



# Arkansas Analytical, Inc.

## Toxicity Test Results

**MAGCOBAR MINE SITE**  
**NPDES PERMIT NUMBER: AR0049794**  
**August 2003**

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

*Ceriodaphnia dubia*, Survival and Reproduction Test  
Test 1002.0

Prepared for: **Mr. Alan B. Brown**  
**Weston Solutions**  
**Magcobar Mine Site**  
**2000 Darby Lane**  
**Malvern, AR 72104**

Prepared by: Arkansas Analytical, Inc.  
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**Lab Number K308578**

Tuesday, September 9, 2003



## **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 once per month for both *Ceriodaphnia dubia* and *Pimephales promelas*. The test results in this report represent the testing for August of 2003.

## **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:	8-21-03, 1000	8-22-03, 1000
Sample #2:	8-24-03, 0930	8-25-03, 0930
Sample #3:	8-25-03, 0930	8-26-03, 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	Temperature Upon Receipt (°C)
Sample #1:	8-22-03, 1534	Not Taken
Sample #2:	8-25-03, 1525	3
Sample #3:	8-26-03, 1531	5

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	25.4	X	
At least 60% of surviving females should have produced 3 broods	90%	X	
The percent coefficient of variation between replicates must be 40% or less for the young of surviving females	13.8%	X	

#### TEST ACCEPTANCE CRITERIA for *Pimephales promelas*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	100%	X	
The percent coefficient of variation between replicates must be 40% or less for survival	0%	X	
Minimum of 0.25 mg average dry weight of surviving controls	0.556	X	
The percent coefficient of variation between replicates must be 40% or less for growth	17.3%	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)	20.4	%CV survival (critical dilution)	0%
%CV Reproduction (critical dilution)	27.3%	Mean dry weight (critical dilution) in milligrams	0.693
		%CV growth (critical dilution)	14.7%

### 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, 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:	8-21-03, 1000	8-22-03, 1000
Sample #2:	8-24-03, 0930	8-25-03, 0930
Sample #3:	8-25-03, 0930	8-26-03, 0930

Test initiated (date, time): 8-22-03, 1600      Test terminated (date, time): 8-29-03, 1600

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	Mean Dry Weight	CV%
0%	0.616	0.620	0.390	0.595	0.557	0.556	17.3
32%	0.702	0.780	0.817	0.663	0.700	0.732	
42%	0.643	0.793	0.743	0.599	0.707	0.697	
56%	0.953	0.671	0.703	0.633	0.649	0.722	
75%	0.733	0.803	0.619	0.650	0.675	0.696	
100%	0.712	0.848	0.600	0.602	0.701	0.693	14.7

**DATA TABLE FOR GROWTH OF FATHEAD MINNOWS**

Effluent Conc %	Average Dry Weight in milligrams in replicate chambers					Mean Percent Survival	
	A	B	C	D	E	Mean Dry Weight	CV%
0%	0.616	0.620	0.390	0.595	0.557	0.556	17.3
32%	0.702	0.780	0.817	0.663	0.700	0.732	
42%	0.643	0.793	0.743	0.599	0.707	0.697	
56%	0.953	0.671	0.703	0.633	0.649	0.722	
75%	0.733	0.803	0.619	0.650	0.675	0.696	
100%	0.712	0.848	0.600	0.602	0.701	0.693	14.7

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)= \_\_\_\_\_   0   %



**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:	8-21-03, 1000	8-22-03, 1000
Sample #2:	8-24-03, 0930	8-25-03, 0930
Sample #3:	8-25-03, 0930	8-26-03, 0930

Test initiated (date, time): 8-22-03, 1600      Test terminated (date, time): 8-28-03, 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	28	20	24	13	21	11
B	24	30	29	24	23	23
C	25	27	32	33	23	19
D	30	27	30	25	27	17
E	28	24	23	27	24	28
F	21	33	28	23	16	X0
G	22	9	24	25	28	27
H	20	28	27	28	26	X0
I	28	X0	25	25	12	18
J	28	32	33	25	28	20
Mean	25.4	23.0	27.5	24.8	22.8	16.6
Mean/surviving female	25.4	25.6	27.5	24.8	22.8	20.4
CV%*	13.8					27.3

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	100	90	100	100	100	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)=   27.3   %




APPENDIX A

Chain of Custody Forms

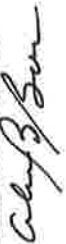



# CHAIN OF CUSTODY RECORD

CLIENT INFORMATION		Project Description		Turnaround Time		Preservation Codes:	
Weston Solutions 2006 Derby Lane Malvern, AR 72104		Reporting Information		(CIRCLE ONE) 24 hour 48 hour routine		1. Cool, 4 degrees Centigrade 2. Sulfuric Acid, pH <2 3. Nitric Acid, pH <2 4. Thio sulfate for dechlorination 5. Hydrochloric Acid for VOA 6. Sodium Hydroxide, pH >12	
Attn: Alan Brown		Telephone: FAX: Bill to P.O.		Preservative Code: Bottle Type		Bottle type code G=glass; P=HDPE W=stump; A=number	
Alan B Brown		SAMPLERS: (Printed)		SAMPLE IDENTIFICATION/ DESCRIPTION		Arkansas Analytical Lab #	
Cely B. Brown		Sample Matrix		Facility Discharge		k308578A	
Field Number	Sample Collection Date/s	Time/s	Grab	Comp	# of Containers	TEST PARAMETERS	
FD0822604	6/22	10:00	X	5	1. Cool, 4 degrees Centigrade 2. Sulfuric Acid, pH <2 3. Nitric Acid, pH <2 4. Thio sulfate for dechlorination 5. Hydrochloric Acid for VOA 6. Sodium Hydroxide, pH >12		
1. Relinquished by: (Signature)		Date/Time		1. Received by: (Signature)		REMARKS	
Cely B. Brown		8/22/03 15:34		[Signature]		Condition of samples: A. Containers Correct? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO B. Preservation Correct? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO C. Seals Intact? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO	
2. Relinquished by: (Signature)		Date/Time		2. Received by: (Signature)			
[Signature]		8/22/03 1534		Barney			

# CHAIN OF CUSTODY RECORD

CLIENT INFORMATION				Project Description		Turnaround Time		Preservation Codes:					
Weston Solutions, Inc. P.O. Box 699 2000 Derby Lane Malvern, AR 72104 Attn: Alan Brown				MAGCOBAR Mine Site Reporting Information Telephone: 501/467-8355 FAX: 501/467-8687 Bill to P.O.		(CIRCLE ONE) 24 hour 48 hour routine		1. Cool, 4 degrees Centigrade 2. Sulfuric Acid, pH <2 3. Nitric Acid, pH <2	4. Thiocyanate for dechlorination 5. Hydrochloric Acid for VO4 6. Sodium Hydroxide, pH >12				
 Alan B. Brown				Samplers: (Printed) Sample Matrix Facility Discharge		Bottle Type P		<b>TEST PARAMETERS</b> Bottle type code G=glass; P=HDPE V=septum; A=amber			Arkansas Analytical Lab # K308578B		
Field Number	Sample Collection Date/s	Time/s	Grab	Comp	# of Containers	IDENTIFICATION/ DESCRIPTION							
FD0825COMP	25-Aug	9:30	X		3	Chronic Bio							

1. Relinquished by: (Signature)		1. Received by: (Signature)		For completion by laboratory		REMARKS	
				Condition of samples: YES <input checked="" type="checkbox"/> NO <input type="checkbox"/> A. Containers Correct? <input checked="" type="checkbox"/> B. Preservation Correct? <input checked="" type="checkbox"/> C. Seals Intact? <input type="checkbox"/> N/A			Temp - 30C
							





## APPENDIX B

### Effluent and Dilution Water Data



CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING

Fathead Minnow

Lab # / Sample ID		K308578							Test Start (Date/Time)		8-22-03/1600	
Client		Weston							Test End (Date/Time)		8-29-03/1600	
		Day of Test										
		1	2	3	4	5	6	7	notes/remarks			
<b>Control</b>		8/22	8-23	8-24	8-25	8/26	8/27	8/28	8/22	SS 69		
D.O (mg/L)	INITIAL	7.3	9.1	9.0	8.6	7.4	7.9	7.5	8/26	SS 10		
	FINAL	6.9	8.5	7.0	6.8	6.7	6.8	6.9				
pH(mg/L)	INITIAL	7.2	7.4	7.7	6.1	6.6	6.7	6.5				
	FINAL	6.6	6.9	6.1	6.1	6.0	7.7	7.2				
temp(C)	INITIAL	23.8	21.3	20.9	21.3	21.0	21.7	21.3				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
ALKALINITY(mg/L)		28				22						
HARDNESS(mg/L)		82				39						
CONDUCTIVITY(umhos/cm)		160				153						
CHLORINE(mg/L)		<0.05				40.05						
<b>CONC:</b>		32%	32%	32%	32%	32%	32%	32%				
D.O (mg/L)	INITIAL	7.5	9.8	9.3	7.7	7.1	7.9	7.5				
	FINAL	6.9	8.3	7.1	6.8	6.7	6.7	7.0				
pH(mg/L)	INITIAL	7.1	7.2	7.4	6.5	6.4	6.8	6.7				
	FINAL	6.5	6.8	6.0	6.0	6.0	7.4	7.1				
temp(C)	INITIAL	23.6	21.6	20.9	22.5	21.0	22.0	22.4				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		42%	42%	42%	42%	42%	42%	42%				
D.O (mg/L)	INITIAL	7.3	9.8	9.1	7.6	7.5	7.9	7.5				
	FINAL	6.9	8.4	7.0	6.9	6.8	6.7	7.0				
pH(mg/L)	INITIAL	7.2	7.2	7.4	6.6	6.4	6.9	6.8				
	FINAL	6.5	6.8	6.0	6.0	6.0	7.3	7.1				
temp(C)	INITIAL	23.5	21.6	20.9	22.9	22.0	22.2	23.2				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		56%	56%	56%	56%	56%	56%	56%				
D.O (mg/L)	INITIAL	7.3	9.9	9.2	7.5	7.6	8.0	7.4				
	FINAL	6.9	8.8	7.0	6.9	6.8	6.7	7.0				
pH(mg/L)	INITIAL	7.2	7.1	7.4	6.7	6.4	6.9	6.9				
	FINAL	6.5	6.8	6.0	6.1	6.1	7.3	7.0				
temp(C)	INITIAL	23.5	21.5	20.9	23.7	21.9	22.5	24.3				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		75%	75%	75%	75%	75%	75%	75%				
D.O (mg/L)	INITIAL	7.5	10.0	9.3	7.4	7.6	8.0	7.5				
	FINAL	7.0	8.9	7.0	7.0	6.8	6.8	6.9				
pH(mg/L)	INITIAL	7.2	7.1	7.3	6.8	6.4	6.9	7.0				
	FINAL	6.5	6.8	6.0	6.1	6.1	7.3	7.0				
temp(C)	INITIAL	23.5	21.2	21.1	24.7	21.9	23.4	25.2				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		100%	100%	100%	100%	100%	100%	100%				
D.O (mg/L)	INITIAL	7.3	9.6	9.4	7.2	7.7	8.6	7.4				
	FINAL	7.0	8.7	7.0	7.1	6.8	6.9	6.9				
pH(mg/L)	INITIAL	7.3	7.2	7.2	7.0	6.4	7.0	7.1				
	FINAL	6.4	6.7	5.9	6.0	6.1	7.2	7.0				
temp(C)	INITIAL	24.0	22.0	21.1	26.3	21.9	24.3	26.8				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC: 100%</b>		A	A	A	B	B	C	C				
ALKALINITY(mg/L)		11			12		13					
HARDNESS(mg/L)		1146			1368		1356					
CONDUCTIVITY(umhos/cm)		2220			2250		2230					
CHLORINE(mg/L)		<0.05			<0.05		<0.05					



CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING

Ceriodaphnia dubia

Lab # / Sample ID		K308578							Test Start (Date/Time)	8-22-03 / 11:00	
Client		Weston							Test End (Date/Time)	8-28-03 / 0920	
		Day of Test									
		1	2	3	4	5	6	7	8	notes/remarks	
<b>Control</b>		8-22	8-23	8-24	8-25	8/26	8/27			8-22 SS#69	
D.O (mg/L)	INITIAL	7.3	9.1	7.0	8.0	7.4	7.9			8-26 SS#70	
	FINAL	7.0	9.3	8.1	8.2	7.7	8.3				
pH	INITIAL	7.2	7.4	7.7	6.1	6.6	6.7				
	FINAL	7.5	7.4	7.1	6.8	6.4	7.9				
temp(C)	INITIAL	28.8	21.3	20.9	21.3	21.0	21.7				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0				
ALKALINITY(mg/L)		28				22					
HARDNESS(mg/L)		82				39					
CONDUCTIVITY(umhos/cm)		160				153					
CHLORINE(mg/L)		<0.05			<del>40.05</del>	40.05					
<b>CONC:</b>		32%	32%	32%	32%	32%	32%				
D.O (mg/L)	INITIAL	7.5	9.8	9.3	7.7	7.7	7.9				
	FINAL	7.0	9.2	8.1	8.3	7.7	8.3				
pH	INITIAL	7.8	7.2	7.4	6.5	6.4	6.8				
	FINAL	7.5	7.3	7.1	6.8	6.3	8.1				
temp(C)	INITIAL	23.6	21.6	20.9	22.5	21.0	22.0				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		42%	42%	42%	42%	42%	42%				
D.O (mg/L)	INITIAL	7.3	9.8	9.1	7.6	7.5	7.9				
	FINAL	7.0	9.2	8.1	8.3	7.8	8.6				
pH	INITIAL	7.2	7.2	7.4	6.6	6.4	6.9				
	FINAL	7.5	7.4	7.0	6.8	6.3	7.9				
temp(C)	INITIAL	23.5	21.6	20.9	22.9	22.0	22.2				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		56%	56%	56%	56%	56%	56%				
D.O (mg/L)	INITIAL	7.3	9.9	9.2	7.5	7.6	8.0				
	FINAL	7.0	9.2	8.1	8.4	7.9	8.6				
pH	INITIAL	7.2	7.1	7.4	6.7	6.4	6.9				
	FINAL	7.4	7.4	7.0	6.7	6.2	7.8				
temp(C)	INITIAL	23.5	21.5	20.9	23.7	21.9	22.5				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		75%	75%	75%	75%	75%	75%				
D.O (mg/L)	INITIAL	7.5	10.0	9.3	7.4	7.6	8.0				
	FINAL	7.1	9.3	8.1	8.4	8.0	8.7				
pH	INITIAL	7.2	7.1	7.3	6.8	6.4	6.9				
	FINAL	7.4	7.4	7.0	6.7	6.2	7.8				
temp(C)	INITIAL	23.5	21.2	21.1	24.7	21.9	23.4				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>		100%	100%	100%	100%	100%	100%				
D.O (mg/L)	INITIAL	7.3	9.6	9.4	7.2	7.7	8.0				
	FINAL	7.1	9.3	8.2	8.4	8.0	8.5				
pH	INITIAL	7.3	7.2	7.2	7.0	6.4	7.0				
	FINAL	7.3	7.2	6.9	6.7	6.1	7.5				
temp(C)	INITIAL	24.0	22.0	21.1	24.3	21.9	24.3				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0				
<b>CONC:</b>	100%	A	A	A	B	B	C				
ALKALINITY(mg/L)		11			12		13				
HARDNESS(mg/L)		1146			1368		1356				
CONDUCTIVITY(umhos/cm)		2220			2250		2230				
CHLORINE(mg/L)		40.05			40.05		40.05				



## APPENDIX C

### Fathead Minnow Raw Data and Statistics

**SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST**

LAB #/ SAMPLE ID K308578 TEST START DATE 8-22-05 TIME 1600  
 CLIENT Weston TEST END DATE 8-29 TIME 1600  
 AGE AND SOURCE OF MINNOWS 224 hrs; Aquatex

CONC:	REP #	DAY (NUMBER SURVIVING)								SURVIVAL		
		start	1	2	3	4	5	6	7	%	MEAN %	CV
Control	A	10	10	10	10	10	10	10	10	100	100%	0%
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
32%	A	10	10	10	10	10	10	10	10	100	100%	
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
42%	A	10	10	10	10	10	10	10	10	100	98%	
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	9	9	9	90		
	E	10	10	10	10	10	10	10	10	100		
50%	A	10	10	10	10	10	10	10	10	100	100%	
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
75%	A	10	10	10	10	10	10	10	10	100	96%	
	B	10	10	10	10	10	9	9	9	90		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	9	9	9	90		
	E	10	10	10	10	10	10	10	10	100		
100%	A	10	10	10	10	10	10	10	10	100	100%	0%
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
ANALYST:		TC	AD	AD	TC	TC	TC	TC	mg			
DATE:		8-22	8-23	8-24	8-25	8-26	8-27	8-28	8-29			
TIME:		1600	1015	1020	1635	1415	1530	1620	1600			

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

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WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB #/S: <u>K508578</u>	TEST DATES (BEGIN/END): <u>8-21-03/8-28-03</u>
CLIENT: <u>Weston</u>	WEIGHING DATE/TIME:
ANALYST/S: <u>AF, AD, TC, MWJ</u>	DRYING TEMPERATURE (DEGREES C): <u>60°C</u>
SAMPLE ID:	DRYING TIME (HOURS): <u>24 hrs.</u>

	REP #	FINAL DRY WEIGHT TIN+LARVAE (g)	INITIAL WEIGHT TIN (g)	TOTAL DRY WEIGHT OF LARVAE (g)	NUMBER OF LARVAE	DRY WEIGHT OF LARVA (mg)	REMARKS
CONTROL	A65	0.98198	0.97582	0.00616	10	0.616	AVG DRY
	B66	0.97571	0.96951	0.00620	10	0.620	WEIGHT (mg)
	C67	0.97899	0.97509	0.00390	10	0.390	0.556
	D68	0.98555	0.97940	0.00595	10	0.595	CV
	E69	0.97805	0.97248	0.00557	10	0.557	17.3
CONC: 32%	A70	0.98806	0.98104	0.00702	10	0.702	AVG DRY
	B71	0.98162	0.97382	0.00780	10	0.780	WEIGHT(MG)
	C72	0.98221	0.97403	0.00817	10	0.817	0.452
	D73	0.98515	0.97852	0.00663	10	0.663	CV
	E74	0.98438	0.97738	0.00700	10	0.700	0.752
CONC: 42%	A75	0.98242	0.97599	0.00643	10	0.643	AVG DRY
	B76	0.98275	0.97482	0.00793	10	0.793	WEIGHT(MG)
	C77	0.98815	0.98072	0.00743	10	0.743	0.697
	D78	0.98198	0.97599	0.00599	10	0.599	CV
	E79	0.98265	0.97558	0.00707	10	0.707	
CONC: 56%	A80	0.98472	0.97519	0.00953	10	0.953	AVG DRY
	B81	0.98513	0.97842	0.00671	10	0.671	WEIGHT(MG)
	C82	0.98715	0.98012	0.00703	10	0.703	0.722
	D83	0.98926	0.98293	0.00633	10	0.633	CV
	E84	0.98827	0.98178	0.00649	10	0.649	
CONC: 75%	A85	0.987327	0.98004	0.00733	10	0.733	AVG DRY
	B86	0.98550	0.97747	0.00803	10	0.803	WEIGHT(MG)
	C87	0.97854	0.97235	0.00619	10	0.619	0.696
	D88	0.98965	0.98315	0.00650	10	0.650	CV
	E89	0.98923	0.98248	0.00675	10	0.675	
CONC: 100%	A90	0.99067	0.98355	0.00712	10	0.712	AVG DRY
	B91	0.98216	0.97368	0.00848	10	0.848	WEIGHT(MG)
	C92	0.97900	0.97300	0.00600	10	0.600	0.693
	D93	0.97708	0.97106	0.00602	10	0.602	CV
	E94	0.97894	0.97193	0.00701	10	0.701	14.69

CV = (STANDARD DEVIATION/MEAN)\*100

AA# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03  
File: K308578S Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro = Wilk's test for normality

---

D = 0.053

W = 0.714

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# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03  
File: K308578S 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# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03

FILE: K308578S

TRANSFORM: ARC SINE(SQUARE ROOT(Y))

NUMBER OF GROUPS: 6

---

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	1.0000	1.4120
1	CONTROL	2	1.0000	1.4120
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	1.0000	1.4120
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	0.9000	1.2490
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	1.0000	1.4120
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	0.9000	1.2490
5	75 % EFFLUENT	3	1.0000	1.4120
5	75 % EFFLUENT	4	0.9000	1.2490
5	75 % EFFLUENT	5	1.0000	1.4120
6	100 % EFFLUENT	1	1.0000	1.4120
6	100 % EFFLUENT	2	1.0000	1.4120
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# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03  
File: K308578S 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	1.412				
2	32 % EFFLUENT	1.412	27.50	16.00	5.00	
3	42 % EFFLUENT	1.379	25.00	16.00	5.00	
4	56 % EFFLUENT	1.412	27.50	16.00	5.00	
5	75 % EFFLUENT	1.347	22.50	16.00	5.00	
6	100 % EFFLUENT	1.412	27.50	16.00	5.00	

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

AA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
File: K308578G Transform: NO TRANSFORMATION

Shapiro - Wilk's test for normality

---

D = 0.209

W = 0.971

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 # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
File: K308578G Transform: NO TRANSFORMATION

---

Bartlett's test for homogeneity of variance

Calculated B1 statistic = 2.62

---

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 # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
FILE: K308578G  
TRANSFORM: NO TRANSFORMATION

NUMBER OF GROUPS: 6

---

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.6160	0.6160
1	CONTROL	2	0.6200	0.6200
1	CONTROL	3	0.3900	0.3900
1	CONTROL	4	0.5950	0.5950
1	CONTROL	5	0.5570	0.5570
2	32 % EFFLUENT	1	0.7020	0.7020
2	32 % EFFLUENT	2	0.7800	0.7800
2	32 % EFFLUENT	3	0.8170	0.8170
2	32 % EFFLUENT	4	0.6630	0.6630
2	32 % EFFLUENT	5	0.7000	0.7000
3	42 % EFFLUENT	1	0.6430	0.6430
3	42 % EFFLUENT	2	0.7930	0.7930
3	42 % EFFLUENT	3	0.7430	0.7430
3	42 % EFFLUENT	4	0.5990	0.5990
3	42 % EFFLUENT	5	0.7070	0.7070
4	56 % EFFLUENT	1	0.9530	0.9530
4	56 % EFFLUENT	2	0.6710	0.6710
4	56 % EFFLUENT	3	0.7030	0.7030
4	56 % EFFLUENT	4	0.6330	0.6330
4	56 % EFFLUENT	5	0.6490	0.6490
5	75 % EFFLUENT	1	0.7330	0.7330
5	75 % EFFLUENT	2	0.8030	0.8030
5	75 % EFFLUENT	3	0.6190	0.6190
5	75 % EFFLUENT	4	0.6500	0.6500
5	75 % EFFLUENT	5	0.6750	0.6750
6	100 % EFFLUENT	1	0.7120	0.7120
6	100 % EFFLUENT	2	0.8480	0.8480
6	100 % EFFLUENT	3	0.6000	0.6000
6	100 % EFFLUENT	4	0.6020	0.6020
6	100 % EFFLUENT	5	0.7010	0.7010

---

AA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
File: K308578G Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.103	0.021	2.367
Within (Error)	24	0.209	0.009	
Total	29	0.312		

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

AA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
 File: K308578G 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	0.556	0.556		
2	32 % EFFLUENT	0.732	0.732	-2.994	
3	42 % EFFLUENT	0.697	0.697	-2.395	
4	56 % EFFLUENT	0.722	0.722	-2.815	
5	75 % EFFLUENT	0.696	0.696	-2.378	
6	100 % EFFLUENT	0.693	0.693	-2.320	

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

AA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
 File: K308578G 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	5			
2	32 % EFFLUENT	5	0.139	25.1	-0.177
3	42 % EFFLUENT	5	0.139	25.1	-0.141
4	56 % EFFLUENT	5	0.139	25.1	-0.166
5	75 % EFFLUENT	5	0.139	25.1	-0.140
6	100 % EFFLUENT	5	0.139	25.1	-0.137



APPENDIX D

*Ceriodaphnia dubia* Raw Data and Statistics

*Ceriodaphnia dubia*

**SURVIVAL AND REPRODUCTION TEST**

Discharger: Wiston Analyst: MG AD, TC  
 Location: K308578 Test Start-Date/Time: 8-27-03/1600  
 Date Sample Collected: See CDC Test Stop-Date/Time: 8-28-03/0920

Conc 1	Replicate													No. of Young Adults	No. of Young Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day 1	0	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
Day 2	0	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
Day 3	5	0	4	5	0	3	6	6	6	6	6	6	6	40	10	TC
Day 4	0	5	0	0	1	5	8	0	10	0	0	0	0	29	10	TC
Day 5	10	9	10	12	10	8	0	8	0	11	7	8	10	78	10	TC
Day 6	13	10	11	13	12	8	11	6	12	11	10	7	10	107	10	TC
Day 7																
Day 8																
Total	28	24	25	30	28	21	22	20	28	28	28	28	28	254		
% <u>Control</u> 90% 3rd brood $\bar{X} = 25.4$ CV = 13.8%																

Conc 2	Replicate													No. of Young Adults	No. of Young Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day 1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10	AD
Day 2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10	AD
Day 3	4	7	6	0	0	0	0	5	0	5	5	5	5	27	9	TC
Day 4	0	0	0	5	5	7	3	0	0	0	0	0	0	20	9	TC
Day 5	8	12	9	9	9	11	5	10	0	12	8	5	9	85	9	TC
Day 6	8	11	12	13	10	15	1	13	0	15	11	11	9	98	9	TC
Day 7																
Day 8																
Total	20	30	27	27	24	33	9	28	0	32	28	28	28	230		
% <u>32</u>																

Conc 3	Replicate													No. of Young Adults	No. of Young Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day 1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10	AD
Day 2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10	AD
Day 3	4	6	7	6	0	0	0	6	5	6	6	6	6	40	10	TC
Day 4	0	0	0	0	5	5	9	0	0	0	0	0	0	16	10	TC
Day 5	7	11	11	12	8	9	9	10	10	13	10	10	10	100	10	TC
Day 6	13	12	14	12	10	14	9	11	10	14	11	10	11	119	10	TC
Day 7																
Day 8																
Total	24	29	32	30	23	28	24	27	25	33	27	25	27	275		
% <u>100</u>																

X=DEAD; Y=MALE  
 $\bar{X} = 20.4$  CV = 27.3%

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
32% 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	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
42% 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

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
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	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
100%	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	1	
2	42% EFFLUENT	10	0	
3	56% EFFLUENT	10	0	
4	75% EFFLUENT	10	0	
5	100%	10	2	



AA # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
File: k308578c 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 # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
File: k308578c Transform: NO TRANSFORMATION

---

Bartlett's test for homogeneity of variance  
Calculated B1 statistic = 21.95

---

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

Data FAIL B1 homogeneity test at 0.01 level. Try another transformation.

TITLE: AA # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
 FILE: k308578c  
 TRANSFORM: NO TRANSFORMATION

NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	28.0000	28.0000
1	CONTROL	2	24.0000	24.0000
1	CONTROL	3	25.0000	25.0000
1	CONTROL	4	30.0000	30.0000
1	CONTROL	5	28.0000	28.0000
1	CONTROL	6	21.0000	21.0000
1	CONTROL	7	22.0000	22.0000
1	CONTROL	8	20.0000	20.0000
1	CONTROL	9	28.0000	28.0000
1	CONTROL	10	28.0000	28.0000
2	32 % EFFLUENT	1	20.0000	20.0000
2	32 % EFFLUENT	2	30.0000	30.0000
2	32 % EFFLUENT	3	27.0000	27.0000
2	32 % EFFLUENT	4	27.0000	27.0000
2	32 % EFFLUENT	5	24.0000	24.0000
2	32 % EFFLUENT	6	33.0000	33.0000
2	32 % EFFLUENT	7	9.0000	9.0000
2	32 % EFFLUENT	8	28.0000	28.0000
2	32 % EFFLUENT	9	0.0000	0.0000
2	32 % EFFLUENT	10	32.0000	32.0000
3	42 % EFFLUENT	1	24.0000	24.0000
3	42 % EFFLUENT	2	29.0000	29.0000
3	42 % EFFLUENT	3	32.0000	32.0000
3	42 % EFFLUENT	4	30.0000	30.0000
3	42 % EFFLUENT	5	23.0000	23.0000
3	42 % EFFLUENT	6	28.0000	28.0000
3	42 % EFFLUENT	7	24.0000	24.0000
3	42 % EFFLUENT	8	27.0000	27.0000
3	42 % EFFLUENT	9	25.0000	25.0000
3	42 % EFFLUENT	10	33.0000	33.0000
4	56 % EFFLUENT	1	13.0000	13.0000
4	56 % EFFLUENT	2	24.0000	24.0000
4	56 % EFFLUENT	3	33.0000	33.0000
4	56 % EFFLUENT	4	25.0000	25.0000
4	56 % EFFLUENT	5	27.0000	27.0000
4	56 % EFFLUENT	6	23.0000	23.0000
4	56 % EFFLUENT	7	25.0000	25.0000
4	56 % EFFLUENT	8	28.0000	28.0000
4	56 % EFFLUENT	9	25.0000	25.0000
4	56 % EFFLUENT	10	25.0000	25.0000
5	75 % EFFLUENT	1	21.0000	21.0000
5	75 % EFFLUENT	2	23.0000	23.0000
5	75 % EFFLUENT	3	23.0000	23.0000
5	75 % EFFLUENT	4	27.0000	27.0000
5	75 % EFFLUENT	5	24.0000	24.0000
5	75 % EFFLUENT	6	16.0000	16.0000
5	75 % EFFLUENT	7	28.0000	28.0000
5	75 % EFFLUENT	8	26.0000	26.0000
5	75 % EFFLUENT	9	12.0000	12.0000
5	75 % EFFLUENT	10	28.0000	28.0000

6	100 %	EFFLUENT	1	31.0000	31.0000
6	100 %	EFFLUENT	2	23.0000	23.0000
6	100 %	EFFLUENT	3	19.0000	19.0000
6	100 %	EFFLUENT	4	17.0000	17.0000
6	100 %	EFFLUENT	5	28.0000	28.0000
6	100 %	EFFLUENT	6	0.0000	0.0000
6	100 %	EFFLUENT	7	27.0000	27.0000
6	100 %	EFFLUENT	8	0.0000	0.0000
6	100 %	EFFLUENT	9	18.0000	18.0000
6	100 %	EFFLUENT	10	20.0000	20.0000

---

AA # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
File: k308578c Transform: NO TRANSFORMATION

STEEL'S MANY-ONE RANK TEST

- Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	25.400				
2	32 % EFFLUENT	23.000	105.50	75.00	10.00	
3	42 % EFFLUENT	27.500	120.00	75.00	10.00	
4	56 % EFFLUENT	24.800	101.50	75.00	10.00	
5	75 % EFFLUENT	22.800	90.00	75.00	10.00	
6	100 % EFFLUENT	18.300	80.50	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 8-21-03 Arkansas Analytical

SPECIES Pimephales promelas

QUANTITY SHIPPED 900+

AGE/LIFE STAGE 44 hrs 8/21 1500 CST

BROODSTOCK SOURCE Anderson Farms, AR

CULTURE WATER groundwater

ALKALINITY (Mg/l as CaCO<sub>3</sub>) = 180

HARDNESS (Mg/l as CaCO<sub>3</sub>)/Salinity (ppt) = 160

FEEDING A13min

COMMENTS \_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_

PACKAGED BY WU

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: 1/17/01

SPECIES: Ceriodaphnia dubia

AGE: Variable

LIFE STAGE: Adult

HATCH DATE: Variable

BEGAN FEEDING: Immediately

FOOD: YTC, Selenastrum

### Water Chemistry Record:

	Mean	Range
TEMPERATURE:	<u>24 °C</u>	<u>21-24°C</u>
SALINITY/CONDUCTIVITY:	<u>--</u>	<u>--</u>
TOTAL HARDNESS (as CaCO <sub>3</sub> ):	<u>112 mg/l</u>	<u>90-124 mg/l</u>
TOTAL ALKALINITY (as CaCO <sub>3</sub> ):	<u>85 mg/l</u>	<u>50-85 mg/l</u>
pH:	<u>8.09</u>	<u>7.68-8.14</u>

### Comments:

  
\_\_\_\_\_  
*Facility Supervisor*

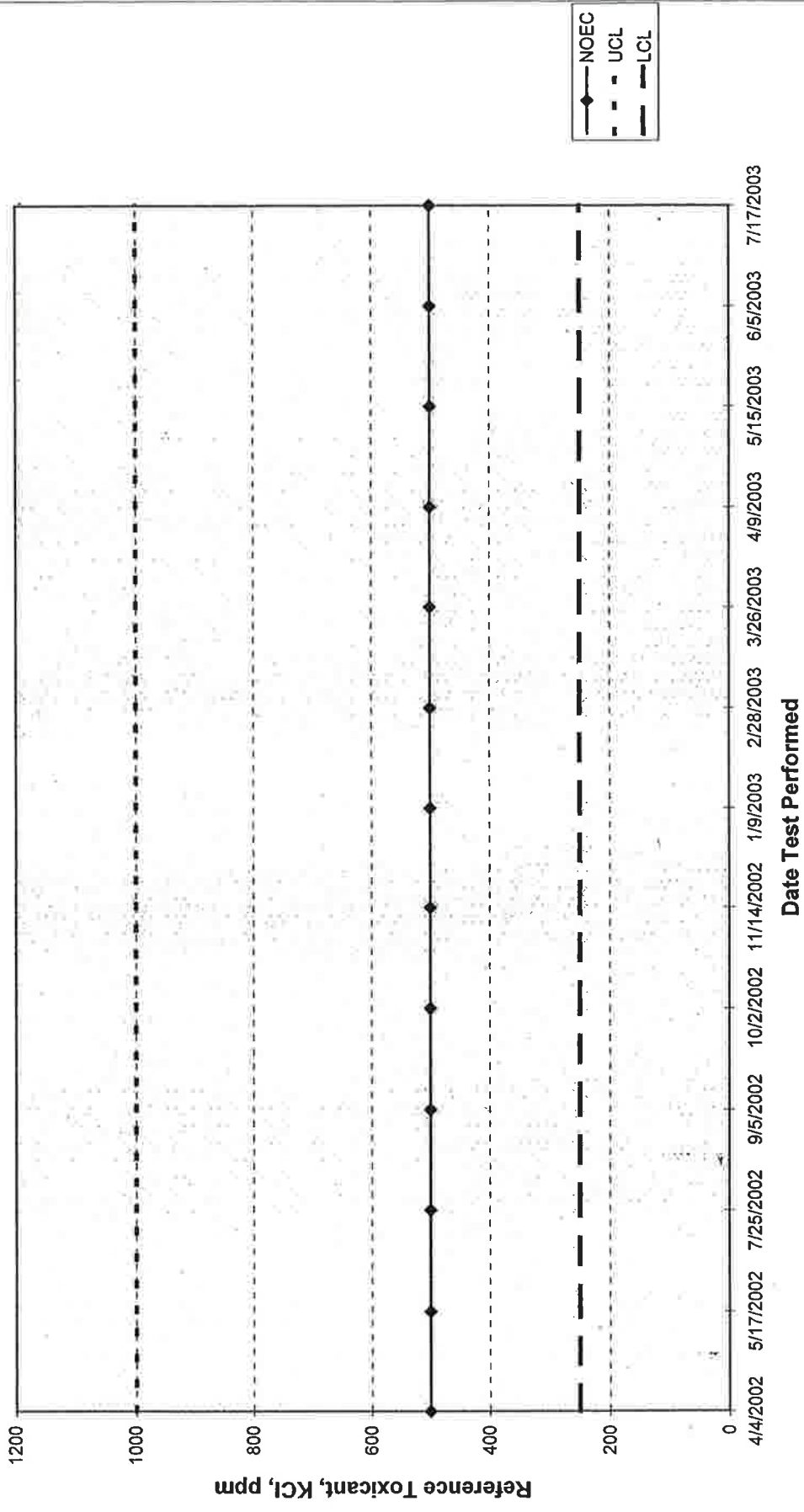


## APPENDIX F

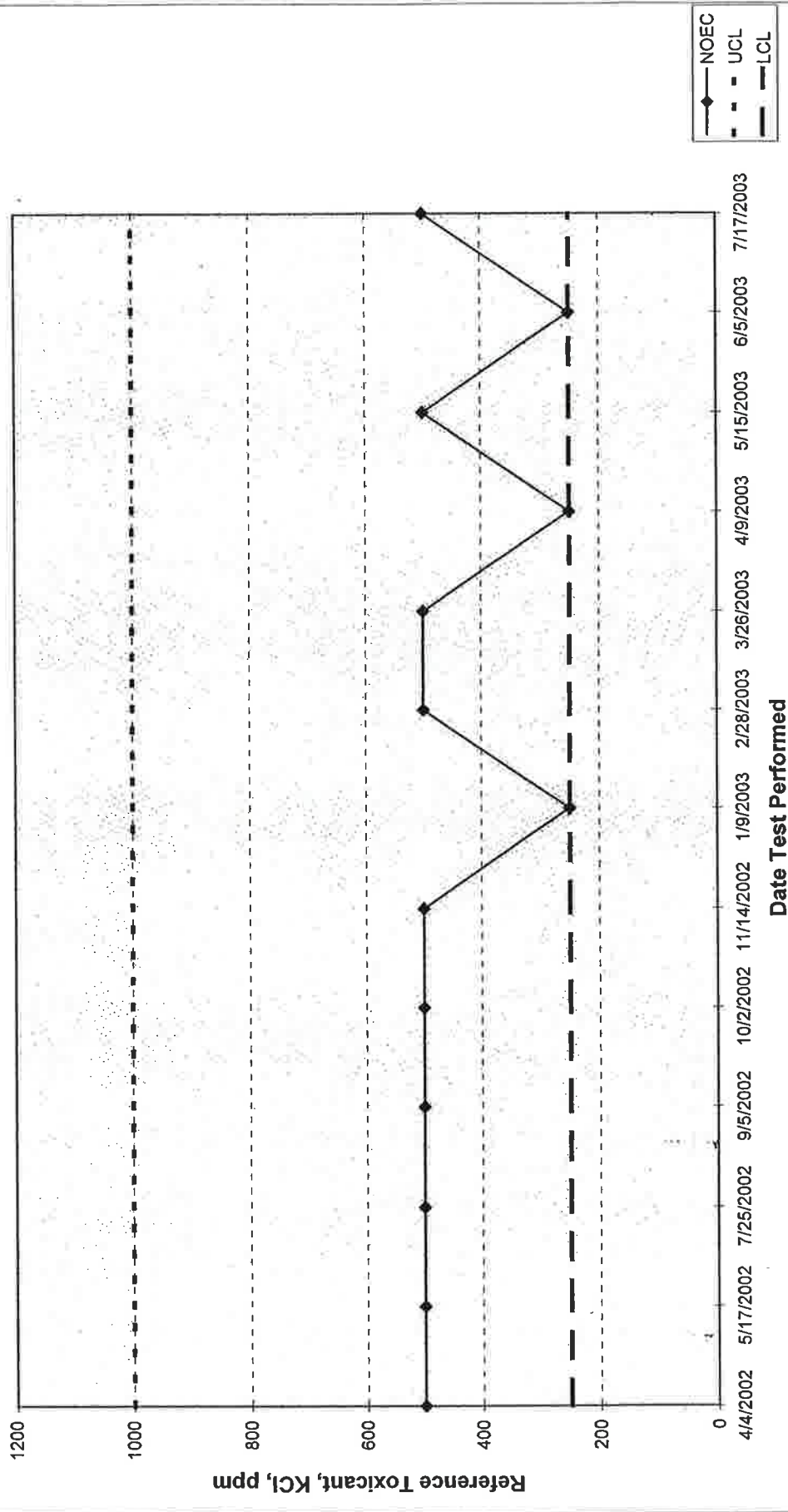
### Quality Assurance Charts



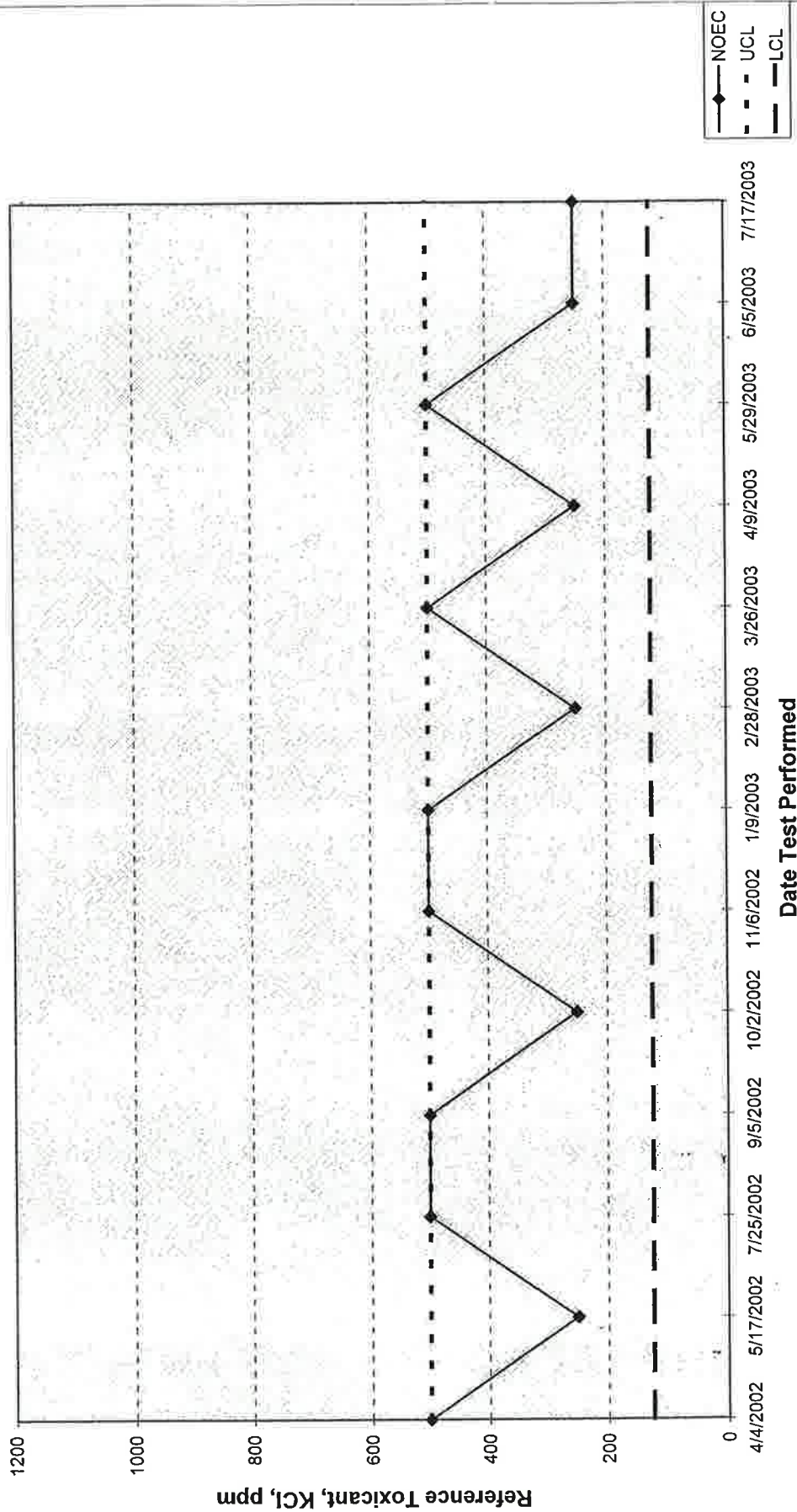
**ARKANSAS ANALYTICAL, INC.**  
**FATHEAD MINNOW SURVIVAL**  
**QUALITY ASSURANCE**



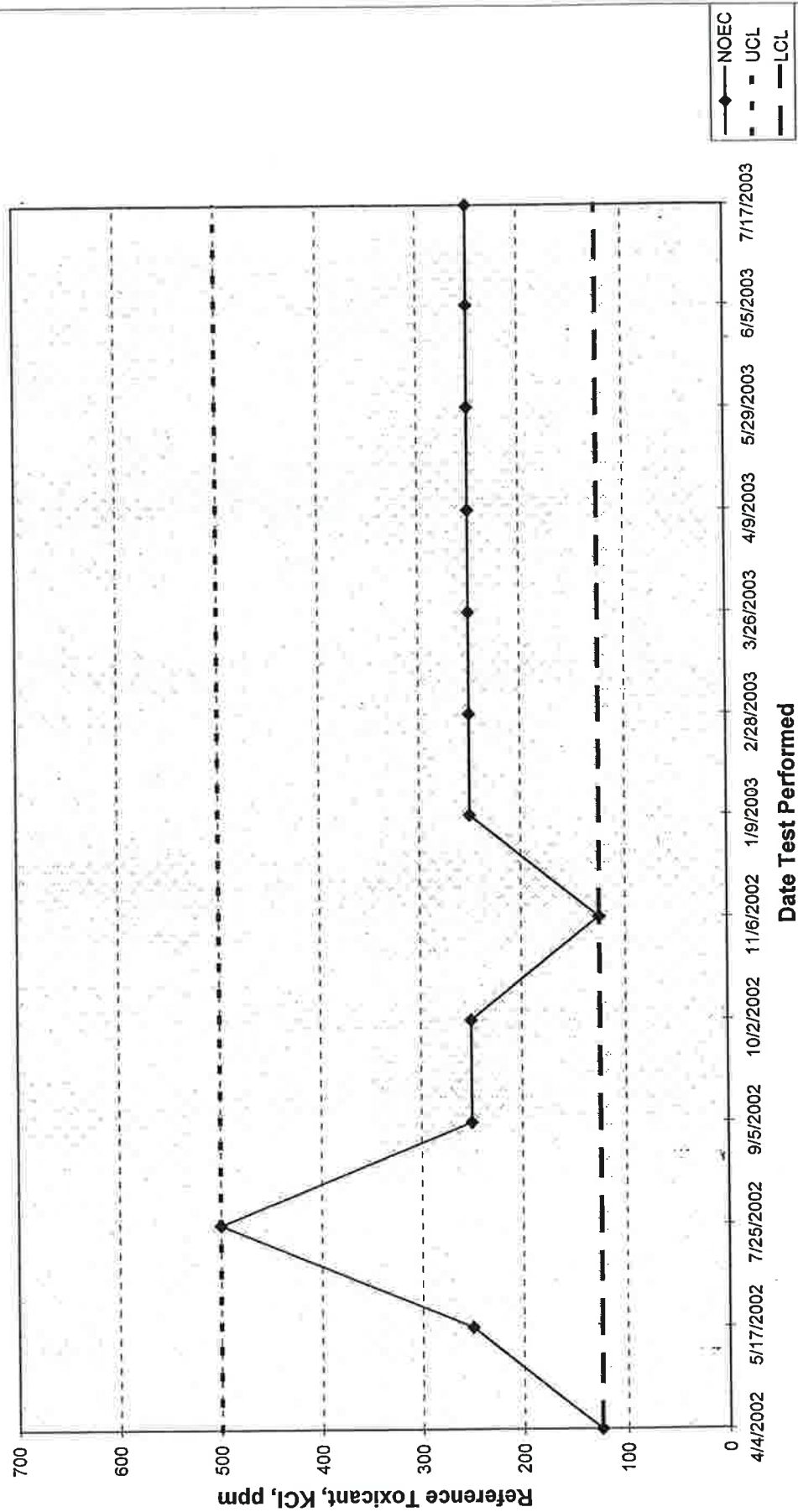
**ARKANSAS ANALYTICAL, INC.**  
**FATHEAD MINNOW GROWTH**  
**QUALITY ASSURANCE**



**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



Be it known that  
**Arkansas Analytical, Inc**  
Little Rock, Arkansas  
has earned certification by this Department for the period of

May 14, 2003 to October 30, 2003

Laboratory ID # 60-1754

Certificate # 03-031-1

The following parameters are certified:

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

*May 15, 2003* Date

*J.A. Sembrski*  
Quality Assurance Officer

ARKANSAS ANALYTICAL, INCORPORATED

11701 I-30, BUILDING 1, SUITE 115  
LITTLE ROCK, AR 72209

Laboratory Control Number: K308578 Date: 9-9-03

Client: Weston Sample ID: effluent

**Pass**                      **Fail**

Fathead Minnow Survival Test      ✓                      \_\_\_\_\_

Fathead Minnow Growth Test        ✓                      \_\_\_\_\_

*Ceriodaphnia dubia* Survival Test    ✓                      \_\_\_\_\_

*Ceriodaphnia dubia* Reproduction Test    ✓                      \_\_\_\_\_ Analyst Initials MA



# Arkansas Analytical, Inc.

## Toxicity Test Results

**MAGCOBAR MINE SITE**  
**NPDES PERMIT NUMBER: AR0049794**  
**August 2003**

12/3/03  
42/6

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

*Ceriodaphnia dubia*, Survival and Reproduction Test  
Test 1002.0

Prepared for: **Mr. Alan B. Brown**  
**Weston Solutions**  
**Magcobar Mine Site**  
**2000 Darby Lane**  
**Malvern, AR 72104**

Prepared by: Arkansas Analytical, Inc.  
11701 I-30, Bldg 1, Suite 115  
Little Rock, Arkansas 72209  
**Lab Number K308578**

Tuesday, September 9, 2003

## **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 once per month for both *Ceriodaphnia dubia* and *Pimephales promelas*. The test results in this report represent the testing for August of 2003.

## **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:	8-21-03, 1000	8-22-03, 1000
Sample #2:	8-24-03, 0930	8-25-03, 0930
Sample #3:	8-25-03, 0930	8-26-03, 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	Temperature Upon Receipt (°C)
Sample #1:	8-22-03, 1534	Not Taken
Sample #2:	8-25-03, 1525	3
Sample #3:	8-26-03, 1531	5

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	25.4	X	
At least 60% of surviving females should have produced 3 broods	90%	X	
The percent coefficient of variation between replicates must be 40% or less for the young of surviving females	13.8%	X	

#### TEST ACCEPTANCE CRITERIA for *Pimephales promelas*

Control Criteria	Results	Pass	Fail
Greater than or equal to 80% survival	100%	X	
The percent coefficient of variation between replicates must be 40% or less for survival	0%	X	
Minimum of 0.25 mg average dry weight of surviving controls	0.556	X	
The percent coefficient of variation between replicates must be 40% or less for growth	17.3%	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)	20.4	%CV survival (critical dilution)	0%
%CV Reproduction (critical dilution)	27.3%	Mean dry weight (critical dilution) in milligrams	0.693
		%CV growth (critical dilution)	14.7%

### Conclusion

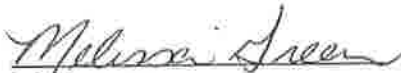
Chronic static renewal larval survival and growth test using fathead minnow, *Pimephales promelas*, (Method 1000.0).

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, 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:

  
Melissa Green

  
Andrea Fox

  
Teresa Canfield

  
Amy Daniel

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:	8-21-03, 1000	8-22-03, 1000
Sample #2:	8-24-03, 0930	8-25-03, 0930
Sample #3:	8-25-03, 0930	8-26-03, 0930

Test initiated (date, time): 8-22-03, 1600      Test terminated (date, time): 8-29-03, 1600

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		Mean Dry Weight	CV%
0%	0.616	0.620	0.390	0.595	0.557		0.556	17.3
32%	0.702	0.780	0.817	0.663	0.700		0.732	
42%	0.643	0.793	0.743	0.599	0.707		0.697	
56%	0.953	0.671	0.703	0.633	0.649		0.722	
75%	0.733	0.803	0.619	0.650	0.675		0.696	
100%	0.712	0.848	0.600	0.602	0.701		0.693	14.7

**DATA TABLE FOR GROWTH OF FATHEAD MINNOWS**

Effluent Conc %	Average Dry Weight in milligrams in replicate chambers					Mean Percent Survival		
	A	B	C	D	E		Mean Dry Weight	CV%
0%	0.616	0.620	0.390	0.595	0.557		0.556	17.3
32%	0.702	0.780	0.817	0.663	0.700		0.732	
42%	0.643	0.793	0.743	0.599	0.707		0.697	
56%	0.953	0.671	0.703	0.633	0.649		0.722	
75%	0.733	0.803	0.619	0.650	0.675		0.696	
100%	0.712	0.848	0.600	0.602	0.701		0.693	14.7

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)= \_\_\_\_\_   0   %

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:	8-21-03, 1000	8-22-03, 1000
Sample #2:	8-24-03, 0930	8-25-03, 0930
Sample #3:	8-25-03, 0930	8-26-03, 0930

Test initiated (date, time): 8-22-03, 1600      Test terminated (date, time): 8-28-03, 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	28	20	24	13	21	11
B	24	30	29	24	23	23
C	25	27	32	33	23	19
D	30	27	30	25	27	17
E	28	24	23	27	24	28
F	21	33	28	23	16	X0
G	22	9	24	25	28	27
H	20	28	27	28	26	X0
I	28	X0	25	25	12	18
J	28	32	33	25	28	20
Mean	25.4	23.0	27.5	24.8	22.8	16.6
Mean/surviving female	25.4	25.6	27.5	24.8	22.8	20.4
CV%*	13.8					27.3

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	100	90	100	100	100	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)= 27.3 %

APPENDIX A

Chain of Custody Forms

# CHAIN OF CUSTODY RECORD

<b>CLIENT INFORMATION</b> Western Solutions 2000 Derby Lane Malvern, AR 72104 Attn: Alan Braun  Alan Braun		<b>Project Description</b>  <b>Reporting Information</b> Telephone: FAX: Bill to/P.O.	<b>Turnaround Time</b> (CIRCLE ONE) 24 hour 48 hour <i>routine</i> Preservative Code: Bottle Type	<b>Preservation Codes:</b> 1. Cool, 4 degrees Centigrade 2. Sulfuric Acid, pH < 2 3. Nitric Acid, pH < 2 4. Thio sulfate for dechlorination 5. Hydrochloric Acid for VOA 6. Sodium Hydroxide, pH > 12  <b>TEST PARAMETERS</b>	Bottle type code G=glass; P=HDPE V=septum; A=amber
<b>Samp. (Signature/s)</b>  Cathy B. Braun		<b>Samp. (Printed)</b> Alan B. Braun		Arkansas Analytical Lab # K308578A	
<b>Field Number</b> FDB22260	<b>Sample Collection Date/s</b> 8/22	<b>Sample Collection Time/s</b> 10:00	<b># of Containers</b> 5	<b>Orb Comp</b> X	<b>SAMPLE IDENTIFICATION/DESCRIPTION</b> Facility Discharge
<b>1. Relinquished by: (Signature)</b> Cathy B. Braun			<b>2. Received by: (Signature)</b> Alan Braun		
<b>Date/Time</b> 8/22/03 15:34			<b>Date/Time</b> 8/22/03 1534		
<b>1. Received by: (Signature)</b>			<b>2. Received by: (Signature)</b>		
<b>For completion by laboratory</b> Condition of samples: A. Containers Correct? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO B. Preservation Correct? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO C. Seals Intact? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO			<b>REMARKS</b>		





# CHAIN OF CUSTODY RECORD

**CLIENT INFORMATION**  
 Weston Solutions, Inc.  
 P.O. Box 699  
 2000 Darby Lane  
 Malvern, AR 72104  
 ATTN: Beverly Bismore Alan Brown

**Project Description**  
 Reporting Information  
 Telephone: 501.467.8355  
 FAX: 501.467.9887  
 Bill to P.O.

**Turnaround Time (CIRCLE ONE)**  
 24 hour  
 48 hour  
 routine

**Preservative Code:**  
 Bottle Type

**Preservation Codes:**  
 1. Cool, 4 degrees Centigrade  
 2. Sulfuric Acid, pH < 3  
 3. Nitric Acid, pH < 2  
 4. Thiouate for dechlorination  
 5. Hydrochloric Acid for VOA  
 6. Sodium Hydroxide, pH > 12

Field Number	Sample Collection		Time/s	Grab	Comp	# of Containers	# of Samples	Sample Identification/Description	Bottle type code
	Date/s	Date/Time							
FD082606	08/26	9:30		X			3	Facility Discharge	Arkansas Analytical Lab # 1308578C

**SIGNATURES:**  
 1. Relinquished by: (Signature) *Alan Brown*  
 Date/Time: 8/26/03 15:31

**RECEIVED:**  
 2. Received by: (Signature) *Beverly Bismore*  
 Date/Time: 8/26/03 15:31

**For completion by laboratory:**  
 Condition of samples: YES  NO   
 A. Containers Correct?   
 B. Preservation Correct?   
 C. Seals Intact?   
 Remarks: *Person in SOC*



## APPENDIX B

### Effluent and Dilution Water Data

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING

Fathead Minnow

Lab # / Sample ID		K308578							Test Start (Date/Time)		8-22-03/1600	
Client		Weston							Test End (Date/Time)		8-29-03/1600	
		Day of Test										
		1	2	3	4	5	6	7	notes/remarks			
<b>Control</b>		8/22	8-23	8-24	8-25	8/26	8/27	8/28	8/22	SS 69		
D.O (mg/L)	INITIAL	7.3	9.1	9.0	8.6	7.4	7.9	7.5	8/26	SS 10		
	FINAL	6.9	8.5	7.0	6.8	6.7	6.8	6.9				
pH(mg/L)	INITIAL	7.2	7.4	7.7	6.1	6.6	6.7	6.5				
	FINAL	6.6	6.9	6.1	6.1	6.0	7.7	7.2				
temp(C)	INITIAL	23.8	21.3	20.9	21.3	21.0	21.7	21.3				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
ALKALINITY(mg/L)		28				22						
HARDNESS(mg/L)		82				39						
CONDUCTIVITY(umhos/cm)		160				153						
CHLORINE(mg/L)		0.05				0.05						
CONC:		32%	32%	32%	32%	32%	32%	32%				
D.O (mg/L)	INITIAL	7.5	9.8	9.3	7.7	7.7	7.9	7.5				
	FINAL	6.9	8.3	7.1	6.8	6.7	6.7	7.0				
pH(mg/L)	INITIAL	7.1	7.2	7.4	6.5	6.4	6.8	6.7				
	FINAL	6.5	6.8	6.0	6.0	6.0	7.4	7.1				
temp(C)	INITIAL	23.6	21.6	20.9	22.5	21.0	22.0	22.4				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
CONC:		42%	42%	42%	42%	42%	42%	42%				
D.O (mg/L)	INITIAL	7.3	9.8	9.1	7.6	7.5	7.9	7.5				
	FINAL	6.9	8.4	7.0	6.9	6.8	6.7	7.0				
pH(mg/L)	INITIAL	7.2	7.2	7.4	6.6	6.4	6.9	6.8				
	FINAL	6.5	6.8	6.0	6.0	6.0	7.3	7.1				
temp(C)	INITIAL	23.5	21.6	20.9	22.9	22.0	22.2	23.2				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
CONC:		56%	56%	56%	56%	56%	56%	56%				
D.O (mg/L)	INITIAL	7.3	9.9	9.2	7.5	7.6	8.0	7.4				
	FINAL	6.9	8.8	7.0	6.9	6.8	6.7	7.0				
pH(mg/L)	INITIAL	7.2	7.1	7.4	6.7	6.4	6.9	6.9				
	FINAL	6.5	6.8	6.0	6.1	6.1	7.3	7.0				
temp(C)	INITIAL	23.5	21.5	20.9	23.7	21.9	22.5	24.3				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
CONC:		75%	75%	75%	75%	75%	75%	75%				
D.O (mg/L)	INITIAL	7.5	10.0	9.3	7.4	7.6	8.0	7.5				
	FINAL	7.0	8.9	7.9	7.0	6.8	6.8	6.9				
pH(mg/L)	INITIAL	7.2	7.1	7.2	6.8	6.4	6.9	7.0				
	FINAL	6.5	6.8	6.0	6.1	6.1	7.3	7.0				
temp(C)	INITIAL	23.5	21.2	21.1	24.7	21.9	23.4	25.2				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
CONC:		100%	100%	100%	100%	100%	100%	100%				
D.O (mg/L)	INITIAL	7.3	9.6	9.4	7.2	7.7	8.6	7.4				
	FINAL	7.0	8.7	7.0	7.1	6.8	6.9	6.9				
pH(mg/L)	INITIAL	7.3	7.2	7.2	7.0	6.4	7.0	7.1				
	FINAL	6.4	6.7	5.9	6.0	6.1	7.2	7.0				
temp(C)	INITIAL	24.0	22.0	21.1	26.3	21.9	24.3	26.8				
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0	25.0				
CONC:		100%	A	A	A	B	B	C				
ALKALINITY( mg/L)		11				12		13				
HARDNESS(mg/L)		1146				1368		1356				
CONDUCTIVITY(umhos/cm)		2220				2250		2220				

CHEMICAL DATA SHEET FOR CHRONIC TOXICITY TESTING

Ceriodaphnia dubia

Lab # / Sample ID		K308578							Test Start (Date/Time)	8-22-03 / 11:00
Client		Weston							Test End (Date/Time)	8-28-03 / 10:20
		Day of Test								
		1	2	3	4	5	6	7	8	notes/remarks
<b>Control</b>		8-22	8-23	8-24	8-25	8-26	8-27			8-22 SS#69
D.O (mg/L)	INITIAL	7.3	9.1	9.0	8.0	7.4	7.9			8-26 SS#70
	FINAL	7.0	9.3	8.1	8.2	7.7	8.3			
pH	INITIAL	7.2	7.4	7.7	6.1	6.6	6.7			
	FINAL	7.5	7.4	7.1	6.8	6.4	7.9			
temp(C)	INITIAL	23.8	21.3	20.9	21.3	21.0	21.7			
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0			
ALKALINITY(mg/L)		28				22				
HARDNESS(mg/L)		82				39				
CONDUCTIVITY(umhos/cm)		160				153				
CHLORINE(mg/L)		<0.05			<del>40.05</del>	20.05				
CONC:		32%	32%	32%	32%	32%	32%			
D.O (mg/L)	INITIAL	7.5	9.8	9.3	7.7	7.7	7.9			
	FINAL	7.0	9.2	8.1	8.3	7.7	8.3			
pH	INITIAL	7.8	7.2	7.4	6.5	6.4	6.8			
	FINAL	7.5	7.3	7.1	6.8	6.3	8.1			
temp(C)	INITIAL	23.6	21.6	20.9	22.5	21.0	22.0			
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0			
CONC:		42%	42%	42%	42%	42%	42%			
D.O (mg/L)	INITIAL	7.3	9.8	9.1	7.6	7.5	7.9			
	FINAL	7.0	9.2	8.1	8.3	7.8	8.6			
pH	INITIAL	7.2	7.2	7.4	6.6	6.4	6.9			
	FINAL	7.5	7.4	7.0	6.8	6.3	7.9			
temp(C)	INITIAL	23.5	21.6	20.9	22.9	22.0	22.2			
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0			
CONC:		56%	56%	56%	56%	56%	56%			
D.O (mg/L)	INITIAL	7.3	9.9	9.2	7.5	7.6	8.0			
	FINAL	7.0	9.2	8.1	8.4	7.9	8.6			
pH	INITIAL	7.2	7.1	7.4	6.7	6.4	6.9			
	FINAL	7.4	7.4	7.0	6.7	6.2	7.8			
temp(C)	INITIAL	23.5	21.5	20.9	23.7	21.9	22.5			
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0			
CONC:		75%	75%	75%	75%	75%	75%			
D.O (mg/L)	INITIAL	7.5	10.0	9.3	7.4	7.6	8.0			
	FINAL	7.1	9.3	8.1	8.4	8.0	8.7			
pH	INITIAL	7.2	7.1	7.3	6.8	6.4	6.9			
	FINAL	7.4	7.4	7.0	6.7	6.2	7.8			
temp(C)	INITIAL	23.5	21.2	21.1	24.7	21.9	23.4			
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0			
CONC:		100%	100%	100%	100%	100%	100%			
D.O (mg/L)	INITIAL	7.3	9.6	9.4	7.2	7.7	8.0			
	FINAL	7.1	9.3	8.2	8.4	8.0	8.5			
pH	INITIAL	7.3	7.2	7.2	7.0	6.4	7.0			
	FINAL	7.3	7.2	6.9	6.7	6.1	7.5			
temp(C)	INITIAL	24.0	22.0	21.1	26.3	21.9	24.3			
	FINAL	25.0	25.0	25.0	25.0	25.0	25.0			
CONC:	100%	A	A	A	B	B	C			
ALKALINITY(mg/L)		11			12		13			
HARDNESS(mg/L)		114.6			136.8		135.6			
CONDUCTIVITY(umhos/cm)		2220			2250		2230			
CHLORINE(mg/L)		20.05			20.05		20.05			

## APPENDIX C

### Fathead Minnow Raw Data and Statistics



**SURVIVAL DATA FOR FATHEAD MINNOW LARVAL SURVIVAL AND GROWTH TEST**

LAB #/ SAMPLE ID K308578 TEST START DATE 8-22-05 TIME 1600  
 CLIENT Weston TEST END DATE 8-29 TIME 1600  
 AGE AND SOURCE OF MINNOWS 224 hrs; Aquatox

CONC:	REP #	DAY (NUMBER SURVIVING)								SURVIVAL		
		start	1	2	3	4	5	6	7	%	MEAN %	CV
Control	A	10	10	10	10	10	10	10	10	100	100%	0%
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
32%	A	10	10	10	10	10	10	10	10	100	100%	
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
42%	A	10	10	10	10	10	10	10	10	100	98%	
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	9	9	9	90		
	E	10	10	10	10	10	10	10	10	100		
50%	A	10	10	10	10	10	10	10	10	100	100%	
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
75%	A	10	10	10	10	10	10	10	10	100	96%	
	B	10	10	10	10	10	9	9	9	90		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	9	9	9	90		
	E	10	10	10	10	10	10	10	10	100		
100%	A	10	10	10	10	10	10	10	10	100	100%	0%
	B	10	10	10	10	10	10	10	10	100		
	C	10	10	10	10	10	10	10	10	100		
	D	10	10	10	10	10	10	10	10	100		
	E	10	10	10	10	10	10	10	10	100		
ANALYST:		TC	AD	AD	TC	TC	TC	TC	mg			
DATE:		8-22	8-23	8-24	8-25	8-26	8-27	8-28	8-29			
TIME:		1600	1015	1020	1135	1415	1530	1620	1600			

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

WEIGHT DATA FOR LARVAL SURVIVAL AND GROWTH TEST

LAB #/S: <u>K308578</u>	TEST DATES (BEGIN/END): <u>8-21-03/8-28-03</u>
CLIENT: <u>Weston</u>	WEIGHING DATE/TIME:
ANALYST/S: <u>AF, AD, TC, MJ</u>	DRYING TEMPERATURE (DEGREES C): <u>60°C</u>
SAMPLE ID:	DRYING TIME (HOURS): <u>24 hrs.</u>

	REF #	FINAL DRY WEIGHT TIN+LARVAE (g)	INITIAL WEIGHT TIN (g)	TOTAL DRY WEIGHT OF LARVAE (g)	NUMBER OF LARVAE	DRY WEIGHT OF LARVA (mg)		REMARKS
CONTROL	A65	0.98198	0.97582	0.00616	10	0.616	AVG DRY	
	B66	0.97571	0.96951	0.00620	10	0.620	WEIGHT (mg)	
	C67	0.97899	0.97509	0.00390	10	0.390	0.556	
	D68	0.98555	0.97940	0.00595	10	0.595	CV	
	E69	0.97809	0.97248	0.00557	10	0.557	17.3	
321	A70	0.98806	0.98104	0.00702	10	0.702	AVG DRY	
	B71	0.98162	0.97382	0.00780	10	0.780	WEIGHT (MG)	
	C72	0.98221	0.97403	0.00817	10	0.817	0.452	
	D73	0.98519	0.97852	0.00663	10	0.663	CV	0.732
	E74	0.98438	0.97738	0.00700	10	0.700		
421	A75	0.98242	0.97599	0.00643	10	0.643	AVG DRY	
	B76	0.98275	0.97482	0.00793	10	0.793	WEIGHT (MG)	
	C77	0.98819	0.98012	0.00743	10	0.743	0.697	
	D78	0.98198	0.97599	0.00599	10	0.599	CV	
	E79	0.98265	0.97558	0.00707	10	0.707		
561	A80	0.98472	0.97519	0.00953	10	0.953	AVG DRY	
	B81	0.98513	0.97842	0.00671	10	0.671	WEIGHT (MG)	
	C82	0.98719	0.98012	0.00703	10	0.703	0.722	
	D83	0.98926	0.98293	0.00633	10	0.633	CV	
	E84	0.98827	0.98178	0.00649	10	0.649		
751	A85	0.98737	0.98004	0.00733	10	0.733	AVG DRY	
	B86	0.98590	0.97747	0.00803	10	0.803	WEIGHT (MG)	
	C87	0.97854	0.97235	0.00619	10	0.619	0.696	
	D88	0.98965	0.98315	0.00650	10	0.650	CV	
	E89	0.98923	0.98248	0.00675	10	0.675		
1001	A90	0.99067	0.98355	0.00712	10	0.712	AVG DRY	
	B91	0.98216	0.97368	0.00848	10	0.848	WEIGHT (MG)	
	C92	0.97900	0.97300	0.00600	10	0.600	0.693	
	D93	0.97708	0.97106	0.00602	10	0.602	CV	
	E94	0.97894	0.97193	0.00701	10	0.701	14.69	

CV = (STANDARD DEVIATION/MEAN)\*100

AA# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03  
File: K308578S Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro - Wilk's test for normality

---

D = 0.053

W = 0.714

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# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03  
File: K308578S 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# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03  
FILE: K308578S  
TRANSFORM: ARC SINE(SQUARE ROOT(Y))

NUMBER OF GROUPS: 6

---

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	1.0000	1.4120
1	CONTROL	2	1.0000	1.4120
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	1.0000	1.4120
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	0.9000	1.2490
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	1.0000	1.4120
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	0.9000	1.2490
5	75 % EFFLUENT	3	1.0000	1.4120
5	75 % EFFLUENT	4	0.9000	1.2490
5	75 % EFFLUENT	5	1.0000	1.4120
6	100 % EFFLUENT	1	1.0000	1.4120
6	100 % EFFLUENT	2	1.0000	1.4120
6	100 % EFFLUENT	3	1.0000	1.4120
6	100 % EFFLUENT	4	1.0000	1.4120
6	100 % EFFLUENT	5	1.0000	1.4120

---

LA# K308578 FATHEAD MINNOW SURVIVAL, 8-21-03

file: K308578S

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	1.412				
2	32 % EFFLUENT	1.412	27.50	16.00	5.00	
3	42 % EFFLUENT	1.379	25.00	16.00	5.00	
4	56 % EFFLUENT	1.412	27.50	16.00	5.00	
5	75 % EFFLUENT	1.347	22.50	16.00	5.00	
6	100 % EFFLUENT	1.412	27.50	16.00	5.00	

---

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

AA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
File: K308578G Transform: NO TRANSFORMATION

Shapiro - Wilk's test for normality

---

D = 0.209

W = 0.971

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 # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
File: K308578G Transform: NO TRANSFORMATION

---

Bartlett's test for homogeneity of variance

Calculated B1 statistic = 2.62

---

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 # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
FILE: K308578G  
TRANSFORM: NO TRANSFORMATION

NUMBER OF GROUPS: 6

---

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	0.6160	0.6160
1	CONTROL	2	0.6200	0.6200
1	CONTROL	3	0.3900	0.3900
1	CONTROL	4	0.5950	0.5950
1	CONTROL	5	0.5570	0.5570
2	32 % EFFLUENT	1	0.7020	0.7020
2	32 % EFFLUENT	2	0.7800	0.7800
2	32 % EFFLUENT	3	0.8170	0.8170
2	32 % EFFLUENT	4	0.6630	0.6630
2	32 % EFFLUENT	5	0.7000	0.7000
3	42 % EFFLUENT	1	0.6430	0.6430
3	42 % EFFLUENT	2	0.7930	0.7930
3	42 % EFFLUENT	3	0.7430	0.7430
3	42 % EFFLUENT	4	0.5990	0.5990
3	42 % EFFLUENT	5	0.7070	0.7070
4	56 % EFFLUENT	1	0.9530	0.9530
4	56 % EFFLUENT	2	0.6710	0.6710
4	56 % EFFLUENT	3	0.7030	0.7030
4	56 % EFFLUENT	4	0.6330	0.6330
4	56 % EFFLUENT	5	0.6490	0.6490
5	75 % EFFLUENT	1	0.7330	0.7330
5	75 % EFFLUENT	2	0.8030	0.8030
5	75 % EFFLUENT	3	0.6190	0.6190
5	75 % EFFLUENT	4	0.6500	0.6500
5	75 % EFFLUENT	5	0.6750	0.6750
6	100 % EFFLUENT	1	0.7120	0.7120
6	100 % EFFLUENT	2	0.8480	0.8480
6	100 % EFFLUENT	3	0.6000	0.6000
6	100 % EFFLUENT	4	0.6020	0.6020
6	100 % EFFLUENT	5	0.7010	0.7010

---

VA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
File: K308578G Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	0.103	0.021	2.367
Within (Error)	24	0.209	0.009	
Total	29	0.312		

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

AA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
 File: K308578G 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	0.556	0.556		
2	32 % EFFLUENT	0.732	0.732	-2.994	
3	42 % EFFLUENT	0.697	0.697	-2.395	
4	56 % EFFLUENT	0.722	0.722	-2.815	
5	75 % EFFLUENT	0.696	0.696	-2.378	
6	100 % EFFLUENT	0.693	0.693	-2.320	

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

AA # K309578, FATHEAD MINNOW GROWTH, 8-21-03  
 File: K308578G 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	5			
2	32 % EFFLUENT	5	0.139	25.1	-0.177
3	42 % EFFLUENT	5	0.139	25.1	-0.141
4	56 % EFFLUENT	5	0.139	25.1	-0.166
5	75 % EFFLUENT	5	0.139	25.1	-0.140
6	100 % EFFLUENT	5	0.139	25.1	-0.137



APPENDIX D

*Ceriodaphnia dubia* Raw Data and Statistics

**SURVIVAL AND REPRODUCTION TEST**

*Caridodaphnia dubia*

Discharger: Winston

Location: See CDC

Date Sample Collected: See CDC

Lab Number/s: K308578

Analyst: MG, AD, TC

Test Start-Date/Time: 8-27-03/1600

Test Stop-Date/Time: 8-28-03/0920

Conc 1	Replicate												No. of Young	No. of Adults	Young/Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day	1	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
%	2	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
	3	5	0	4	5	0	3	6	6	6	6	6	40	10	4.0	TC
	4	0	5	0	0	1	5	8	0	10	0	2	29	10	2.9	TC
	5	10	9	10	12	10	8	0	8	0	11	7	78	10	7.8	TC
	6	13	10	11	13	12	8	11	6	12	11	10	107	10	10.7	TC
	7															
	8															
Total	28	24	25	30	28	21	22	20	28	28	28	28	254			

10% 3rd brood

Conc 2	Replicate												No. of Young	No. of Adults	Young/Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day	1	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
%	2	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
	3	4	7	6	0	0	0	0	0	5	2	1	27	9	2.7	TC
	4	0	0	0	5	7	3	0	-	0	2	0	20	9	2.2	TC
	5	8	12	9	9	11	5	10	-	12	8	5	85	9	9.4	TC
	6	8	11	12	13	10	15	1	13	-	15	9	98	9	10.9	TC
	7															
	8															
Total	20	30	27	27	24	33	9	28	28	28	28	28	230			

32

Conc 3	Replicate												No. of Young	No. of Adults	Young/Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day	1	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
%	2	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
	3	4	6	7	6	0	0	0	0	5	6	6	40	10	4.0	TC
	4	0	0	0	5	5	6	9	0	0	0	1	16	10	1.6	TC
	5	7	11	11	12	8	9	9	10	10	13	10	100	10	10.0	TC
	6	13	12	14	12	10	14	9	11	10	14	11	119	10	11.9	TC
	7															
	8															
Total	24	29	32	30	23	28	24	27	25	33	27	27	275			

42

Conc 4	Replicate												No. of Young	No. of Adults	Young/Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day	1	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
%	2	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
	3	0	2	6	5	6	0	0	4	5	4	1	16	8	1.6	TC
	4	5	0	0	0	0	6	9	10	8	0	7	26	8	2.0	TC
	5	0	11	12	9	11	8	9	10	9	10	9	57	8	7.1	TC
	6	8	11	15	11	10	9	10	14	12	12	10	77	8	9.6	TC
	7															
	8															
Total	13	24	33	25	27	23	25	28	25	25	25	25	248			

Conc 5	Replicate												No. of Young	No. of Adults	Young/Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day	1	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
%	2	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
	3	4	2	6	0	5	0	0	5	5	5	3	32	10	3.2	TC
	4	0	3	0	4	0	0	0	0	0	0	1	13	10	1.3	TC
	5	6	10	7	10	7	8	11	10	7	11	8	87	10	8.7	TC
	6	11	8	10	13	12	8	11	11	0	12	9	96	10	9.6	TC
	7															
	8															
Total	21	23	23	27	24	16	28	26	12	28	28	28	228			

Conc 6	Replicate												No. of Young	No. of Adults	Young/Adult	Analyst
	A	B	C	D	E	F	G	H	I	J						
Day	1	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
%	2	0	0	0	0	0	0	0	0	0	0	0	0	10	0	AD
	3	0	3	0	2	4	X	0	X	3	0	4	16	8	1.6	TC
	4	10	0	3	1	1	-	5	-	6	0	2	26	8	2.0	TC
	5	15	11	7	7	11	-	10	-	0	6	5	57	8	7.1	TC
	6	6	9	9	7	12	-	12	-	12	10	7	77	8	9.6	TC
	7															
	8															
Total	11	23	19	17	28	X	0	27	X	0	18	20	160			

X=DEAD; Y=MALE

Y=20:4 CH=27:31

FISHER'S EXACT TEST

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
32% 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	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
42% 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

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
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

=====

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
75% 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

=====

IDENTIFICATION	NUMBER OF		
	ALIVE	DEAD	TOTAL ANIMALS
CONTROL	10	0	10
100%	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	1	
2	42% EFFLUENT	10	0	
3	56% EFFLUENT	10	0	
4	75% EFFLUENT	10	0	
5	100%	10	2	

AA # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
File: k308578c 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 # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
File: k308578c Transform: NO TRANSFORMATION

---

Bartlett's test for homogeneity of variance  
Calculated B1 statistic = 21.95

---

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

Data FAIL B1 homogeneity test at 0.01 level. Try another transformation.



TITLE: AA # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
 FILE: k308578c  
 TRANSFORM: NO TRANSFORMATION

NUMBER OF GROUPS: 6

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	CONTROL	1	28.0000	28.0000
1	CONTROL	2	24.0000	24.0000
1	CONTROL	3	25.0000	25.0000
1	CONTROL	4	30.0000	30.0000
1	CONTROL	5	28.0000	28.0000
1	CONTROL	6	21.0000	21.0000
1	CONTROL	7	22.0000	22.0000
1	CONTROL	8	20.0000	20.0000
1	CONTROL	9	28.0000	28.0000
1	CONTROL	10	28.0000	28.0000
2	32 % EFFLUENT	1	20.0000	20.0000
2	32 % EFFLUENT	2	30.0000	30.0000
2	32 % EFFLUENT	3	27.0000	27.0000
2	32 % EFFLUENT	4	27.0000	27.0000
2	32 % EFFLUENT	5	24.0000	24.0000
2	32 % EFFLUENT	6	33.0000	33.0000
2	32 % EFFLUENT	7	9.0000	9.0000
2	32 % EFFLUENT	8	28.0000	28.0000
2	32 % EFFLUENT	9	0.0000	0.0000
2	32 % EFFLUENT	10	32.0000	32.0000
3	42 % EFFLUENT	1	24.0000	24.0000
3	42 % EFFLUENT	2	29.0000	29.0000
3	42 % EFFLUENT	3	32.0000	32.0000
3	42 % EFFLUENT	4	30.0000	30.0000
3	42 % EFFLUENT	5	23.0000	23.0000
3	42 % EFFLUENT	6	28.0000	28.0000
3	42 % EFFLUENT	7	24.0000	24.0000
3	42 % EFFLUENT	8	27.0000	27.0000
3	42 % EFFLUENT	9	25.0000	25.0000
3	42 % EFFLUENT	10	33.0000	33.0000
4	56 % EFFLUENT	1	13.0000	13.0000
4	56 % EFFLUENT	2	24.0000	24.0000
4	56 % EFFLUENT	3	33.0000	33.0000
4	56 % EFFLUENT	4	25.0000	25.0000
4	56 % EFFLUENT	5	27.0000	27.0000
4	56 % EFFLUENT	6	23.0000	23.0000
4	56 % EFFLUENT	7	25.0000	25.0000
4	56 % EFFLUENT	8	28.0000	28.0000
4	56 % EFFLUENT	9	25.0000	25.0000
4	56 % EFFLUENT	10	25.0000	25.0000
5	75 % EFFLUENT	1	21.0000	21.0000
5	75 % EFFLUENT	2	23.0000	23.0000
5	75 % EFFLUENT	3	23.0000	23.0000
5	75 % EFFLUENT	4	27.0000	27.0000
5	75 % EFFLUENT	5	24.0000	24.0000
5	75 % EFFLUENT	6	16.0000	16.0000
5	75 % EFFLUENT	7	28.0000	28.0000
5	75 % EFFLUENT	8	26.0000	26.0000
5	75 % EFFLUENT	9	12.0000	12.0000
5	75 % EFFLUENT	10	28.0000	28.0000

6	100	%	EFFLUENT	1	31.0000	31.0000
6	100	%	EFFLUENT	2	23.0000	23.0000
6	100	%	EFFLUENT	3	19.0000	19.0000
6	100	%	EFFLUENT	4	17.0000	17.0000
6	100	%	EFFLUENT	5	28.0000	28.0000
6	100	%	EFFLUENT	6	0.0000	0.0000
6	100	%	EFFLUENT	7	27.0000	27.0000
6	100	%	EFFLUENT	8	0.0000	0.0000
6	100	%	EFFLUENT	9	18.0000	18.0000
6	100	%	EFFLUENT	10	20.0000	20.0000

---

AA # K308578, CERIODAPHNIA DUBIA REPRODUCTION, 8-21-03  
File: k308578c Transform: NO TRANSFORMATION

STEEL'S MANY-ONE RANK TEST

Ho: Control < Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	RANK SUM	CRIT. VALUE	df	SIG
1	CONTROL	25.400				
2	32 % EFFLUENT	23.000	105.50	75.00	10.00	
3	42 % EFFLUENT	27.500	120.00	75.00	10.00	
4	56 % EFFLUENT	24.800	101.50	75.00	10.00	
5	75 % EFFLUENT	22.800	90.00	75.00	10.00	
6	100 % EFFLUENT	18.300	80.50	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 8-21-03 Arkansas Analytical

SPECIES Pimephales promelas

QUANTITY SHIPPED 900+

AGE/LIFE STAGE 44 hrs 8/21 1500 LST

BROODSTOCK SOURCE Anderson Farms, AR

CULTURE WATER groundwater

ALKALINITY (Mg/l as CaCO<sub>3</sub>) = 180

**HARDNESS** (Mg/l as CaCO<sub>3</sub>)/Salinity (ppt) = 160

FEEDING Artemia

COMMENTS \_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_

PACKAGED BY lll

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: 1/17/01

SPECIES: Ceriodaphnia dubia

AGE: Variable

LIFE STAGE: Adult

HATCH DATE: Variable

BEGAN FEEDING: Immediately

FOOD: YTC, Selenastrum

### Water Chemistry Record:

	Mean	Range
TEMPERATURE:	<u>24 °C</u>	<u>21-24°C</u>
SALINITY/CONDUCTIVITY:	<u>--</u>	<u>--</u>
TOTAL HARDNESS (as CaCO <sub>3</sub> ):	<u>112 mg/l</u>	<u>90-124 mg/l</u>
TOTAL ALKALINITY (as CaCO <sub>3</sub> ):	<u>85 mg/l</u>	<u>50-85 mg/l</u>
pH:	<u>8.09</u>	<u>7.68-8.14</u>

Comments:

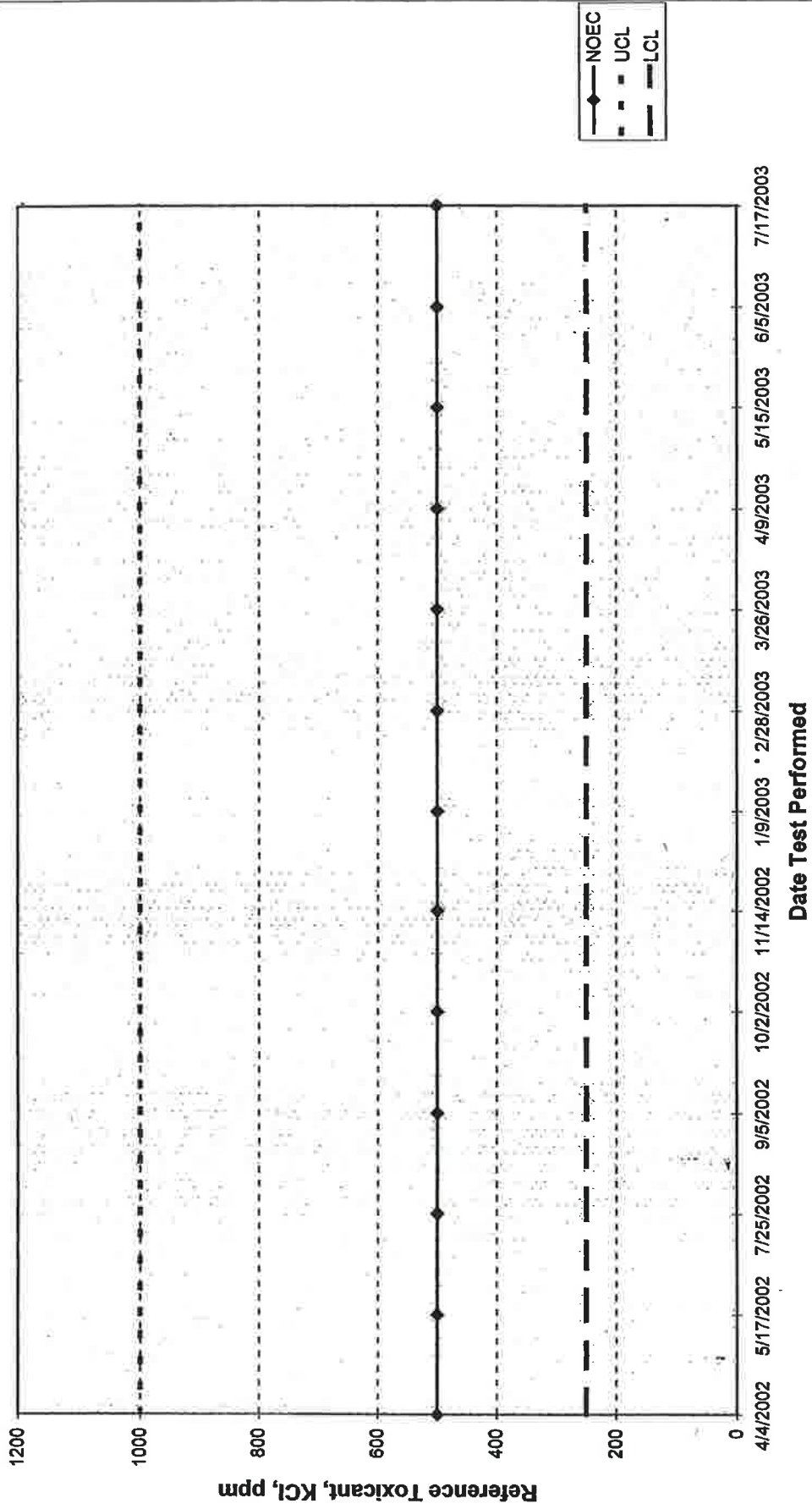
  
\_\_\_\_\_  
Facility Supervisor



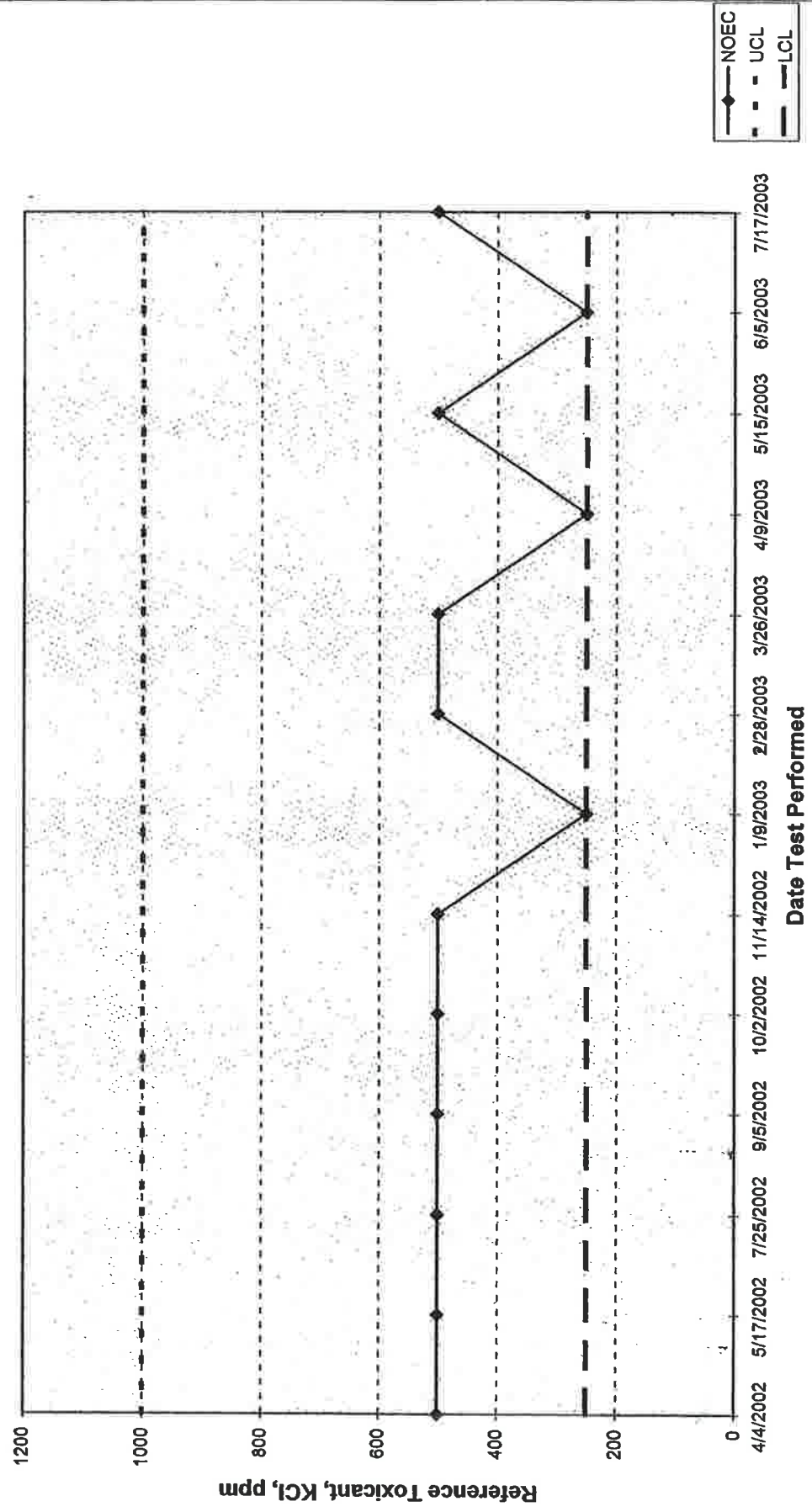
## APPENDIX F

### Quality Assurance Charts

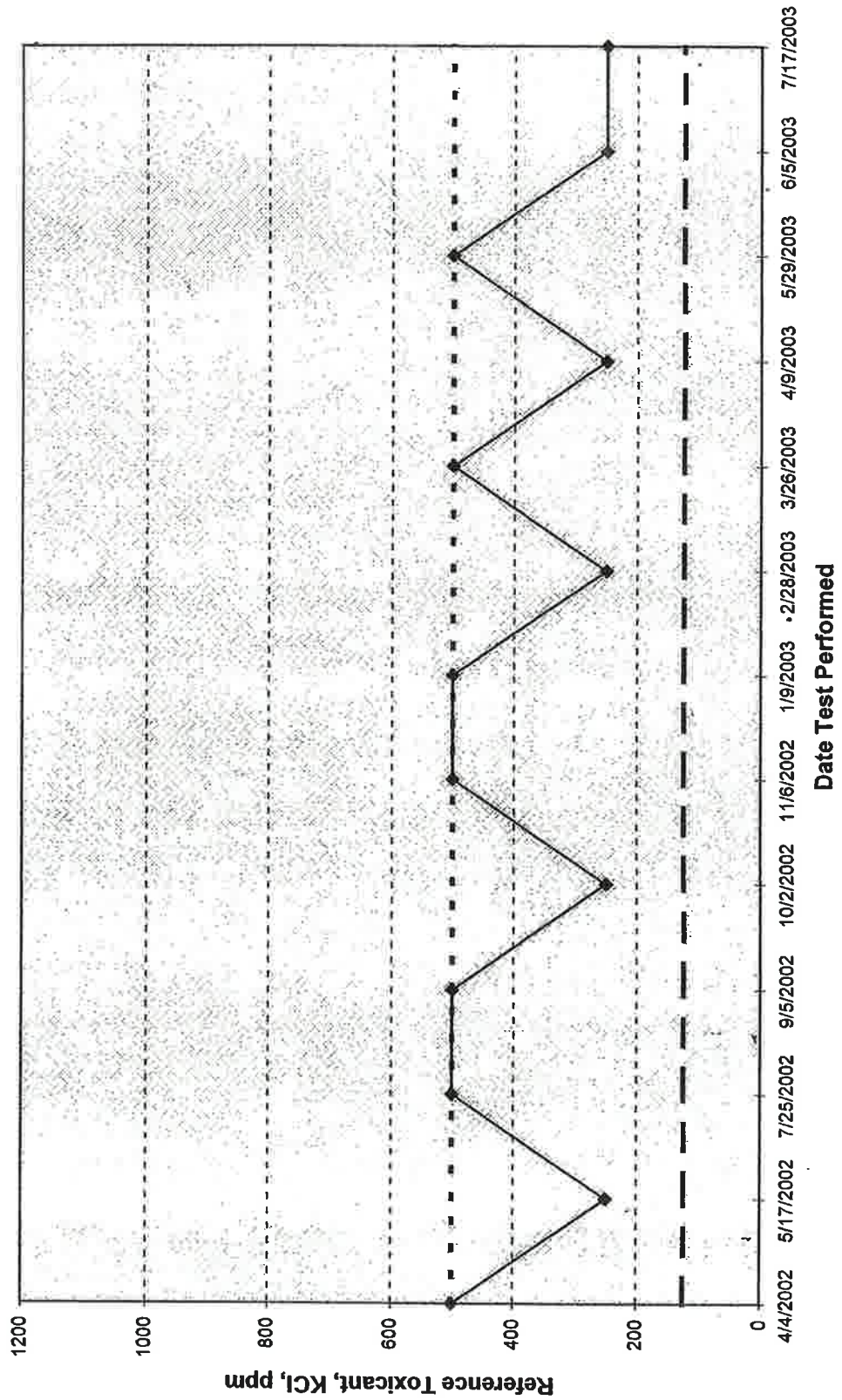
**ARKANSAS ANALYTICAL, INC.**  
**FATHEAD MINNOW SURVIVAL**  
**QUALITY ASSURANCE**



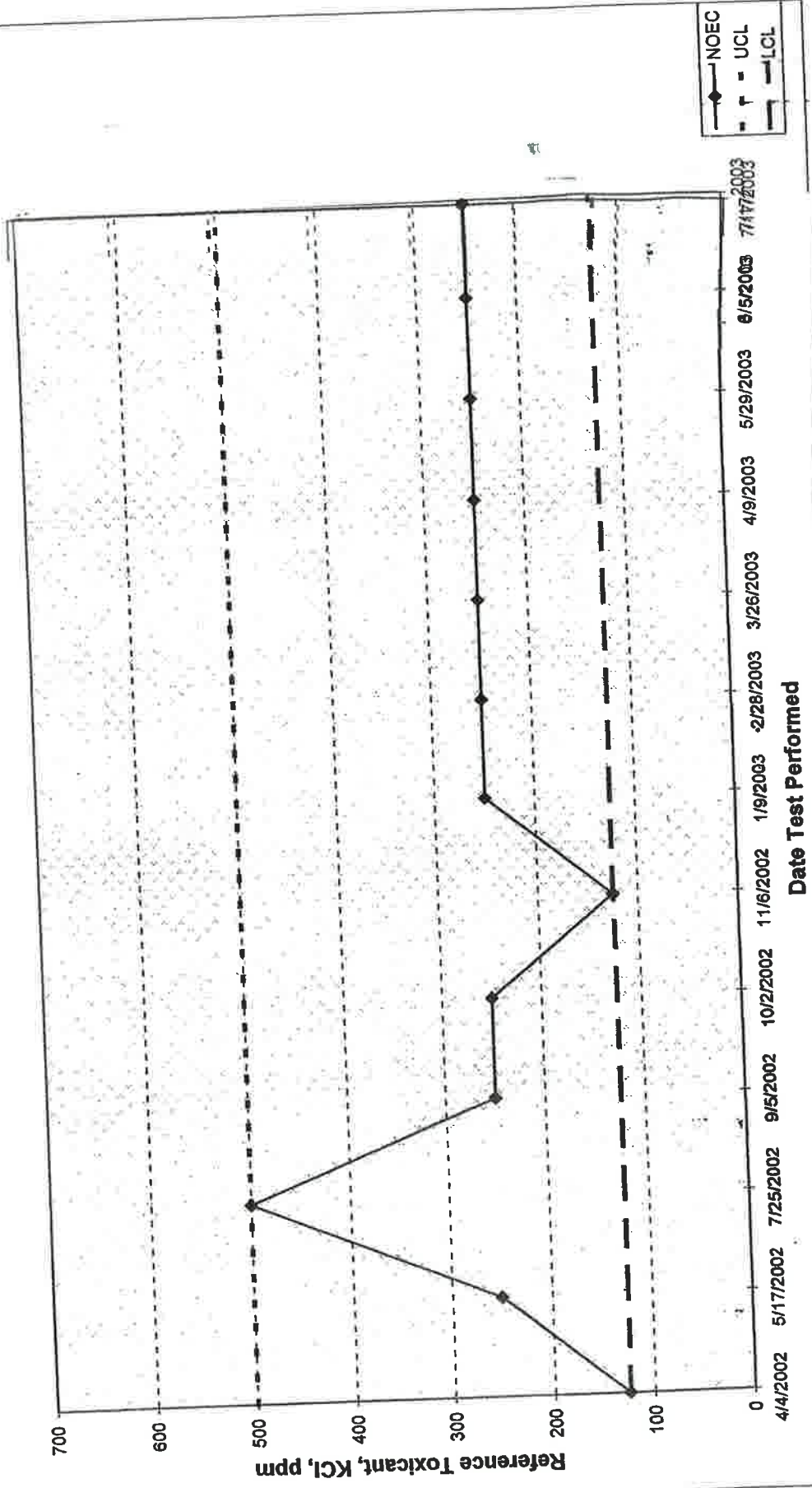
**ARKANSAS ANALYTICAL, INC.**  
**FATHEAD MINNOW GROWTH**  
**QUALITY ASSURANCE**



**ARKANSAS ANALYTICAL, INC.**  
**CERIODAPHNIA DUBIA SURVIVAL**  
**QUALITY ASSURANCE**



**ARKANSAS ANALYTICAL, INC.**  
**CERIODAPHNIA DUBIA REPRODUCTION**  
**QUALITY ASSURANCE**



—◆— NOEC  
 - - - UCL  
 - - - LCL

Date Test Performed

APPENDIX G

Lab Certification





State of Arkansas  
 Department of Environmental Quality  
 Laboratory Certification Program



Be it known that  
 Arkansas Analytical, Inc  
 Little Rock, Arkansas  
 has earned certification by this Department for the period of

May 14, 2003 to October 30, 2003

Laboratory ID # 60-1754

Certificate # 03-031-1

The following parameters are certified:

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

Date May 15 2003

*J.A. Semberick*  
 Quality Assurance Officer