



Ecotoxicology Research Facility



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March 9, 2015

Jonathan Kopp
Walnut Ridge Wastewater Treatment Plant
216 Southwest 4th Street
Walnut Ridge, AR 72476

Dear Jon,

Please find enclosed the results of the 7-day chronic tests using water collected from Walnut Ridge's plant facilities during the week of February 24, 2015. No lethal or sublethal effects were measured in *Ceriodaphnia dubia* exposed to the critical flow concentration (100%) or other treated dilutions from this outfall.

All test conditions and acceptability criteria as suggested by our laboratory and the US EPA were met during these tests.

Please call if you have any questions regarding this particular test series or any other tests conducted in the past.

Sincerely,

Jennifer L. Bouldin, PhD
Director Ecotoxicology Research Facility
PO Box 847
Arkansas State University
State University, AR 72467

Facility Director / Contact: Jennifer L. Bouldin, PhD
Phone: (870) 972-2570

Client: Walnut Ridge Wastewater Treatment Plant
216 Southwest 4th Street
Walnut Ridge, AR 72476

Contact: Jon Kopp
870-866-2312

NPDES Permit #: AR0046566 AFIN#: 38-00040

Effluent Sampling Point/Type: 24hr Composite

Samples Collected:

Sample #	Sampling Times	Received	Arrival Temp
1	02/24/15 0900 hrs to 02/25/15 0900 hrs	02/25/15 1150 hrs	0.0°C
2	02/26/15 0900 hrs to 02/27/15 0900 hrs	02/27/15 1121 hrs	-0.9°C
3	03/01/15 0900 hrs to 03/02/15 0900 hrs	03/02/15 1118 hrs	1.9°C

Test Methods:

7-Day Chronic Toxicity, Static renewal, Cladoceran, EPA 821/R-02/013, Section 13

Organisms: *C. dubia* <24hrs

Culture Source: ASU ERF

Dilutions: 0%, 32%, 42%, 56%, 80%, 100%

Critical Dilution: 100%

Statistical Method: Toxcalc 5.0.25

Results:

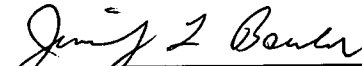
C. dubia

NOEC Survival:	100%
Pass/Fail (0=pass; 1=fail):	0
NOEC Growth/Reproduction:	100%
Pass/Fail (0=pass; 1=fail):	0
Control Survival:	100%
Control % CV Growth/Reproduction:	28.8
Critical Dilution % CV Growth/Reproduction:	23.8
Mean Weight/ # Neonates in Control:	18.9
Mean Weight/ # of Neonates in Critical Dilution:	17.7
MSDp Growth/ Reproduction	0.2921
Daily Average Minimum NOEC:	100%
7-Day Minimum NOEC:	100%

Results Summary: Neither lethal nor sublethal effects were measured in *C. dubia* exposed to treated effluent or dilution mixtures.

QA/Reference Testing: Data attached

Reviewed By:


Jennifer L. Bouldin, PhD
Director ASU Ecotoxicology Research Facility

Toxicity Test Performed: 7-day *Ceriodaphnia dubia* Survival and Reproduction
 Effluent Sampling Point: Walnut Ridge WWT Plant
 Date Test Started: 02/25/15 *C. dubia*
 Time Test Started: 1335 *C. dubia*
 Date Test Terminated: 03/04/15 *C. dubia*
 Time Test Terminated: 1125 *C. dubia*
 Laboratory Analyst: Griffin/Bouldin

I. Test Methods

A. Physical and Chemical Testing - APHA, Standard Methods for the Examination of Water and Wastewater; Vol. 21, 2005.

<u>Test</u>	<u>Method</u>
Alkalinity	2320B
Conductivity	2510B
Dissolved Oxygen (mg/L, DO)	4500-O-G
Hardness (mg/L CaCO ₃)	2340C
pH	4500-H ⁺ B
Temperature (°C)	2550B

B. Toxicity Testing – EPA 821/R-02/013: Short Term Methods for Estimating the Chronic Toxicity of Effluents to Freshwater Organisms

<u>Test</u>	<u>Method</u>
Cladoceran Survival and Reproduction	Section 13

II. Test Organisms

A. Name: *Ceriodaphnia dubia* (Cladoceran)
 Source: Laboratory Culture
 Age: <24 hours
 Life Stage: Neonate

III. External Factors

A. Incubator

Temperature (°C)

Average: Average: 24.6

Range: 24.5 – 24.8

Light Cycle: 16 hours light/ 8 hours dark

Light Intensity: 100 footcandles

Control Water: Moderately Hard Synthetic Water (#935)

B. *Ceriodaphnia dubia*

Test Chambers: 30 ml Solo cups

Volume per Chamber: 15-20 ml

Number of Organisms per Chamber: 1

Number of Replicates per Concentration: 10

Acclimation: Laboratory control water was added to cultures until >50% of the culture water consisted of control water.

Food: Cladocera were fed *Selenastrum* (#ABS 011915 & 012615) and yeast/cereal/trout chow mix (#YCT 122214-11/12/13) one hour prior to test setup and once daily thereafter.

IV. Quality Assurance

A. Standard Toxicant: Sodium Chloride

B. Organism: *Ceriodaphnia dubia*

Date and time of Reference Toxicant Test

Start: 02/22/15

Terminated: 03/03/15

Time of Reference Toxicant Test

Start: 1405

Terminated: 1355

Laboratory Analyst: McCauley/Vogt

Dilution Water Used: Moderately Hard Synthetic Water #934/935

Results: Survival and Reproduction within control limits

	<u>Survival</u>	<u>Reproduction</u>
LOEC:	1.82 g/L NaCl	0.62 g/L NaCl
EC50:	1.94 g/L NaCl	0.71 g/L NaCl

V. Physical and Chemical Data - See Attached

VI. Survival and Growth Data - See Attached

VII. Statistical Methods - See Attached

SUMMARY REPORTING FORM
WET Testing
Ceriodaphnia dubia Survival and Reproduction

Permittee: Walnut Ridge WWT Plant

NPDES No.: AR0046566

		<u>Time</u>	<u>Date</u>	to	<u>Time</u>	<u>Date</u>
Composite 1:	Collected from	0900	02/24/15		0900	02/25/15
Composite 2:	Collected From	0900	02/26/15		0900	02/27/15
Composite 3:	Collected From	0900	03/01/15		0900	03/02/15

Test Initiated: 1335

Date: 02/25/15

Time Terminated: 1125

Date: 03/04/15

Dilution H₂O: MH 935

PERCENT SURVIVAL
Percent Effluent

<u>Time of Reading</u>	<u>Control</u>	<u>32%</u>	<u>42%</u>	<u>56%</u>	<u>80%</u>	<u>100%</u>
24h	100	100	100	100	100	100
48h	100	100	100	100	100	100
7 day	100	90	100	90	100	100

NUMBER OF YOUNG/FEMALE @ 7 DAYS
Percent Effluent

<u>REP</u>	<u>0%</u>	<u>32%</u>	<u>42%</u>	<u>56%</u>	<u>80%</u>	<u>100%</u>
A	26	16	20	25	26	17
B	21	15	17	22	8	11
C	20	13	19	19	18	19
D	20	13	12	12	26	23
E	18	8	15	13	14	19
F	27	0	17	17	9	18
G	16	18	14	5	15	12
H	8	15	17	21	21	22
I	15	15	20	7	15	22
J	18	22	21	11	13	14
Mean	18.9	13.5	17.2	15.2	16.5	17.7
CV%*	28.8	44.1	16.9	43.7	38.0	23.8

*Coefficient of Variation% = Standard Deviation x 100/Mean

***Ceriodaphnia dubia* Survival and Reproduction**

1. FISHER'S EXACT TEST:
Is the mean survival for the critical dilution (100%) at 7 days significantly different ($p=0.05$) than the control survival?
 Yes X No

2. DUNNETT'S PROCEDURE OR STEEL'S MANY-ONE RANK TEST AS APPROPRIATE:
Is the mean number of young produced per female by the critical dilution (100%) significantly different ($p=0.05$) than the control's number of young per female?
 Yes X No

3. If the NOEC for survival is less than the critical dilution, enter [1], otherwise enter [0] for parameter #TGP3B: 0

4. If the NOEC for reproduction is less than the critical dilution, enter [1], otherwise enter [0] for parameter #TLP3B: 0

5. Report the NOEC value for survival, Parameter #TOP3B:
NOEC survival 100 % effluent

6. Report the NOEC value for reproduction, Parameter #TPP3B:
NOEC reproduction 100 % effluent

7. Report the % coefficient of variation (largest of critical and control dilutions), Parameter #TQP3B:
CV % reproduction 28.8% (control)

Whole Effluent Lethality Values for *Ceriodaphnia dubia*

1. Report the Whole Effluent Lethality values for the 30-Day average minimum, Parameter #22414:
Daily Average Minimum NOEC: 100%

2. Report the Whole Effluent Lethality values for the 7-day minimum, Parameter #22414:
7-Day Minimum NOEC: 100%

CHRONIC TOXICITY SUMMARY FORM
WET Testing *Ceriodaphnia dubia* (Cladoceran)
CHEMICAL PARAMETERS CHART

Permittee: Walnut Ridge WWT Plant

Sample No. 1 Collected Ending Date: 02/25/15 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 02/27/15 Time: 0900

Contact: Jon Kopp

Sample No. 3 Collected Ending Date: 03/02/15 Time: 0900

Analyst: Griffin/Bouldin

Test Begin: Date: 02/25/15 Time: 1335 Test End: Date: 03/04/15 Time: 1125

Initial Water Chemistry for Chronic Tests								
Project: Walnut Ridge – <i>C. dubia</i>								
Test day		0	1	2	3	4	5	6
Date		2/25/2015	2/26/2015	2/27/2015	2/28/2015	3/1/2015	3/2/2015	3/3/2015
H ₂ O #		MH 935	MH 935	MH 935	MH 935	MH 935	MH 935	MH 935
Temp (°C)	Control	22.5	22.5	23.0	22.5	22.2	23.1	23.1
	32%	22.5	22.5	23.2	22.6	22.2	23.1	23.1
	42%	22.5	22.7	23.0	22.5	22.2	23.1	23.1
	56%	22.5	22.7	23.0	22.5	22.2	23.1	23.1
	80%	22.5	22.8	23.0	22.5	22.2	23.1	23.1
	100%	22.5	22.6	23.0	22.5	22.1	23.1	23.1
pH (Standard Units)	Control	7.98	8.38	7.98	8.05	8.07	7.92	8.11
	32%	8.00	7.71	7.87	7.84	7.74	7.64	7.99
	42%	8.02	7.70	7.85	7.74	7.84	7.70	7.96
	56%	8.00	7.71	7.81	7.81	7.85	7.68	7.93
	80%	7.98	7.71	7.78	7.77	7.82	7.63	7.88
	100%	7.97	7.71	7.73	7.76	7.78	7.58	7.83
DO (mg/L)	Control	9.0	9.2	9.1	8.6	8.6	8.5	8.5
	32%	9.0	8.9	8.4	8.7	8.5	8.6	8.3
	42%	8.9	8.8	8.5	8.7	8.3	8.4	8.3
	56%	8.9	8.7	8.5	8.7	8.3	8.5	8.2
	80%	8.9	8.7	8.5	8.7	8.3	8.6	8.2
	100%	8.9	8.6	8.6	8.7	8.3	8.8	8.2
Cond (µS/cm)	Control	288	304	290	289	293	300	293
	32%	293	297	331	326	330	348	357
	42%	295	299	343	339	343	368	377
	56%	297	301	361	357	361	395	406
	80%	300	304	384	403	394	443	458
	100%	306	310	420	418	423	485	504
Alk (mg/L)	Control	57		57			57	
	100%	52		72			82	
Hard (mg/L)	Control	100		100			100	
	100%	110		180			200	

CHRONIC TOXICITY SUMMARY FORM
WET Testing *Ceriodaphnia dubia* (Cladoceran)

CHEMICAL PARAMETERS CHART

Permittee: Walnut Ridge WWT Plant

Sample No. 1 Collected Ending Date: 02/25/15 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 02/27/15 Time: 0900

Contact: Jon Kopp

Sample No. 3 Collected Ending Date: 03/02/15 Time: 0900

Analyst: Griffin/Bouldin

Test Begin: Date: 02/25/15 Time: 1335 Test End: Date: 03/04/15 Time: 1125

Final Water Chemistry for Chronic Tests

Project: Walnut Ridge - *C. dubia*

Test day		1	2	3	4	5	6	7
Date:		2/26/2015	2/27/2015	2/28/2015	3/1/2015	3/2/2015	3/3/2015	3/4/2015
H ₂ O #		MH 935	MH 935	MH 935	MH 935	MH 935	MH 935	MH 935
Temp (°C)	Control	23.0	22.0	22.4	22.2	23.5	23.0	24.0
	32%	23.0	22.5	22.3	22.1	23.4	22.9	23.9
	42%	22.2	22.2	22.4	22.1	23.5	23.0	23.9
	56%	22.5	22.5	22.4	22.1	23.5	23.0	24.0
	80%	22.6	22.3	22.3	22.2	23.6	22.9	24.0
	100%	23.0	22.7	22.4	22.1	23.6	22.9	23.9
pH (Standard Units)	Control	8.17	8.20	8.20	8.37	8.20	8.22	8.67
	32%	8.29	8.39	8.29	8.44	8.58	8.20	8.52
	42%	8.27	8.40	8.29	8.45	8.57	8.26	8.54
	56%	8.28	8.39	8.30	8.44	8.68	8.22	8.63
	80%	8.25	8.38	8.29	8.47	8.62	8.25	8.46
	100%	8.28	8.40	8.33	8.43	8.70	8.24	8.28
DO (mg/L)	Control	9.4	9.3	9.0	9.0	9.0	8.7	9.4
	32%	9.2	9.0	9.1	9.4	9.8	8.5	9.5
	42%	9.3	9.3	9.1	9.4	9.6	8.5	9.2
	56%	9.3	9.3	9.0	9.4	9.6	8.5	9.6
	80%	9.3	9.3	9.0	9.2	9.7	8.5	9.6
	100%	9.3	9.3	9.1	9.0	9.6	8.5	9.7

Ceriodaphnia Survival and Reproduction Test-7 Day Survival

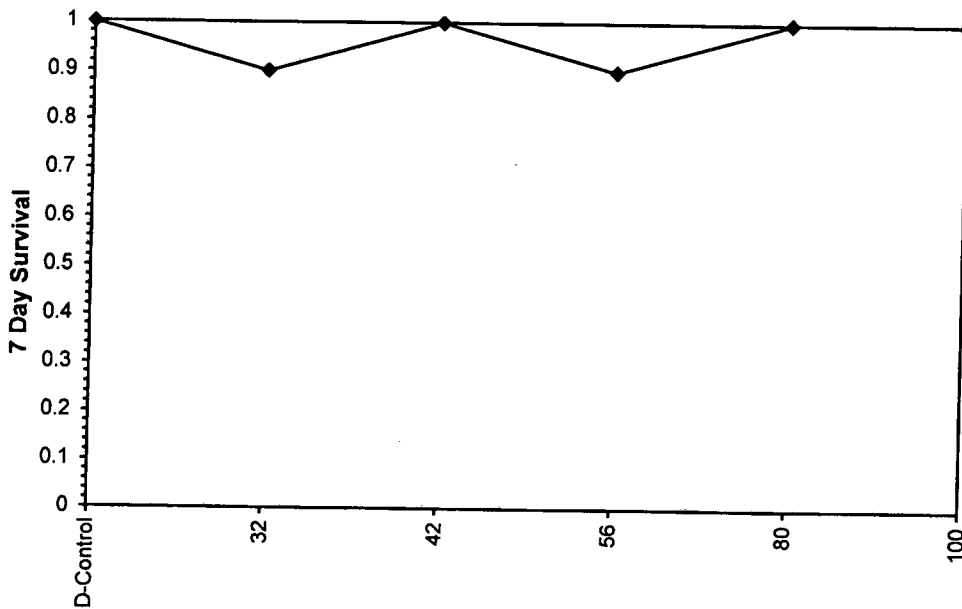
Start Date: 2/25/2015 13:35 Test ID: WR Sample ID: NPDES Permit # AR0046566
 End Date: 3/4/2015 11:25 Lab ID: ASU-ERF Sample Type: EFF1-POTW
 Sample Date: 2/25/2015 Protocol: EPAF 02-EPA Freshwater Test Species: CD-Ceriodaphnia dubia
 Comments:

Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
32	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000
42	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
56	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000
80	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
100	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000

Conc-%	Mean	N-Mean	Resp	Not Resp	Total	N	Fisher's Exact P	1-Tailed Critical
D-Control	1.0000	1.0000	0	10	10	10		
32	0.9000	0.9000	1	9	10	10	0.5000	0.0500
42	1.0000	1.0000	0	10	10	10	1.0000	0.0500
56	0.9000	0.9000	1	9	10	10	0.5000	0.0500
80	1.0000	1.0000	0	10	10	10	1.0000	0.0500
100	1.0000	1.0000	0	10	10	10	1.0000	0.0500

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Fisher's Exact Test	100	>100		1

Dose-Response Plot



Ceriodaphnia Survival and Reproduction Test-Reproduction

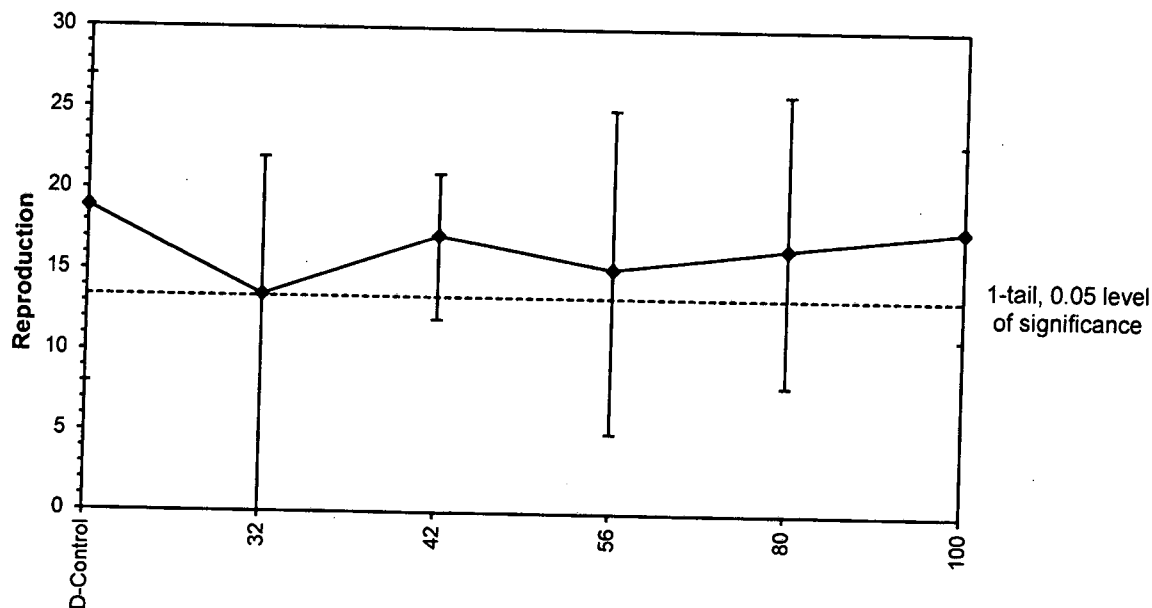
Start Date: 2/25/2015 13:35 Test ID: WR Sample ID: NPDES Permit # AR0046566
 End Date: 3/4/2015 11:25 Lab ID: ASU-ERF Sample Type: EFF1-POTW
 Sample Date: 2/25/2015 Protocol: EPAF 02-EPA Freshwater Test Species: CD-Ceriodaphnia dubia
 Comments:

Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	26.000	21.000	20.000	20.000	18.000	27.000	16.000	8.000	15.000	18.000
32	16.000	15.000	13.000	13.000	8.000	0.000	18.000	15.000	15.000	22.000
42	20.000	17.000	19.000	12.000	15.000	17.000	14.000	17.000	20.000	21.000
56	25.000	22.000	19.000	12.000	13.000	17.000	5.000	21.000	7.000	11.000
80	26.000	8.000	18.000	26.000	14.000	9.000	15.000	21.000	15.000	13.000
100	17.000	11.000	19.000	23.000	19.000	18.000	12.000	22.000	22.000	14.000

Conc-%	Mean	N-Mean	Transform: Untransformed				N	t-Stat	1-Tailed	
			Mean	Min	Max	CV%			Critical	MSD
D-Control	18.900	1.0000	18.900	8.000	27.000	28.813	10			
32	13.500	0.7143	13.500	0.000	22.000	44.066	10	2.237	2.287	5.520
42	17.200	0.9101	17.200	12.000	21.000	16.850	10	0.704	2.287	5.520
56	15.200	0.8042	15.200	5.000	25.000	43.728	10	1.533	2.287	5.520
80	16.500	0.8730	16.500	8.000	26.000	38.037	10	0.994	2.287	5.520
100	17.700	0.9365	17.700	11.000	23.000	23.829	10	0.497	2.287	5.520

Auxiliary Tests		Statistic	Critical	Skew	Kurt						
Kolmogorov D Test indicates normal distribution (p > 0.01)		0.53099	1.035	-0.27883	0.02998						
Bartlett's Test indicates equal variances (p = 0.22)		6.95992	15.0863								
Hypothesis Test (1-tail, 0.05)		NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test		100	>100		1	5.51967	0.29205	36.76	29.1333	0.29384	5, 54

Dose-Response Plot



CHRONIC TEST DATA SHEET

Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 022515 Time: 1335 Test Species: C. dubia
 Dilution H₂O: MH935 Ending Date: 030415 Time: 1125 Age: 224h

Test Type: (*)Static Renewal () Flowthrough Toxicant/Effluent: _____

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
Control	1	0	0	0	6	0	5	15	26
	2				5	0	2	14	21
	3				2	5	0	13	20
	4				4	0	3	13	20
	5				5	0	0	13	18
	6				2	6	1	18	27
	7				4	0	0	12	16
	8				0	2	0	6	8
	9				4	2	0	9	15
	10	↓	↓	↓	3	3	1	11	18
32%	1	0	0	0	0	6	0	10	16
	2				0	0	4	11	15
	3				1	7	0	5	13
	4				0	3	0	10	13
	5				0	0	0	8	8
	6				X/0	-----			X/0
	7				0	0	8	10	18
	8				0	4	5	6	15
	9				0	2	0	13	15
	10	↓	↓	↓	0	0	10	12	22
Date		022615	022715	022815	030115	030215	030315	030415	
Initials		MG	MG	JB	JB	JB	JB	JB	

CHRONIC TEST DATA SHEET
Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 022515 Time: 1335 Test Species: C.dubia
Dilution H₂O: MH935 Ending Date: 030415 Time: 1125 Age: <24h

Test Type: (*)Static Renewal () Flowthrough Toxicant/Effluent: _____

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
42%	1	0	0	0	0	2	8	10	20
	2	↓	↓	↓	3	5	0	9	19
	3	↓	↓	↓	0	5	3	11	19
	4	↓	↓	↓	0	4	0	8	12
	5	↓	↓	↓	1	0	2	12	15
	6	↓	↓	↓	3	6	0	8	17
	7	↓	↓	↓	3	6	0	5	14
	8	↓	↓	↓	0	0	7	10	17
	9	↓	↓	↓	4	0	6	10	20
	10	↓	↓	↓	0	2	8	11	21
56%	1	0	0	0	5	3	9	8	25
	2	↓	↓	↓	1	3	8	10	22
	3	↓	↓	↓	3	0	8	8	19
	4	↓	↓	↓	1	0	0	11	12
	5	↓	↓	↓	1	0	0	12	13
	6	↓	↓	↓	0	5	0	12	17
	7	↓	↓	↓	0	2	0	3	5
	8	↓	↓	↓	0	4	8	9	21
	9	↓	↓	↓	0	2	0	5	7
	10	↓	↓	↓	0	1	10	X/0	X/11
Date		022615	022715	022815	030115	030215	030315	030415	
Initials		MB	MB	B	B	B	B	B	B

CHRONIC TEST DATA SHEET
Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 022515 Time: 1335 Test Species: C. dubia

Dilution H₂O: MH035 Ending Date: 030415 Time: 1125 Age: 224h

Test Type: (*)Static Renewal () Flowthrough Toxicant/Effluent: _____

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
80%	1	0	0	0	5	0	8	13	26
	2	↓	↓	↓	0	5	3	0	8
	3	↓	↓	↓	3	5	0	10	18
	4	↓	↓	↓	0	7	7	12	26
	5	↓	↓	↓	0	2	0	12	14
	6	↓	↓	↓	0	0	4	5	9
	7	↓	↓	↓	4	3	8	0	15
	8	↓	↓	↓	0	6	0	15	21
	9	↓	↓	↓	0	7	0	8	15
	10	↓	↓	↓	0	0	3	10	13
100%	1	0	0	0	0	0	5	12	17
	2	↓	↓	↓	0	4	1	6	11
	3	↓	↓	↓	0	0	9	10	19
	4	↓	↓	↓	3	0	8	12	23
	5	↓	↓	↓	0	0	8	11	19
	6	↓	↓	↓	2	3	0	13	18
	7	↓	↓	↓	0	2	7	3	12
	8	↓	↓	↓	0	0	7	15	22
	9	↓	↓	↓	0	2	8	12	22
	10	↓	↓	↓	0	0	0	14	14
Date		022615	022715	022815	023015	030215	030315	030415	
Initials		MG	MG	g	g	g	g	g	g

Initial Water Chemistry for Chronic Tests
Project: Walnut Ridge - *C. dubia*

Test Day:		0	1	2	3	4	5	6
Date:		022515	022615	022715	022815	030115	030415	030315
H ₂ O Batch #:		MH935	MH935	MH935	MH935	MH935	MH935	MH935
Temp. (°C)	Control	22.5	22.5	23.0	22.5	22.2	23.1	23.1
	32%	22.5	22.5	23.2	22.6	22.2	23.1	23.1
	42%	22.5	22.7	23.0	22.5	22.2	23.1	23.1
	56%	22.5	22.7	23.0	22.5	22.2	23.1	23.1
	80%	22.5	22.8	23.0	22.5	22.2	23.1	23.1
	100%	22.5	22.4	23.0	22.5	22.1	23.1	23.1
pH	Control	7.98	8.38	7.98	8.05	8.07	7.92	8.11
	32%	8.00	7.71	7.87	7.84	7.74	7.64	7.99
	42%	8.02	7.70	7.85	7.74	7.84	7.70	7.96
	56%	8.00	7.71	7.81	7.81	7.85	7.68	7.93
	80%	7.98	7.71	7.78	7.77	7.82	7.63	7.88
	100%	7.97	7.71	7.73	7.76	7.78	7.58	7.83
DO (mg/L)	Control	9.0	9.2	9.1	8.6	8.6	8.5	8.5
	32%	9.0	8.9	8.4	8.5	8.5	8.6	8.3
	42%	8.9	8.8	8.5	8.7	8.3	8.4	8.3
	56%	8.9	8.7	8.5	8.7	8.3	8.5	8.2
	80%	8.9	8.7	8.5	8.7	8.3	8.6	8.2
	100%	8.9	8.6	8.6	8.7	8.3	8.8	8.2
Cond. (µS/cm)	Control	288	304	290	289	293	300	293
	32%	293	297	331	339 ²²⁶	330	348	357
	42%	295	299	343	339	343	368	377
	56%	297	301	361	357	361	395	406
	80%	300	304	384	403	394	443	458
	100%	306	310	420	418	423	485	504
Alk. (mg/L)	Control	57		57			57	
	100%	52		72			82	
Hard. (mg/L)	Control	100		100			100	
	100%	110		180			200	
Initials		MG	MG	MG	P	P	P	P

Final Water Chemistry for Chronic Tests
Project: Walnut Ridge - *C. dubia*

Test Day:		1	2	3	4	5	6	7
Date:		022615	022715	022815	030115	030215	030315	031415
H ₂ O Batch #:		MH935	MH935	MH935	MH935	MH935	MH935	MH935
Temp. (°C)	Control	23.0	22.0	22.4	22.2	23.5	23.0	24.0
	32%	23.0	22.5	22.3	22.1	23.4	22.9	23.9
	42%	22.2	22.2	22.4	22.1	23.5	23.0	23.9
	56%	22.5	22.5	22.4	22.1	23.5	23.0	24.0
	80%	22.4	22.3	22.3	22.2	23.6	22.9	24.0
	100%	23.0	22.7	22.4	22.1	23.6	22.5	23.9
pH	Control	8.17	8.20	8.20	8.27	8.20	8.22	8.67
	32%	8.29	8.39	8.29	8.44	8.58	8.20	8.52
	42%	8.27	8.40	8.29	8.45	8.57	8.26	8.54
	56%	8.29	8.39	8.30	8.44	8.68	8.22	8.63
	80%	8.25	8.38	8.29	8.47	8.62	8.25	8.46
	100%	8.28	8.40	8.33	8.43	8.70	8.28	8.28
DO (mg/L)	Control	9.4	9.3	9.0	9.0	9.0	8.58	9.4
	32%	9.2	9.0	9.1	9.4	9.8	8.5	9.5
	42%	9.3	9.3	9.1	9.4	9.6	8.5	9.2
	56%	9.3	9.3	9.0	9.4	9.6	8.5	9.6
	80%	9.3	9.3	9.0	9.2	9.7	8.5	9.6
	100%	9.3	9.3	9.1	9.0	9.6	8.5	9.7
Initials		MG	MG	8	8	8	8	8



Ecotoxicology Research Facility

SAMPLE CHECK IN

Sample ID Number: 1

Fill out this information with each effluent or river water sample coming in for testing. Keep completed sheets with test data and file with the lab QA/QC officer.

Date: 022515 Sampling Date: 022415 Arrival Time: 1150

Field Identification Number: _____ Description: effluent

Shipped by: Federal Express _____ UPS _____ Hand delivered by: Walnut Ridge personnell

Drop-Off Location: ASU-ERF

Storage While Shipped: Cooler w/ice

Analysis Requested: C.dubia

Initial Water Chemistry Analysis:

Sample Received by: T. Woodruff

Temperature (°C): 0.0

Ice Present upon delivery: YES NO

Date: 022515

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	TW	022515	✓	
Refrigerated at 4°C	TW	022515	✓	
Field Record Received	TW	022515		✓
Sample Label Affixed Properly	TW	022515	✓	
Project Leader Informed	TW	022515	✓	

Comments: _____



SAMPLE CHECK IN

Sample ID Number: WR #2

Fill out this information with each effluent or river water sample coming in for testing. Keep completed sheets with test data and file with the lab QA/QC officer.

Date: 022715 Sampling Date: 02/26 - 02/27 Arrival Time: 1121

Field Identification Number: _____ Description: Effluent

Shipped by: Federal Express _____ UPS _____ Hand delivered by: WR personnel

Drop-Off Location: ASU-ERF

Storage While Shipped: cooler w/ ice

Analysis Requested: chronic C. dubia

Initial Water Chemistry Analysis:

Sample Received by: SV

Temperature (°C): -0.9 Ice Present upon delivery: YES NO

Date: 022715

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	SV	022715	✓	
Refrigerated at 4°C	↓	↓	✓	
Field Record Received				✓
Sample Label Affixed Properly	↓	↓	✓	
Project Leader Informed	↓	↓	✓	

Comments: _____



Ecotoxicology Research Facility

SAMPLE CHECK IN

Sample ID Number: WR #3

Fill out this information with each effluent or river water sample coming in for testing. Keep completed sheets with test data and file with the lab QA/QC officer.

Date: 3/2/15 Sampling Date: 3/1 → 3/2/15 Arrival Time: 1118

Field Identification Number: _____ Description: POTW effluent

Shipped by: Federal Express _____ UPS _____ Hand delivered by: WR person

Drop-Off Location: ASU-ERF

Storage While Shipped: cooled w/ ice

Analysis Requested: chemi c data

Initial Water Chemistry Analysis:

Sample Received by: JS

Temperature (°C): 1.9 Ice Present upon delivery: YES NO

Date: 3/2/15

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	JS	3/2/15	—	
Refrigerated at 4°C	JS	3/2/15	—	
Field Record Received	JS	3/2/15		—
Sample Label Affixed Properly	JS	3/2/15	—	
Project Leader Informed	JS	3/2/15	—	

Comments: _____



Ecotoxicology Research Facility

Ecotoxicology Research Facility

Arkansas State University

2645 Caddo Drive

State University, AR 72467

(870) 972-2570 Fax (870) 972-2577

CHAIN OF CUSTODY RECORD



Client Name Walnut Ridge Wastewater Treatment			Phone: (870) 886-2312							Analyses (List Below)											
Project #			Fax:																		
Sampler (sign) <i>[Signature]</i>			PO #:							<table border="1"> <tr> <td>Chronic <i>C. dubia</i></td> <td>Chronic <i>P. promelas</i></td> <td></td> <td></td> </tr> <tr> <td></td> <td></td> <td></td> <td></td> </tr> </table>				Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>						
Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>																				
Remarks: Contact: Jonathan Kopp			Sample Date																		
Cont. #	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix														
					Comp	Grab	Aqueous	Soil	Other												
1			2-24-15	9 A.M.	X					X											
			2-25-15	9 A.M.																	
Ice present at delivery:		<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No																			
Temp:		0.0 °C		W initials																	
1. Relinquished By (sign) <i>[Signature]</i>			Date	Time	1. Received By (sign)				Date	Time											
2. Relinquished By (sign) <i>[Signature]</i>			Date	Time	2. Received By (sign) <i>[Signature]</i>				Date	Time											



Ecotoxicology Research Facility

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Arkansas State University

501 Iroquois Street

State University, AR 72467

(870) 972-2570 Fax (870) 972-2577

CHAIN OF CUSTODY RECORD



Client Name Walnut Ridge Wastewater Treatment			Phone: (870) 886-2312					Analyses (List Below)							
Project #			Fax:									Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>		
Sampler (sign) 			PO #:												
Remarks:			Contact: Jonathan Kopp												
Cont. #	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix								
					Comp	Grab	Aqueous	Soil	Other						
			2-26-15	9:20am	X										
			2-27-15	9:40am								X			
Ice present at delivery: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No															
Temp: 70.9°C			SV		Initials										
1. Relinquished By (sign) 			Date 2-27-15		Time 9:30 A.M.		1. Received By (sign)				Date		Time		
2. Relinquished By (sign) 			Date 022715		Time 1121		2. Received By (sign) 				Date		Time		



Ecotoxicology Research Facility

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Arkansas State University

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CHAIN OF CUSTODY RECORD



Client Name Walnut Ridge Wastewater Treatment			Phone: (870) 886-2312			Analyses (List Below)										
Project #			Fax:													
Sampler (sign) <i>[Signature]</i>			PO #:			<table border="1"> <tr> <td>Chronic <i>C. dubia</i></td> <td>Chronic <i>P. promelas</i></td> <td></td> <td></td> </tr> <tr> <td></td> <td></td> <td></td> <td></td> </tr> </table>			Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>						
Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>															
Remarks: Contact: Jonathan Kopp			Sample Date			Sample Time										
Cont.#	Sample ID Number	Location	Sample Type		Matrix											
			Comp	Grab	Aqueous	Soil	Other									
			X					X								
Ice present at delivery:		<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No														
Temp:		19°C		Initials												
1. Relinquished By (sign) <i>[Signature]</i>		Date	3-2-15	Time	11:18	1. Received By (sign) <i>[Signature]</i>		Date	3/2/15							
2. Relinquished By (sign)		Date		Time		2. Received By (sign)		Date								
								Time	1118							



Ecotoxicology Research Facility



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College of Sciences & Mathematics
www.astate.edu

February 26, 2015

Jonathan Kopp
Walnut Ridge Wastewater Treatment Plant
216 Southwest 4th Street
Walnut Ridge, AR 72476

Dear Jon,

Please find enclosed the results of the 7-day chronic tests using water collected from Walnut Ridge's plant facilities during the week of February 1, 2015. No lethal or sublethal effects were measured in *Pimephales promelas* exposed to the critical flow concentration (100%) or other treated dilutions from this outfall. However, due to low survival (50%) in the controls, the *Ceriodaphnia dubia* test has been deemed invalid and will need to be repeated.

With the exception of the invalid *C. dubia* test, all test conditions and acceptability criteria as suggested by our laboratory and the US EPA were met during these tests.

Please call if you have any questions regarding this particular test series or any other tests conducted in the past.

Sincerely,

Jennifer L. Bouldin, PhD
Director Ecotoxicology Research Facility
PO Box 847
Arkansas State University
State University, AR 72467

Facility Director / Contact: Jennifer L. Bouldin, PhD
Phone: (870) 972-2570

Client: Walnut Ridge Wastewater Treatment Plant
216 Southwest 4th Street
Walnut Ridge, AR 72476

Contact: Jon Kopp
870-866-2312

NPDES Permit #: AR0046566 AFIN#: 38-00040
Effluent Sampling Point/Type: 24hr Composite
Samples Collected:

Sample #	Sampling Times	Received	Arrival Temp
1	02/01/15 0900 hrs to 02/02/15 0900 hrs	02/02/15 1115 hrs	2.0°C
2	02/03/15 0900 hrs to 02/04/15 0900 hrs	02/04/15 1106 hrs	1.0°C
3	02/05/15 0900 hrs to 02/06/15 0900 hrs	02/06/15 1040 hrs	1.0°C

Test Methods:

7-Day Chronic Toxicity, Static renewal, Fathead minnow, EPA 821/R-02/013, Section 11
7-Day Chronic Toxicity, Static renewal, Cladoceran, EPA 821/R-02/013, Section 13

Organisms: *P. promelas* <24hrs, *C. dubia* <24hrs

Culture Source: ASU ERF

Dilutions: 0%, 32%, 42%, 56%, 80%, 100%

Critical Dilution: 100%

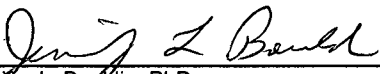
Statistical Method: Toxcalc 5.0.25

Results:	<i>P. promelas</i>	<i>C. dubia</i>
NOEC Survival:	100%	100%
Pass/Fail (0=pass; 1=fail):	0	0
NOEC Growth/Reproduction:	100%	100%
Pass/Fail (0=pass; 1=fail):	0	0
Control Survival:	100%	50%
Control % CV Growth/Reproduction:	10.2	18.5
Critical Dilution % CV Growth/Reproduction:	7.9	60.8
Mean Weight/ # Neonates in Control:	0.4648 mg	16.8
Mean Weight/ # of Neonates in Critical Dilution:	0.5888 mg	12.1
MSDp Growth/ Reproduction	0.1405	0.5550
Daily Average Minimum NOEC:	100%	100%
7-Day Minimum NOEC:	100%	100%

Results Summary: Neither lethal nor sublethal effects were measured in *P. promelas*; however due to only 50% control survival the *C. dubia* test was deemed invalid.

QA/Reference Testing: Data attached

Reviewed By:


Jennifer L. Bouldin, PhD
Director ASU Ecotoxicology Research Facility

Toxicity Test Performed: 7-day *Pimephales promelas* Survival and Growth
 Effluent Sampling Point: Walnut Ridge WWT Plant
 Date Test Started: 02/02/15 *P. promelas*
 Time Test Started: 1230 *P. promelas*
 Date Test Terminated: 02/09/15 *P. promelas*
 Time Test Terminated: 1230 *P. promelas*
 Laboratory Analyst: Rosado-Berrios

Toxicity Test Performed: 7-day *Ceriodaphnia dubia* Survival and Reproduction
 Effluent Sampling Point: Walnut Ridge WWT Plant
 Date Test Started: 02/02/15 *C. dubia*
 Time Test Started: 1230 *C. dubia*
 Date Test Terminated: 02/09/15 *C. dubia*
 Time Test Terminated: 1230 *C. dubia*
 Laboratory Analyst: Griffin/Kilmer

I. Test Methods

A. Physical and Chemical Testing - APHA, Standard Methods for the Examination of Water and Wastewater; Vol. 21, 2005.

<u>Test</u>	<u>Method</u>
Alkalinity	2320B
Conductivity	2510B
Dissolved Oxygen (mg/L, DO)	4500-O-G
Hardness (mg/L CaCO ₃)	2340C
pH	4500-H ⁺ B
Temperature (°C)	2550B

B. Toxicity Testing – EPA 821/R-02/013: Short Term Methods for Estimating the Chronic Toxicity of Effluents to Freshwater Organisms

<u>Test</u>	<u>Method</u>
Fathead Minnow Survival and Growth	Section 11
Cladoceran Survival and Reproduction	Section 13

II. Test Organisms

- A. Name: *Pimephales promelas* (Fathead minnow)
 Source: Laboratory Culture
 Age: <24 hours
 Life Stage: Larval

- B. Name: *Ceriodaphnia dubia* (Cladoceran)
- Source: Laboratory Culture
- Age: <24 hours
- Life Stage: Neonate

III. External Factors

A. Incubator

Temperature (°C)

Average: Average: 24.6

Range: 24.5 – 24.6

Light Cycle: 16 hours light/ 8 hours dark

Light Intensity: 100 footcandles

Control Water: Moderately Hard Synthetic Water (#932/933)

B. *Pimephales promelas*

Test Chambers: 250 ml storage dishes

Volume per Chamber: 200 ml

Number of Organisms per Chamber: 8

Number of Replicates per Concentration: 5

Acclimation: Laboratory control water was added to cultures until >50% of the culture water consisted of control water.

Food: Larval fish were fed 0.15ml of laboratory-cultured *Artemia* brine shrimp one hour prior to test setup and then 3X daily thereafter.

C. *Ceriodaphnia dubia*

Test Chambers: 30 ml Solo cups

Volume per Chamber: 15-20 ml

Number of Organisms per Chamber: 1

Number of Replicates per Concentration: 10

Acclimation: Laboratory control water was added to cultures until >50% of the culture water consisted of control water.

Food: Cladocera were fed *Selenastrum* (#ABS 121514) and yeast/cereal/trout chow mix (#YCT 122214-7/8) one hour prior to test setup and once daily thereafter.

IV. Quality Assurance

A. Standard Toxicant: Sodium Chloride

B. Organism: *Pimephales promelas*

Date of Reference Toxicant Test

Start: 02/10/15

Terminated: 02/17/15

Time of Reference Toxicant Test

Start: 1520

Terminated: 1510

Laboratory Analyst: Vogt/McCauley

Dilution Water Used: Moderately Hard Synthetic Water #933/934

Results: Survival and Growth within control limits

SurvivalGrowth

LOEC: 4.22 g/L NaCl

LOEC: 5.63 g/L NaCl

EC50: 4.83 g/L NaCl

IC25: 4.55 g/L NaCl

C. Organism: *Ceriodaphnia dubia*

Date and time of Reference Toxicant Test

Start: 02/22/15

Terminated: 03/03/15

Time of Reference Toxicant Test

Start: 1405

Terminated: 1355

Laboratory Analyst: Vogt/McCauley

Dilution Water Used: Moderately Hard Synthetic Water #934/935

Results: Survival and Reproduction within control limits

SurvivalReproduction

LOEC: 1.82 g/L NaCl

LOEC: 0.62 g/L NaCl

EC50: 1.94 g/L NaCl

IC25: 0.71 g/L NaCl

V. Physical and Chemical Data - See Attached

VI. Survival and Growth Data - See Attached

VII. Statistical Methods - See Attached

SUMMARY REPORTING FORM
WET Testing
Fathead Minnow Larvae (*Pimephales promelas*) Survival and Growth

Permittee: Walnut Ridge WWT Plant

NPDES No.: AR0046566

		<u>Time</u>	<u>Date</u>		<u>Time</u>	<u>Date</u>
Composite 1:	Collected from	0900	02/01/15	to	0900	02/02/15
Composite 2:	Collected From	0900	02/03/15	to	0900	02/04/15
Composite 3:	Collected From	0900	02/05/15	to	0900	02/06/15

Test Initiated: 1230

Date: 02/02/15

Time Terminated: 1230

Date: 02/09/15

Dilution H₂O: MH 932/933

DATA TABLE FOR SURVIVAL

Effluent Conc. %	Replicate Chambers					Mean % Survival			CV%
	A	B	C	D	E	24h	48h	7days	
Control	100	100	100	100	100	100	100	100	0.0
32	87.5	100	100	100	100	100	100	97.5	6.1
42	100	100	100	100	100	100	100	100	0.0
56	87.5	100	100	100	100	100	100	97.5	6.1
80	100	100	100	100	100	100	100	100	0.0
100	100	100	100	100	100	100	100	100	0.0

DATA TABLE FOR GROWTH

Effluent Conc %	Replicate Chambers (mg)					Mean Dry Weight (mg) CV%	
	A	B	C	D	E		
Control	0.4525	0.5038	0.4538	0.3975	0.5162	0.4648	10.2
32	0.6014	0.5650	0.6100	0.6513	0.6200	0.6095	5.1
42	0.6500	0.5475	0.5813	0.6012	0.5200	0.5800	8.6
56	0.5429	0.5375	0.4850	0.6400	0.5800	0.5571	10.3
80	0.5575	0.5688	0.5675	0.5813	0.6013	0.5753	2.9
100	0.5713	0.6013	0.6637	0.5637	0.5437	0.5888	7.9

Coefficient of Variation = Standard Deviation x 100/Mean

Fathead Minnow Larvae (*Pimephales promelas*) Survival and Growth

1. FISHER'S EXACT TEST:

Is the mean survival for the critical dilution (100%) at 7 days significantly different ($p=0.05$) than the control survival?

_____ Yes X No

2. DUNNETT'S PROCEDURE OR STEEL'S MANY-ONE RANK TEST AS APPROPRIATE:

Is the mean growth by *P. promelas* in the critical dilution (100%) significantly different ($p=0.05$) than the growth in control exposures?

_____ Yes X No

3. If the NOEC for survival is less than the critical dilution, enter [1], otherwise enter [0] for parameter #TGP6C: 0
4. If the NOEC for growth is less than the critical dilution, enter [1], otherwise enter [0] for parameter #TLP6C: 0
5. Report the NOEC value for survival, Parameter #TOP6C:
NOEC survival 100 % effluent
6. Report the NOEC value for growth, Parameter #TPP6C:
NOEC growth 100 % effluent
7. Report the % coefficient of variation (largest of critical and control dilutions), Parameter #TQP6C: CV % growth 10.2% (control)

Whole Effluent Lethality Values

1. Report the Whole Effluent Lethality values for the 30-Day average minimum, Parameter #22414:
Daily Average Minimum NOEC: 100%
2. Report the Whole Effluent Lethality values for the 7-day minimum, Parameter #22414:
7-Day Minimum NOEC: 100%

SUMMARY REPORTING FORM

WET Testing

Fathead Minnow Larvae (*Pimephales promelas*) Survival and Growth

Permittee: Walnut Ridge WWT Plant

Sample No. 1 Collected Ending Date: 02/02/15 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 02/04/15 Time: 0900

Contact: Jon Kopp

Sample No. 3 Collected Ending Date: 02/06/15 Time: 0900

Analyst: Rosado-Berrios

Test Begin: Date: 02/02/15 Time: 1230 Test End: Date: 02/09/15 Time: 1230

Initial Water Chemistry for Chronic Tests

Project: Walnut Ridge – *P. promelas*

Test day		0	1	2	3	4	5	6
Date		2/2/2015	2/3/2015	2/4/2015	2/5/2015	2/6/2015	2/7/2015	2/8/2015
H ₂ O #		MH 932	MH 932	MH 932	MH 932	MH 932	MH 932	MH 932
Temp (°C)	Control	22.0	22.0	23.5	22.8	23.0	22.5	23.5
	32%	22.0	21.5	23.5	23.0	23.1	22.5	23.5
	42%	22.0	21.5	23.7	23.2	23.0	23.0	23.5
	56%	22.0	21.5	23.7	23.2	23.0	23.0	24.0
	80%	22.0	21.8	23.7	23.2	23.5	23.5	24.0
	100%	22.0	21.5	23.7	23.3	23.5	23.5	24.0
pH (Standard Units)	Control	8.12	8.35	8.03	7.53	7.88	8.11	8.14
	32%	8.06	7.92	8.00	7.51	7.88	8.09	7.99
	42%	8.03	7.85	8.00	7.50	7.88	8.10	7.95
	56%	7.99	7.79	7.94	7.47	7.90	8.10	7.90
	80%	7.85	7.68	7.90	7.42	7.88	8.14	7.88
	100%	7.79	7.67	7.88	7.35	7.81	8.09	7.79
DO (mg/L)	Control	8.9	8.8	8.8	9.1	8.9	9.1	8.3
	32%	8.9	8.7	8.7	8.9	9.0	9.2	8.3
	42%	8.8	8.7	8.6	8.8	8.8	8.8	8.3
	56%	8.8	8.7	8.6	8.8	8.8	8.9	8.3
	80%	8.9	8.6	8.6	8.8	8.7	9.0	8.3
	100%	8.9	8.6	8.6	9.0	8.7	8.8	8.5
Cond (μS/cm)	Control	314	317	319	317	316	312	306
	32%	391	393	409	407	408	402	412
	42%	416	422	431	433	438	434	440
	56%	455	459	470	473	485	477	486
	80%	514	522	541	543	560	551	557
	100%	566	576	592	598	616	615	625
Alk (mg/L)	Control	60		60		61		
	100%	99		96		98		
Hard (mg/L)	Control	95		95		90		
	100%	200		310		200		

SUMMARY REPORTING FORM

WET Testing

Fathead Minnow Larvae (*Pimephales promelas*) Survival and Growth

Permittee: Walnut Ridge WWT Plant

Sample No. 1 Collected Ending Date: 02/02/15 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 02/04/15 Time: 0900

Contact: Jon Kopp

Sample No. 3 Collected Ending Date: 02/06/15 Time: 0900

Analyst: Rosado-Berrios

Test Begin: Date: 02/02/15 Time: 1230 Test End: Date: 02/09/15 Time: 1230

Final Water Chemistry for Chronic Tests

Project: Walnut Ridge – *P. promelas*

Test day		1	2	3	4	5	6	7
Date		2/3/2015	2/4/2015	2/5/2015	2/6/2015	2/7/2015	2/8/2015	2/9/2015
H ₂ O #		MH 932	MH 932	MH 932	MH 932	MH 932	MH 932	MH 932
Temp (°C)	Control	21.0	23.0	22.0	22.1	22.0	23.0	22.2
	32%	21.0	23.0	22.0	22.0	22.0	22.8	22.1
	42%	21.0	23.0	22.0	22.1	22.2	22.9	22.1
	56%	21.5	23.0	22.0	22.2	22.5	23.0	22.2
	80%	21.8	23.0	22.0	22.3	22.8	23.0	22.2
	100%	21.8	23.0	22.0	22.0	23.0	23.0	22.2
pH (Standard Units)	Control	7.91	7.66	7.84	7.58	7.61	7.50	7.46
	32%	7.92	7.61	7.86	7.66	7.70	7.56	7.50
	42%	7.91	7.62	7.87	7.69	7.66	7.58	7.53
	56%	7.90	7.64	7.88	7.68	7.73	7.63	7.57
	80%	7.92	7.67	7.88	7.73	7.74	7.67	7.58
	100%	7.93	7.68	7.89	7.76	7.71	7.71	7.62
DO (mg/L)	Control	8.3	7.2	8.2	6.8	7.1	5.5	5.4
	32%	8.5	6.4	7.8	6.7	7.0	5.6	5.9
	42%	7.9	6.2	7.6	6.6	6.8	5.5	5.4
	56%	7.8	6.2	7.6	6.4	6.6	5.3	5.4
	80%	8.0	6.0	7.4	6.4	6.6	5.3	5.3
	100%	7.6	5.6	7.2	6.2	5.9	5.4	5.0

SUMMARY REPORTING FORM
WET Testing
Ceriodaphnia dubia Survival and Reproduction

Permittee: Walnut Ridge WWT Plant

NPDES No.: AR0046566

		<u>Time</u>	<u>Date</u>	to	<u>Time</u>	<u>Date</u>
Composite 1:	Collected from	0900	02/01/15		0900	02/02/15
Composite 2:	Collected From	0900	02/03/15		0900	02/04/15
Composite 3:	Collected From	0900	02/05/15		0900	02/06/15

Test Initiated: 1230

Date: 02/02/15

Time Terminated: 1230

Date: 02/09/15

Dilution H₂O: MH 932/933

PERCENT SURVIVAL
Percent Effluent

<u>Time of Reading</u>	<u>Control</u>	<u>32%</u>	<u>42%</u>	<u>56%</u>	<u>80%</u>	<u>100%</u>
24h	100	100	100	100	100	100
48h	80	100	100	100	100	100
7 day	50	50	70	50	80	80

NUMBER OF YOUNG/FEMALE @ 7 DAYS
Percent Effluent

<u>REP</u>	<u>0%</u>	<u>32%</u>	<u>42%</u>	<u>56%</u>	<u>80%</u>	<u>100%</u>
A	20	X/4	11	3	16	X/7
B	16	17	X/6	X/11	X/0	X/7
C	20	X/13	2	2	5	15
D	15	X/13	17	21	9	1
E	13	19	4	X/9	12	22
F	X/0	X/8	X/2	X/7	10	14
G	X/0	10	7	11	15	8
H	X/0	21	X/10	X/0	X/13	5
I	X/0	X/6	3	11	12	11
J	X/0	1	23	X/3	4	21
Mean	16.8	13.6	9.6	9.6	10.4	12.1
CV%*	18.5	60.1	82.7	79.9	41.5	60.8

*Coefficient of Variation% = Standard Deviation x 100/Mean

***Ceriodaphnia dubia* Survival and Reproduction**

1. FISHER'S EXACT TEST:
Is the mean survival for the critical dilution (100%) at 7 days significantly different ($p=0.05$) than the control survival?
 Yes X No

2. DUNNETT'S PROCEDURE OR STEEL'S MANY-ONE RANK TEST AS APPROPRIATE:
Is the mean number of young produced per female by the critical dilution (100%) significantly different ($p=0.05$) than the control's number of young per female?
 Yes X No

3. If the NOEC for survival is less than the critical dilution, enter [1], otherwise enter [0] for parameter #TGP3B: 0

4. If the NOEC for reproduction is less than the critical dilution, enter [1], otherwise enter [0] for parameter #TLP3B: 0

5. Report the NOEC value for survival, Parameter #TOP3B:
NOEC survival 100 % effluent

6. Report the NOEC value for reproduction, Parameter #TPP3B:
NOEC reproduction 100 % effluent

7. Report the % coefficient of variation (largest of critical and control dilutions), Parameter #TQP3B:
CV % reproduction 60.8% (critical)

Whole Effluent Lethality Values for *Ceriodaphnia dubia*

1. Report the Whole Effluent Lethality values for the 30-Day average minimum, Parameter #22414:
Daily Average Minimum NOEC: 100%

2. Report the Whole Effluent Lethality values for the 7-day minimum, Parameter #22414:
7-Day Minimum NOEC: 100%

CHRONIC TOXICITY SUMMARY FORM
WET Testing *Ceriodaphnia dubia* (Cladoceran)
CHEMICAL PARAMETERS CHART

Permittee: Walnut Ridge WWT Plant

Sample No. 1 Collected Ending Date: 02/02/15 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 02/04/15 Time: 0900

Contact: Jon Kopp

Sample No. 3 Collected Ending Date: 02/06/15 Time: 0900

Analyst: Griffin/Kilmer

Test Begin: Date: 02/02/15 Time: 1230 Test End: Date: 02/09/15 Time: 1230

Initial Water Chemistry for Chronic Tests								
Project: Walnut Ridge – <i>C. dubia</i>								
Test day		0	1	2	3	4	5	6
Date		2/2/2015	2/3/2015	2/4/2015	2/5/2015	2/6/2015	2/7/2015	2/8/2015
H ₂ O #		MH 932	MH 932	MH 932	MH 932	MH 932	MH 932	MH 932
Temp (°C)	Control	22.0	22.0	23.5	22.8	23.0	22.5	23.5
	32%	22.0	21.5	23.5	23.0	23.1	22.5	23.5
	42%	22.0	21.5	23.7	23.2	23.0	23.0	23.5
	56%	22.0	21.5	23.7	23.2	23.0	23.0	24.0
	80%	22.0	21.8	23.7	23.2	23.5	23.5	24.0
	100%	22.0	21.5	23.7	23.3	23.5	23.5	24.0
pH (Standard Units)	Control	8.12	8.35	8.03	7.53	7.88	8.11	8.14
	32%	8.06	7.92	8.00	7.51	7.88	8.09	7.99
	42%	8.03	7.85	8.00	7.50	7.88	8.10	7.95
	56%	7.99	7.79	7.94	7.47	7.90	8.10	7.90
	80%	7.85	7.68	7.90	7.42	7.88	8.14	7.88
	100%	7.79	7.67	7.88	7.35	7.81	8.09	7.79
DO (mg/L)	Control	8.9	8.8	8.8	9.1	8.9	9.1	8.3
	32%	8.9	8.7	8.7	8.9	9.0	9.2	8.3
	42%	8.8	8.7	8.6	8.8	8.8	8.8	8.3
	56%	8.8	8.7	8.6	8.8	8.8	8.9	8.3
	80%	8.9	8.6	8.6	8.8	8.7	9.0	8.3
	100%	8.9	8.6	8.6	9.0	8.7	8.8	8.5
Cond (µS/cm)	Control	314	317	319	317	316	312	306
	32%	391	393	409	407	408	402	412
	42%	416	422	431	433	438	434	440
	56%	455	459	470	473	485	477	486
	80%	514	522	541	543	560	551	557
	100%	566	576	592	598	616	615	625
Alk (mg/L)	Control	60		60		61		
	100%	99		96		98		
Hard (mg/L)	Control	95		95		90		
	100%	200		310		200		

CHRONIC TOXICITY SUMMARY FORMWET Testing *Ceriodaphnia dubia* (Cladoceran)
CHEMICAL PARAMETERS CHART

Permittee: Walnut Ridge WWT Plant

Sample No. 1 Collected Ending Date: 02/02/15 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 02/04/15 Time: 0900

Contact: Jon Kopp

Sample No. 3 Collected Ending Date: 02/06/15 Time: 0900

Analyst: Griffin/Kilmer

Test Begin: Date: 02/02/15 Time: 1230 Test End: Date: 02/09/15 Time: 1230

Final Water Chemistry for Chronic Tests								
Project: Walnut Ridge - <i>C. dubia</i>								
Test day		1	2	3	4	5	6	7
Date:		2/3/2015	2/4/2015	2/5/2015	2/6/2015	2/7/2015	2/8/2015	2/9/2015
H ₂ O #		MH 932	MH 932	MH 932	MH 932	MH 932	MH 932	MH 932
Temp (°C)	Control	21.5	23.5	23.5	24.0	22.5	23.5	23.5
	32%	22.0	23.7	23.5	23.8	22.5	23.8	23.5
	42%	21.5	23.7	23.5	23.6	22.5	24.0	23.5
	56%	21.8	23.8	23.5	23.6	22.5	24.0	23.5
	80%	21.8	23.8	23.5	23.9	22.5	24.0	23.5
	100%	21.5	23.9	23.5	23.9	22.5	24.0	23.5
pH (Standard Units)	Control	8.22	8.33	8.42	8.16	8.09	8.22	8.36
	32%	8.35	8.69	8.49	8.83	8.30	8.78	8.47
	42%	8.39	8.69	8.57	8.76	8.28	8.90	8.58
	56%	8.36	8.63	8.50	8.86	8.36	8.70	8.46
	80%	8.38	8.68	8.54	8.72	8.35	8.87	8.60
	100%	8.36	8.62	8.51	8.80	8.36	8.77	8.54
DO (mg/L)	Control	9.1	9.3	9.2	9.4	9.3	9.0	8.9
	32%	9.3	9.6	9.3	9.8	9.2	8.9	8.7
	42%	9.3	9.7	9.5	9.9	9.2	9.3	8.9
	56%	9.2	9.7	9.6	9.9	9.2	9.3	8.9
	80%	9.2	9.6	9.5	9.9	9.1	9.4	8.9
	100%	9.2	9.6	9.5	9.9	9.1	9.4	9.1

Larval Fish Growth and Survival Test-7 Day Survival

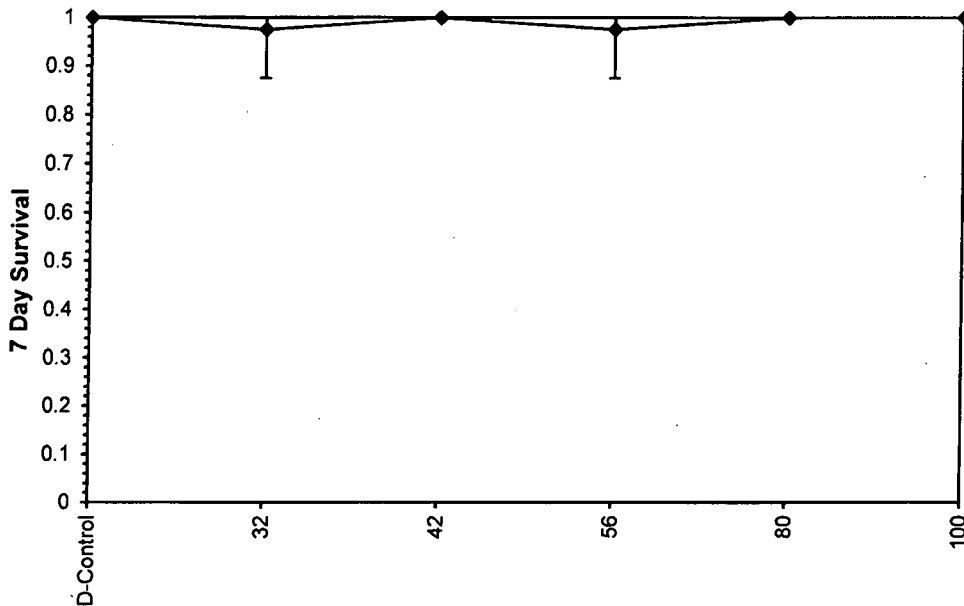
Start Date: 2/2/2015 12:30	Test ID: WR	Sample ID: NPDES Permit #AR0046566
End Date: 2/9/2015 12:30	Lab ID: ASU ERF	Sample Type: EFF1-POTW
Sample Date: 02/02/2015	Protocol: EPAF 02-EPA Freshwater	Test Species: PP-Pimephales promelas
Comments: 1st Quarter WET Testing		

Conc-%	1	2	3	4	5
D-Control	1.0000	1.0000	1.0000	1.0000	1.0000
32	0.8750	1.0000	1.0000	1.0000	1.0000
42	1.0000	1.0000	1.0000	1.0000	1.0000
56	0.8750	1.0000	1.0000	1.0000	1.0000
80	1.0000	1.0000	1.0000	1.0000	1.0000
100	1.0000	1.0000	1.0000	1.0000	1.0000

Conc-%	Mean	N-Mean	Transform: Arcsin Square Root				N	Rank Sum	1-Tailed Critical
			Mean	Min	Max	CV%			
D-Control	1.0000	1.0000	1.3931	1.3931	1.3931	0.000	5		
32	0.9750	0.9750	1.3564	1.2094	1.3931	6.055	5	25.00	16.00
42	1.0000	1.0000	1.3931	1.3931	1.3931	0.000	5	27.50	16.00
56	0.9750	0.9750	1.3564	1.2094	1.3931	6.055	5	25.00	16.00
80	1.0000	1.0000	1.3931	1.3931	1.3931	0.000	5	27.50	16.00
100	1.0000	1.0000	1.3931	1.3931	1.3931	0.000	5	27.50	16.00

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.5466	0.9	-2.73687	8.25694
Equality of variance cannot be confirmed				
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	100	>100		1

Dose-Response Plot



Larval Fish Growth and Survival Test-7 Day Growth

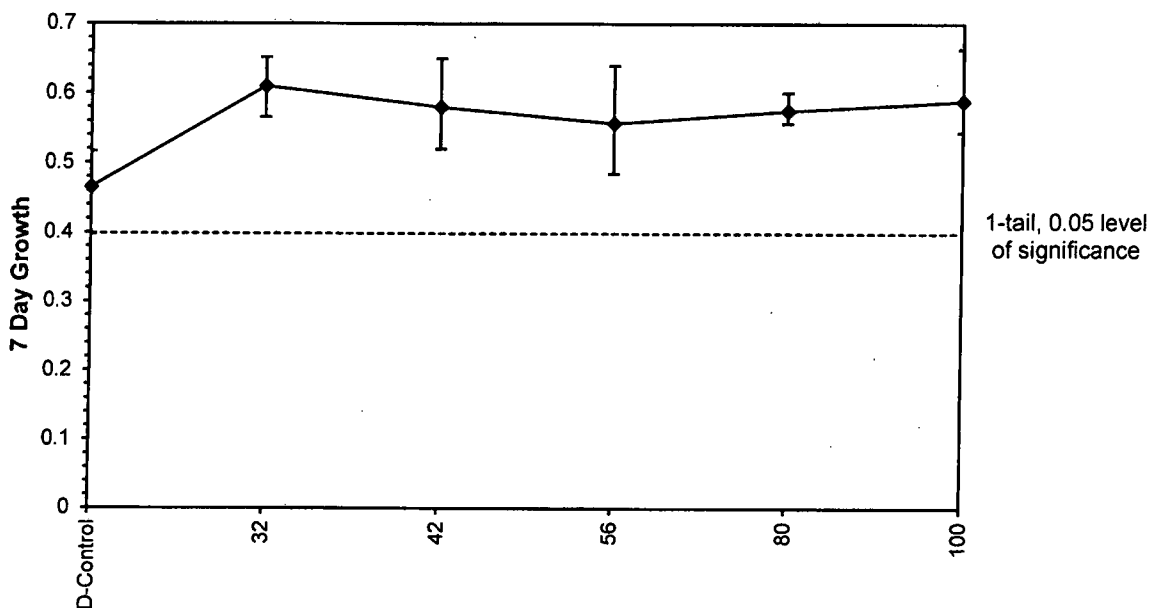
Start Date: 2/2/2015 12:30	Test ID: WR	Sample ID: NPDES Permit #AR0046566
End Date: 2/9/2015 12:30	Lab ID: ASU ERF	Sample Type: EFF1-POTW
Sample Date: 02/02/2015	Protocol: EPAF 02-EPA Freshwater	Test Species: PP-Pimephales promelas
Comments: 1st Quarter WET Testing		

Conc-%	1	2	3	4	5
D-Control	0.4525	0.5038	0.4538	0.3975	0.5162
32	0.6014	0.5650	0.6100	0.6513	0.6200
42	0.6500	0.5475	0.5813	0.6012	0.5200
56	0.5429	0.5375	0.4850	0.6400	0.5800
80	0.5575	0.5688	0.5675	0.5813	0.6013
100	0.5713	0.6013	0.6637	0.5637	0.5437

Conc-%	Mean	N-Mean	Transform: Untransformed					N	t-Stat	1-Tailed Critical	MSD
			Mean	Min	Max	CV%					
D-Control	0.4648	1.0000	0.4648	0.3975	0.5162	10.188	5				
32	0.6095	1.3115	0.6095	0.5650	0.6513	5.122	5	-5.233	2.360	0.0653	
42	0.5800	1.2480	0.5800	0.5200	0.6500	8.625	5	-4.166	2.360	0.0653	
56	0.5571	1.1986	0.5571	0.4850	0.6400	10.305	5	-3.337	2.360	0.0653	
80	0.5753	1.2378	0.5753	0.5575	0.6013	2.921	5	-3.994	2.360	0.0653	
100	0.5888	1.2668	0.5888	0.5437	0.6637	7.938	5	-4.482	2.360	0.0653	

Auxiliary Tests	Statistic	Critical	Skew	Kurt						
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.96992	0.9	0.29111	-0.17995						
Bartlett's Test indicates equal variances (p = 0.37)	5.41544	15.0863								
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	100	>100		1	0.06529	0.14049	0.01295	0.00191	4.6E-04	5, 24

Dose-Response Plot



CHRONIC TEST DATA SHEET
Pimephales promelas

Project: Walnut Ridge Beginning Date: 020215 Time: 1230 Test Species: *P. promelas*

Dilution H₂O: MH432 Ending Date: 020915 Time: 1230 Age: < 24 hrs
MH983

Test Type: (*)Static Renewal () Flowthrough Toxicant/Effluent: _____

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Pan #
Control	1	8/0	8/0	8/0	8/0	8/0	8/0	8/0	1
	2	8/0	8/0	8/0	8/0	8/0	8/0	8/0	2
	3	8/0	8/0	8/0	8/0	8/0	8/0	8/0	3
	4	8/0	8/0	8/0	8/0	8/0	8/0	8/0	4
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	5
32%	1	8/0	7/0	7/0	7/0	7/0	7/0	7/0	6
	2	8/0	8/0	8/0	8/0	8/0	8/0	8/0	7
	3	8/0	8/0	8/0	8/0	8/0	8/0	8/0	8
	4	8/0	8/0	8/0	8/0	8/0	8/0	8/0	9
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	10
42%	1	8/0	8/0	8/0	8/0	8/0	8/0	8/0	11
	2	8/0	8/0	8/0	8/0	8/0	8/0	8/0	12
	3	8/0	8/0	8/0	8/0	8/0	8/0	8/0	13
	4	8/0	8/0	8/0	8/0	8/0	8/0	8/0	14
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	15
56%	1	8/0	8/0	8/0	8/0	8/0	8/1	7/0	16
	2	8/0	8/0	8/0	8/0	8/0	8/0	8/0	17
	3	8/0	8/0	8/0	8/0	8/0	8/0	8/0	18
	4	8/0	8/0	8/0	8/0	8/0	8/0	8/0	19
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	20
Date		020315	020415	020515	020615	020715	020815	020915	
Initials		LARB	LARB	LARB	LARB	LARB	LARB	SN/CARB	

CHRONIC TEST DATA SHEET
Pimephales promelas

Project: Walnut Ridge Beginning Date: 020215 Time: 1230 Test Species: P.promelas

Dilution H₂O: MH932 Ending Date: 020915 Time: 1230 Age: < 24 hrs.
MH933

Test Type: (*)Static Renewal () Flowthrough Toxicant/Effluent: _____

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Pan #
80%	1	8/0	8/0	8/0	8/0	8/0	8/0	8/0	21
	2	8/0	8/0	8/0	8/0	8/0	8/0	8/0	22
	3	8/0	8/0	8/0	8/0	8/0	8/0	8/0	23
	4	8/0	8/0	8/0	8/0	8/0	8/0	8/0	24
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	25
100%	1	8/0	8/0	8/0	8/0	8/0	8/0	8/0	26
	2	8/0	8/0	8/0	8/0	8/0	8/0	8/0	27
	3	8/0	8/0	8/0	8/0	8/0	8/0	8/0	28
	4	8/0	8/0	8/0	8/0	8/0	8/0	8/0	29
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	30
Date		020315	020415	020515	020615	020715	020815	020915	
Initials		LAB	LAB	LAB	LAB	LAB	LAB	SV/CARB	

020215

AF: 1600 02/01

BF: 1050 02/02

012315 T: 5,9,12,16,19,24

012415 T: 4,5,6,7,8,10,11,12,14,16,17,19,23

012515 T: 2,3,4,6,7,8,9,10,12,13,16,17,18,20,23

012615 T: 1,2,3,5,7,9,10,11,12,13,14,18,23,24

CARB

Ceriodaphnia Survival and Reproduction Test-7 Day Survival

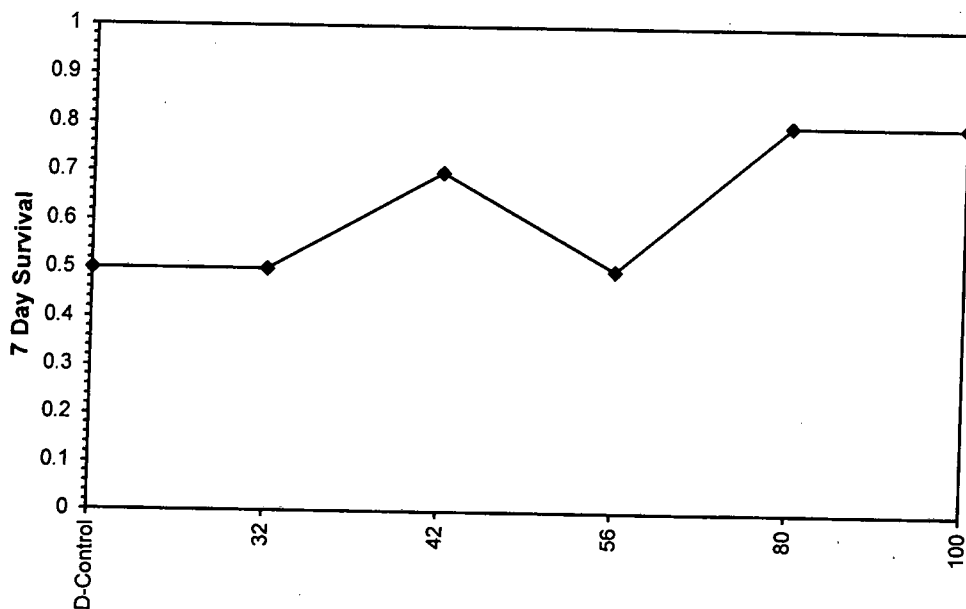
Start Date: 2/2/2015 12:30	Test ID: WR	Sample ID: NPDES Permit #AR0046566
End Date: 2/9/2015 12:30	Lab ID: ASU ERF	Sample Type: EFF1-POTW
Sample Date: 02/02/2015	Protocol: EPAF 02-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia
Comments: 1st Quarter WET Testing		

Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.0000	0.0000
32	0.0000	1.0000	0.0000	0.0000	1.0000	0.0000	1.0000	1.0000	0.0000	1.0000
42	1.0000	0.0000	1.0000	1.0000	1.0000	0.0000	1.0000	0.0000	1.0000	1.0000
56	1.0000	0.0000	1.0000	1.0000	0.0000	0.0000	1.0000	0.0000	1.0000	0.0000
80	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	1.0000	1.0000
100	0.0000	0.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000

Conc-%	Mean	N-Mean	Resp	Not Resp	Total	N	Fisher's Exact P	1-Tailed Critical
D-Control	0.5000	1.0000	5	5	10	10		
32	0.5000	1.0000	5	5	10	10	0.6719	0.0500
42	0.7000	1.4000	3	7	10	10	0.3250	0.0500
56	0.5000	1.0000	5	5	10	10	0.6719	0.0500
80	0.8000	1.6000	2	8	10	10	0.1749	0.0500
100	0.8000	1.6000	2	8	10	10	0.1749	0.0500

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Fisher's Exact Test	100	>100		1

Dose-Response Plot



Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date: 2/2/2015 12:30	Test ID: WR	Sample ID: NPDES Permit #AR00
End Date: 2/9/2015 12:30	Lab ID: ASU ERF	Sample Type: EFF1-POTW
Sample Date:	Protocol: EPAF 02-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia

Comments:

Conc-%	1	2	3	4	5	6	7	8
D-Control	20.000	16.000	20.000	15.000	13.000			
32	17.000	19.000	10.000	21.000	1.000			
42	11.000	2.000	17.000	4.000	7.000	3.000	23.000	
56	3.000	2.000	21.000	11.000	11.000			
80	16.000	5.000	9.000	12.000	10.000	15.000	12.000	4.000
100	15.000	1.000	22.000	14.000	8.000	5.000	11.000	21.000

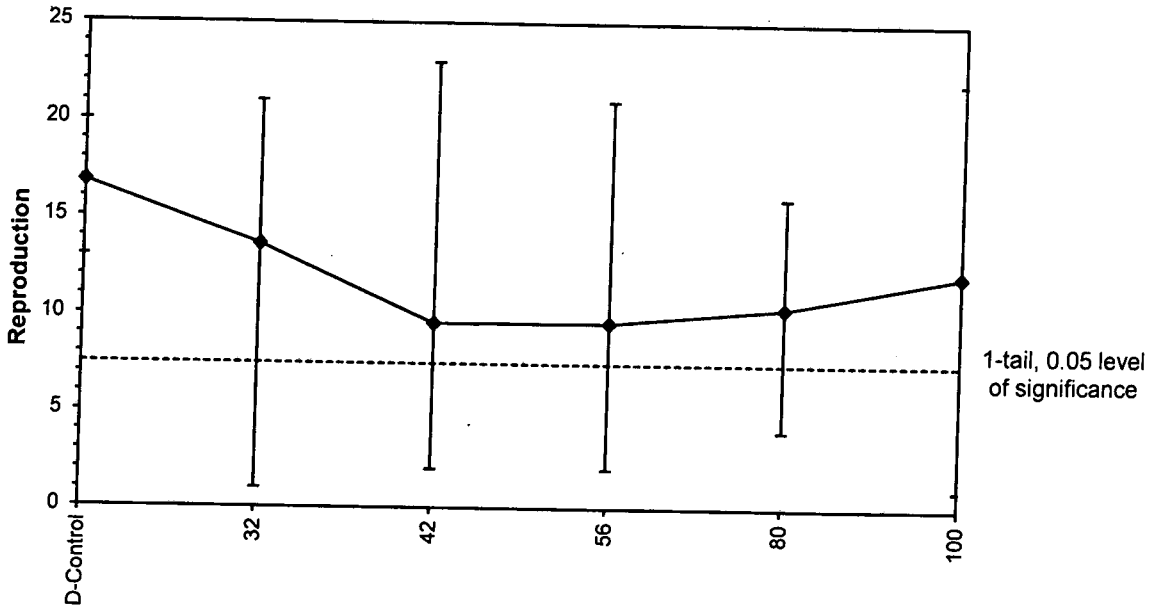
Conc-%	Mean	N-Mean	Transform: Untransformed					t-Stat	1-Tailed Critical	MSD
			Mean	Min	Max	CV%	N			
D-Control	16.800	1.0000	16.800	13.000	20.000	18.539	5			
32	13.600	0.8095	13.600	1.000	21.000	60.097	5	0.757	2.449	10.344
42	9.571	0.5697	9.571	2.000	23.000	82.675	7	1.848	2.449	9.577
56	9.600	0.5714	9.600	2.000	21.000	79.876	5	1.704	2.449	10.344
80	10.375	0.6176	10.375	4.000	16.000	41.517	8	1.687	2.449	9.324
100	12.125	0.7217	12.125	1.000	22.000	60.836	8	1.228	2.449	9.324

Auxiliary Tests

Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ($p > 0.01$)	0.98207	0.916	0.11868
Bartlett's Test indicates equal variances ($p = 0.34$)	5.66104	15.0863	-0.45002

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Bonferroni t Test	100	>100		1	9.32419	0.55501	43.4408	44.6145	0.44867	5, 32

Dose-Response Plot



CHRONIC TEST DATA SHEET

Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 020215 Time: 1230 Test Species: C. dubia

Dilution H₂O: MH932 Ending Date: 020915 Time: 1230 Age: <24h

Test Type: (*)Static Renewal () Flowthrough Toxicant/Effluent: _____

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
Control	1	0	0	0	3	0	6	11	20
	2		0	0	2	0	7	7	16
	3		0	0	3	1	6	10	20
	4		0	0	1	0	4	10	15
	5		0	0	1	0	1	11	13
	6		0	X/0					X/0
	7		X/0						X/0
	8		0	X/0					X/0
	9		0	X/0					X/0
	10	↓	X/0						X/0
32%	1	0	0	0	0	0	4	X/0	X/4
	2				1	0	7	9	17
	3				0	9	0	X/4	X/13
	4				0	0	1	X/0	X/1
	5				0	8	10	1	19
	6				1	7	X/0		X/8
	7				0	6	4	0	10
	8				2	10	0	9	21
	9				0	4	2	X/0	X/6
	10	↓	↓	↓	0	0	1	0	1
Date		020315	020415	020515	020615	020715	020815	020915	
Initials		KL	MG	MG	MG	KL	KL	MG	

CHRONIC TEST DATA SHEET
Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 020215 Time: 1230 Test Species: C. dubia

Dilution H₂O: MH932 Ending Date: 020915 Time: 1230 Age: 624h
MH933

Test Type: (*)Static Renewal () Flowthrough Toxicant/ Effluent:

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
42%	1	0	0	0	2	8	0	1	11
	2	↓	↓	↓	0	6	X/0	—	→ X/6
	3	↓	↓	↓	0	0	2	0	2
	4	↓	↓	↓	3	3	0	11	17
	5	↓	↓	↓	1	3	0	0	4
	6	↓	↓	↓	2	0	X/0	—	→ X/2
	7	↓	↓	↓	2	0	5	0	7
	8	↓	↓	↓	2	8	0	X/0	X/10
	9	↓	↓	↓	0	1	2	0	3
	10	↓	↓	↓	4	2	7	10	23
56%	1	0	0	0	0	0	0	3	3
	2	↓	↓	↓	1	0	10	X/0	X/11
	3	↓	↓	↓	0	0	0	2	2
	4	↓	↓	↓	3	1	7	10	21
	5	↓	↓	↓	3	6	X/0	—	→ X/9
	6	↓	↓	↓	0	7	0	X/0	X/7
	7	↓	↓	↓	4	7	0	0	11
	8	↓	↓	↓	X/0	—	—	—	→ X/0
	9	↓	↓	↓	0	0	5	6	11
	10	↓	↓	↓	X/3	—	—	—	→ X/3
Date		020315	020415	020515	020615	020715	020815	020915	
Initials		KL	MG	MG	MG	KL	KL	MG	

CHRONIC TEST DATA SHEET
Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 010215 Time: 1230 Test Species: C. dubia

Dilution H₂O: MH932 Ending Date: 020915 Time: 1230 Age: 424h
MH933

Test Type: () Static Renewal () Flowthrough Toxicant/Effluent: _____

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
80%	1	0	0	0	0	0	8	8	16
	2	↓	↓	↓	0	0	X/0	—	X/0
	3	↓	↓	↓	0	5	0	0	5
	4	↓	↓	↓	1	X/0	8	0	9
	5	↓	↓	↓	1	0	1	10	12
	6	↓	↓	↓	0	7	3	0	10
	7	↓	↓	↓	0	0	7	8	15
	8	↓	↓	↓	1	0	8	X/4	X/13
	9	↓	↓	↓	0	0	6	6	12
	10	↓	↓	↓	0	0	4	0	4
100%	1	0	0	0	1	0	6	X/0	X/7
	2	↓	↓	↓	0	1	6	X/0	X/7
	3	↓	↓	↓	4	6	0	5	15
	4	↓	↓	↓	0	0	0	1	1
	5	↓	↓	↓	4	9	0	9	22
	6	↓	↓	↓	0	0	6	8	14
	7	↓	↓	↓	0	0	4	4	8
	8	↓	↓	↓	0	0	4	1	5
	9	↓	↓	↓	0	0	0	11	11
	10	↓	↓	↓	0	1	7	13	21
Date		020315	020415	020515	020615	020715	020815	020915	
Initials		KK	LMG	LMG	LMG	KK	KK	LMG	

Initial Water Chemistry for Chronic Tests
Project: Walnut Ridge - *C. dubia* / *P. promelas*

Test Day:		0	1	2	3	4	5	6
Date:		020215	020315	020415	020515	020615	020715	020815
H ₂ O Batch #:		MH931	MH932	MH932	MH933	MH933	MH933	MH933
Temp. (°C)	Control:	22.0	22.5	23.5	22.8	23.0	22.5	23.5
	32%:	22.0	22.5	23.5	23.0	23.1	22.5	23.5
	42%:	22.0	21.5	23.7	23.2	23.0	23.0	23.5
	56%:	22.0	21.5	23.7	23.2	23.0	23.0	24.0
	80%:	22.0	21.8	23.7	23.2	23.5	23.5	24.0
	100%:	22.0	21.5	23.7	23.3	23.5	23.5	24.0
pH	Control:	8.12	8.35	8.03	7.53	7.88	8.11	8.14
	32%:	8.00	7.92	8.00	7.51	7.88	8.09	7.99
	42%:	8.03	7.85	8.00	7.50	7.88	8.10	7.95
	56%:	7.99	7.79	7.94	7.47	7.90	8.10	7.90
	80%:	7.85	7.68	7.90	7.42	7.80	8.14	7.88
	100%:	7.79	7.67	7.88	7.35	7.81	8.09	7.79
DO (mg/L)	Control:	8.9	8.8	8.8	9.1	8.9	9.1	8.3
	32%:	8.9	8.7	8.7	8.9	9.0	9.2	8.3
	42%:	8.8	8.7	8.6	8.8	8.8	8.8	8.3
	56%:	8.8	8.7	8.6	8.8	8.8	8.9	8.3
	80%:	8.9	8.6	8.6	8.8	8.7	9.0	8.3
	100%:	8.9	8.6	8.6	9.0	8.7	8.8	8.5
Cond. (µS/cm)	Control:	314	317	319	317	316	312	306
	32%:	391	393	409	407	408	402	412
	42%:	414	422	431	433	438	434	440
	56%:	455	459	470	473	485	477	486
	80%:	514	522	541	543	560	551	557
	100%:	566	576	592	598	616	613	625
Alk. (mg/L)	Control:	60		60		61		
	100%:	99		96		98		
Hard. (mg/L)	Control:	95		95		90		
	100%:	200		310		200		
Initials:		MG/KL	MG	MG	MG	MG	KL	MG/KL

Final Water Chemistry for Chronic Tests
Project: Walnut Ridge - *C. dubia*

Test Day:		1	2	3	4	5	6	7
Date:		020315	020415	020515	020615	020715	020815	020915
H ₂ O Batch #:		MH932	MH932	MH932	MH932	MH933	MH933	MH933
Temp (°C)	Control	22.5	23.5	23.5	24.0	22.5	23.5	23.5
	32%	22.9	23.7	23.5	23.8	22.5	23.8	23.5
	42%	21.5	23.7	23.5	23.6	22.5	24.0	23.5
	56%	21.8	23.8	23.5	23.6	22.5	24.0	23.5
	80%	21.8	23.8	23.5	23.9	22.5	24.0	23.5
	100%	21.5	23.9	23.5	23.9	22.5	24.0	23.5
pH	Control	8.22	8.33	8.42	8.16	8.09	8.22	8.36
	32%	8.35	8.69	8.49	8.83	8.30	8.78	8.47
	42%	8.39	8.69	8.57	8.76	8.28	8.90	8.59
	56%	8.36	8.63	8.50	8.86	8.36	8.70	8.46
	80%	8.38	8.68	8.54	8.72	8.35	8.87	8.60
	100%	8.36	8.62	8.51	8.80	8.36	8.77	8.54
DO (mg/L)	Control	9.1	9.3	9.2	9.4	9.3	9.0	8.9
	32%	9.3	9.4	9.3	9.8	9.2	8.9	8.7
	42%	9.3	9.7	9.5	9.9	9.2	9.3	8.9
	56%	9.2	9.7	9.6	9.9	9.2	9.3	8.9
	80%	9.2	9.6	9.5	9.9	9.1	9.4	8.9
	100%	9.2	9.6	9.5	9.9	9.1	9.4	9.1
Initials		KL/SV	MG	MG	MG	KL	KL	MG



Ecotoxicology Research Facility

SAMPLE CHECK IN

Sample ID Number: WR#1

Fill out this information with each effluent or river water sample coming in for testing. Keep completed sheets with test data and file with the lab QA/QC officer.

Date: 020215 Sampling Date: 02/1-2/15 Arrival Time: 1115

Field Identification Number: _____ Description: effluent

Shipped by: Federal Express _____ UPS _____ Hand delivered by: Walnut Ridge personnel

Drop-Off Location: ASU-ERF

Storage While Shipped: cooler w/ice

Analysis Requested: chronic Cd, Cu, & Pb

Initial Water Chemistry Analysis:

Sample Received by: MG

Temperature (°C): 20

Ice Present upon delivery: YES NO

Date: 020215

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	<u>MG</u>	<u>020215</u>	<input checked="" type="checkbox"/>	
Refrigerated at 4°C	<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/>	
Field Record Received	<input checked="" type="checkbox"/>			<input checked="" type="checkbox"/>
Sample Label Affixed Properly	<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/>	
Project Leader Informed	<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/>	

Comments: _____



Ecotoxicology Research Facility

SAMPLE CHECK IN

Sample ID Number: WR#2

Fill out this information with each effluent or river water sample coming in for testing. Keep completed sheets with test data and file with the lab QA/QC officer.

Date: 020415 Sampling Date: 02/3-4/15 Arrival Time: 1106

Field Identification Number: _____ Description: effluent

Shipped by: Federal Express _____ UPS _____ Hand delivered by: WR personnel

Drop-Off Location: ASU-ERF

Storage While Shipped: cooler w/ ice

Analysis Requested: chronic Cdutra + Ppmanelas

Initial Water Chemistry Analysis: _____

Sample Received by: JH

Temperature (°C): 1.0 Ice Present upon delivery: YES NO

Date: 020415

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	<u>LMC</u>	<u>020415</u>	<input checked="" type="checkbox"/>	
Refrigerated at 4°C	<u>↓</u>	<u>↓</u>	<input checked="" type="checkbox"/>	
Field Record Received				<input checked="" type="checkbox"/>
Sample Label Affixed Properly			<input checked="" type="checkbox"/>	
Project Leader Informed	<u>↓</u>	<u>↓</u>	<input checked="" type="checkbox"/>	

Comments: _____



Ecotoxicology Research Facility

SAMPLE CHECK IN

Sample ID Number: WR#3

Fill out this information with each effluent or river water sample coming in for testing. Keep completed sheets with test data and file with the lab QA/QC officer.

Date: 020615 Sampling Date: 02/5-6/15 Arrival Time: 1040

Field Identification Number: _____ Description: Effluent

Shipped by: Federal Express _____ UPS _____ Hand delivered by: WR personnel

Drop-Off Location: ASU-ERF

Storage While Shipped: cooler w/ice

Analysis Requested: chronic Cadabra + Ppmular

Initial Water Chemistry Analysis:

Sample Received by: LMG

Temperature (°C): 1.0 Ice Present upon delivery: YES NO

Date: 020615

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	<u>LMG</u>	<u>020615</u>	<input checked="" type="checkbox"/>	<input type="checkbox"/>
Refrigerated at 4°C	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>
Field Record Received	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>
Sample Label Affixed Properly	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>
Project Leader Informed	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>

Comments: _____



Ecotoxicology Research Facility

Ecotoxicology Research Facility

Arkansas State University

501 Iroquois Street

State University, AR 72467

(870) 972-2570 Fax (870) 972-2577

CHAIN OF CUSTODY RECORD



Client Name Walnut Ridge Wastewater Treatment			Phone: (870) 886-2312					Analyses (List Below)				
Project #			Fax:									
Sampler (sign) <i>Jonathan Kopp</i>			PO #:									
Remarks: Contact: Jonathan Kopp								Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>			
Cont.#	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix					
					Comp	Grab	Aqueous	Soil	Other			
			<i>2-1-15</i>	<i>9 A.M.</i>						<i>X</i>	<i>X</i>	
			<i>2-2-15</i>	<i>9 A.M.</i>								
Ice present at delivery: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No												
Temp: <i>2.0 °C</i>			<i>LMC</i> Initials									
1. Relinquished By (sign) <i>Jonathan Kopp</i>			Date <i>2-2-15</i>	Time <i>11:15 AM</i>	1. Received By (sign) <i>Melanie Giffen</i>					Date <i>02/02/15</i>	Time <i>11:15</i>	
2. Relinquished By (sign)			Date	Time	2. Received By (sign)					Date	Time	



Ecotoxicology Research Facility

Ecotoxicology Research Facility

Arkansas State University

501 Iroquois Street

State University, AR 72467

(870) 972-2570 Fax (870) 972-2577

CHAIN OF CUSTODY RECORD



Client Name Walnut Ridge Wastewater Treatment			Phone: (870) 886-2312					Analyses (List Below)											
Project #			Fax:																
Sampler (sign) <i>[Signature]</i>			PO #:					<table border="1"> <tr> <td>Chronic <i>C. dubia</i></td> <td>Chronic <i>P. promelas</i></td> <td></td> <td></td> </tr> <tr> <td>X</td> <td>X</td> <td></td> <td></td> </tr> </table>				Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>			X	X		
Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>																		
X	X																		
Remarks: Contact: Jonathan Kopp			Sample Date					Sample Time											
Cont.#	Sample ID Number	Location	Sample Type		Matrix														
			Comp	Grab	Aqueous	Soil	Other												
			X					X	X										
Ice present at delivery: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No																			
Temp: <u>10°C</u>			Initials: <u>JK</u>																
1. Relinquished By (sign) <i>[Signature]</i>			Date <u>2-4-15</u>		Time <u>11:00 AM</u>		1. Received By (sign) <i>[Signature]</i>			Date <u>2-4-15</u>									
2. Relinquished By (sign) <i>[Signature]</i>			Date		Time		2. Received By (sign)			Date <u>11:06</u>									



Ecotoxicology Research Facility

Ecotoxicology Research Facility

Arkansas State University

2645 Caddo Drive

State University, AR 72467

(870) 972-2570 Fax (870) 972-2577

CHAIN OF CUSTODY RECORD



ARKANSAS STATE UNIVERSITY

Client Name Walnut Ridge Wastewater Treatment			Phone: (870) 886-2312				Analyses (List Below)									
Project #			Fax:													
Sampler (sign) <i>[Signature]</i>			PO #:				<table border="1"> <tr> <td rowspan="2">Chronic C. dubia</td> <td rowspan="2">Chronic P. promelas</td> <td></td> <td></td> </tr> <tr> <td></td> <td></td> </tr> </table>				Chronic C. dubia	Chronic P. promelas				
Chronic C. dubia	Chronic P. promelas															
Remarks:			Contact: Jonathan Kopp													
Cont.#	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix									
					Comp	Grab	Aqueous	Soil	Other							
1		outfall 1001	2-5-15	9 A.M.							X	X				
			2-6-15	9 A.M.												
Ice present at delivery: <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No			Temp: <u>1.0 °C</u>				Initials: <u>MG</u>									
1. Relinquished By (sign) <i>[Signature]</i>			Date 2-6-15	Time 10:00 AM	1. Received By (sign) <i>[Signature]</i>			Date 02/06/15	Time 1040							
2. Relinquished By (sign) <i>[Signature]</i>			Date	Time	2. Received By (sign) <i>[Signature]</i>			Date	Time							

City Water Works
216 S.W. Fourth
Walnut Ridge, AR 72476



ADEQ
Water Division-Enforcement Division
5301 NorthShore Drive
North Little Rock, AR 72118-5317

