	2	25.0000	25.0000
2	32 % EFFLUENT	3	25.0000	25.0000
2	32 % EFFLUENT	4	0.0000	0.0000
2	32 % EFFLUENT	5	14.0000	14.0000
2	32 % EFFLUENT	6	15.0000	15.0000
2	32 % EFFLUENT	7	16.0000	16.0000
2	32 % EFFLUENT	8	17.0000	17.0000
2	32 % EFFLUENT	9	8.0000	8.0000
2	32 % EFFLUENT	10	22.0000	22.0000
3	42 % EFFLUENT	1	26.0000	26.0000
3	42 % EFFLUENT	2	19.0000	19.0000
3	42 % EFFLUENT	3	13.0000	13.0000
3	42 % EFFLUENT	4	22.0000	22.0000
3	42 % EFFLUENT	5	12.0000	12.0000
3	42 % EFFLUENT	6	12.0000	12.0000
3	42 % EFFLUENT	7	13.0000	13.0000
3	42 % EFFLUENT	8	15.0000	15.0000
3	42 % EFFLUENT	9	16.0000	16.0000
3	42 % EFFLUENT	10	10.0000	10.0000
4	56 % EFFLUENT	1	25.0000	25.0000
4	56 % EFFLUENT	2	14.0000	14.0000
4	56 % EFFLUENT	3	14.0000	14.0000
4	56 % EFFLUENT	4	15.0000	15.0000
4	56 % EFFLUENT	5	12.0000	12.0000
4	56 % EFFLUENT	6	15.0000	15.0000
4	56 % EFFLUENT	7	19.0000	19.0000
4	56 % EFFLUENT	8	25.0000	25.0000
4	56 % EFFLUENT	9	4.0000	4.0000
4	56 % EFFLUENT	10	13.0000	13.0000
5	75 % EFFLUENT	1	19.0000	19.0000

5	75 % EFFLUENT	2	16.0000	16.0000
5	75 % EFFLUENT	3	21.0000	21.0000
5	75 % EFFLUENT	4	13.0000	13.0000
5	75 % EFFLUENT	5	8.0000	8.0000
5	75 % EFFLUENT	6	12.0000	12.0000
5	75 % EFFLUENT	7	20.0000	20.0000
5	75 % EFFLUENT	8	3.0000	3.0000
5	75 % EFFLUENT	9	19.0000	19.0000
5	75 % EFFLUENT	10	15.0000	15.0000
6	100 % EFFLUENT	1	14.0000	14.0000
6	100 % EFFLUENT	2	20.0000	20.0000
6	100 % EFFLUENT	3	11.0000	11.0000
6	100 % EFFLUENT	4	0.0000	0.0000
6	100 % EFFLUENT	5	12.0000	12.0000
6	100 % EFFLUENT	6	15.0000	15.0000
6	100 % EFFLUENT	7	12.0000	12.0000
6	100 % EFFLUENT	8	17.0000	17.0000
6	100 % EFFLUENT	9	13.0000	13.0000
6	100 % EFFLUENT	10	15.0000	15.0000

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
 File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	76.133	15.227	0.426
Within (Error)	54	1928.200	35.707	
Total	59	2004.333		

Critical F value = 2.45 (0.05,5,40)
 Since F < Critical F FAIL TO REJECT Ho: All equal

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12
 File: Z:\TOXSTAT\MONTE\CD. Transform: NO TRANSFORMATION

DUNNETT'S TEST - TABLE 1 OF 2 Ho: Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	CONTROL	14.000	14.000		
2	32 % EFFLUENT	16.100	16.100	-0.786	
3	42 % EFFLUENT	15.800	15.800	-0.674	
4	56 % EFFLUENT	15.600	15.600	-0.599	
5	75 % EFFLUENT	14.600	14.600	-0.225	
6	100 % EFFLUENT	12.900	12.900	0.412	

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

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12

File: Z:\TOXSTAT\MONTE\CD.

Transform: NO TRANSFORMATION

DUNNETT'S TEST

TABLE 2 OF 2

Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	CONTROL	10			
2	32 % EFFLUENT	10	6.173	44.1	-2.100
3	42 % EFFLUENT	10	6.173	44.1	-1.800
4	56 % EFFLUENT	10	6.173	44.1	-1.600
5	75 % EFFLUENT	10	6.173	44.1	-0.600
6	100 % EFFLUENT	10	6.173	44.1	1.100

AA # K1206003, C. DUBIA CHRONIC, REPRODUCCION, 6-7-12

File: Z:\TOXSTAT\MONTE\CD.

Transform: NO TRANSFORMATION

STEEL'S MANY-ONE RANK TEST

Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	14.000				
2	32 % EFFLUENT	16.100	117.50	75.00	10.00	
3	42 % EFFLUENT	15.800	105.00	75.00	10.00	
4	56 % EFFLUENT	15.600	107.00	75.00	10.00	
5	75 % EFFLUENT	14.600	111.00	75.00	10.00	
6	100 % EFFLUENT	12.900	92.50	75.00	10.00	

Critical values use $k = 5$, are 1 tailed, and $\alpha = 0.05$

APPENDIX E

Organism History

AQUATOX, INC.

416 TWIN POINTS ROAD
HOT SPRINGS, ARKANSAS 71913
501-520-0560

TEST ORGANISM HISTORY

DATE SHIPPED 6/7/12 CLIENT AR Analytical
Ken

Purchase Order #: _____

SPECIES: Pimephales promelas

Quantity Shipped: 300

Age: hatched 6/6/12 1500 EST

Brood Stock Source: Anderson Farms, AR

Culture Water: Groundwater

Hardness (Mg/l CaCO₃): 160

Dissolved Oxygen (Mg/l): 8.2

Temperature (°C): 25.1°C

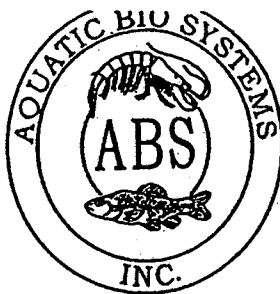
Feeding: Artemia

Comments: _____

Shipped Via: Federal Express UPS Overnight Shuttle

Packaged By: _____

1300 Blue Spruce Drive, Suite C
Fort Collins, Colorado 80524



Toll Free: 800/331-5916
Tel: 970/484-5091 Fax: 970/484-2514

ORGANISM HISTORY

DATE: 6/22/09

SPECIES: Ceriodaphnia dubia

AGE: Variable

LIFE STAGE: Adult

HATCH DATE: Variable

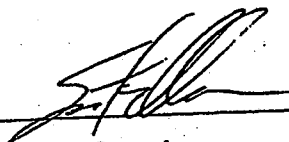
BEGAN FEEDING: Immediately

FOOD: YTC, Selenastrum sp.

Water Chemistry Record:

	Current	Range
TEMPERATURE:	<u>25°C</u>	<u>20-25°C</u>
SALINITY/CONDUCTIVITY:	<u>-</u>	<u>-</u>
TOTAL HARDNESS (as CaCO ₃):	<u>142 mg/l</u>	<u>86-124 mg/l</u>
TOTAL ALKALINITY (as CaCO ₃):	<u>100 mg/l</u>	<u>65-130 mg/l</u>
pH:	<u>7.92</u>	<u>7.56-8.35</u>

Comments:

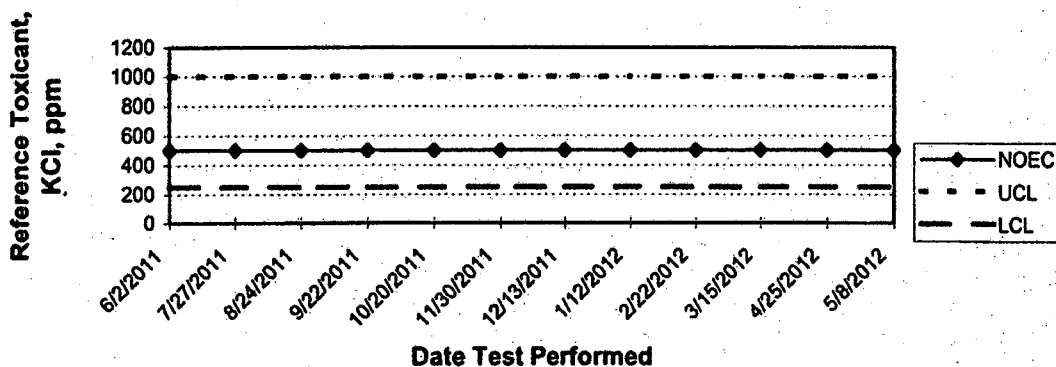


Facility Supervisor

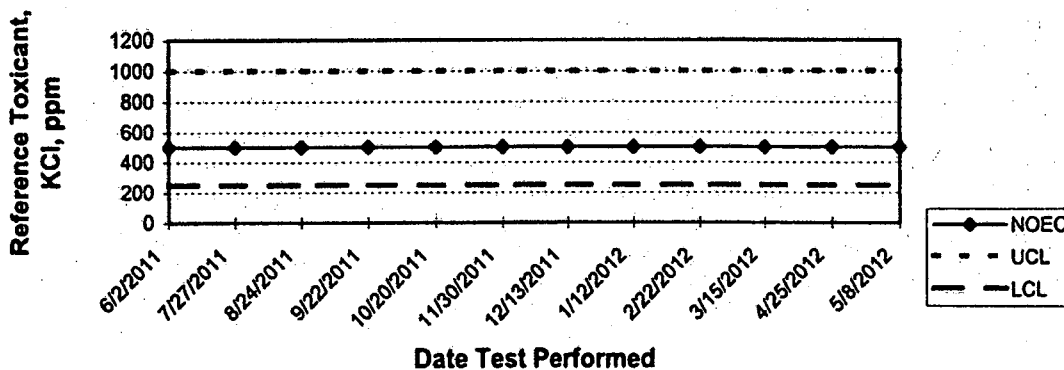
APPENDIX F

Quality Assurance Charts

ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW SURVIVAL
QUALITY ASSURANCE



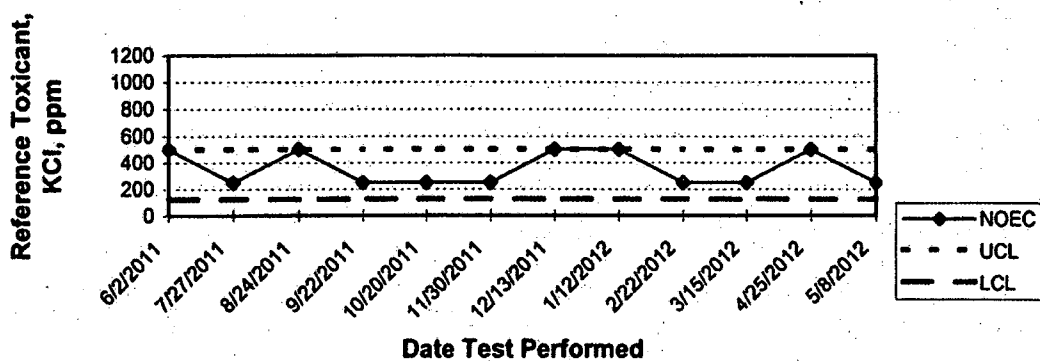
ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW GROWTH
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.

CERIODAPHNIA DUBIA SURVIVAL

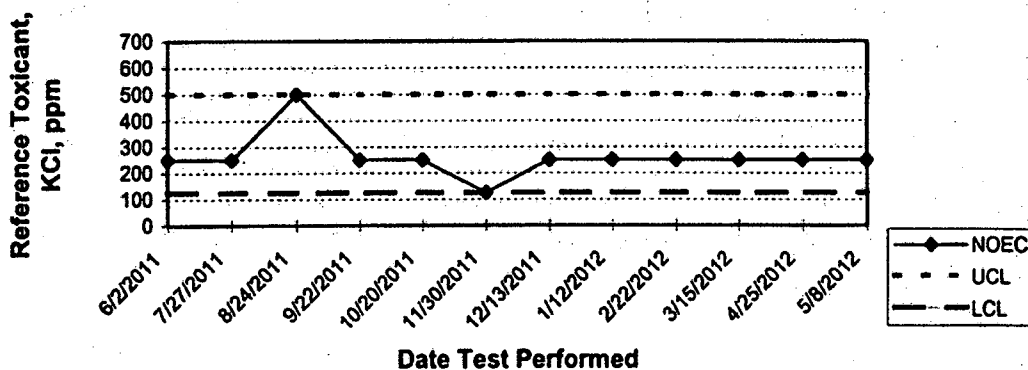
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.

CERIODAPHNIA DUBIA REPRODUCTION

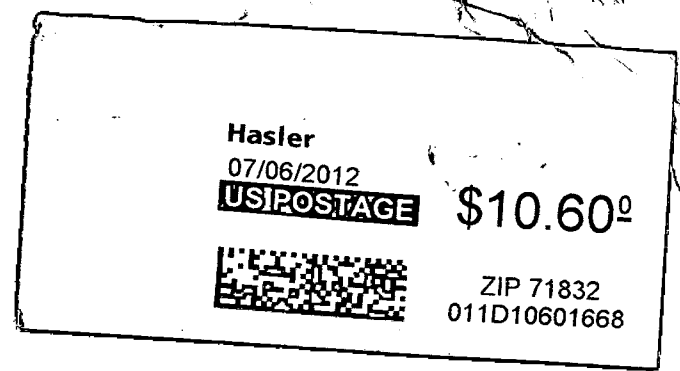
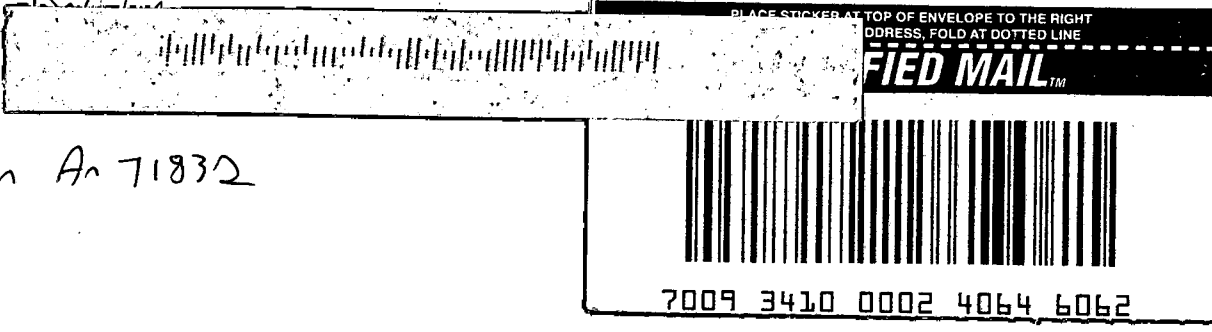
QUALITY ASSURANCE



City of DeQueen

P.O. Box

DeQueen Ar 71832



ADEQ
 NPDES Enforcement Section
 5301 Northshore Dr.
 North Little Rock Ar. 72118-5317