

Arkansas Analytical, Inc.

Toxicity Test Results

MAGCOBAR MINE SITE
NPDES PERMIT NUMBER: AR0049794
March 2009
AFIN# 00-00348
Retest due to PMSD Failure

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 K903009

Tuesday, April 07, 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 March 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:	3-23-09, 0845	3-24-09, 0845
Sample #2:	3-24-09, 0720	3-25-09, 0720
Sample #3:	3-26-09, 0920	3-27-09, 0920

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:	3-24-09, 1502	3
Sample #2:	3-25-09, 1501	4
Sample #3:	3-27-09, 1320	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. 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	90%	X	
Average of 15 or more young per surviving female	17.0	X	
At least 60% of surviving females should have produced 3 broods	79%	X	
The percent coefficient of variation between replicates must be 40% or less for the young of surviving females	29.6%	X	

TEST ACCEPTANCE CRITERIA for *Pimephales promelas*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	97.5%	X	
The percent coefficient of variation between replicates must be 40% or less for survival	5.73%	X	
Minimum of 0.25 mg average dry weight of surviving controls	0.329	X	
The percent coefficient of variation between replicates must be 40% or less for growth	9.78%	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> 2/5-12/09		<i>Pimephales promelas</i> 2/5-12/09	
NOEC Survival:	500 ppm KCl	NOEC Survival:	500 ppm KCl
LOEC Survival:	1000 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 Magcobar Mine Site

<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)	13.2	%CV survival (critical dilution)	5.73%
%CV Reproduction (critical dilution)	28.5%	Mean dry weight (critical dilution) in milligrams	0.655
		%CV growth (critical dilution)	13.3%
PMSD Reproduction	31.7	PMSD Growth	30.7

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


Chris Turney

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:	3-23-09, 0845	3-24-09, 0845
Sample #2:	3-24-09, 0720	3-25-09, 0720
Sample #3:	3-26-09, 0920	3-27-09, 0920

Test initiated (date, time): 3-25-09, 1650 Test terminated (date, time): 4-1-09, 1030

Dilution water used: Soft Synthetic

DATA TABLE FOR FATHEAD MINNOW SURVIVAL

Effluent Conc %	Percent Survival in Replicate Chambers						Mean Percent Survival			
	A	B	C	D	E		24 hours	48 hours	7 days	CV %
0%	87.5	100	100	100	100		100	100	97.5	5.73
32%	87.5	100	87.5	100	100		100	100	95	
42%	100	100	100	100	100		100	100	100	
56%	100	87.5	100	100	100		100	100	97.5	
75%	75	100	87.5	100	100		100	100	92.5	
100%	100	100	100	87.5	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%	0.284	0.326	0.348	0.369	0.317		0.329	9.78
32%	0.331	0.484	0.395	0.363	0.323		0.379	
42%	0.466	0.467	0.499	0.516	0.397		0.469	
56%	0.496	0.423	0.482	0.561	0.448		0.482	
75%	0.491	0.595	0.518	0.784	0.604		0.598	
100%	0.789	0.618	0.668	0.654	0.550		0.656	13.3

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)= 13.3 %

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:	3-23-09, 0845	3-24-09, 0845
Sample #2:	3-24-09, 0720	3-25-09, 0720
Sample #3:	3-26-09, 0920	3-27-09, 0920

Test initiated (date, time): 3-25-09, 0920 Test terminated (date, time): 4-1-09, 1150

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	22	17	14	23	18	9
B	16	15	10	16	12	11
C	10	16	15	23	10	16
D	26	19	19	17	14	19
E	13	10	14	7	17	8
F	15	12	13	17	13	13
G	16	10	14	17	15	15
H	14	20	19	12	11	11
I	21	10	14	x0	x2	x0
J	x14	17	24	19	18	17
Mean	16.7	14.6	15.6	15.1	13.0	11.9
Mean/surviving female	17.0	14.6	15.6	16.8	14.2	13.2
CV%*	29.6					28.5

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	90	100	100	90	90	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)= 29.6 %

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		Project Description		Turnaround Time	Preservation Codes:									
EEMA O & M Services Group		EEMA O & M Services Group		Magcobar Mine Site		24 Hour	1. Cool, 4 Degrees Centigrade			4. Thiosulfate for Dechlorination				
Magcobar Mine Site		P.O. Box 699		Reporting Information		48 Hour	2. Sulfuric Acid (H ₂ SO ₄), pH < 2			5. Hydrochloric Acid(HCl)				
2000 Darby Lane		Malvern, AR 72104		Telephone: 501-467-8355		72 Hour	3. Nitric Acid (HNO ₃), pH < 2			6. Sodium Hydroxide (NaOH), pH > 12				
Malvern, AR 72104		FAX: 501-467-8687		Bill to/P.O. #:		Routine (5 Day)	TEST PARAMETERS					Bottle Type Code		
Attn: Bill Mc Alister		Preservative Code		Bottle Type		1							G = Glass; P = Plastic V = Septum; A = Amber	
		P												

Bill Mc Alister

Bill Mc Alister




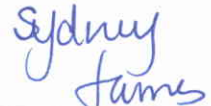

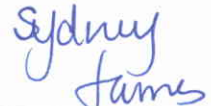
Sampler(s) Signature		Sampler(s) Printed						Chronic Biomonitoring	Arkansas Analytical Work Order Number:
Field Number	SAMPLE COLLECTION Date/s Time/s	Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION			
FD1Comp	3/24/2009 8:45 AM		X	5	W	Facility Discharge FD-1	X	K903009 A	

1. Relinquished by: (Signature) <i>Bill Mc Alister</i>	Date/Time 3-24-09 1502	2. Received by: (Signature) <i>Sydney James</i>	SAMPLE CONDITION UPON RECEIPT IN LAB		REMARKS / SAMPLE COMMENTS
	3. Relinquished by: (Signature)		Date/Time	4. Received by lab: (Signature)	
			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: 3°C		
FOR COMPLETION BY LAB ONLY					



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

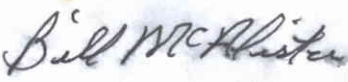


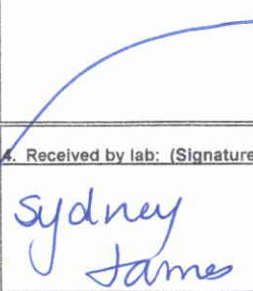

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION				Project Description			Turnaround Time	Preservation Codes:															
EEMA O & M Services Group		EEMA O & M Services Group		Magcobar Mine Site			24 Hour	1. Cool, 4 Degrees Centigrade					4. Thiosulfate for Dechlorination										
Magcobar Mine Site		P.O. Box 699		Reporting Information			48 Hour	2. Sulfuric Acid (H ₂ SO ₄), pH < 2					5. Hydrochloric Acid(HCl)										
2000 Darby Lane		Malvern, AR 72104		Telephone: 501-467-8355			72 Hour	3. Nitric Acid (HNO ₃), pH < 2					6. Sodium Hydroxide (NaOH), pH > 12										
Malvern, AR 72104				FAX: 501-467-8687			Routine (5 Day)	TEST PARAMETERS										Bottle Type Code					
Attn: Bill Mc Alister				Bill to/P.O. #:			Preservative Code:	1															
							Bottle Type:	P															
 Sampler(s) Signature				 Sampler(s) Printed																			
Field Number	SAMPLE COLLECTION		Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION															Chronic Biomonitoring	Arkansas Analytical Work Order Number:
FD-1 Comp	3/25/2009	7:20 AM		X	3	W	Facility Discharge FD-1															X	K903009 B
1. Relinquished by: (Signature)				Date/Time		2. Received by: (Signature)				SAMPLE CONDITION UPON RECEIPT IN LAB										REMARKS / SAMPLE COMMENTS			
				3-25-09 1501						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: 4°C													
3. Relinquished by: (Signature)				Date/Time		4. Received by lab: (Signature)				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			Project Description				Turnaround Time		Preservation Codes:											
EEMA O & M Services Group	EEMA O & M Services Group		Magcobar Mine Site				24 Hour		1. Cool, 4 Degrees Centigrade				4. Thiosulfate for Dechlorination							
Magcobar Mine Site	P.O. Box 699		Reporting Information				48 Hour		2. Sulfuric Acid (H ₂ SO ₄), pH < 2				5. Hydrochloric Acid(HCl)							
2000 Darby Lane	Malvern, AR 72104		Telephone: 501-467-8355				72 Hour		3. Nitric Acid (HNO ₃), pH < 2				6. Sodium Hydroxide (NaOH), pH > 12							
Malvern, AR 72104			FAX: 501-467-8687				Routine (5 Day)		TEST PARAMETERS								Bottle Type Code			
Attn: Bill Mc Alister			Bill to/P.O. #:				Preservative Code:		1	2	3	4	5	6	7	8	9	10	G = Glass: P = Plastic	
							Bottle Type:		P										V = Septum: A = Amber	
									Chronic Biomonitoring											Arkansas Analytical Work Order Number:
Sampler(s) Signature			Sampler(s) Printed																	K903009
Field Number	SAMPLE COLLECTION		Grab	Comp	Number of Bottles	Sample Matrix	SAMPLE IDENTIFICATION/ DESCRIPTION													
FD-2 Comp	3/27/2009	9:20 AM		X	3	W	Facility Discharge FD-2			X										C
1. Relinquished by: (Signature)		Date/Time	2. Received by: (Signature)		SAMPLE CONDITION UPON RECEIPT IN LAB					REMARKS / SAMPLE COMMENTS										
		3-27-09 1320			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 checked="" type="checkbox"/> Yes <input type="checkbox"/> No 6. TEMPERATURE ON RECEIPT: 3°C															
3. Relinquished by: (Signature)		Date/Time	4. Received by lab: (Signature)		FOR COMPLETION BY LAB ONLY															
																				

APPENDIX B

Effluent and Dilution Water Data

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING

Fathead Minnow

Lab # / Sample ID K903004

Test Start (Date/Time) 3/25/09

Client Weston

Test End (Date/Time) 4/1/09

		Day of Test							notes/remarks
		1	2	3	4	5	6	7	
Control		3/25	3/26	3/27	3/28	3/29	3/30	3/31	
D.O. (mg/L)	INITIAL	73	74	81	84	8.3	79	77	
	FINAL	74	73	8.0	8.4	80	76	26	
pH (s.u.)	INITIAL	77	78	79	80	7.4	78	77	
	FINAL	75	78	7.5	7.7	78	78	78	
temp (C)	INITIAL	220	221	220	214	214	212	222	
	FINAL	250	250	25	25	250	250	250	
ALKALINITY (mg/L)		40							
HARDNESS (mg/L)		48							
CONDUCTIVITY (umhos/cm)		165							
CHLORINE (mg/L)		0.05							
CONC:		<u>432</u>							
D.O. (mg/L)	INITIAL	73	74	82	85	8.4	79	82	
	FINAL	72	73	7.8	8.5	80	75	74	
pH (s.u.)	INITIAL	75	75	8.6	70	7.4	77	76	
	FINAL	72	74	7.5	7.6	76	75	74	
temp (C)	INITIAL	218	215	222	214	214	210	226	
	FINAL	250	250	25	25	250	250	250	
CONC:		<u>47</u>							
D.O. (mg/L)	INITIAL	73	74	83	85	8.6	79	83	
	FINAL	72	72	7.6	8.2	78	74	75	
pH (mg/L)	INITIAL	75	75	8.76	73	7.4	77	75	
	FINAL	72	74	7.6	7.6	76	74	74	
temp (C)	INITIAL	218	215	223	214	21.4	211	226	
	FINAL	250	250	25	25	250	250	250	
CONC:		<u>56</u>							
D.O. (mg/L)	INITIAL	74	74	82	85	8.9	80	85	
	FINAL	69	50	7.4	8.1	79	79	78	
pH (s.u.)	INITIAL	75	75	8.76	73	7.2	76	75	
	FINAL	72	74	7.6	7.5	76	74	74	
temp (C)	INITIAL	219	215	223	214	21.4	211	227	
	FINAL	250	250	25	25	250	250	250	
CONC:		<u>75</u>							
D.O. (mg/L)	INITIAL	75	74	82	85	9.2	80	85	
	FINAL	68	70	7.6	8.0	79	74	78	
pH (s.u.)	INITIAL	75	75	8.75	73	7.2	75	74	
	FINAL	72	73	7.6	7.3	75	73	73	
temp (C)	INITIAL	219	215	223	214	21.4	211	229	
	FINAL	250	250	25	25	250	250	250	
CONC:		<u>100</u>							
D.O. (mg/L)	INITIAL	77	76	82	85	9.5	80	86	
	FINAL	70	69	7.7	7.9	80	73	80	
pH (s.u.)	INITIAL	73	7.3	8.73	72	7.1	74	73	
	FINAL	71	71	7.5	7.2	73	72	72	
temp (C)	INITIAL	220	215	223	214	21.4	212	228	
	FINAL	250	250	25	25	250	250	250	
CONC:		<u>100%</u>							
ALKALINITY (mg/L)		16			8		16	4	
HARDNESS (mg/L)		2600			2600		2600		
CONDUCTIVITY (umhos/cm)		22000			22000		22000		
CHLORINE (mg/L)		0.05			0.05		0.05		

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING

Cerodaphnia Dubia

Lab # / Sample ID 1903009

Test Start (Date/Time) 3/25/09

Client Westar

Test End (Date/Time) 4/1/09

		Day of Test							notes/remarks
		1	2	3	4	5	6	7	
Control		3/25	3/26	3/27	3/28	3/29	3/30	3/31	
D.O. (mg/L)	INITIAL	73	74	81	84	83	79	77	
	FINAL	70	75	75	74	79	77		
pH (s.u.)	INITIAL	77	78	79	80	79	78	77	
	FINAL	80	77	75	75	75	80		
temp (C)	INITIAL	220	221	220	218	214	212	222	
	FINAL	250	250	25	250	250	250		
ALKALINITY (mg/L)		46							
HARDNESS (mg/L)		48							
CONDUCTIVITY (umhos/cm)		165							
CHLORINE (mg/L)		0.05							
CONC:		30							
D.O. (mg/L)	INITIAL	73	74	82	8.5	84	79	82	
	FINAL	70	75	76	74	74	77		
pH (s.u.)	INITIAL	75	75	76	70	74	77	76	
	FINAL	76	74	78	76	75	77		
temp (C)	INITIAL	218	215	222	214	214	210	226	
	FINAL	250	250	25	250	250	250		
CONC:		42							
D.O. (mg/L)	INITIAL	73	74	83	8.5	86	79	83	
	FINAL	71	75	76	74	73	74		
pH (mg/L)	INITIAL	75	75	76	7.3	74	77	75	
	FINAL	76	74	78	75	75	76		
temp (C)	INITIAL	218	215	223	214	214	211	226	
	FINAL	250	250	25	250	250	250		
CONC:		50							
D.O. (mg/L)	INITIAL	74	74	82	8.5	89	80	85	
	FINAL	71	75	76	74	73	74		
pH (s.u.)	INITIAL	75	75	76	7.3	72	76	75	
	FINAL	75	74	76	75	75	76		
temp (C)	INITIAL	219	215	223	214	214	211	227	
	FINAL	250	250	25	250	250	250		
CONC:		75							
D.O. (mg/L)	INITIAL	75	74	82	8.5	92	80	85	
	FINAL	73	75	76	75	75	73		
pH (s.u.)	INITIAL	75	75	75	7.3	72	75	74	
	FINAL	75	73	76	74	74	75		
temp (C)	INITIAL	219	215	223	214	214	211	229	
	FINAL	250	250	25	250	250	250		
CONC:		100							
D.O. (mg/L)	INITIAL	77	76	82	8.5	95	80	86	
	FINAL	73	75	77	77	74	73		
pH (s.u.)	INITIAL	73	73	73	7.2	71	74	73	
	FINAL	74	72	75	73	73	73		
temp (C)	INITIAL	220	215	203	214	214	212	228	
	FINAL	250	250	25	250	250	250		
CONC:		100%	A	A	B	B	C	C	
ALKALINITY (mg/L)		16			8		16		
HARDNESS (mg/L)		2600			2600		2600		
CONDUCTIVITY (umhos/cm)		22000			22000		22000		
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	K90309		TEST START DATE	3/25	TIME	1656					
CLIENT	Weston		TEST END DATE	4/1	TIME	1030					
Summary							AGE AND SOURCE OF MINNOWS				
							DAY (NUMBER SURVIVING)				
							SURVIVAL				
CONC:	REP #	start	1	2	3	4	5	6	7 %	MEAN %	CV
Control	A	8	8	8	8	8	8	8	7 87.5	97.5	5.73
	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	7	7	7	7 87.5	95.0	
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	87.5		
	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	87.5		
	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	7	7	7	6 75	95.0	
	B	8	8	8	8	8	8	8	100		
	C	8	8	8	8	8	8	8	87.5		
	D	8	8	8	8	8	8	8	100		
	E	8	8	8	8	8	8	8	100		
100	A	8	8	8	8	8	8	8	8 100	97.5	5.73
	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	87.5		
	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										
DAY (NUMBER SURVIVING)						SURVIVAL				
REP #	start	1	2	3	4	5	6	7 %	MEAN %	CV
C	CONC: 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		
32	CONC: 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		
42	CONC: 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		
56	CONC: 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		
75	CONC: 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		
100	CONC: 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	KP	KP	CT	CT	KP	KP	KP	
DATE:		3/25	3/26	3/27	3/28	3/29	3/30	3/31	4/1	
TIME:		1650	1530	1030		11:30	1640	1405	1030	

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)										
REP #	start	1	2	3	4	5	6	7 %	MEAN %	CV
C	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
32	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
42	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
56	CONC: A	2	2	2	2	2	2	1		
	B	1	1	1	1	1	1	2		
	C	1	1	1	1	1	1	2		
	D	1	1	1	1	1	1	2		
	E	1	1	1	1	1	1	2		
75	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
100	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
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					SURVIVAL				
		DAY (NUMBER SURVIVING)									
CONC:	REP #	start	1	2	3	4	5	6	7 %	MEAN %	CV
C	A	2	2	2	3	2	2	2	2		
	B	1	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1	1		
32	A	2	2	2	2	2	2	2	1		
	B	1	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1	1		
42	A	2	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1	1		
56	A	2	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1	1		
75	A	2	2	2	2	2	2	2	1		
	B	1	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1	1		
100	A	2	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1	1		
ANALYST											
DATE:					3-29						
TIME:					11:30						

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							
Weston D		AGE AND SOURCE OF MINNOWS									
		DAY (NUMBER SURVIVING)				SURVIVAL					
CONC:	REP #	start	1	2	3	4	5	6	7 %	MEAN %	CV
C	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			
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			
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			
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			
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			
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											
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										
DAY (NUMBER SURVIVING)							SURVIVAL			
REP #	start	1	2	3	4	5	6	7 %	MEAN %	CV
C	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
32	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
42	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
56	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
75	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
100	CONC: A	2	2	2	2	2	2	2		
	B	1	1	1	1	1	1	1		
	C	1	1	1	1	1	1	1		
	D	1	1	1	1	1	1	1		
	E	1	1	1	1	1	1	1		
ANALYST					cl					
DATE:					3-29					
TIME:					11:10					

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

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB # / #s:		K903009		TEST DATES (BEGIN / END):		3/25/09, 4/1/09	
CLIENT:		EEMA		WEIGHING DATE / TIME:		4/6/09, 1520	
ANALYSTS:		KP		DRYING TEMP (DEGREES C):		60	
SAMPLE ID:		SEE COC		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	0.95351	0.95124	0.00227	8	0.284	AVG DRY
	B	0.99254	0.98993	0.00261	8	0.326	WEIGHT (mg)
	C	0.97188	0.96910	0.00278	8	0.348	0.329
	D	0.95509	0.95214	0.00295	8	0.369	CV
	E	0.97889	0.97635	0.00254	8	0.317	9.7
CONC: 32%	A	0.93840	0.93575	0.00265	8	0.331	AVG DRY
	B	0.98136	0.97749	0.00387	8	0.484	WEIGHT (mg)
	C	0.94360	0.94044	0.00316	8	0.395	0.379
	D	0.99625	0.99335	0.00290	8	0.363	CV
	E	0.95046	0.94788	0.00258	8	0.323	
CONC: 42%	A	0.94246	0.93873	0.00373	8	0.466	AVG DRY
	B	0.95690	0.95316	0.00374	8	0.467	WEIGHT (mg)
	C	0.96686	0.96287	0.00399	8	0.499	0.469
	D	0.96385	0.95972	0.00413	8	0.516	CV
	E	0.99220	0.98902	0.00318	8	0.397	
CONC: 56%	A	0.99195	0.98798	0.00397	8	0.496	AVG DRY
	B	0.98862	0.98524	0.00338	8	0.423	WEIGHT (mg)
	C	1.00393	1.00007	0.00386	8	0.482	0.482
	D	0.96100	0.95651	0.00449	8	0.561	CV
	E	0.96090	0.95732	0.00358	8	0.448	
CONC: 75%	A	1.00850	1.00457	0.00393	8	0.491	AVG DRY
	B	0.98929	0.98453	0.00476	8	0.595	WEIGHT (mg)
	C	0.99563	0.99149	0.00414	8	0.518	0.598
	D	1.00776	1.00149	0.00627	8	0.784	CV
	E	0.95758	0.95275	0.00483	8	0.604	
CONC: 100%	A	1.03154	1.02523	0.00631	8	0.789	AVG DRY
	B	0.94874	0.94380	0.00494	8	0.618	WEIGHT (mg)
	C	0.94664	0.94130	0.00534	8	0.668	0.655
	D	0.98616	0.98093	0.00523	8	0.654	CV
	E	0.97229	0.96789	0.00440	8	0.550	13.3

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

Pimephales promelas

FATHEAD MINNOW

TEST 1000.0

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB # / #s: <u>K90309</u>	TEST DATES (BEGIN / END): <u>3/25/09 4/1/09</u>
CLIENT: <u>Weston</u>	WEIGHING DATE / TIME: <u>4/6/09 1520</u>
ANALYSTS: <u>KP</u>	DRYING TEMP (DEGREES C): <u>60</u>
SAMPLE ID:	DRYING TIME (HOURS): <u>24</u>

	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	0.95351	0.95124				AVG DRY WEIGHT (mg)
	B	0.99254	0.98993				
	C	0.97188	0.96910				
	D	0.95509	0.95214				CV
	E	0.97889	0.97635				
CONC: 31	A	0.93840	0.93575				AVG DRY WEIGHT (mg)
	B	0.98136	0.97749				
	C	0.94360	0.94044				
	D	0.99675	0.99335				CV
	E	0.95046	0.94788				
CONC: 42	A	0.94246	0.93873				AVG DRY WEIGHT (mg)
	B	0.95690	0.95316				
	C	0.96686	0.96287				
	D	0.96385	0.95972				CV
	E	0.99270	0.98902				
CONC: 56	A	0.99195	0.98798				AVG DRY WEIGHT (mg)
	B	0.98862	0.98574				
	C	1.00393	1.00067				
	D	0.96100	0.95051				CV
	E	0.96090	0.95732				
CONC: 75	A	1.00856	1.00457				AVG DRY WEIGHT (mg)
	B	0.98929	0.98453				
	C	0.99563	0.99149				
	D	1.00776	1.00149				CV
	E	0.96758	0.95775				
CONC: 100	A	1.03154	1.02523				AVG DRY WEIGHT (mg)
	B	0.94874	0.94380				
	C	0.94664	0.94130				
	D	0.98616	0.98093				CV
	E	0.97679	0.96789				

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

AA# K903009, FATHEAD MINNOW SURVIVAL, CHRONIC 3-25-09
File: J:\TOXSTAT\MONTE\FHSURV~1. Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.219

W = 0.839

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# K903009, FATHEAD MINNOW SURVIVAL, CHRONIC 3-25-09
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# K903009, FATHEAD MINNOW SURVIVAL, CHRONIC 3-25-09
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	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	0.8750	1.2094
2	32 % EFFLUENT	2	1.0000	1.3931
2	32 % EFFLUENT	3	0.8750	1.2094
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	0.8750	1.2094
4	56 %	EFFLUENT	3	1.0000	1.3931
4	56 %	EFFLUENT	4	1.0000	1.3931
4	56 %	EFFLUENT	5	1.0000	1.3931
5	75 %	EFFLUENT	1	0.7500	1.0472
5	75 %	EFFLUENT	2	1.0000	1.3931
5	75 %	EFFLUENT	3	0.8750	1.2094
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	0.8750	1.2094
6	100 %	EFFLUENT	5	1.0000	1.3931

AA# K903009, FATHEAD MINNOW SURVIVAL, CHRONIC 3-25-09

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	1.356				
2	32 % EFFLUENT	1.320	25.00	16.00	5.00	
3	42 % EFFLUENT	1.393	30.00	16.00	5.00	
4	56 % EFFLUENT	1.356	27.50	16.00	5.00	
5	75 % EFFLUENT	1.287	24.50	16.00	5.00	
6	100 % EFFLUENT	1.356	27.50	16.00	5.00	

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

AA# K903009, FATHEAD MINNOW GROWTH CHRONIC, 3-25-09

File: J:/toxstat/monte\FHGGROWTH.

Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.137

W = 0.931

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# K903009, FATHEAD MINNOW GROWTH CHRONIC, 3-25-09

File: J:/toxstat/monte\FHGGROWTH.

Transform: ARC SINE(SQUARE ROOT(Y))

Bartlett's test for homogeneity of variance

Calculated B1 statistic = 7.83

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# K903009, FATHEAD MINNOW GROWTH CHRONIC, 3-25-09

FILE: J:/toxstat/monte\FHGGROWTH.

TRANSFORM: ARC SINE(SQUARE ROOT(Y))

NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.2840	0.5620
1	CONTROL	2	0.3260	0.6077
1	CONTROL	3	0.3480	0.6310
1	CONTROL	4	0.3690	0.6529
1	CONTROL	5	0.3170	0.5980
2	32 % EFFLUENT	1	0.3310	0.6130
2	32 % EFFLUENT	2	0.4840	0.7694
2	32 % EFFLUENT	3	0.3950	0.6796
2	32 % EFFLUENT	4	0.3620	0.6456
2	32 % EFFLUENT	5	0.3230	0.6045
3	42 % EFFLUENT	1	0.4660	0.7514
3	42 % EFFLUENT	2	0.4670	0.7524
3	42 % EFFLUENT	3	0.4990	0.7844
3	42 % EFFLUENT	4	0.5160	0.8014
3	42 % EFFLUENT	5	0.3970	0.6817
4	56 % EFFLUENT	1	0.4960	0.7814

4	56 % EFFLUENT	2	0.4230	0.7081
4	56 % EFFLUENT	3	0.4820	0.7674
4	56 % EFFLUENT	4	0.5610	0.8466
4	56 % EFFLUENT	5	0.4480	0.7333
5	75 % EFFLUENT	1	0.4910	0.7764
5	75 % EFFLUENT	2	0.5950	0.8810
5	75 % EFFLUENT	3	0.5180	0.8034
5	75 % EFFLUENT	4	0.7840	1.0874
5	75 % EFFLUENT	5	0.6040	0.8902
6	100 % EFFLUENT	1	0.7890	1.0935
6	100 % EFFLUENT	2	0.6180	0.9045
6	100 % EFFLUENT	3	0.6680	0.9567
6	100 % EFFLUENT	4	0.6540	0.9419
6	100 % EFFLUENT	5	0.5500	0.8355

AA# K903009, FATHEAD MINNOW GROWTH CHRONIC, 3-25-09

File: J:/toxstat/monte\FHGGROWTH.

Transform: ARC SINE(SQUARE ROOT(Y))

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.411	0.082	14.378
Within (Error)	24	0.137	0.006	
Total	29	0.549		

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

AA# K903009, FATHEAD MINNOW GROWTH CHRONIC, 3-25-09

File: J:/toxstat/monte\FHGGROWTH.

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	0.610	0.329		
2	32 % EFFLUENT	0.662	0.379	-1.089	
3	42 % EFFLUENT	0.754	0.469	-3.008	
4	56 % EFFLUENT	0.767	0.482	-3.282	
5	75 % EFFLUENT	0.888	0.598	-5.797	
6	100 % EFFLUENT	0.946	0.656	-7.025	

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

AA# K903009, FATHEAD MINNOW GROWTH CHRONIC, 3-25-09

File: J:/toxstat/monte\FHGGROWTH.

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	0.101	30.7	-0.050
3	42 % EFFLUENT	5	0.101	30.7	-0.140
4	56 % EFFLUENT	5	0.101	30.7	-0.153
5	75 % EFFLUENT	5	0.101	30.7	-0.270
6	100 % EFFLUENT	5	0.101	30.7	-0.327

APPENDIX D

Ceriodaphnia dubia Raw Data and Statistics

SURVIVAL AND REPRODUCTION TEST

Cerodaphnia dubia
 Discharger: Neston
 Lab Number/s: N0309
 Date Sample Collected: _____

Analyst: RP

Test Start - Date/Time: 3/25/09, 0930
 Test Stop - Date/Time: 4/08, 1150

Conc 1	Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
		A	B	C	D	E	F	G	H	I	J				
Control	1	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
	2	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
	3	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
	4	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
	5	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
	6	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
	7	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
	8	0	0	0	0	0	0	0	0	0	0	0	10	0	RP
Total		0	0	0	0	0	0	0	0	0	0	0	100	0	

Conc 2	Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
		A	B	C	D	E	F	G	H	I	J				
N	1	0	0	0	0	0	0	0	0	0	0	0	16	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	0	10	0	
	4	0	0	0	0	0	0	0	0	0	0	0	10	0	
	5	0	0	0	0	0	0	0	0	0	0	0	10	0	
	6	0	0	0	0	0	0	0	0	0	0	0	10	0	
	7	0	0	0	0	0	0	0	0	0	0	0	10	0	
	8	0	0	0	0	0	0	0	0	0	0	0	10	0	
Total		0	0	0	0	0	0	0	0	0	0	0	146	0	

Conc 3	Day	Replicate										No. of Young	No. of Adult	Young/Adult	Analyst
		A	B	C	D	E	F	G	H	I	J				
1/2	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	0	10	0	
	4	0	0	0	0	0	0	0	0	0	0	0	10	0	
	5	0	0	0	0	0	0	0	0	0	0	0	10	0	
	6	0	0	0	0	0	0	0	0	0	0	0	10	0	
	7	0	0	0	0	0	0	0	0	0	0	0	10	0	
	8	0	0	0	0	0	0	0	0	0	0	0	10	0	
Total		0	0	0	0	0	0	0	0	0	0	0	156	0	

X = DEAD, Y = MALE
 $\bar{X} = 13.2$
 $CV = 28.5$

AA # K903009, C. DUBIA CHRONIC, REPRODUCCION, 3-25-09
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 # K903009, C. DUBIA CHRONIC, REPRODUCCION, 3-25-09
File: J:\TOXSTAT\MONTE\C.DUB Transform: NO TRANSFORMATION

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 4.69

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		
	DEAD	ALIVE	TOTAL ANIMALS
CONTROL	1	9	10
32%	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
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		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	9	1	10
56%	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		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	9	1	10
75%	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		
	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)

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

TITLE: AA # K903009, C. DUBIA CHRONIC, REPRODUCCION, 3-25-09
FILE: J:\TOXSTAT\MONTE\C.DUB
TRANSFORM: NO TRANSFORMATION NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	22.0000	22.0000
1	CONTROL	2	16.0000	16.0000
1	CONTROL	3	10.0000	10.0000
1	CONTROL	4	26.0000	26.0000
1	CONTROL	5	13.0000	13.0000
1	CONTROL	6	15.0000	15.0000
1	CONTROL	7	16.0000	16.0000
1	CONTROL	8	14.0000	14.0000
1	CONTROL	9	21.0000	21.0000
1	CONTROL	10	14.0000	14.0000
2	32 % EFFLUENT	1	17.0000	17.0000
2	32 % EFFLUENT	2	15.0000	15.0000
2	32 % EFFLUENT	3	16.0000	16.0000
2	32 % EFFLUENT	4	19.0000	19.0000
2	32 % EFFLUENT	5	10.0000	10.0000
2	32 % EFFLUENT	6	12.0000	12.0000
2	32 % EFFLUENT	7	10.0000	10.0000
2	32 % EFFLUENT	8	20.0000	20.0000
2	32 % EFFLUENT	9	10.0000	10.0000
2	32 % EFFLUENT	10	17.0000	17.0000
3	42 % EFFLUENT	1	14.0000	14.0000
3	42 % EFFLUENT	2	10.0000	10.0000
3	42 % EFFLUENT	3	15.0000	15.0000
3	42 % EFFLUENT	4	19.0000	19.0000
3	42 % EFFLUENT	5	14.0000	14.0000
3	42 % EFFLUENT	6	13.0000	13.0000
3	42 % EFFLUENT	7	14.0000	14.0000
3	42 % EFFLUENT	8	19.0000	19.0000
3	42 % EFFLUENT	9	14.0000	14.0000
3	42 % EFFLUENT	10	24.0000	24.0000
4	56 % EFFLUENT	1	23.0000	23.0000
4	56 % EFFLUENT	2	16.0000	16.0000
4	56 % EFFLUENT	3	23.0000	23.0000
4	56 % EFFLUENT	4	17.0000	17.0000
4	56 % EFFLUENT	5	7.0000	7.0000
4	56 % EFFLUENT	6	17.0000	17.0000
4	56 % EFFLUENT	7	17.0000	17.0000
4	56 % EFFLUENT	8	12.0000	12.0000
4	56 % EFFLUENT	9	0.0000	0.0000
4	56 % EFFLUENT	10	19.0000	19.0000

5	75 % EFFLUENT	1	18.0000	18.0000
5	75 % EFFLUENT	2	12.0000	12.0000
5	75 % EFFLUENT	3	10.0000	10.0000
5	75 % EFFLUENT	4	14.0000	14.0000
5	75 % EFFLUENT	5	17.0000	17.0000
5	75 % EFFLUENT	6	13.0000	13.0000
5	75 % EFFLUENT	7	15.0000	15.0000
5	75 % EFFLUENT	8	11.0000	11.0000
5	75 % EFFLUENT	9	2.0000	2.0000
5	75 % EFFLUENT	10	18.0000	18.0000
6	100 % EFFLUENT	1	9.0000	9.0000
6	100 % EFFLUENT	2	11.0000	11.0000
6	100 % EFFLUENT	3	16.0000	16.0000
6	100 % EFFLUENT	4	19.0000	19.0000
6	100 % EFFLUENT	5	8.0000	8.0000
6	100 % EFFLUENT	6	13.0000	13.0000
6	100 % EFFLUENT	7	15.0000	15.0000
6	100 % EFFLUENT	8	11.0000	11.0000
6	100 % EFFLUENT	9	0.0000	0.0000
6	100 % EFFLUENT	10	17.0000	17.0000

AA # K903009, C. DUBIA CHRONIC, REPRODUCCION, 3-25-09
 File: J:\TOXSTAT\MONTE\C.DUB Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	154.283	30.857	1.176
Within (Error)	54	1416.700	26.235	
Total	59	1570.983		

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

AA # K903009, C. DUBIA CHRONIC, REPRODUCCION, 3-25-09
 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	16.700	16.700		
2	32 % EFFLUENT	14.600	14.600	0.917	
3	42 % EFFLUENT	15.600	15.600	0.480	
4	56 % EFFLUENT	15.100	15.100	0.698	
5	75 % EFFLUENT	13.000	13.000	1.615	
6	100 % EFFLUENT	11.900	11.900	2.095	

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

AA # K903009, C. DUBIA CHRONIC, REPRODUCCION, 3-25-09

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	5.291	31.7	2.100
3	42 % EFFLUENT	10	5.291	31.7	1.100
4	56 % EFFLUENT	10	5.291	31.7	1.600
5	75 % EFFLUENT	10	5.291	31.7	3.700
6	100 % EFFLUENT	10	5.291	31.7	4.800

AA # K903009, C. DUBIA CHRONIC, REPRODUCCION, 3-25-09

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	16.700				
2	32 % EFFLUENT	14.600	96.00	75.00	10.00	
3	42 % EFFLUENT	15.600	96.50	75.00	10.00	
4	56 % EFFLUENT	15.100	108.00	75.00	10.00	
5	75 % EFFLUENT	13.000	87.50	75.00	10.00	
6	100 % EFFLUENT	11.900	83.00	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 3-25-09 Arkansas Analytical

SPECIES Pimephales promelas

QUANTITY SHIPPED 540+

AGE/LIFE STAGE 4 wks 3/25 1500ct

BROODSTOCK SOURCE Anderson Farms, AR

CULTURE WATER groundwater

ALKALINITY (Mg/l as CaCO₃) 180

HARDNESS (Mg/l as CaCO₃)/Salinity (ppt) 160

FEEDING ARTEMIL

COMMENTS _____

PACKAGED BY llw

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:	<u>24°C</u>	<u>22-25°C</u>
SALINITY/CONDUCTIVITY:	<u>--</u>	<u>--</u>
TOTAL HARDNESS (as CaCO ₃):	<u>126 mg/l</u>	<u>60-138 mg/l</u>
TOTAL ALKALINITY (as CaCO ₃):	<u>60 mg/l</u>	<u>50-110 mg/l</u>
pH:	<u>8.00</u>	<u>6.98-8.32</u>

Comments:

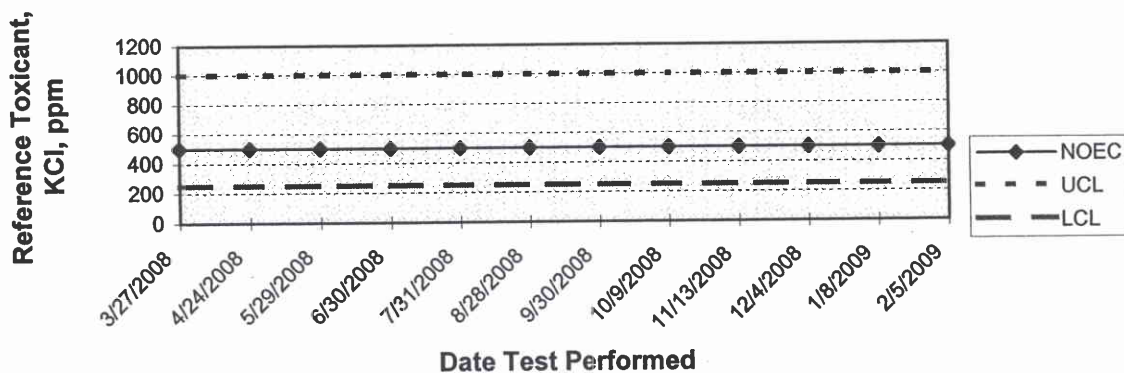


Facility Supervisor

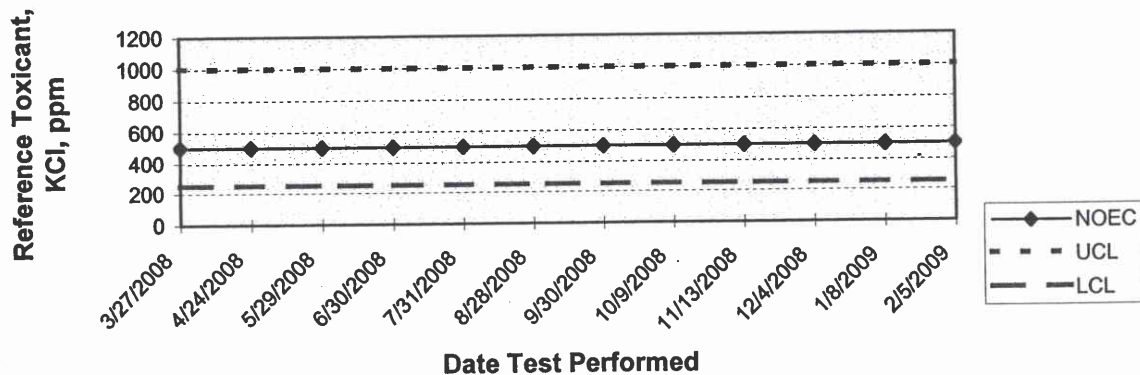
APPENDIX F

Quality Assurance Charts

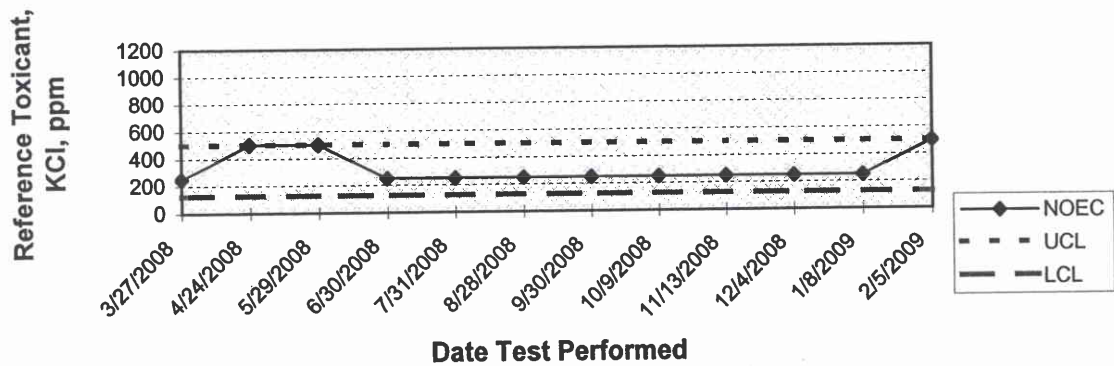
ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW SURVIVAL
QUALITY ASSURANCE



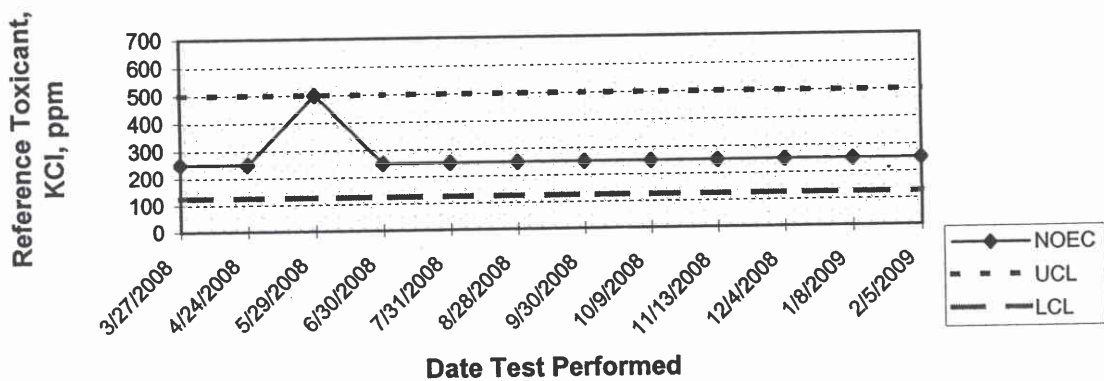
ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW GROWTH
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.
CERIODAPHNIA DUBIA SURVIVAL
QUALITY ASSURANCE



ARKANSAS ANALYTICAL, INC.
CERIODAPHNIA DUBIA REPRODUCTION
QUALITY ASSURANCE



APPENDIX G

Lab Certification



State of Arkansas
Department of Environmental Quality
Laboratory Certification Program

Arkansas Analytical, Inc.

Little Rock, AR

has earned certification by law in accordance with Code Annotated §8-2-201 et seq., the State Environmental Laboratory Certification Program Act for the following parameters:

Alkalinity	Orthophosphate	Antimony	Mercury	Explosives
Ammonia	Perchlorate	Arsenic	Molybdenum	GRO
BOD	pH	Barium	Nickel	TPH
Bromide	Phenol	Beryllium	Potassium	Acute Toxicity
CBOD	Sulfate	Boron	Selenium	Chronic Toxicity
Chloride	Sulfide	Cadmium	Silver	Herbicides
Chlorine	TDS	Calcium	Sodium	Pesticides & PCBs
COD	TKN	Chromium	Strontium	Semi-volatiles
Conductivity	TOC	Cobalt	Thallium	Volatile Organics
Cyanide	Total Phosphorus	Copper	Tin	
Fluoride	Total Solids	Hex. Chromium	Titanium	
Hardness	TSS	Iron	Vanadium	
Nitrate	Turbidity	Lead	Zinc	
Nitrite	Vol Solids	Magnesium	Fecal Coliform	
Oil & Grease	Aluminum	Manganese	DRO	

Laboratory ID: 60-1754

Certificate Number: 08-073-0

Issued Date: 30 October 2008

Expired Date: 30 October 2009

Jeresa Maibe

ADEQ Director