



Arkansas Analytical, Inc.

Toxicity Test Results

**MAGCOBAR MINE SITE
NPDES PERMIT NUMBER: AR0049794
July 2005**

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
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Tuesday, August 16, 2005



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 Quachita 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 July of 2005.

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:	7-19-05, 0930	7-20-05, 0930
Sample #2:	7-20-05, 0930	7-21-05, 0930
Sample #3:	7-25-05, 0930	7-26-05, 0930

The sample was a composite 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	Storage Temperature (°C)
Sample #1:	7-20-05, 1120	4
Sample #2:	7-21-05, 1119	4
Sample #3:	7-26-05, 1142	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. There were no deviations from the reference method. The test chambers were 500 ml plastic cups, and each chamber contained ten organisms in a test solution volume of 250 mls. 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	100%	X	
Average of 15 or more young per surviving female	15.7	X	
At least 60% of surviving females should have produced 3 broods	60%	X	
The percent coefficient of variation between replicates must be 40% or less for the young of surviving females	30.2%	X	

TEST ACCEPTANCE CRITERIA for *Pimephales promelas*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	96%	X	
The percent coefficient of variation between replicates must be 40% or less for survival	9.32%	X	
Minimum of 0.25 mg average dry weight of surviving controls	0.353	X	
The percent coefficient of variation between replicates must be 40% or less for growth	17.9%	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>		<i>Pimephales promelas</i>	
NOEC Survival:	250 ppm KCl	NOEC Survival:	500 ppm KCl
LOEC Survival:	500 ppm KCl	LOEC Survival:	1000 ppm KCl
NOEC Reproduction:	250 ppm KCl	NOEC Growth:	500 ppm KCl
LOEC Reproduction:	500 ppm KCl	LOEC Growth:	1000 ppm KCl

Quality Assurance charts are provided in Appendix F.



Summary of Results

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)	15.7	%CV survival (critical dilution)	5.71%
%CV Reproduction (critical dilution)	30.2%	Mean dry weight (critical dilution) in milligrams	0.565
		%CV growth (critical dilution)	17.9%

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, 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:

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**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:	7-19-05, 0930	7-20-05, 0930
Sample #2:	7-20-05, 0930	7-21-05, 0930
Sample #3:	7-25-05, 0930	7-26-05, 0930

Test initiated (date, time): 7-21-05, 1530 Test terminated (date, time): 7-28-05, 1135

Dilution water used: Soft Synthetic

DATA TABLE FOR FATHEAD MINNOW SURVIVAL

Effluent Conc %	Percent Survival in Replicate Chambers					Mean Percent Survival				CV %
	A	B	C	D	E	24 hours	48 hours	7 days		
0%	100	100	100	80	100	100	100	96	9.32	
32%	100	90	100	100	100	100	100	98		
42%	100	100	100	100	80	100	100	96		
56%	90	100	90	100	100	98	98	96		
75%	100	100	90	100	100	100	100	98		
100%	100	90	90	100	100	98	98	96	5.71	

DATA TABLE FOR GROWTH OF FATHEAD MINNOWS

Average Dry Weight in milligrams in replicate chambers

Effluent Conc %	A	B	C	D	E	Mean Dry Weight	CV%
0%	0.437	0.417	0.371	0.234	0.304	0.353	23.7
32%	0.436	0.395	0.436	0.512	0.451	0.446	
42%	0.523	0.533	0.476	0.487	0.472	0.498	
56%	0.550	0.503	0.478	0.453	0.490	0.495	
75%	0.532	0.536	0.402	0.520	0.461	0.490	
100%	0.613	0.499	0.420	0.636	0.655	0.565	17.9

Coefficient of Variation = standard deviation / mean * 100



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

1. Dunnett's procedure or Steel's Many-One Rank Test as appropriate:
Is the mean survival at 7 days significantly different ($p=0.05$) than the control survival for:
a) LOW FLOW OR CRITICAL DILUTION, (100%) YES _____ NO **X** _____
2. Dunnett's Procedure
Is the mean dry weight (growth) at 7 days significantly different ($p=0.05$) than the control's dry weight (growth) for:
a) LOW FLOW OR CRITICAL DILUTION, (100%) YES _____ NO **X** _____
3. If NO was answered to 1.a) enter [0] otherwise enter [1] (parameter TLP6C): **0** _____
4. If NO was answered to 2.a) enter [0] otherwise enter [1] (parameter TGP6C): **0** _____
5. Enter percentage corresponding to each parameter below:
 - a) NOEC survival (parameter TOP6C)= **100** % effluent
 - b) NOEC growth (parameter TPP6C)= **100** % effluent
 - c) Coefficient of variation (parameter TQP6C)= **9.32** %



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:	7-19-05, 0930	7-20-05, 0930
Sample #2:	7-20-05, 0930	7-21-05, 0930
Sample #3:	7-25-05, 0930	7-26-05, 0930

Test initiated (date, time): 7-21-05, 1000 Test terminated (date, time): 7-28-05, 0920

Dilution water used: Soft Synthetic

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

Replicate	0%	32%	42%	52%	75%	100%
A	21	14	17	22	14	15
B	11	X0	X3	18	18	14
C	21	18	19	17	13	19
D	13	20	11	21	18	22
E	22	X0	20	16	14	X0
F	13	13	16	20	12	18
G	12	10	17	14	11	11
H	19	10	13	20	19	X0
I	16	X2	12	5	X12	14
J	9	14	17	9	X13	14
Mean	15.7	9.8	14.5	16.2	14.4	12.7
Mean/surviving female	15.7	14.1	15.8	16.2	14.9	15.9
CV%*	30.2					22.2

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: Magobar Mine Site

NPDES #: AR0049794

PERCENT SURVIVAL

PERCENT EFFLUENT	0%	32%	42%	56%	75%	100%
Time of Reading: 24 HOURS	100	80	100	100	100	80
48 HOURS	100	80	100	100	100	80
Test termination	100	70	90	100	80	80

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)= 30.2 %

APPENDIX A

Chain of Custody Forms

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		Project Description		Turnaround Time		Preservation Codes:	
EEMA O&M P.O. Box 699 2000 Darby Lane Malvern, AR 72104 Attn: Darrel Scott		MAGCOBAR Mine Site Reporting Information Telephone: 501/467-8355 FAX: 501/467-8687 Bill to/P.O.		(CIRCLE ONE) 24 hour 48 hour Touring Preservative Code: <input checked="" type="checkbox"/> P		1. Cool, 4 degrees Centigrade 2. Sulfuric Acid, pH <2 3. Nitric Acid, pH <2 4. Thiosulfate for dechlorination 5. Hydrochloric Acid for VO4 6. Sodium Hydroxide, pH >12	
TEST PARAMETERS							
<i>Darrel Scott</i>		<i>Darrel Scott</i> Samplers/(Signatures)		Chronic Bio		Arkansas Analytical Lab #	
		Field Number	Sample Collection Date/s	# of Containers	Sample Matrix	SAMPLE IDENTIFICATION/DESCRIPTION	<input checked="" type="checkbox"/> K507556A
FD720Comp	7/20/2005	X	8	Facility Discharge			
1 Relinquished by/(Signature) <i>Bill Horton</i>				1 Received by/(Signature) <i>10.15 11:20</i>		For completion by laboratory REMARKS Temp. on Receipt - 17°C	
2 Relinquished by/(Signature) <i> </i>				2 Received by laboratory/(Signature) <i>7-20-05 1120+015</i>		Condition of samples: <input checked="" type="checkbox"/> A. Containers Correct? <input checked="" type="checkbox"/> B. Preservation Correct? <input checked="" type="checkbox"/> C. Seals Intact?	

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		Project Description		Turnaround Time (CIRCLE ONE)		Preservation Codes:	
EEMA O&M P.O. Box 698 2000 Derby Lane Malvern, AR 72104 Attn: Darrel Scott		MAGCOBAR Mine Site Reporting Information Telephone: 501/467-8355 FAX: 501/467-8687 Bill to/P.O.: Darrel Scott		24 hour 48 hour <u>Routine</u> Preservative Code:		1. Cool, 4 degrees Centigrade 2. Sulfuric Acid, pH <2 3. Nitric Acid, pH <2 4. Thiosulfate for dechlorination 5. Hydrochloric Acid for VOA 6. Sodium Hydroxide, pH >12	
TEST PARAMETERS							
<u>Darrel Scott</u> <u>Darrel Scott</u>		Samplers (Printed) Field Number Sample Collection Date/s # of Containers		Sample Matrix Chronic Bio		Arkansas Analytical Lab #	
		Date/s 7/21/2005 9:30		# of Containers X Facility Discharge		K5075506	
Samplers (Signature/s)		Sample Collection Date/s		SAMPLE IDENTIFICATION/ DESCRIPTION		For completion by laboratory	
<u>Biff Horner</u>		<u>7-21-05 10:00 119</u>		<u>1. Received by (Signature)</u> <u>7-21-05</u>		Condition of samples: A. Containers Correct?: <input checked="" type="checkbox"/> B. Preservation Correct?: <input checked="" type="checkbox"/> C. Seals Intact?: <input checked="" type="checkbox"/>	
<u>Sydney James</u>		<u>119</u>		<u>2. Received by laboratory (Signature)</u> <u>7-21-05</u>		Remarks <u>Temp - 10C</u>	
Relinquished by: (Signature)		Date/Time		Date/Time		Date/Time	
<u>Biff Horner</u>		<u>7-21-05 10:00 119</u>		<u>7-21-05 119</u>		<u>7-21-05 119</u>	
Relinquished by: (Signature)		Date/Time		Date/Time		Date/Time	
<u>Sydney James</u>		<u>119</u>		<u>119</u>		<u>119</u>	

CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		Project Description		Turnaround Time (CIRCLE ONE)		Preservation Codes:	
EEMA Q&M INC. P.O. Box 698 2000 Derby Lane Malvern, AR 72104 Attn: Darel Scott		MAGCOBAR Mine Site Reporting Information Telephone: 501/467-8355 FAX: 601/467-8687 Billed to/P.O.		24 hour 48 hour routine Preservative Code: Bottle Type: P		1. Cool, 4 degrees Centigrade 2. Sulfuric Acid, pH <2 3. Nitric Acid, pH <2 4. Thiosulfate for dechlorination 5. Hydrochloric Acid for VOA 6. Sodium Hydroxide, pH >12 G=glass,P=HDPE V=septum,A=amber	
TEST PARAMETERS							
<i>Darel Scott</i>						Arkansas	
						Analytical Lab #	
Samplers(Signature/s)		SAMPLE IDENTIFICATION/ DESCRIPTION		Chromic Bi ₆		REMARKS	
Field Number	Sample Collection Date/s	Time/s	Grab Comp	# of Containers	Sample Matrix	X	
FD726COMP	7/26/2005	9:30	X	3	Facility Discharge	X	
1. Received by/ <i>Signature</i> : Bell Harbor Date/Time: 7-26-05 10:30 2. Received by/laboratory/ <i>Signature</i> : Sydney James Date/Time: 7-26-05 1142							
For completion by laboratory Condition of samples: A. Containers Correct? <input checked="" type="checkbox"/> No <input type="checkbox"/> B. Preservation Correct? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> C. Seals intact? <input checked="" type="checkbox"/> Yes <input type="checkbox"/>							

APPENDIX B

Effluent and Dilution Water Data

APPENDIX C

Fathead minnow raw data and statistics

Pimephales promelas

FATHEAD MINNOW

TEST 1000.0

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB # / #'s:	K507556			TEST DATES (BEGIN / END):	7/21-28/05		
CLIENT:	Weston			WEIGHING DATE / TIME:			
ANALYSTS:				DRYING TEMP (DEGREES C):	60		
SAMPLE ID:				DRYING TIME (HOURS):	24		
REP #	FINAL DRY WEIGHT TIN+LARVAE (g)	INITIAL WEIGHT TIN (g)	TOTAL DRY WEIGHT OF LARVAE (g)	NUMBER OF LARVAE	DRY WEIGHT OF LARVAE (mg)		
CONTROL	A	0.97485	0.97048	0.00437	10	0.437	
	B	0.97600	0.97183	0.00417	10	0.417	
	C	0.97227	0.96856	0.00371	10	0.371	
	D	0.97216	0.96982	0.00234	10	0.234	
	E	0.97352	0.97048	0.00304	10	0.304	
CONC:	A	0.97624	0.97188	0.00436	10	0.436	
	B	0.97745	0.97350	0.00395	10	0.395	
	C	0.97781	0.97345	0.00436	10	0.436	
	D	0.98073	0.97561	0.00512	10	0.512	
	E	0.97653	0.97202	0.00451	10	0.451	
CONC:	A	0.97778	0.97255	0.00523	10	0.523	
	B	0.97735	0.97202	0.00533	10	0.533	
	C	0.97972	0.97496	0.00476	10	0.476	
	D	0.97671	0.97184	0.00487	10	0.487	
	E	0.97760	0.97288	0.00472	10	0.472	
CONC:	A	0.97838	0.97288	0.00550	10	0.550	
	B	0.97613	0.97110	0.00503	10	0.503	
	C	0.98025	0.97547	0.00478	10	0.478	
	D	0.97443	0.96990	0.00453	10	0.453	
	E	0.98130	0.97640	0.00490	10	0.490	
CONC:	A	0.98023	0.97491	0.00532	10	0.532	
	B	0.98030	0.97494	0.00536	10	0.536	
	C	0.97455	0.97053	0.00402	10	0.402	
	D	0.97715	0.97195	0.00520	10	0.520	
	E	0.97307	0.96846	0.00461	10	0.461	
CONC:	A	0.97259	0.96646	0.00613	10	0.613	
	B	1.24670	1.24171	0.00499	10	0.499	
	C	1.27652	1.27232	0.00420	10	0.420	
	D	1.26227	1.25591	0.00636	10	0.636	
	E	1.25883	1.25228	0.00655	10	0.655	

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB # / #s:	K5C7554			TEST DATES (BEGIN / END):	7-21-05 / 7-28-05		
CLIENT:	Weston			WEIGHING DATE / TIME:			
ANALYSTS:	HH, JM, TLT			DRYING TEMP (DEGREES C):	60°C		
SAMPLE ID:				DRYING TIME (HOURS):	24 hrs.		
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 31	0.97485	0.97048			Avg Dry Weight (mg)	
	B 32	0.97400	0.97183				
	C 33	0.97227	0.96856				
	D 34	0.97210	0.96982				
	E 35	0.97352	0.97048			CV	
CONC:	A 36	0.971024	0.97188			Avg Dry Weight (mg)	
	B 37	0.97745	0.97350				
32%	C 38	0.97781	0.97345				
	D 39	0.98073	0.97561				
	E 40	0.971053	0.97202			CV	
CONC:	A 41	0.97778	0.97255			Avg Dry Weight (mg)	
	B 42	0.97735	0.97202				
42%	C 43	0.97912	0.97496				
	D 44	0.97671	0.97134				
	E 45	0.97700	0.97288			CV	
CONC:	A 46	0.97838	0.97288			Avg Dry Weight (mg)	
	B 47	0.97613	0.97110				
56%	C 48	0.98025	0.97547				
	D 49	0.97443	0.96990				
	E 50	0.98130	0.97640			CV	
CONC:	A 51	0.98023	0.97491			Avg Dry Weight (mg)	
	B 52	0.98030	0.97494				
75%	C 53	0.97455	0.97053				
	D 54	0.97715	0.97195				
	E 55	0.97307	0.96846			CV	
CONC:	A 56	0.97259	0.96646			Avg Dry Weight (mg)	
	B 57	1.24670	1.24171				
100%	C 58	1.27452	1.27232				
	D 59	1.26227	1.25591				
	E 60	1.25883	1.25228			CV	

CV = (STANDARD DEVIATION/MEAN)*100

REMARKS:

AA# K507556, FATHEAD MINNOW SURVIVAL, 7-21-05
File: k507556s Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

D = 0.255

W = 0.700

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# K507556, FATHEAD MINNOW SURVIVAL, 7-21-05
File: k507556s Transform: ARC SINE(SQUARE ROOT(Y))

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 3.07

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# K507556, FATHEAD MINNOW SURVIVAL, 7-21-05

FILE: C:\TOXSTAT\WESTON\K507556.S.

TRANSFORM: NO TRANSFORMATION

NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	1.0000	1.0000
1	CONTROL	2	1.0000	1.0000
1	CONTROL	3	1.0000	1.0000
1	CONTROL	4	0.8000	0.8000
1	CONTROL	5	1.0000	1.0000
2	32 % EFFLUENT	1	1.0000	1.0000
2	32 % EFFLUENT	2	0.9000	0.9000
2	32 % EFFLUENT	3	1.0000	1.0000
2	32 % EFFLUENT	4	1.0000	1.0000
2	32 % EFFLUENT	5	1.0000	1.0000
3	42 % EFLLUENT	1	1.0000	1.0000
3	42 % EFLLUENT	2	1.0000	1.0000
3	42 % EFLLUENT	3	1.0000	1.0000
3	42 % EFLLUENT	4	1.0000	1.0000
3	42 % EFLLUENT	5	0.8000	0.8000
4	56 % EFFLUENT	1	0.9000	0.9000
4	56 % EFFLUENT	2	1.0000	1.0000
4	56 % EFFLUENT	3	0.9000	0.9000
4	56 % EFFLUENT	4	1.0000	1.0000
4	56 % EFFLUENT	5	1.0000	1.0000
5	75 % EFFLUENT	1	1.0000	1.0000
5	75 % EFFLUENT	2	1.0000	1.0000
5	75 % EFFLUENT	3	0.9000	0.9000
5	75 % EFFLUENT	4	1.0000	1.0000
5	75 % EFFLUENT	5	1.0000	1.0000
6	100 % EFFLUENT	1	1.0000	1.0000
6	100 % EFFLUENT	2	0.9000	0.9000
6	100 % EFFLUENT	3	0.9000	0.9000
6	100 % EFFLUENT	4	1.0000	1.0000
6	100 % EFFLUENT	5	1.0000	1.0000

AA# K507556, FATHEAD MINNOW SURVIVAL, 7-21-05
File: C:\TOXSTAT\WESTON\K507556S. Transform: NO TRANSFORMATION

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

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	0.960				
2	32 % EFFLUENT	0.980	28.00	16.00	5.00	
3	42 % EFLLUENT	0.960	27.50	16.00	5.00	
4	56 % EFFLUENT	0.960	26.00	16.00	5.00	
5	75 % EFFLUENT	0.980	28.00	16.00	5.00	
6	100 % EFFLUENT	0.960	26.00	16.00	5.00	

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

AA # K507556, FATHEAD MINNOW GROWTH, 7-21-05
File: k507556g Transform: NO TRANSFORMATION

Shapiro - Wilk's test for normality

D = 0.098

W = 0.963

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 # K507556, FATHEAD MINNOW GROWTH, 7-21-05
File: k507556g Transform: NO TRANSFORMATION

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 8.57

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 # K507556, FATHEAD MINNOW GROWTH, 7-21-05

FILE: k507556g

TRANSFORM: NO TRANSFORMATION

NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.4370	0.4370
1	CONTROL	2	0.4170	0.4170
1	CONTROL	3	0.3710	0.3710
1	CONTROL	4	0.2340	0.2340
1	CONTROL	5	0.3040	0.3040
2	32 % EFFLUENT	1	0.4360	0.4360
2	32 % EFFLUENT	2	0.3950	0.3950
2	32 % EFFLUENT	3	0.4360	0.4360
2	32 % EFFLUENT	4	0.5120	0.5120
2	32 % EFFLUENT	5	0.4510	0.4510
3	42 % EFFLUENT	1	0.5230	0.5230
3	42 % EFFLUENT	2	0.5330	0.5330
3	42 % EFFLUENT	3	0.4760	0.4760
3	42 % EFFLUENT	4	0.4870	0.4870
3	42 % EFFLUENT	5	0.4720	0.4720
4	56 % EFFLUENT	1	0.5500	0.5500
4	56 % EFFLUENT	2	0.5030	0.5030
4	56 % EFFLUENT	3	0.4780	0.4780
4	56 % EFFLUENT	4	0.4530	0.4530
4	56 % EFFLUENT	5	0.4900	0.4900
5	75 % EFFLUENT	1	0.5320	0.5320
5	75 % EFFLUENT	2	0.5360	0.5360
5	75 % EFFLUENT	3	0.4020	0.4020
5	75 % EFFLUENT	4	0.5200	0.5200
5	75 % EFFLUENT	5	0.4610	0.4610
6	100 % EFFLUENT	1	0.6130	0.6130
6	100 % EFFLUENT	2	0.4990	0.4990
6	100 % EFFLUENT	3	0.4200	0.4200
6	100 % EFFLUENT	4	0.6360	0.6360
6	100 % EFFLUENT	5	0.6550	0.6550

AA # K507556, FATHEAD MINNOW GROWTH, 7-21-05
File: k507556g Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.125	0.025	6.143
Within (Error)	24	0.098	0.004	
Total	29	0.223		

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

AA # K507556, FATHEAD MINNOW GROWTH, 7-21-05
 File: k507556g Transform: NO TRANSFORMATION

DUNNETT'S TEST - TABLE 1 OF 2

Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED	MEAN CALCULATED IN	T STAT	SIG
		MEAN	ORIGINAL UNITS		
1	CONTROL	0.353	0.353		
2	32 % EFFLUENT	0.446	0.446	-2.315	
3	42 % EFFLUENT	0.498	0.498	-3.608	
4	56 % EFFLUENT	0.495	0.495	-3.524	
5	75 % EFFLUENT	0.490	0.490	-3.410	
6	100 % EFFLUENT	0.565	0.565	-5.254	

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

AA # K507556, FATHEAD MINNOW GROWTH, 7-21-05
 File: k507556g Transform: NO TRANSFORMATION

DUNNETT'S TEST - TABLE 2 OF 2

Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF	Minimum Sig Diff	% of	DIFFERENCE
		REPS	(IN ORIG. UNITS)	CONTROL	FROM CONTROL
1	CONTROL	5			
2	32 % EFFLUENT	5	0.095	27.0	-0.093
3	42 % EFFLUENT	5	0.095	27.0	-0.146
4	56 % EFFLUENT	5	0.095	27.0	-0.142
5	75 % EFFLUENT	5	0.095	27.0	-0.138
6	100 % EFFLUENT	5	0.095	27.0	-0.212

APPENDIX D

Ceriodaphnia dubia Raw Data and Statistics

FISHER'S EXACT TEST

IDENTIFICATION			TOTAL ANIMALS
	ALIVE	DEAD	
CONTROL	10	0	10
32% effluent	7	3	10
TOTAL	17	3	20

CRITICAL FISHER'S VALUE (10,10,10) (p=0.05) IS 6. b VALUE IS 7.
Since b is greater than 6 there is no significant difference
between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

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

CRITICAL FISHER'S VALUE (10,10,10) (p=0.05) IS 6. b VALUE IS 9.
Since b is greater than 6 there is no significant difference
between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

IDENTIFICATION			TOTAL ANIMALS
	ALIVE	DEAD	
CONTROL	10	0	10
56% effluent	10	0	10

TOTAL	20	0	20
-------	----	---	----

CRITICAL FISHER'S VALUE (10,10,10) (p=0.05) IS 6. b VALUE IS 10.

Since b is greater than 6 there is no significant difference between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

NUMBER OF

IDENTIFICATION	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
75% effluent	8	2	10
TOTAL	18	2	20

CRITICAL FISHER'S VALUE (10,10,10) (p=0.05) IS 6. b VALUE IS 8.

Since b is greater than 6 there is no significant difference between CONTROL and TREATMENT at the 0.05 level.

FISHER'S EXACT TEST

NUMBER OF

IDENTIFICATION	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
100% effluent	8	2	10
TOTAL	18	2	20

CRITICAL FISHER'S VALUE (10,10,10) (p=0.05) IS 6. b VALUE IS 8.

Since b is greater than 6 there is no significant difference between CONTROL and TREATMENT at the 0.05 level.

SUMMARY OF FISHER'S EXACT TESTS

GROUP	IDENTIFICATION	NUMBER EXPOSED	NUMBER DEAD	SIG (P=.05)
	CONTROL	10	0	
1	32% effluent	10	3	
2	42% effluent	10	1	
3	56% effluent	10	0	
4	75% effluent	10	2	
5	100% effluent	10	2	

AA# K507556, C DUBIA REPRODUCTION, 7-21-05
File: C:\TOXSTAT\WESTON\K507556C. 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# K507556, C DUBIA REPRODUCTION, 7-21-05
File: C:\TOXSTAT\WESTON\K507556C. Transform: NO TRANSFORMATION

Bartlett's test for homogeneity of variance
Calculated B1 statistic = 8.53

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# K507556, C DUBIA REPRODUCTION, 7-21-05

FILE: C:\TOXSTAT\WESTON\K507556.C.

TRANSFORM: NO TRANSFORMATION

NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	21.0000	21.0000
1	CONTROL	2	11.0000	11.0000
1	CONTROL	3	21.0000	21.0000
1	CONTROL	4	13.0000	13.0000
1	CONTROL	5	22.0000	22.0000
1	CONTROL	6	13.0000	13.0000
1	CONTROL	7	12.0000	12.0000
1	CONTROL	8	19.0000	19.0000
1	CONTROL	9	16.0000	16.0000
1	CONTROL	10	9.0000	9.0000
2	32 % EFFLUENT	1	11.0000	11.0000
2	32 % EFFLUENT	2	0.0000	0.0000
2	32 % EFFLUENT	3	18.0000	18.0000
2	32 % EFFLUENT	4	20.0000	20.0000
2	32 % EFFLUENT	5	0.0000	0.0000
2	32 % EFFLUENT	6	13.0000	13.0000
2	32 % EFFLUENT	7	10.0000	10.0000
2	32 % EFFLUENT	8	10.0000	10.0000
2	32 % EFFLUENT	9	2.0000	2.0000
2	32 % EFFLUENT	10	14.0000	14.0000
3	42 % EFFLUENT	1	17.0000	17.0000
3	42 % EFFLUENT	2	3.0000	3.0000
3	42 % EFFLUENT	3	19.0000	19.0000
3	42 % EFFLUENT	4	11.0000	11.0000
3	42 % EFFLUENT	5	20.0000	20.0000
3	42 % EFFLUENT	6	16.0000	16.0000
3	42 % EFFLUENT	7	17.0000	17.0000
3	42 % EFFLUENT	8	13.0000	13.0000
3	42 % EFFLUENT	9	12.0000	12.0000
3	42 % EFFLUENT	10	17.0000	17.0000
4	56 % EFFLUENT	1	22.0000	22.0000
4	56 % EFFLUENT	2	18.0000	18.0000
4	56 % EFFLUENT	3	17.0000	17.0000
4	56 % EFFLUENT	4	21.0000	21.0000
4	56 % EFFLUENT	5	16.0000	16.0000
4	56 % EFFLUENT	6	20.0000	20.0000
4	56 % EFFLUENT	7	14.0000	14.0000
4	56 % EFFLUENT	8	20.0000	20.0000
4	56 % EFFLUENT	9	5.0000	5.0000
4	56 % EFFLUENT	10	9.0000	9.0000
5	75 % EFFLUENT	1	14.0000	14.0000
5	75 % EFFLUENT	2	18.0000	18.0000
5	75 % EFFLUENT	3	13.0000	13.0000
5	75 % EFFLUENT	4	18.0000	18.0000
5	75 % EFFLUENT	5	14.0000	14.0000
5	75 % EFFLUENT	6	12.0000	12.0000
5	75 % EFFLUENT	7	11.0000	11.0000
5	75 % EFFLUENT	8	19.0000	19.0000
5	75 % EFFLUENT	9	12.0000	12.0000

5	75	%	EFFLUENT	10	13.0000	13.0000
6	100	%	EFFLUENT	1	15.0000	15.0000
6	100	%	EFFLUENT	2	14.0000	14.0000
6	100	%	EFFLUENT	3	19.0000	19.0000
6	100	%	EFFLUENT	4	22.0000	22.0000
6	100	%	EFFLUENT	5	0.0000	0.0000
6	100	%	EFFLUENT	6	18.0000	18.0000
6	100	%	EFFLUENT	7	11.0000	11.0000
6	100	%	EFFLUENT	8	0.0000	0.0000
6	100	%	EFFLUENT	9	14.0000	14.0000
6	100	%	EFFLUENT	10	14.0000	14.0000

AA# K507556, C DUBIA REPRODUCTION, 7-21-05

File: C:\TOXSTAT\WESTON\K507556C.

Transform: NO TRANSFORMATION

STEEL'S MANY-ONE RANK TEST

Ho: Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	15.700				
2	32 % EFFLUENT	9.800	80.50	75.00	10.00	
3	42 % EFFLUENT	14.500	100.00	75.00	10.00	
4	56 % EFFLUENT	16.200	109.50	75.00	10.00	
5	75 % EFFLUENT	14.400	98.00	75.00	10.00	
6	100 % EFFLUENT	12.700	98.50	75.00	10.00	

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

APPENDIX E

Organism History

AQUATOX, INC.

100 Springwood Drive #15

Hot Springs, Arkansas 71913

(501) 767-9120

TEST ORGANISM HISTORY

DATE SHIPPED 7-21-05 *Arkansas Anthonotus*

SPECIES *Pimephales promelas*

QUANTITY SHIPPED 750+

AGE/LIFE STAGE 124 ms *7/21 1500ct*

BROODSTOCK SOURCE Anderson Farm, AR

CULTURE WATER Graduator

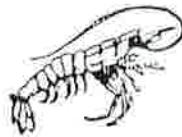
ALKALINITY (Mg/l as CaCO₃) =180

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

FEEDING Artemia

COMMENTS _____

PACKAGED BY lmc



Aquatic Research Organisms

DATA SHEET

I. Organism History

Species: Cerio daphnia dubia
Source: Lab reared Hatchery reared _____ Field collected _____
Hatch date 01/05 Receipt date _____
Lot number 02 07 05CD Strain ARO
Brood Origination EPA OH

II. Water Quality

Temperature 24 °C Salinity — ppt DO SAT
pH 7.4 Hardness ≈ 75 ppm

III. Culture Conditions

System: F₂ static renewal
Diet: Flake Food _____ Phytoplankton Trout Chow _____
Brine Shrimp _____ Rotifers _____ Other CT
Prophylactic Treatments: _____
Comments: All gravid as of 2:00pm
EOT

IV. Shipping Information

Client: Arkansas Analytical # of Organisms: 1 culture

Carrier: Fed Ex Date Shipped: 2/7/05

Biologist: [Signature]

1 - 800 - 927 - 1650

PO Box 1271 • One Lafayette Road • Hampton, NH 03842 • (603) 926-1650



Aquatic Research Organisms

DATA SHEET

I. Organism History

Species: Cerco daphnia dubia
Source: Lab reared Hatchery reared _____ Field collected _____
Hatch date 01/05 Receipt date _____
Lot number 02 07 05 CD Strain ARC
Brood Origination EPA OH

II. Water Quality

Temperature 24 °C Salinity — ppt DO SAT
pH 7.4 Hardness ≈ 75 ppm

III. Culture Conditions

System: Fu static renewal
Diet: Flake Food _____ Phytoplankton Trout Chow _____
Brine Shrimp _____ Rotifers _____ Other YCT
Prophylactic Treatments: _____
Comments: All graded as of 2:00pm
EST

IV. Shipping Information

Client: Arkansas Analytical # of Organisms: 1000
Carrier: Fed Ex Date Shipped: 2/7/05

Biologist: [Signature]

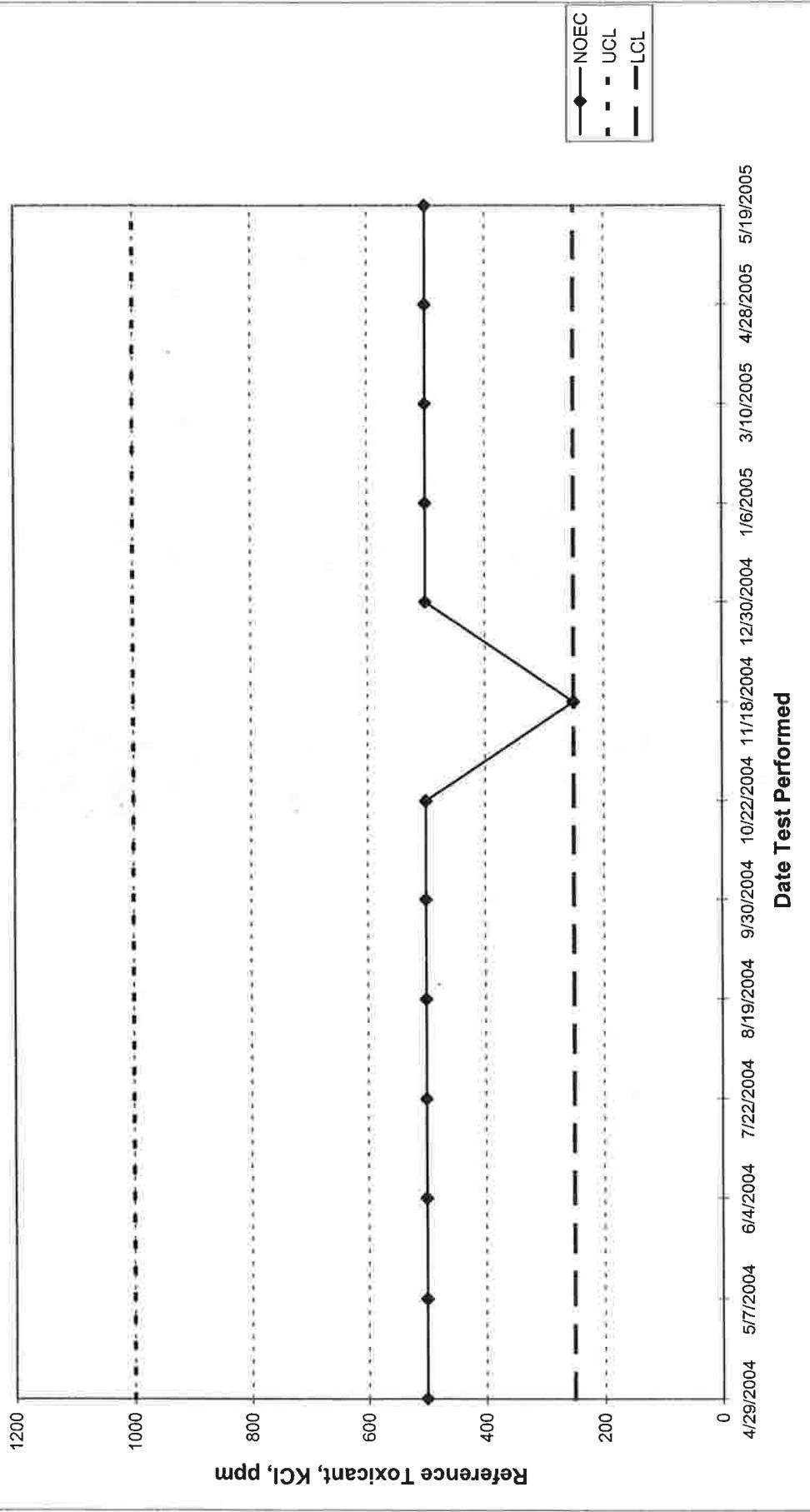
1 - 800 - 927 - 1650

PO Box 1271 • One Lafayette Road • Hampton, NH 03842 • (603) 926-1650

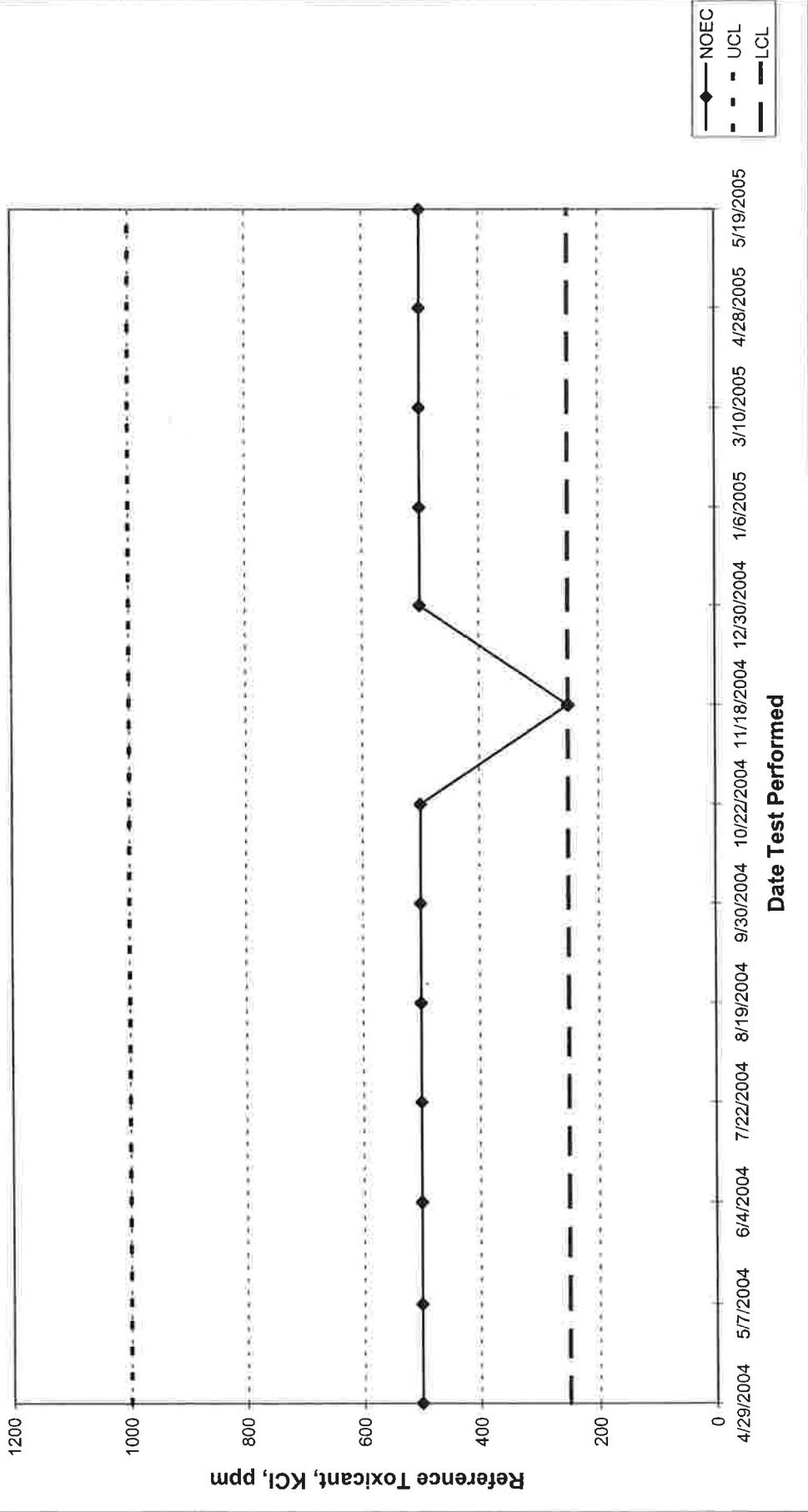
APPENDIX F

Quality Assurance Charts

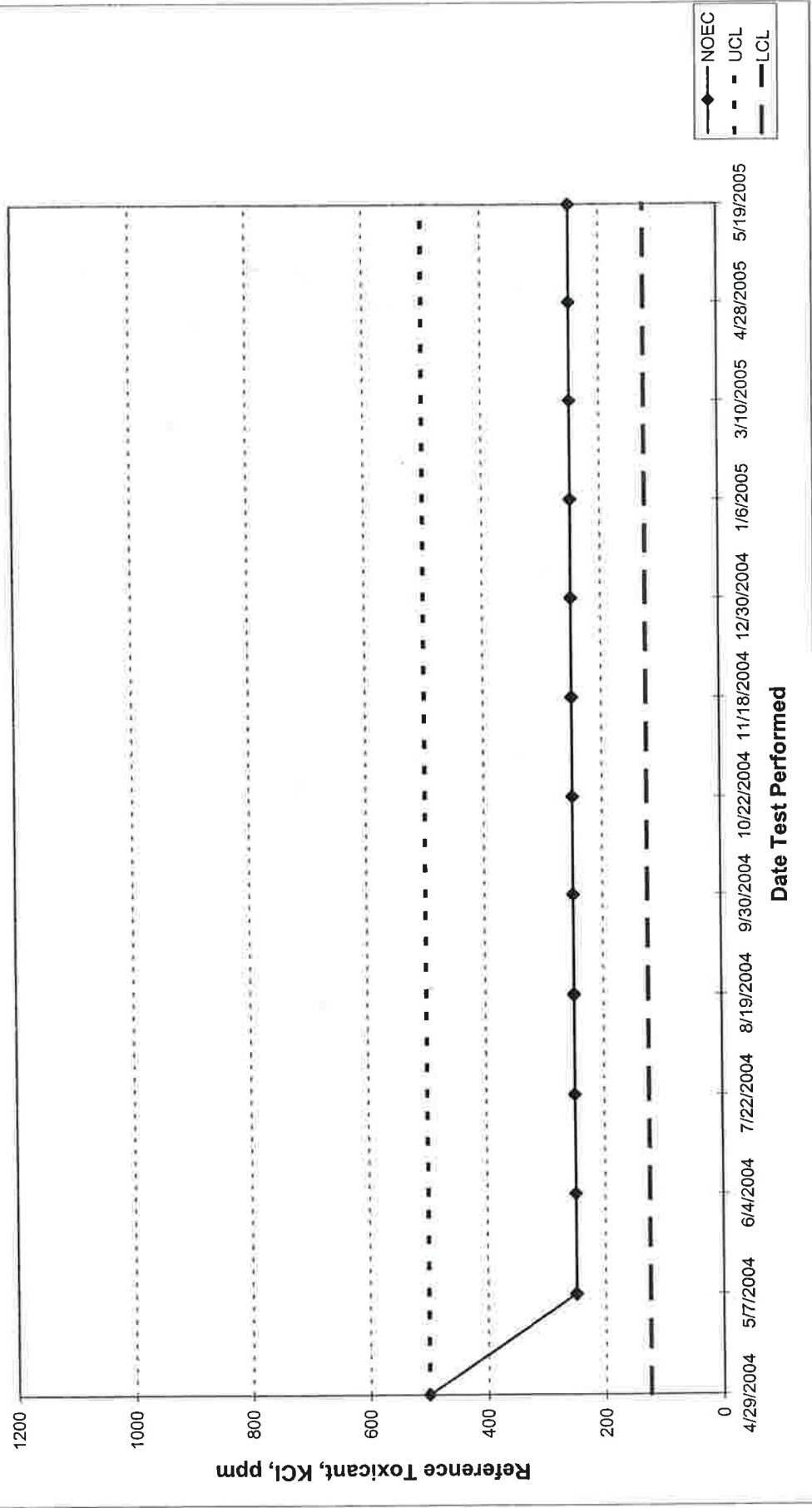
ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW SURVIVAL
QUALITY ASSURANCE



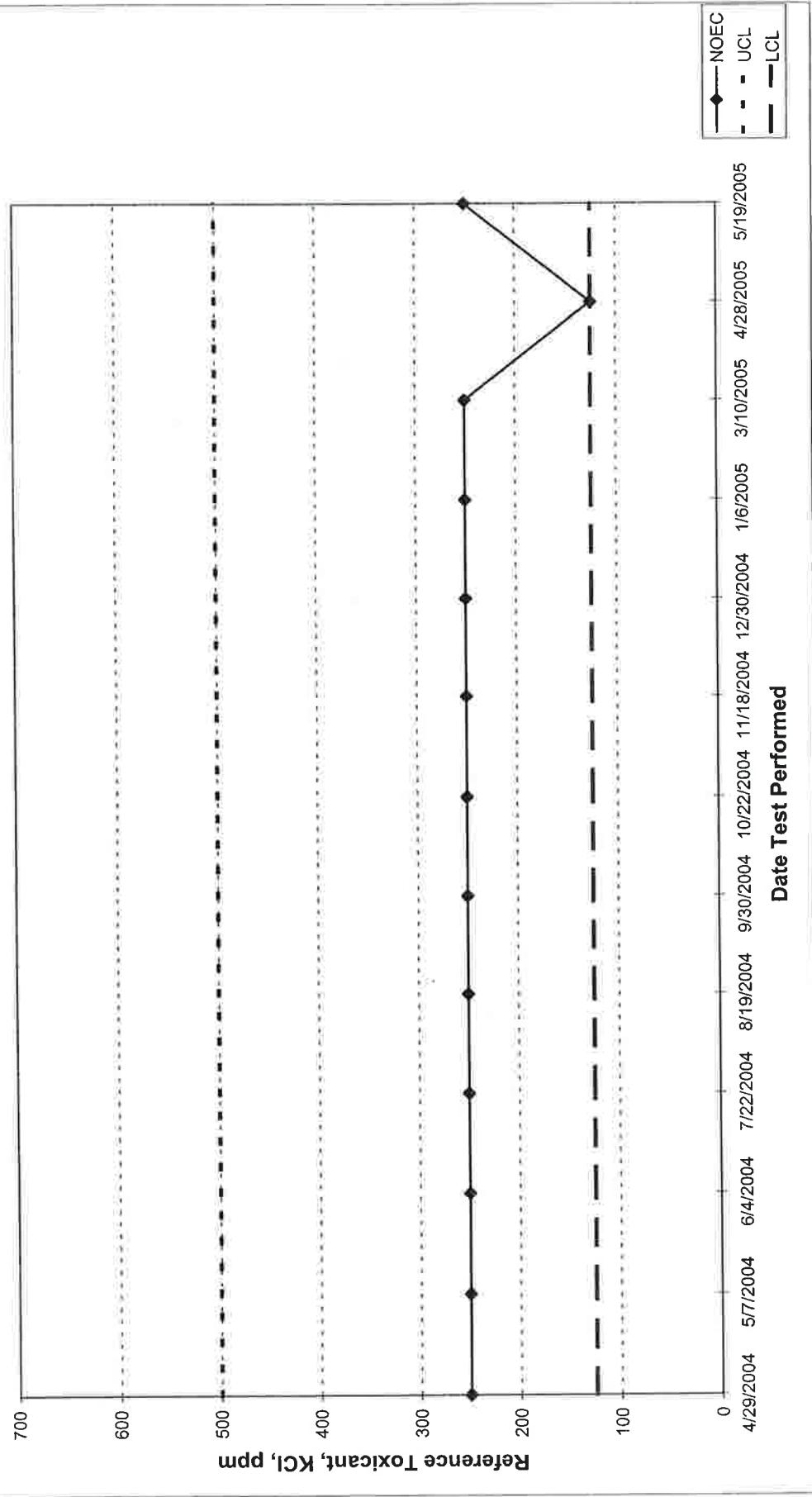
ARKANSAS ANALYTICAL, INC.
FATHEAD MINNOW GROWTH
QUALITY ASSURANCE



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

Laboratory ID: 60-1754

Certificate Number: 04-075-0

Issued Date: 30 October 2004

Expired Date: 30 October 2005

J.A. Lemanske

ADEQ Quality Assurance Officer

Date October 27, 2004