Arkansas Analytical, Inc.

Toxicity Test Results

MAGCOBAR MINE SITE
NPDES PERMIT NUMBER: AR0049794
June 2009
AFIN# 00-00348

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

Ceriodaphnia dubia, Survival and Reproduction Test
Test 1002.0

Prepared for: **Mr. David Friedman**
EEMA O&M Services Group
P.O. Box 232
Kulpsville, PA 19443

Prepared by: Arkansas Analytical, Inc.
11701 I-30, Bldg 1, Suite 115
Little Rock, Arkansas 72209
Lab Number K906006

Wednesday, July 08, 2009

Introduction

This report contains test results for toxicity testing for the Magcobar Mine Site. The NPDES permit number is AR0049794. The facility is located one mile northeast of Magnet Cove in Sections 10, 11, 14, & 15, Township 3 South, Range 17 West in Hot Springs County, Arkansas. The facility discharges into Chamberlain Creek, thence to Cove Creek, thence to Ouachita River in Segment 2F of the Ouachita River Basin.

The permit requires chronic biomonitoring testing bi-monthly for both *Ceriodaphnia dubia* and *Pimephales promelas*. The test results in this report represent the testing for June of 2009.

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:	6-3-09, 0905	6-4-09, 0905
Sample #2:	6-4-09, 0837	6-5-09, 0837
Sample #3:	6-8-09, 0730	6-9-09, 0730

The samples were composites collected at the final discharge from the Magcobar mine site.

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:	6-4-09, 1344	4
Sample #2:	6-5-09, 1256	4
Sample #3:	6-9-09, 1329	4

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. Synthetic dilution water was substituted either because zero flow conditions existed or due to an earlier characterization of the receiving water as being toxic.

Each sample 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	80%	X	
Average of 15 or more young per surviving female	16.6	X	
At least 60% of surviving females should have produced 3 broods	100%	X	
The percent coefficient of variation between replicates must be 40% or less for the young of surviving females	11.1%	X	

TEST ACCEPTANCE CRITERIA for *Pimephales promelas*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	95%	X	
The percent coefficient of variation between replicates must be 40% or less for survival	7.21%	X	
Minimum of 0.25 mg average dry weight of surviving controls	0.555	X	
The percent coefficient of variation between replicates must be 40% or less for growth	25.8%	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

Ceriodaphnia dubia 5/28/09, 6/4/09		*Pimephales promelas* 5/28/09, 6/4/09	
NOEC Survival:	125 ppm KCl	NOEC Survival:	500 ppm KCl
LOEC Survival:	250 ppm KCl	LOEC Survival:	1000 ppm KCl
NOEC Reproduction:	125 ppm KCl	NOEC Growth:	500 ppm KCl
LOEC Reproduction:	250 ppm KCl	LOEC Growth:	1000 ppm KCl

Quality Assurance charts are provided in Appendix F.

Summary of Results
Magcobar Mine Site

Ceriodaphnia dubia		*Pimephales promelas*	
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)	16.9	%CV survival (critical dilution)	5.73%
%CV Reproduction (critical dilution)	16.1%	Mean dry weight (critical dilution) in milligrams	0.670
		%CV growth (critical dilution)	12.4%
PMSD Reproduction	46.4	PMSD Growth	30.6

Conclusion

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

The permit issued to the Magcobar Mine Site, AR0049794, 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.

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

The permit issued to the Magcobar Mine Site, AR0049794, 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:

Ken Pigue

Melissa Green

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

PERMITTEE: Magcobar Mine Site **NPDES #: AR0049794**

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-3-09, 0905	6-4-09, 0905
Sample #2:	6-4-09, 0837	6-5-09, 0837
Sample #3:	6-8-09, 0730	6-9-09, 0730

Test initiated (date, time): 6-5-09, 1215 Test terminated (date, time): 6-12-09, 1420

Dilution water used: Soft Synthetic

DATA TABLE FOR FATHEAD MINNOW SURVIVAL

	Percent Survival in Replicate Chambers						**Mean Percent Survival**			
Effluent Conc %	A	B	C	D	E		24 hours	48 hours	7 days	CV %
0%	87.5	87.5	100	100	100		100	97.5	95	7.21
32%	100	87.5	100	100	100		100	100	97.5	
42%	100	100	100	100	100		100	100	100	
56%	100	100	87.5	100	100		100	100	97.5	
75%	100	100	87.5	100	100		100	100	97.5	
100%	100	87.5	100	100	100		100	100	97.5	5.73

DATA TABLE FOR GROWTH OF FATHEAD MINNOWS

SUMMARY

Effluent Conc %	A	B	C	D	E		Mean Dry Weight	CV%
0%	0.765	0.449	0.425	0.500	0.636		0.555	25.8
32%	0.384	0.430	0.564	0.442	0.629		0.490	
42%	0.723	0.458	0.598	0.541	0.512		0.566	
56%	0.510	0.600	0.438	0.694	0.653		0.579	
75%	0.815	0.581	0.600	0.607	0.534		0.627	
100%	0.661	0.560	0.624	0.750	0.754		0.670	12.4

Coefficient of Variation = standard deviation / mean * 100

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)= _____**25.8**_____%

SUMMARY REPORTING FORMS FOR CHRONIC BIOMONITORING
Ceriodaphnia dubia SURVIVAL AND REPRODUCTION

Permittee: Magcobar Mine Site **NPDES #: AR0049794**

Sample Collection:	Date, Time Started	Date, Time Ended
Sample #1:	6-3-09, 0905	6-4-09, 0905
Sample #2:	6-4-09, 0837	6-5-09, 0837
Sample #3:	6-8-09, 0730	6-9-09, 0730

Test initiated (date, time): 6-5-09, 1205 Test terminated (date, time): 6-12-09, 1345

Dilution water used: Soft Synthetic

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

Replicate	0%	32%	42%	56%	75%	100%
A	14	17	19	x0	18	15
B	15	16	20	11	13	19
C	17	11	x13	7	21	x7
D	15	20	17	20	27	21
E	19	21	23	16	26	16
F	17	x5	9	26	x0	12
G	19	19	18	16	19	18
H	x10	x0	x3	19	13	17
I	x10	18	17	21	x0	15
J	17	17	26	x1	16	19
Mean	15.3	14.4	16.5	13.7	15.3	15.9
Mean/surviving female	16.6	17.4	18.6	17.0	19.1	16.9
CV%*	11.1					16.1

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: Magcobar Mine Site **NPDES #: AR0049794**

PERCENT SURVIVAL

PERCENT EFFLUENT	0%	32%	42%	56%	75%	100%
Time of Reading: 24 HOURS	100	100	100	100	100	100
48 HOURS	100	100	100	100	100	100
Test termination	80	80	80	80	80	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)= _____**16.1**_____%

APPENDIX A

Chain of Custody Forms

Arkansas Analytical Inc.
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		Project Description	Turnaround Time
EEMA O & M Services Group	EEMA O & M Services Group	Magcobar Mine Site	24 Hour
Magcobar Mine Site	P.O. Box 699	Reporting Information	48 Hour
2000 Darby Lane	Malvern, AR 72104	Telephone: 501-467-8355	72 Hour
Malvern, AR 72104		FAX: 501-467-8687	Routine (5 Day)
Attn: Bill Mc Alister		Bill to/P.O. #:	Preservative Code: 1
			Bottle Type: P

Preservation Codes:

1. Cool, 4 Degrees Centigrade
2. Sulfuric Acid (H_2SO_4), pH < 2
3. Nitric Acid (HNO_3), pH < 2
4. Thiosulfate for Dechlorination
5. Hydrochloric Acid(HCl)
6. Sodium Hydroxide (NaOH), pH > 12

Bottle Type Code: G = Glass; P = Plastic; V = Septum; A = Amber

Sampler(s) Signature: Bill McAlister

Sampler(s) Printed: Bill McAlister

Field Number	SAMPLE COLLECTION Date/s	Time/s	Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION	Chronic Biomonitoring	TEST PARAMETERS	Arkansas Analytical Work Order Number:
									K906006 A	~~01~~
FD1Comp	6/4/2009	9:05 AM		X	3	W	Facility Discharge FD-1	X		~~01~~

1. Relinquished by: (Signature)	Date/Time	2. Received by: (Signature)
Bill McAlister	6-4-09	
3. Relinquished by: (Signature)	Date/Time	4. Received by lab: (Signature)
	6-4-09 1344	Sarah E Rouse

SAMPLE CONDITION UPON RECEIPT IN LAB

1. CUSTODY SEALS:	✓ Yes	No
2. CONTAINERS CORRECT:	Yes	No
3. COC/LABELS AGREE:	Yes	No
4. PRESERVATION CONFIRMED:	Yes	No
5. RECEIVED ON ICE:	Yes	No
6. TEMPERATURE ON RECEIPT:	4°C	

FOR COMPLETION BY LAB ONLY

REMARKS / SAMPLE COMMENTS

Arkansas Analytical Inc.

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		Project Description	Turnaround Time
EEMA O & M Services Group	EEMA O & M Services Group	Magcobar Mine Site	24 Hour
Magcobar Mine Site	P.O. Box 699	Reporting Information	48 Hour
2000 Darby Lane	Malvern, AR 72104	Telephone: 501-467-8355	72 Hour
Malvern, AR 72104		FAX: 501-467-8687	Routine (5 Day)
Attn: Bill Mc Alister		Bill to/P.O. #:	Preservative Code
			Bottle Type

Preservation Codes:

1. Cool, 4 Degrees Centigrade
2. Sulfuric Acid (H_2SO_4), pH < 2
3. Nitric Acid (HNO_3), pH < 2
4. Thiosulfate for Dechlorination
5. Hydrochloric Acid(HCl)
6. Sodium Hydroxide (NaOH), pH > 12

Bill McAlister
Sampler(s) Signature

Bill McAlister
Sampler(s) Printed

Field Number	SAMPLE COLLECTION Date/s	Time/s	Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION	TEST PARAMETERS: Chronic Biomonitoring	Bottle Type Code
								1	G = Glass; P = Plastic
								P	V = Septum; A = Amber
FD2Comp	6/5/2009	8:37 AM		X	3	W	Facility Discharge FD-2	X	Arkansas Analytical Work Order Number:

1. Relinquished by: (Signature)	Date/Time	2. Received by: (Signature)
Bill McAlister	6-5-09 1256	
3. Relinquished by: (Signature)	Date/Time	4. Received by lab: (Signature)

SAMPLE CONDITION UPON RECEIPT IN LAB

1. CUSTODY SEALS:	Yes	No
2. CONTAINERS CORRECT:	Yes	No
3. COC/LABELS AGREE:	Yes	No
4. PRESERVATION CONFIRMED:	Yes	No
5. RECEIVED ON ICE:	Yes	No
6. TEMPERATURE ON RECEIPT:	4°C	

FOR COMPLETION BY LAB ONLY

REMARKS / SAMPLE COMMENTS

Arkansas Analytical Inc.

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		Project Description	Turnaround Time
EEMA O & M Services Group	EEMA O & M Services Group	Magcobar Mine Site	24 Hour
Magcobar Mine Site	P.O. Box 699	Reporting Information	48 Hour
2000 Darby Lane	Malvern, AR 72104	Telephone: 501-467-8355	72 Hour
Malvern, AR 72104		FAX: 501-467-8687	Routine (5 Day)
Attn: Bill Mc Alister		Bill to/P.O. #:	Preservative Code
			Bottle Type

Preservation Codes:

1. Cool, 4 Degrees Centigrade
2. Sulfuric Acid (H_2SO_4), pH < 2
3. Nitric Acid (HNO_3), pH < 2
4. Thiosulfate for Dechlorination
5. Hydrochloric Acid(HCl)
6. Sodium Hydroxide (NaOH), pH > 12

Bottle Type Code: G = Glass, P = Plastic, V = Septum, A = Amber

Sampler(s) Signature: Bill McAlister

Sampler(s) Printed: Bill McAlister

Arkansas Analytical Work Order Number: K906004

Field Number	SAMPLE COLLECTION Date/s	Time/s	Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION	TEST PARAMETERS: Chronic Biomonitoring	Preservative Code	Bottle Type	Bottle Type Code
FD1Comp	6/9/2009	7:30 AM		X	3	W	Facility Discharge FD-1	X	1	P	C

1. Relinquished by: (Signature)	Date/Time	2. Received by: (Signature)
Bill McAlister	6-9-09 1329	
3. Relinquished by: (Signature)	Date/Time	4. Received by lab: (Signature)
		Sydney James

SAMPLE CONDITION UPON RECEIPT IN LAB

1. CUSTODY SEALS:	✓ Yes	No
2. CONTAINERS CORRECT:	Yes	No
3. COC/LABELS AGREE:	Yes	No
4. PRESERVATION CONFIRMED:	Yes	No
5. RECEIVED ON ICE:	Yes	No
6. TEMPERATURE ON RECEIPT:	4° C	

FOR COMPLETION BY LAB ONLY

REMARKS / SAMPLE COMMENTS

APPENDIX B

Effluent and Dilution Water Data

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING — Fathead Minnow

Lab # / Sample ID: 1C906066 — Test Start (Date/Time): 6/5/09

Client: Weston — Test End (Date/Time): 6/12/09

		Day of Test 1	2	3	4	5	6	7	notes/remarks
Control	SS 207	6/5	6/6	6/7	6/8	6/9	6/10	6/11	
D.O. (mg/L)	INITIAL	7.1	7.9	7.8	7.6	7.4	7.5	7.8	
	FINAL	8.2	7.8	7.2	7.3	7.3	7.6	7.5	
pH (s.u.)	INITIAL	7.9	7.0	7.5	7.5	7.7	7.7	7.3	
	FINAL	7.2	7.5	7.7	7.0	7.4	7.3	7.3	
temp (C)	INITIAL	22.6	24.0	22.9	22.8	22.2	22.3	21.5	
	FINAL	24.1	25.0	25.0	25.0	25.0	25.0	25.0	
ALKALINITY (mg/L)		36							
HARDNESS (mg/L)		48							
CONDUCTIVITY (umhos/cm)		180							
CHLORINE (mg/L)		<0.05							
CONC:	32								
D.O. (mg/L)	INITIAL	7.2	8.1	7.9	7.7	7.1	6.9	7.8	
	FINAL	8.1	7.5	7.2	7.1	7.2	7.6	7.3	
pH (s.u)	INITIAL	7.9	7.2	7.4	7.1	7.4	7.1	7.0	
	FINAL	7.1	7.1	7.3	6.8	6.9	7.0.6	7.2	
temp (C)	INITIAL	22.8	22.9	23.0	23.5	22.0	22.4	21.5	
	FINAL	23.0	25.0	25.0	25.0	25.0	25.0	25.0	
CONC:	42								
D.O. (mg/L)	INITIAL	7.4	8.3	8.1	7.8	7.5	7.1	7.8	
	FINAL	7.9	7.6	7.2	7.1	7.2	7.3	7.4	
pH (mg/L)	INITIAL	8.7	7.2	7.5	7.1	7.4	7.1	6.9	
	FINAL	7.2	7.1	7.3	6.9	7.0	6.7	7.0	
temp (C)	INITIAL	22.8	22.6	23.2	23.8	22.0	22.5	21.4	
	FINAL	22.4	25.0	25.0	25.0	25.0	25.0	25.0	
CONC:	56								
D.O. (mg/L)	INITIAL	7.6	8.3	8.3	7.9	7.4	7.4	7.8	
	FINAL	8.1	7.6	7.2	7.2	7.2	7.3	7.4	
pH (s.u.)	INITIAL	8.0	7.2	7.4	7.6	7.4	7.1	6.9	
	FINAL	7.3	7.1	7.3	6.8	6.9	6.7	7.0	
temp (C)	INITIAL	22.8	22.3	23.2	24.1	22.1	22.4	21.7	
	FINAL	22.8	25.0	25.0	25.0	25.0	25.0	25.0	
CONC:	75								
D.O. (mg/L)	INITIAL	7.6	8.5	8.3	7.9	7.5	7.5	8.1	
	FINAL	8.0	7.7	7.3	7.3	7.2	7.3	7.4	
pH (s.u.)	INITIAL	7.9	7.2	7.4	7.1	7.4	7.0	6.9	
	FINAL	7.2	7.1	7.2	6.9	6.9	6.7	6.9	
temp (C)	INITIAL	22.8	22.4	23.3	24.4	22.1	22.4	21.3	
	FINAL	22.3	25.0	25.0	25.0	25.0	25.0	25.0	
CONC:	100								
D.O. (mg/L)	INITIAL	7.8	8.5	8.9	8.1	7.5	7.7	8.3	
	FINAL	7.8	7.6	7.3	7.3	7.1	7.1	7.2	
pH (s.u.)	INITIAL	8.0	7.2	7.3	7.1	7.3	7.0	6.8	
	FINAL	7.2	7.0	7.7	6.9	6.9	6.6	6.8	
temp (C)	INITIAL	22.0	22.5	23.4	24.6	22.9	22.2	21.5	
	FINAL	22.6	25.0	25.0	25.0	25.0	25.0	25.0	
CONC:	100%	A	A	A	B	B	C	C	
ALKALINITY (mg/L)		18			22		26		
HARDNESS (mg/L)		1040			>600		>600		
CONDUCTIVITY (umhos/cm)		1925			1946		1946		
CHLORINE (mg/L)		<0.05			<0.05		<0.05		

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING — Cerodaphnia Dubia

Lab # / Sample ID: R906006 — Test Start (Date/Time): 6/5/09

Client: Weston — Test End (Date/Time): 6/12/09

		Day of Test 1	2	3	4	5	6	7	notes/remarks
Control	55207	6/5	6/6	6/7	6/8	6/9	6/10	6/11	
D.O. (mg/L)	INITIAL	71	7.9	7.8	76	74	75	78	
	FINAL	7.2	8.0	76	76	75	74	72	
pH (s.u.)	INITIAL	79	7.0	7.5	71	77	72	73	
	FINAL	7.2	7.4	76	75	74	75	76	
temp (C)	INITIAL	22.6	24.0	22.9	22.8	22.2	22.3	21.5	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0	
ALKALINITY (mg/L)		36							
HARDNESS (mg/L)		48							
CONDUCTIVITY (umhos/cm)		180							
CHLORINE (mg/L)		<0.05							
CONC: 32									
D.O. (mg/L)	INITIAL	72	8.1	7.9	77	71	69	78	
	FINAL	7.3	7.6	77	76	73	72	72	
pH (s.u)	INITIAL	79	7.2	7.4	71	74	71	70	
	FINAL	7.0	7.0	72	73	72	76	74	
temp (C)	INITIAL	22.8	22.9	23.0	23.5	22.0	22.4	21.5	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0	
CONC: 42									
D.O. (mg/L)	INITIAL	74	8.3	8.1	78	75	71	78	
	FINAL	7.2	7.3	77	76	75	73	73	
pH (mg/L)	INITIAL	82	7.2	7.5	71	74	71	69	
	FINAL	7.2	7.1	72	73	72	76	74	
temp (C)	INITIAL	22.8	22.6	23.2	23.8	23.0	22.5	21.4	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0	
CONC: 56									
D.O. (mg/L)	INITIAL	76	8.3	8.3	79	74	74	78	
	FINAL	7.3	7.4	76	75	74	72	74	
pH (s.u.)	INITIAL	80	7.3	7.4	71	74	71	69	
	FINAL	7.1	7.2 or	72	71	71	75	72	
temp (C)	INITIAL	22.8	22.3	23.2	24.1	22.1	22.4	21.2	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0	
CONC: 75									
D.O. (mg/L)	INITIAL	76	8.5	8.3	79	75	75	81	
	FINAL	7.2	7.4	76	76	74	72	73	
pH (s.u.)	INITIAL	79	7.2	7.4	71	74	76	69	
	FINAL	7.1	7.1	72	71	72	74	71	
temp (C)	INITIAL	22.8	22.4	23.3	24.4	22.1	22.4	21.3	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0	
CONC: 100									
D.O. (mg/L)	INITIAL	78	8.5	8.9	81	75	77	83	
	FINAL	7.1	7.3	76	77	74	74	73	
pH (s.u.)	INITIAL	80	7.2	7.3	71	73	76	68	
	FINAL	7.1	7.1	71	74	72	74	72	
temp (C)	INITIAL	23.6	22.5	23.4	24.6	22.9	22.7	21.5	
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0	
CONC: 100%		A	A	A	B	B	C	C	
ALKALINITY (mg/L)		18 40			22		60		
HARDNESS (mg/L)		1040			>600		>600		
CONDUCTIVITY (umhos/cm)		1925			1946		1940		
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 K906006 TEST START DATE 6/5/09 TIME 1245

CLIENT Weston TEST END DATE 6/12/09 TIME 1420

AGE AND SOURCE OF MINNOWS

		D A Y (NUMBER SURVIVING)								SURVIVAL		
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: Cont	A	8	8	8	8	8	8	8	7	87.5	95	7.21
	B			7	7	7	7	7	7	87.5		
	C			8	8	8	8	8	8	100		
	D			8	8	8	8	8	8	100		
	E			8	8	8	8	8	8	100		
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 32	A	8	8	8	8	8	8	8	8	100	97.5	
	B							7	7	87.5		
	C							8	8	100		
	D							8	8	100		
	E							8	8	100		
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 42	A	8	8	8	8	8	8	8	8	100	100	
	B								8	100		
	C								8	100		
	D								8	100		
	E								8	100		
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 56	A	8	8	8	8	8	8	8	8	100	97.5	
	B							8	8	100		
	C							7	7	87.5		
	D							8	8	100		
	E							8	8	100		
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 75	A	8	8	8	8	8	8	8	8	100	97.5	
	B							8	8	100		
	C							7	7	87.5		
	D							8	8	100		
	E							8	8	100		
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 100	A	8	8	8	8	8	8	8	8	100	97.5	5.13
	B								7	87.5		
	C								8	100		
	D								8	100		
	E								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 K9060006 TEST START DATE 6/5/09 TIME 1215

CLIENT Weston A TEST END DATE 6/14/09 TIME 1420

AGE AND SOURCE OF MINNOWS

D A Y (NUMBER SURVIVING) SURVIVAL

	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: Control	A	2	2	2	2	2	2	2	1			
	B								2			
	C								2			
	D								2			
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 32	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 42	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 56	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 75	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 100	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
ANALYST		KP	CT	CT	KP	KP	KP	KP	KP			
DATE:		6/5/09	6/6/09	6/7/09	6/8/09	6/9/09	6/10/09	6/11/09	6/12/09			
TIME:		1215	1420	1400	1130	1310	1205	0720	1420			

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

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID K906006 TEST START DATE 6/5/09 TIME 1215

CLIENT Weston 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: Control	A	2	2	1	2	2	2	2	2			
	B	1	1	2	1	1	1	1	2			
	C	1	1	1	1	1	1	1	1			
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 32	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	2	2			
	C								1			
	D							1	2			
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 42	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	1			
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 56	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	1			
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 75	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	1			
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 100	A	2	2	2	2	2	2	1	1			
	B	1	1	1	1	1	1	2	2			
	C							2	2			
	D							2	2			
	E											
ANALYST		KP	CT	CT								
DATE:		6/5/09	6/6/09	6/7/09								
TIME:		1215	1430	1400								

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

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID K906006 TEST START DATE 6/5/09 TIME 1215

CLIENT Weston C 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: Control	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	1			
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 32	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	1			
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 42	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	1			
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 56	A	2	2	2	2	2	2	2	3			
	B	1	1	1	1	1	1	2	2			
	C							1	1			
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 75	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	2			
	C							2	2			
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 100	A	2	2	2	2	2	2	2	2			
	B	1	1	1	1	1	1	1	1			
	C											
	D											
	E											
ANALYST		KP	CK	ct								
DATE:		6/5/09	6/6/09	6/7/09								
TIME:		1215	1430	1400								

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

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID K906006 TEST START DATE 6/5/09 TIME 1215

CLIENT Weston D 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: Control	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
CONC: 32	A	2	1	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
CONC: 42	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
CONC: 56	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
CONC: 75	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
CONC: 100	A	2	2	2	2	2	2	2	2			
	B											
	C											
	D											
	E											
ANALYST		KP	CK	CK								
DATE:		6/5/09	6/6/09	6/7/09								
TIME:		1215	1430	1400								

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

SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST

LAB # / SAMPLE ID K906006 TEST START DATE 6/5/09 TIME 1215

CLIENT Weston F 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: Control	A	7	7	7	7	7	7	7	7			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 32	A	7	7	7	7	7	7	7	7			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 42	A	7	7	7	7	7	7	7	7			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 56	A	7	7	7	7	7	7	7	7			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 75	A	7	7	7	7	7	7	7	7			
	B											
	C											
	D											
	E											
	REP #	start	1	2	3	4	5	6	7	%	MEAN %	CV
CONC: 100	A	7	7	7	7	7	7	7	7			
	B											
	C											
	D											
	E											
ANALYST		KP	CA	CA								
DATE:		6/5/09	6/6/09	6/7/09								
TIME:		1215	1430	1400								

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:	K906006
CLIENT:	EEMA
ANALYSTS:	KP
SAMPLE ID:	SEE COC

TEST DATES (BEGIN / END):	6/5-12/09
WEIGHING DATE / TIME:	6/16/09, 1400
DRYING TEMP (DEGREES C):	60
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	0.99414	0.98802	0.00612	8	0.765	AVG DRY WEIGHT (mg)
	B	1.01321	1.00962	0.00359	8	0.449	0.555
	C	0.98504	0.98164	0.00340	8	0.425	
	D	1.00733	1.00333	0.00400	8	0.500	CV
	E	0.97285	0.96776	0.00509	8	0.636	25.8
CONC: 32%	A	0.97171	0.96864	0.00307	8	0.384	AVG DRY WEIGHT (mg)
	B	0.99828	0.99484	0.00344	8	0.430	0.490
	C	0.99244	0.98793	0.00451	8	0.564	
	D	0.96343	0.95989	0.00354	8	0.442	CV
	E	0.99960	0.99457	0.00503	8	0.629	
CONC: 42%	A	0.94287	0.93709	0.00578	8	0.723	AVG DRY WEIGHT (mg)
	B	0.94655	0.94289	0.00366	8	0.458	0.566
	C	0.98149	0.97671	0.00478	8	0.598	
	D	0.97074	0.96641	0.00433	8	0.541	CV
	E	0.98856	0.98446	0.00410	8	0.512	
CONC: 56%	A	1.01491	1.01083	0.00408	8	0.510	AVG DRY WEIGHT (mg)
	B	1.03875	1.03395	0.00480	8	0.600	0.579
	C	0.98053	0.97703	0.00350	8	0.438	
	D	1.00305	0.99750	0.00555	8	0.694	CV
	E	1.02593	1.02071	0.00522	8	0.653	
CONC: 75%	A	0.98316	0.97664	0.00652	8	0.815	AVG DRY WEIGHT (mg)
	B	0.99472	0.99007	0.00465	8	0.581	0.628
	C	1.01367	1.00887	0.00480	8	0.600	
	D	0.96975	0.96489	0.00486	8	0.607	CV
	E	0.97658	0.97231	0.00427	8	0.534	
CONC: 100%	A	0.94917	0.94388	0.00529	8	0.661	AVG DRY WEIGHT (mg)
	B	0.97311	0.96863	0.00448	8	0.560	0.670
	C	1.01142	1.00643	0.00499	8	0.624	
	D	0.99889	0.99289	0.00600	8	0.750	CV
	E	0.97135	0.96532	0.00603	8	0.754	12.4

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

Pimephales promelas **FATHEAD MINNOW** TEST 1000.0

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB # / #s:	1906.006	TEST DATES (BEGIN / END):	6/5-12/09
CLIENT:	Weston	WEIGHING DATE / TIME:	6/16/09 1400
ANALYSTS:		DRYING TEMP (DEGREES C):	60
SAMPLE ID:		DRYING TIME (HOURS):	24

	REP#	FINAL DRY WEIGHT TIN+LARVAE (g)	INTIAL WEIGHT TIN (g)	TOTAL DRY WEIGHT OF LARVAE (g)	NUMBER OF LARVAE	DRY WEIGHT OF LARVAE (mg)	
CONTROL	A 1	0.99305	0.98802				AVG DRY WEIGHT (mg)
	B 2	1.01321	1.00962				
	C 3	0.98404	0.98164				
	D 4	1.00612	1.00333				CV
	E 5	0.97105	0.96776				
CONC: 0.97171	A 6	0.98077	0.96864				AVG DRY WEIGHT (mg)
	B 7	0.99828	0.99484				
	C 8	0.99244	0.98943				
	D 9	0.96343	0.95989				CV
	E 10	0.99960	0.99457				
CONC:	A 11	0.94287	0.93709				AVG DRY WEIGHT (mg)
	B 12	0.94655	0.94289				
	C 13	0.98149	0.97671				
	D 14	0.97074	0.96641				CV
	E 15	0.98856	0.98446				
CONC:	A 16	1.01491	1.01083				AVG DRY WEIGHT (mg)
	B 17	1.03875	1.03395				
	C 18	0.98053	0.97763				
	D 19	1.00305	0.99750				CV
	E 20	1.02593	1.02071				
CONC:	A 21	0.98316	0.97664				AVG DRY WEIGHT (mg)
	B 22	0.99472	0.99007				
	C 23	1.01367	1.00887				
	D 24	0.96975	0.96489				CV
	E 25	0.97658	0.97231				
CONC:	A 26	0.94917	0.94388				AVG DRY WEIGHT (mg)
	B 27	0.97311	0.96863				
	C 28	1.01142	1.00643				
	D 29	0.99829	0.99289				CV
	E 30	0.97155	0.96532				

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

KP3

AA# K906006, FATHEAD MINNOW SURVIVAL, CHRONIC 6-5-09
File: J:\TOXSTAT\MONTE\FHSURV~1. Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.181

W = 0.705

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# K906006, FATHEAD MINNOW SURVIVAL, CHRONIC 6-5-09
File: J:\TOXSTAT\MONTE\FHSURV~1. 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# K906006, FATHEAD MINNOW SURVIVAL, CHRONIC 6-5-09
FILE: J:\TOXSTAT\MONTE\FHSURV~1.
TRANSFORM: ARC SINE(SQUARE ROOT(Y)) NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.8750	1.2094
1	CONTROL	2	0.8750	1.2094
1	CONTROL	3	1.0000	1.4120
1	CONTROL	4	1.0000	1.4120
1	CONTROL	5	1.0000	1.4120
2	32 % EFFLUENT	1	1.0000	1.4120
2	32 % EFFLUENT	2	0.8750	1.2094
2	32 % EFFLUENT	3	1.0000	1.4120
2	32 % EFFLUENT	4	1.0000	1.4120
2	32 % EFFLUENT	5	1.0000	1.4120
3	42 % EFFLUENT	1	1.0000	1.4120
3	42 % EFFLUENT	2	1.0000	1.4120
3	42 % EFFLUENT	3	1.0000	1.4120
3	42 % EFFLUENT	4	1.0000	1.4120
3	42 % EFFLUENT	5	1.0000	1.4120
4	56 % EFFLUENT	1	1.0000	1.4120
4	56 % EFFLUENT	2	1.0000	1.4120
4	56 % EFFLUENT	3	0.8750	1.2094
4	56 % EFFLUENT	4	1.0000	1.4120
4	56 % EFFLUENT	5	1.0000	1.4120
5	75 % EFFLUENT	1	1.0000	1.4120
5	75 % EFFLUENT	2	1.0000	1.4120
5	75 % EFFLUENT	3	0.8750	1.2094
5	75 % EFFLUENT	4	1.0000	1.4120
5	75 % EFFLUENT	5	1.0000	1.4120
6	100 % EFFLUENT	1	1.0000	1.4120
6	100 % EFFLUENT	2	0.8750	1.2094
6	100 % EFFLUENT	3	1.0000	1.4120
6	100 % EFFLUENT	4	1.0000	1.4120
6	100 % EFFLUENT	5	1.0000	1.4120

AA# K906006, FATHEAD MINNOW SURVIVAL, CHRONIC 6-5-09
File: J:\TOXSTAT\MONTE\FHSURV~1. 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	1.331				
2	32 % EFFLUENT	1.372	30.00	16.00	5.00	
3	42 % EFFLUENT	1.412	32.50	16.00	5.00	
4	56 % EFFLUENT	1.372	30.00	16.00	5.00	
5	75 % EFFLUENT	1.372	30.00	16.00	5.00	
6	100 % EFFLUENT	1.372	30.00	16.00	5.00	

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

KP1

AA# K906013, FATHEAD MINNOW GROWTH CHRONIC, 6-18-09
File: J:/toxstat/monte\FHGROWTH. Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.315

W = 0.929

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# K906013, FATHEAD MINNOW GROWTH CHRONIC, 6-18-09
File: J:/toxstat/monte\FHGROWTH. Transform: ARC SINE(SQUARE ROOT(Y))

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 1.11

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# K906013, FATHEAD MINNOW GROWTH CHRONIC, 6-18-09
FILE: J:/toxstat/monte\FHGROWTH.
TRANSFORM: ARC SINE(SQUARE ROOT(Y)) NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.7650	1.0647
1	CONTROL	2	0.4490	0.7343
1	CONTROL	3	0.4250	0.7101
1	CONTROL	4	0.5000	0.7854
1	CONTROL	5	0.6360	0.9231
2	32 % EFFLUENT	1	0.3590	0.6425
2	32 % EFFLUENT	2	0.4300	0.7152
2	32 % EFFLUENT	3	0.5640	0.8496
2	32 % EFFLUENT	4	0.4420	0.7273
2	32 % EFFLUENT	5	0.6290	0.9159
3	42 % EFFLUENT	1	0.7230	1.0165
3	42 % EFFLUENT	2	0.4580	0.7433
3	42 % EFFLUENT	3	0.5980	0.8840
3	42 % EFFLUENT	4	0.5410	0.8264
3	42 % EFFLUENT	5	0.5120	0.7974
4	56 % EFFLUENT	1	0.5100	0.7954
4	56 % EFFLUENT	2	0.6000	0.8861
4	56 % EFFLUENT	3	0.4380	0.7232
4	56 % EFFLUENT	4	0.6940	0.9846
4	56 % EFFLUENT	5	0.6530	0.9409
5	75 % EFFLUENT	1	0.8150	1.1262
5	75 % EFFLUENT	2	0.5810	0.8668
5	75 % EFFLUENT	3	0.6000	0.8861
5	75 % EFFLUENT	4	0.6070	0.8932
5	75 % EFFLUENT	5	0.5340	0.8194
6	100 % EFFLUENT	1	0.6610	0.9493
6	100 % EFFLUENT	2	0.5600	0.8455
6	100 % EFFLUENT	3	0.6240	0.9107
6	100 % EFFLUENT	4	0.7500	1.0472
6	100 % EFFLUENT	5	0.7540	1.0518

AA# K906013, FATHEAD MINNOW GROWTH CHRONIC, 6-18-09
File: J:/toxstat/monte\FHGROWTH. Transform: ARC SINE(SQUARE ROOT(Y))

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.108	0.022	1.643
Within (Error)	24	0.315	0.013	
Total	29	0.423		

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

KP2

File: J:/toxstat/monte\FHGROWTH. 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	0.844	0.555		
2	32 % EFFLUENT	0.770	0.485	1.014	
3	42 % EFFLUENT	0.854	0.566	-0.138	
4	56 % EFFLUENT	0.866	0.579	-0.311	
5	75 % EFFLUENT	0.918	0.627	-1.032	
6	100 % EFFLUENT	0.961	0.670	-1.620	

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

AA# K906013, FATHEAD MINNOW GROWTH CHRONIC, 6-18-09

File: J:/toxstat/monte\FHGROWTH. 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	0.170	30.6	0.070
3	42 % EFFLUENT	5	0.170	30.6	-0.011
4	56 % EFFLUENT	5	0.170	30.6	-0.024
5	75 % EFFLUENT	5	0.170	30.6	-0.072
6	100 % EFFLUENT	5	0.170	30.6	-0.115

APPENDIX D

Ceriodaphnia dubia Raw Data and Statistics

Cerodaphnia dubia

SURVIVAL AND REPRODUCTION TEST

Analyst: KP

Discharger: Weston

Lab Number/s: K906006

Location:

Test Start - Date/ Time: 6.5.09 1205

Date Sample Collected:

Test Stop - Date/Time: 6.10.09 1345

Conc 1 (Control)

%	Day	A	B	C	D	E	F	G	H	I	J	No. of Young	No. of Adult	Young/ Adult	Analyst
	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	0	0	0	0	0	0	0	0	5	10	0.5	KP
	4	3	2	0	2	4	2	3	4	1	3	22	10	2.2	KP
	5	0	2	5	3	4	3	4	6	2	5	34	9	3.8	KP
	6	5	3	6	6	8	9	8	3	7	7	63	8	7.9	KP
	7	5	3	6	4	3	3	3	—	—	2	29	8	3.6	KP
	8														
	Total	14	15	17	15	19	17	19	X10	X0	17	163		X= 16.6	

CV= 11.1

Conc 2 (32)

%	Day	A	B	C	D	E	F	G	H	I	J	No. of Young	No. of Adult	Young/ Adult	Analyst
	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	0	X	X	0	0	X0	X	0	16	9	1.1	
	4	2	1	0	2	6	3	3	—	4	6	37	9	3.0	
	5	5	5	3	2	4	2	3	—	5	3	32	8	4.0	
	6	3	2	6	9	6	—	9	—	3	1	50	8	6.3	
	7	1	3	2	5	0	—	4	—	3	1	25	8	3.1	
	8														
	Total	17	16	11	20	21	X6	19	X0	18	17	144			

Conc 3 (42)

%	Day	A	B	C	D	E	F	G	H	I	J	No. of Young	No. of Adult	Young/ Adult	Analyst
	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	Y	0	Y	X	0	Y	X	Y	Y	14	9	1.6	
	4	6	0	3	5	4	6	1	—	2	4	23	9	2.8	
	5	3	2	3	4	5	0	5	—	1	7	35	9	3.9	
	6	8	5	X	7	6	9	2	—	8	6	58	8	7.3	
	7	2	5	—	1	3	2	4	—	5	6	33	8	4.1	
	8														
	Total	19	20	X3	17	23	9	18	X3	17	26	165			

Conc 4 (56)

%	Day	A	B	C	D	E	F	G	H	I	J	No. of Young	No. of Adult	Young/ Adult	Analyst
	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	Y	Y	0	0	0	0	Y	Y	11	8	1.4	
	4	X	X	8	8	6	6	7	0	3	—	12	8	1.5	
	5	—	0	1	3	6	7	4	6	4	<	31	8	3.9	
	6	—	6	7	8	5	9	7	9	6	—	52	8	6.5	
	7	—	4	3	5	5	4	3	4	3	—	31	8	3.9	
	8														
	Total	X0	11	12	20	16	26	16	19	21	X1	142			

7 → 137

Conc 5 (18)

%	Day	A	B	C	D	E	F	G	H	I	J	No. of Young	No. of Adult	Young/ Adult	Analyst
	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	0	3	0	0	0	X	0	7	8	0.9	
	4	4	0	5	5	6	—	5	3	—	2	30	8	3.8	
	5	3	5	4	7	5	—	4	3	—	3	34	8	4.3	
	6	9	6	8	8	7	—	4	7	—	7	56	8	7.0	
	7	2	2	3	4	5	—	6	0	—	4	26	8	3.3	
	8														
	Total	18	13	21	27	23	X0	19	13	X0	16	153			

(K)

Conc 6 (100)

%	Day	A	B	C	D	E	F	G	H	I	J	No. of Young	No. of Adult	Young/ Adult	Analyst
	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	Y	Y	0	0	0	0	0	0	0	0	6	10	0.6	
	4	0	5	6	3	4	3	3	4	1	4	33	10	3.3	
	5	4	5	X	4	2	3	5	4	6	3	36	9	4.0	
	6	6	5	—	8	6	4	7	4.5	—	7	57	9	6.3	
	7	1	3	—	6	4	2	3	0	3	5	27	9	3.0	
	8														
	Total	15	19	X7	21	16	12	18	17	15	19	159			

X= DEAD; Y= MALE

$\bar{X} = 16.9$

$CV = 16.1$

KP1

AA # K906006, C. DUBIA CHRONIC, REPRODUCION, 6-5-09
File: J:/toxstat/monte\C.DUB 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 # K906006, C. DUBIA CHRONIC, REPRODUCION, 6-5-09
File: J:/toxstat/monte\C.DUB Transform: NO TRANSFORMATION

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 12.98

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.

KP2

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF ALIVE	DEAD	TOTAL ANIMALS
CONTROL	8	2	10
32%	8	2	10
TOTAL	16	4	20

CRITICAL FISHER'S VALUE (10,10,8) (p=0.05) IS 3. b VALUE IS 8.
Since b is greater than 3 there is no significant difference between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF ALIVE	DEAD	TOTAL ANIMALS
CONTROL	8	2	10
42%	8	2	10
TOTAL	16	4	20

CRITICAL FISHER'S VALUE (10,10,8) (p=0.05) IS 3. b VALUE IS 8.
Since b is greater than 3 there is no significant difference between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF ALIVE	DEAD	TOTAL ANIMALS
CONTROL	8	2	10
56%	8	2	10
TOTAL	16	4	20

KP2

CRITICAL FISHER'S VALUE (10,10,8) (p=0.05) IS 3. b VALUE IS 8.
Since b is greater than 3 there is no significant difference
between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF ALIVE	NUMBER OF DEAD	TOTAL ANIMALS
CONTROL	8	2	10
75%	8	2	10
TOTAL	16	4	20

CRITICAL FISHER'S VALUE (10,10,8) (p=0.05) IS 3. b VALUE IS 8.
Since b is greater than 3 there is no significant difference
between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF DEAD	NUMBER OF ALIVE	TOTAL ANIMALS
CONTROL	2	8	10
100%	1	9	10
TOTAL	3	17	20

CRITICAL FISHER'S VALUE (10,10,2) (p=0.05) IS LESS THAN 0. b VALUE IS 1.
NO SIGNIFICANT DIFFERENCE

SUMMARY OF FISHER'S EXACT TESTS

GROUP	IDENTIFICATION	NUMBER EXPOSED	NUMBER DEAD	SIG (P=.05)
	CONTROL	10	2	
1	32%	10	2	
2	42%	10	2	
3	56%	10	2	
4	75%	10	2	
5	100%	10	1	

TITLE: AA # K906006, C. DUBIA CHRONIC, REPRODUCION, 6-5-09
FILE: J:/toxstat/monte\C.DUB
TRANSFORM: NO TRANSFORMATION NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	14.0000	14.0000
1	CONTROL	2	15.0000	15.0000
1	CONTROL	3	17.0000	17.0000
1	CONTROL	4	15.0000	15.0000
1	CONTROL	5	19.0000	19.0000
1	CONTROL	6	17.0000	17.0000
1	CONTROL	7	19.0000	19.0000
1	CONTROL	8	10.0000	10.0000
1	CONTROL	9	10.0000	10.0000
1	CONTROL	10	17.0000	17.0000
2	32 % EFFLUENT	1	17.0000	17.0000
2	32 % EFFLUENT	2	16.0000	16.0000
2	32 % EFFLUENT	3	11.0000	11.0000
2	32 % EFFLUENT	4	20.0000	20.0000
2	32 % EFFLUENT	5	21.0000	21.0000
2	32 % EFFLUENT	6	5.0000	5.0000
2	32 % EFFLUENT	7	19.0000	19.0000
2	32 % EFFLUENT	8	0.0000	0.0000
2	32 % EFFLUENT	9	18.0000	18.0000
2	32 % EFFLUENT	10	17.0000	17.0000
3	42 % EFFLUENT	1	19.0000	19.0000
3	42 % EFFLUENT	2	20.0000	20.0000
3	42 % EFFLUENT	3	13.0000	13.0000
3	42 % EFFLUENT	4	17.0000	17.0000
3	42 % EFFLUENT	5	23.0000	23.0000
3	42 % EFFLUENT	6	9.0000	9.0000
3	42 % EFFLUENT	7	18.0000	18.0000
3	42 % EFFLUENT	8	3.0000	3.0000
3	42 % EFFLUENT	9	17.0000	17.0000
3	42 % EFFLUENT	10	26.0000	26.0000
4	56 % EFFLUENT	1	0.0000	0.0000
4	56 % EFFLUENT	2	11.0000	11.0000
4	56 % EFFLUENT	3	7.0000	7.0000
4	56 % EFFLUENT	4	20.0000	20.0000
4	56 % EFFLUENT	5	16.0000	16.0000
4	56 % EFFLUENT	6	26.0000	26.0000
4	56 % EFFLUENT	7	16.0000	16.0000
4	56 % EFFLUENT	8	19.0000	19.0000
4	56 % EFFLUENT	9	21.0000	21.0000
4	56 % EFFLUENT	10	1.0000	1.0000
5	75 % EFFLUENT	1	18.0000	18.0000
5	75 % EFFLUENT	2	13.0000	13.0000
5	75 % EFFLUENT	3	21.0000	21.0000
5	75 % EFFLUENT	4	27.0000	27.0000
5	75 % EFFLUENT	5	26.0000	26.0000
5	75 % EFFLUENT	6	0.0000	0.0000
5	75 % EFFLUENT	7	19.0000	19.0000
5	75 % EFFLUENT	8	13.0000	13.0000
5	75 % EFFLUENT	9	0.0000	0.0000
5	75 % EFFLUENT	10	16.0000	16.0000

			KP2	
6	100 % EFFLUENT	1	15.0000	15.0000
6	100 % EFFLUENT	2	19.0000	19.0000
6	100 % EFFLUENT	3	7.0000	7.0000
6	100 % EFFLUENT	4	21.0000	21.0000
6	100 % EFFLUENT	5	16.0000	16.0000
6	100 % EFFLUENT	6	12.0000	12.0000
6	100 % EFFLUENT	7	18.0000	18.0000
6	100 % EFFLUENT	8	17.0000	17.0000
6	100 % EFFLUENT	9	15.0000	15.0000
6	100 % EFFLUENT	10	19.0000	19.0000

AA # K906006, C. DUBIA CHRONIC, REPRODUCION, 6-5-09
File: J:/toxstat/monte\C.DUB Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	50.883	10.177	0.216
Within (Error)	54	2546.100	47.150	
Total	59	2596.983		

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

AA # K906006, C. DUBIA CHRONIC, REPRODUCION, 6-5-09
File: J:/toxstat/monte\C.DUB 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	15.300	15.300		
2	32 % EFFLUENT	14.400	14.400	0.293	
3	42 % EFFLUENT	16.500	16.500	-0.391	
4	56 % EFFLUENT	13.700	13.700	0.521	
5	75 % EFFLUENT	15.300	15.300	0.000	
6	100 % EFFLUENT	15.900	15.900	-0.195	

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

AA # K906006, C. DUBIA CHRONIC, REPRODUCION, 6-5-09
File: J:/toxstat/monte\C.DUB 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	7.094	46.4	0.900
3	42 % EFFLUENT	10	7.094	46.4	-1.200

			KP2		
4	56 % EFFLUENT	10	7.094	46.4	1.600
5	75 % EFFLUENT	10	7.094	46.4	0.000
6	100 % EFFLUENT	10	7.094	46.4	-0.600

AA # K906006, C. DUBIA CHRONIC, REPRODUCION, 6-5-09
File: J:/toxstat/monte\C.DUB Transform: NO TRANSFORMATION

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

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	15.300				
2	32 % EFFLUENT	14.400	112.00	75.00	10.00	
3	42 % EFFLUENT	16.500	117.00	75.00	10.00	
4	56 % EFFLUENT	13.700	106.00	75.00	10.00	
5	75 % EFFLUENT	15.300	111.00	75.00	10.00	
6	100 % EFFLUENT	15.900	112.50	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 6-4-09 Arkansas Analytical

SPECIES Pimephales promelas

QUANTITY SHIPPED 240+

AGE/LIFE STAGE 24hrs 6/4/09 post

BROODSTOCK SOURCE Anderson Farms, AR

CULTURE WATER groundwater

ALKALINITY (Mg/l as $CaCO_3$) =180

HARDNESS (Mg/l as $CaCO_3$)/Salinity (ppt) =160

FEEDING Artemia

COMMENTS

PACKAGED BY

BILL HALL PRINTERS 3171

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: 7/13/06

SPECIES: *Ceriodaphnia dubia*

AGE: Variable

LIFE STAGE: Adult

HATCH DATE: Variable

BEGAN FEEDING: Immediately

FOOD: YTC, *Selenastrum* sp.

Water Chemistry Record:	Current	Range
TEMPERATURE:	24°C	22-25°C
SALINITY/CONDUCTIVITY:	--	--
TOTAL HARDNESS (as $CaCO_3$):	126 mg/l	60-138 mg/l
TOTAL ALKALINITY (as $CaCO_3$):	60 mg/l	50-110 mg/l
pH:	8.00	6.98-8.32

Comments:

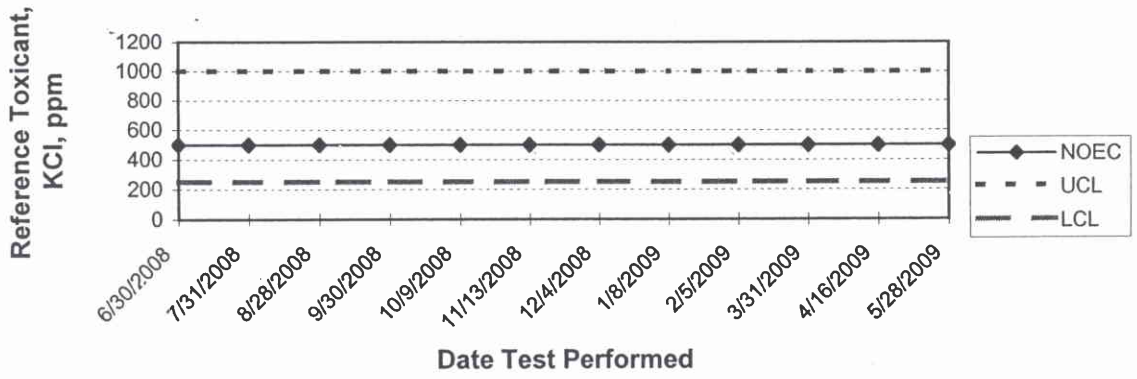
Facility Supervisor

APPENDIX F

Quality Assurance Charts

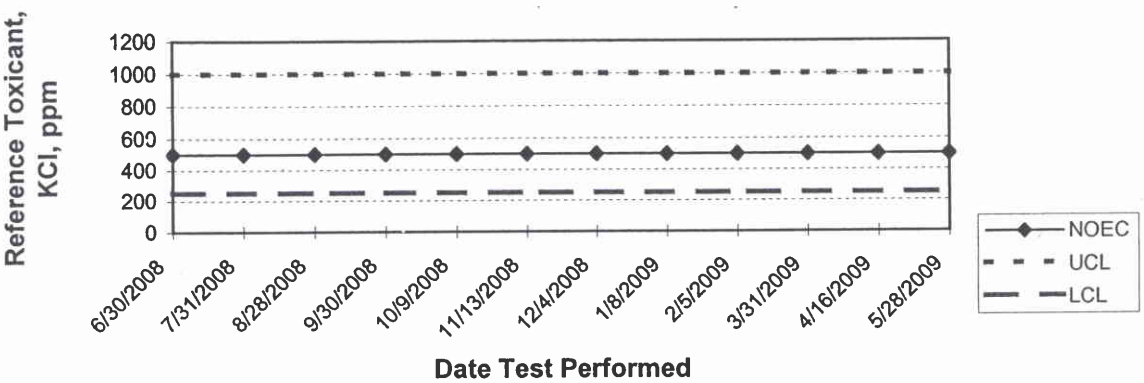
ARKANSAS ANALYTICAL, INC.

FATHEAD MINNOW SURVIVAL
QUALITY ASSURANCE



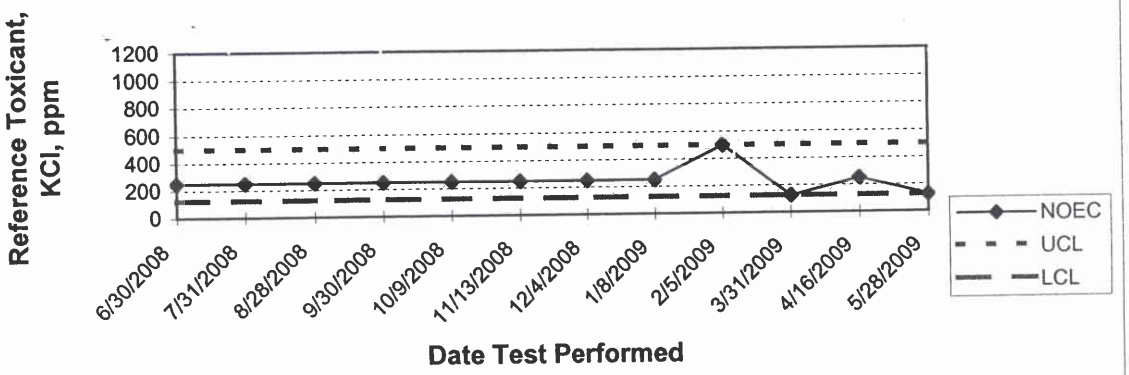
ARKANSAS ANALYTICAL, INC.

FATHEAD MINNOW GROWTH
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.

CERIODAPHNIA DUBIA SURVIVAL
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.

CERIODAPHNIA DUBIA REPRODUCTION
QUALITY ASSURANCE