



Ecotoxicology Research Facility



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November 1, 2016

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 October 17, 2016. No lethal or sublethal effects were measured in *Pimephales promelas*. No lethal effects were measured in *Ceriodaphnia dubia*, however, sublethal effects were measured in all effluent dilutions except the critical flow concentration (100%).

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

Arkansas State University Ecotoxicology Research Facility

Laboratory Report

Facility Director / Lab Contact: Jennifer Bouldin
 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	10/16/16 0900 hrs to 10/17/16 0900 hrs	10/17/16 1133 hrs	1.0 °C
2	10/18/16 0900 hrs to 10/19/16 0900 hrs	10/19/16 1215 hrs	1.0 °C
3	10/20/16 0900 hrs to 10/21/16 0900 hrs	10/21/16 1140 hrs	0.5 °C

Test Methods:

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

Organisms: *C. dubia* <24hrs, *P. promelas* <24hrs Culture Source: ASU Ecotox

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

Statistical Method: Toxcalc 5.0.25

<i>C. dubia</i>			<i>P. promelas</i>		
whole effluent toxicity			whole effluent toxicity		
	lethality	sublethality		lethality	sublethality
DMR Code	22414 10	22414 P0	DMR Code	22414 10	22414 P0
Result	100%	100%	Result	100%	100%
	lethality	sublethality		lethality	sublethality
DMR Code	TGP3B	TLP3B	DMR Code	TGP6C	TLP6C
Result	0	1	Result	0	0
	NOEC lethality	NOEC sublethal		NOEC lethality	NOEC sublethal
DMR Code	TOP3B	TPP3B	DMR Code	TOP6C	TPP6C
Result	100%	<32%	Result	100%	100%
	CV%			CV%	
DMR Code	TQP3B		DMR Code	TQP6C	
Result	18.8%		Result	11.0%	
control survival	control mean reproduction		control survival	control mean weight	
100%	24.1		100%	0.5970	
critical dil. survival	critical mean reproduction		critical dil. survival	critical mean weight	
100%	23.0		90%	0.6197	
			MSDp		
			0.1540		

Results Summary: No lethal or sublethal effects were measured to *P. promelas*; no lethal effects were measured to *C. dubia*, however sublethal effects were measured in all dilutions except the 100%.

QA/Reference Testing: Data attached

Reviewed By:



 Jennifer L. Bouldin, 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: 10/17/16 *C. dubia*
 Time Test Started: 1326 *C. dubia*
 Date Test Terminated: 10/24/16 *C. dubia*
 Time Test Terminated: 1330 *C. dubia*
 Laboratory Analyst: Sweeney/Nicholson

Toxicity Test Performed: 7-day *Pimephales promelas* Survival and Growth
 Effluent Sampling Point: Walnut Ridge WWT Plant
 Date Test Started: 10/17/16 *P. promelas*
 Time Test Started: 1340 *P. promelas*
 Date Test Terminated: 10/24/16 *P. promelas*
 Time Test Terminated: 1400 *P. promelas*
 Laboratory Analyst: McCauley/Sanchez-Gonzales

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
Fathead Minnow Survival and Growth	Section 11

II. Test Organisms

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

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

III. External Factors

A. Incubator

Temperature (°C)

Average: 24.8

Range: 24.5-25.0

Light Cycle: 16 hours light/ 8 hours dark

Light Intensity: 100 footcandles

Control Water: Moderately Hard Synthetic Water (#MH 977)

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 09052016) and yeast/cereal/trout chow mix (#YCT 09142016) one hour prior to test setup and once daily thereafter.

C. *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.

IV. Quality Assurance

A. Standard Toxicant: Sodium Chloride

B. Organism: *Ceriodaphnia dubia*

Date and time of Reference Toxicant Test

Start: 10/10/2016

Terminated: 10/17/2016

Time of Reference Toxicant Test

Start: 16:15

Terminated: 15:30

Laboratory Analyst: McCauley

Dilution Water Used: Moderately Hard Synthetic Water #976

Results: Survival and reproduction were within the acceptable limits

Survival

LOEC: 1.82 g/L NaCl

EC50: 1.73 g/L NaCl

Reproduction

LOEC: 0.89 g/L NaCl

IC25: 0.76 g/L NaCl

C. Organism: *Pimephales promelas*

Date of Reference Toxicant Test

Start: 10/05/2016

Terminated: 10/12/2016

Time of Reference Toxicant Test

Start: 15:40

Terminated: 16:00

Laboratory Analyst: McCauley

Dilution Water Used: Moderately Hard Synthetic Water #976

Results: Survival and growth were within the acceptable limits

Survival

LOEC: 5.63 g/L NaCl

EC50: 5.41 g/L NaCl

Growth

LOEC: 7.50 g/L NaCl

IC25: 6.12 g/L NaCl

V. Physical and Chemical Data - See Attached

VI. Survival and Growth Data - See Attached

VII. Statistical Methods - See Attached

VIII. NPDES Permit Results - 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	10/16/16		0900	10/17/16
Composite 2:	Collected from	0900	10/18/16		0900	10/19/16
Composite 3:	Collected from	0900	10/20/16		0900	10/21/16

Test Initiated: 1326

Date: 10/17/16

Time Terminated: 1330

Date: 10/24/16

Dilution H₂O: MH 977

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	100	100	100	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	18	20	17	10	18
B	24	20	17	18	16	27
C	22	17	23	20	20	22
D	28	14	16	19	20	19
E	24	20	27	21	15	30
F	26	21	19	11	25	17
G	26	24	19	11	11	24
H	22	21	21	17	20	25
I	17	19	17	24	19	21
J	26	19	18	21	21	27
Mean	24.1	19.3	19.7	17.9	17.7	23.0
CV%*	13.0	13.8	16.8	23.5	26.4	18.8

*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 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 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: 1
5. Report the NOEC value for survival, Parameter #TOP3B:
NOEC survival 100% effluent
6. Report the NOEC value for reproduction, Parameter #TPP3B:
NOEC reproduction <32% effluent
7. Report the % coefficient of variation (largest of critical and control dilutions), Parameter #TQP3B:
CV % reproduction 18.8% (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%

WET Testing Summary Form
Ceriodaphnia dubia (Cladoceran)

Chemical Parameters Chart

Permittee: Walnut Ridge Wastewater Plant
 NPDES No.: AR0046566
 Contact: John Kopp
 Analyst: Sweeney/Nicholson

Sample No. 1 Collected Ending Date: 10/17/16 Time: 0900
 Sample No. 2 Collected Ending Date: 10/19/16 Time: 0900
 Sample No. 3 Collected Ending Date: 10/21/16 Time: 0900
 Test Begin: Date: 10/17/16 Time: 1326 Test End: Date: 10/24/16 Time: 1330

Initial Water Chemistry for Chronic Tests								
Project: Walnut Ridge – <i>C. dubia</i>								
Test day		0	1	2	3	4	5	6
Date		10/17/2016	10/18/2016	10/19/2016	10/20/2016	10/21/2016	10/22/2016	10/23/2016
H ₂ O #		MH977	MH977	MH977	MH977	MH977	MH977	MH977
Temp (°C)	Control	23.8	23.7	23.0	22.9	23.0	23.0	22.8
	32%	24.0	23.9	23.0	23.0	23.1	23.1	22.7
	42%	24.2	23.8	23.0	22.8	23.0	23.4	22.9
	56%	24.0	23.8	23.0	23.2	23.5	23.0	23.2
	80%	24.0	23.9	23.5	23.0	23.2	23.3	23.1
	100%	24.2	24.0	23.5	23.2	23.5	23.5	23.1
pH (Standard Units)	Control	7.99	7.97	7.66	8.09	8.16	8.10	8.04
	32%	8.16	8.03	7.90	8.13	8.24	8.19	8.11
	42%	8.16	8.06	7.93	8.16	8.25	8.20	8.13
	56%	8.16	8.07	7.96	8.15	8.25	8.19	8.13
	80%	8.13	8.09	7.96	8.14	8.24	8.14	8.17
	100%	8.10	8.06	7.93	8.07	8.20	8.12	8.15
DO (mg/L)	Control	8.4	8.4	8.5	8.8	8.3	8.5	8.9
	32%	8.4	8.4	8.5	8.8	8.5	8.4	8.8
	42%	8.4	8.0	8.3	8.8	8.5	8.4	8.8
	56%	8.5	8.3	8.3	8.7	8.4	8.4	8.7
	80%	8.6	8.3	8.2	8.7	8.5	8.3	8.6
	100%	8.6	8.3	8.1	8.7	8.5	8.3	8.5
Cond (µS/cm)	Control	310	306	301	305	312	309	311
	32%	380	382	422	412	421	419	416
	42%	400	409	448	449	457	461	450
	56%	432	448	497	500	507	503	499
	80%	485	506	588	587	596	576	588
	100%	535	562	671	651	674	664	660
Alk (mg/L)	Control	62		62		62		
	100%	158		150		162		
Hard (mg/L)	Control	90		90		90		
	100%	210		220		230		

WET Testing Summary Form
***Ceriodaphnia dubia* (Cladoceran)**

Chemical Parameters Chart

Permittee: Walnut Ridge Wastewater Plant Sample No. 1 Collected Ending Date: 10/17/16 Time: 0900
 NPDES No.: AR0046566 Sample No. 2 Collected Ending Date: 10/19/16 Time: 0900
 Contact: Jon Kopp Sample No. 3 Collected Ending Date: 10/21/16 Time: 0900
 Analyst: Sweeney/Nicholson Test Begin: Date: 10/17/16 Time: 1326 Test End: Date: 10/24/16 Time: 1330

Final Water Chemistry for Chronic Tests								
Project: Walnut Ridge - <i>C. dubia</i>								
Test day		1	2	3	4	5	6	7
Date:		10/18/2016	10/19/2016	10/20/2016	10/21/2016	10/22/2016	10/23/2016	10/24/2016
H ₂ O #		MH977	MH977	MH977	MH977	MH977	MH977	MH977
Temp (°C)	Control	23.8	23.9	23.1	22.0	22.5	23.0	24.4
	32%	24.0	23.7	23.1	22.0	22.7	23.2	24.5
	42%	23.9	23.2	23.0	22.3	22.6	22.9	24.4
	56%	23.9	23.0	23.2	22.5	22.9	22.9	24.8
	80%	24.0	23.1	23.0	22.1	23.0	23.0	24.6
	100%	24.0	23.0	23.4	22.3	23.2	23.1	24.6
pH (Standard Units)	Control	8.29	8.16	8.46	8.66	8.41	8.42	8.29
	32%	8.47	8.55	8.44	8.74	8.43	8.53	8.19
	42%	8.44	8.62	8.52	8.74	8.41	8.49	8.50
	56%	8.51	8.59	8.50	8.76	8.46	8.50	8.25
	80%	8.53	8.68	8.56	8.75	8.49	8.57	8.33
	100%	8.54	8.65	8.55	8.75	8.51	8.53	8.41
DO (mg/L)	Control	8.8	8.7	8.9	9.2	8.6	8.9	9.1
	32%	9.0	9.0	9.1	9.5	8.7	9.2	8.8
	42%	9.1	9.4	9.0	9.5	8.6	9.2	8.8
	56%	8.9	9.5	9.1	9.4	8.6	9.1	8.8
	80%	9.0	9.4	9.0	9.4	8.5	9.1	8.5
	100%	9.0	9.5	9.0	9.3	8.5	9.1	8.5

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	10/16/16	to	0900	10/17/16
Composite 2:	Collected from	0900	10/18/16	to	0900	10/18/16
Composite 3:	Collected from	0900	10/20/16	to	0900	10/19/16

Test Initiated: 1326

Date: 10/17/16

Time Terminated: 1400

Date: 10/24/16

Dilution H₂O: MH 977

DATA TABLE FOR SURVIVAL

Effluent Conc. %	% Survival in Replicate Chambers					Mean % Survival			CV%
	A	B	C	D	E	24h	48h	7 days	
Control	100	100	100	100	100	100	100	100	0.0
32	87.5	100	100	87.5	100	100	100	95	7.6
42	62.5	100	100	62.5	75	100	97.5	80	21.7
56	75	100	100	87.5	100	100	100	90	12.1
80	87.5	100	87.5	62.5	100	100	97.5	87.5	16.1
100	100	100	100	62.5	100	100	100	90	16.6

DATA TABLE FOR GROWTH

Effluent Conc %	Average Dry Weight in Replicate Chambers (mg)					Mean Dry Weight (mg)	CV%
	A	B	C	D	E		
Control	0.6125	0.5600	0.6475	0.5025	0.6625	0.5970	11.0
32	0.7229	0.6112	0.6888	0.6357	0.5962	0.6510	8.2
42	0.6220	0.5913	0.5775	0.5900	0.5433	0.5848	4.9
56	0.5267	0.6238	0.8000	0.5829	0.6300	0.6327	16.2
80	0.7186	0.6850	0.6257	0.7120	0.5712	0.6625	9.5
100	0.6137	0.6075	0.5987	0.6360	0.6425	0.6197	3.0

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 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 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 low flow and control dilutions), Parameter #TQP6C: CV % growth 11.0% (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%

WET Testing Summary Form

Fathead Minnow Larvae (*Pimephales promelas*)

Chemical Parameters Chart

Permittee: Walnut Ridge Wastewater Plant

Sample No. 1 Collected Ending Date: 10/17/16 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 10/19/16 Time: 0900

Contact: John Kopp

Sample No. 3 Collected Ending Date: 10/21/16 Time: 0900

Analyst: McCauley/Sanchez-Gonzales Test Begin: Date: 10/17/16 Time: 1340 Test End: Date: 10/24/16 Time: 1400

Initial Water Chemistry for Chronic Tests								
Project: Walnut Ridge – <i>P. promelas</i>								
Test day		0	1	2	3	4	5	6
Date		10/17/2016	10/18/2016	10/19/2016	10/20/2016	10/21/2016	10/22/2016	10/23/2016
H ₂ O #		MH977	MH977	MH977	MH977	MH977	MH977	MH977
Temp (°C)	Control	23.8	23.7	23.0	22.9	23.0	23.0	22.8
	32%	24.0	23.9	23.0	23.0	23.1	23.1	22.7
	42%	24.2	23.8	23.0	22.8	23.0	23.4	22.9
	56%	24.0	23.8	23.0	23.2	23.5	23.0	23.2
	80%	24.0	23.9	23.5	23.0	23.2	23.3	23.1
	100%	24.2	24.0	23.5	23.2	23.5	23.5	23.1
pH (Standard Units)	Control	7.99	7.97	7.66	8.09	8.16	8.10	8.04
	32%	8.16	8.03	7.90	8.13	8.24	8.19	8.11
	42%	8.16	8.06	7.93	8.16	8.25	8.20	8.13
	56%	8.16	8.07	7.96	8.15	8.25	8.19	8.13
	80%	8.13	8.09	7.96	8.14	8.24	8.14	8.17
	100%	8.10	8.06	7.93	8.07	8.20	8.12	8.15
DO (mg/L)	Control	8.4	8.4	8.5	8.8	8.3	8.5	8.9
	32%	8.4	8.4	8.5	8.8	8.5	8.4	8.8
	42%	8.4	8.0	8.3	8.8	8.5	8.4	8.8
	56%	8.5	8.3	8.3	8.7	8.4	8.4	8.7
	80%	8.6	8.3	8.2	8.7	8.5	8.3	8.6
	100%	8.6	8.3	8.1	8.7	8.5	8.3	8.5
Cond (µS/cm)	Control	310	306	301	305	312	309	311
	32%	380	382	422	412	421	419	416
	42%	400	409	448	449	457	461	450
	56%	432	448	497	500	507	503	499
	80%	485	506	588	587	596	576	588
	100%	535	562	671	651	674	664	660
Alk (mg/L)	Control	62		62		62		
	100%	158		150		162		
Hard (mg/L)	Control	90		90		90		
	100%	210		220		230		

WET Testing Summary Form
Fathead Minnow Larvae (*Pimephales promelas*)

Chemical Parameters Chart

Permittee: Walnut Ridge Wastewater Plant

Sample No. 1 Collected Ending Date: 10/17/16 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 10/19/16 Time: 0900

Contact: John Kopp

Sample No. 3 Collected Ending Date: 10/21/16 Time: 0900

Analyst: McCauley/Sanchez-Gonzales

Test Begin: Date: 10/17/16 Time: 1340

Test End: Date: 10/24/16 Time: 1400

Final Water Chemistry for Chronic Tests								
Project: Walnut Ridge – <i>P. promelas</i>								
Test day		1	2	3	4	5	6	7
Date		10/18/2016	10/19/2016	10/20/2016	10/21/2016	10/22/2016	10/23/2016	10/24/2016
H ₂ O #		MH977	MH977	MH977	MH977	MH977	MH977	MH977
Temp (°C)	Control	23.0	22.2	22.0	22.0	22.1	22.0	22.0
	32%	23.0	22.0	22.1	22.2	22.2	22.6	22.3
	42%	23.5	22.0	22.1	22.0	22.6	22.2	22.3
	56%	23.0	22.0	22.0	22.2	22.5	21.6	22.3
	80%	23.5	22.2	22.1	22.2	22.4	22.4	22.6
	100%	23.5	21.9	21.9	21.8	22.5	22.1	22.5
pH (Standard Units)	Control	7.67	7.11	7.65	7.71	7.61	7.50	7.42
	32%	7.86	7.35	7.78	7.83	7.75	7.70	7.61
	42%	7.90	7.42	7.88	7.84	7.89	7.74	7.65
	56%	7.95	7.47	7.89	7.94	7.91	7.76	7.67
	80%	8.03	7.64	8.00	8.03	8.04	7.82	7.76
	100%	8.09	7.71	8.12	8.02	8.04	7.85	7.81
DO (mg/L)	Control	8.3	6.0	7.3	6.7	7.5	6.6	6.2
	32%	7.9	6.4	6.7	6.7	6.7	6.2	6.0
	42%	7.6	6.5	6.6	5.7	6.6	6.1	6.0
	56%	7.6	6.5	6.6	5.7	6.6	5.9	6.0
	80%	7.6	6.3	6.5	5.8	6.6	5.9	5.7
	100%	7.5	6.4	6.4	5.7	6.3	5.4	5.7

Ceriodaphnia Survival and Reproduction Test-7 Day Survival

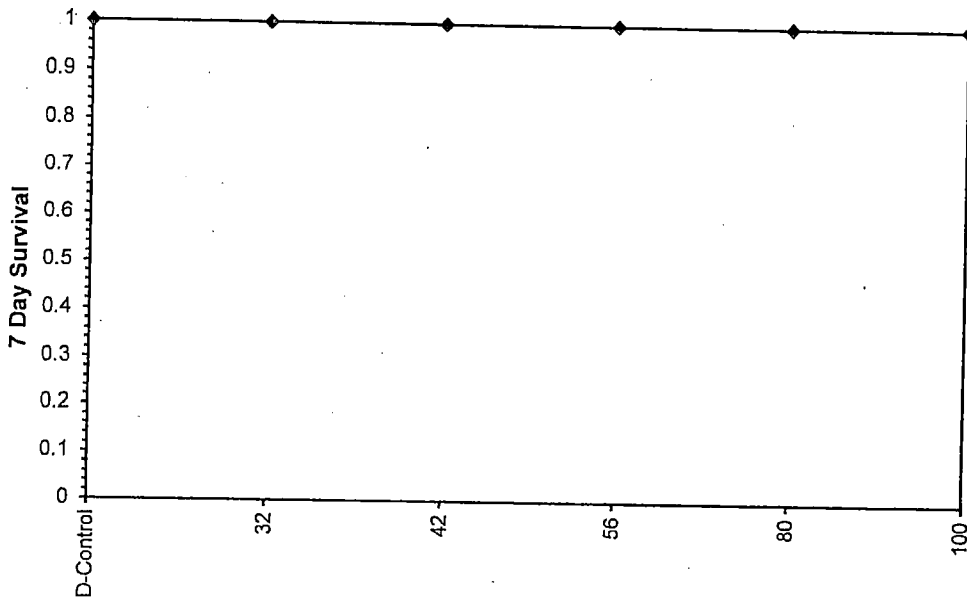
Start Date: 10/17/2016 13:26 Test ID: W. Ridge Sample ID: AR0046566-NPDES Permit #
 End Date: 10/24/2016 13:30 Lab ID: ASU ERF Sample Type: EFF1-POTW
 Sample Date: Protocol: EPAF 02-EPA Freshwater Test Species: CD-Ceriodaphnia dubia
 Comments:

Conc-%	1	2	3	4	5	6	7	8	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	1.0000	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	1.0000
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	1.0000	1.0000	0	10	10	10	1.0000	0.0500
42	1.0000	1.0000	0	10	10	10	1.0000	0.0500
56	1.0000	1.0000	0	10	10	10	1.0000	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

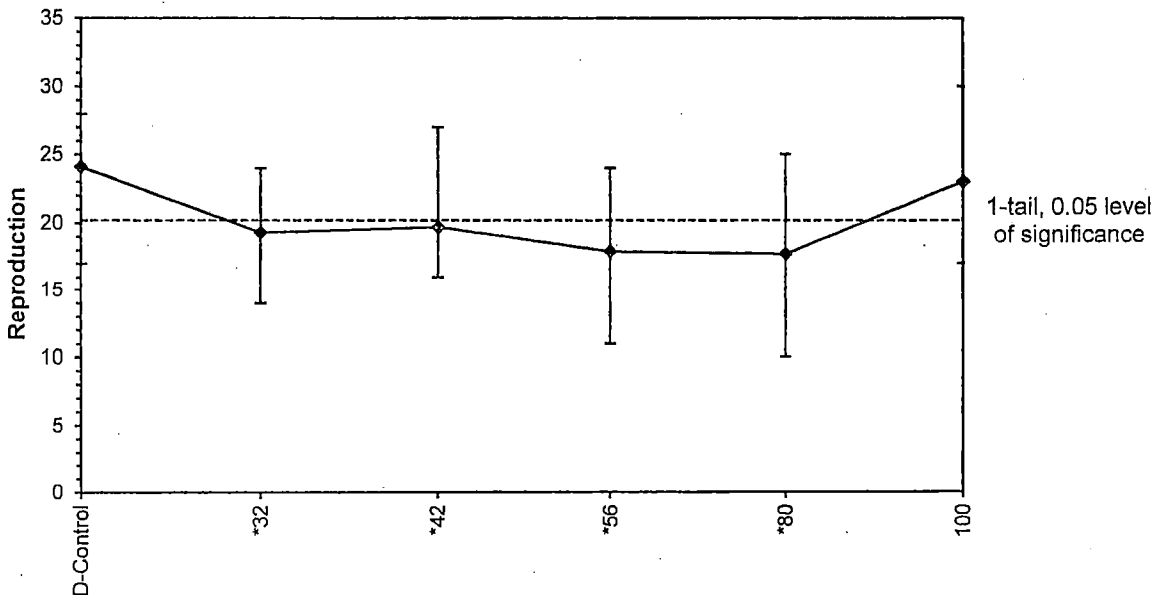
Start Date: 10/17/2016 13:26 Test ID: Wridge Sample ID: AR0046566-NPDES Permit #
 End Date: 10/24/2016 13:30 Lab ID: ASU-ERF Sample Type: EFF1-POTW
 Sample Date: Protocol: EPAF 02-EPA Freshwater Test Species: CD-Ceriodaphnia dubia
 Comments:

Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	26.000	24.000	22.000	28.000	24.000	26.000	26.000	22.000	17.000	26.000
32	18.000	20.000	17.000	14.000	20.000	21.000	24.000	21.000	19.000	19.000
42	20.000	17.000	23.000	16.000	27.000	19.000	19.000	21.000	17.000	18.000
56	17.000	18.000	20.000	19.000	21.000	11.000	11.000	17.000	24.000	21.000
80	10.000	16.000	20.000	20.000	15.000	25.000	11.000	20.000	19.000	21.000
100	18.000	27.000	22.000	19.000	30.000	17.000	24.000	25.000	21.000	27.000

Conc-%	Mean	N-Mean	Transform: Untransformed					N	t-Stat	1-Tailed Critical	MSD
			Mean	Min	Max	CV%					
D-Control	24.100	1.0000	24.100	17.000	28.000	13.041	10				
*32	19.300	0.8008	19.300	14.000	24.000	13.828	10	2.835	2.287	3.872	
*42	19.700	0.8174	19.700	16.000	27.000	16.759	10	2.599	2.287	3.872	
*56	17.900	0.7427	17.900	11.000	24.000	23.474	10	3.662	2.287	3.872	
*80	17.700	0.7344	17.700	10.000	25.000	26.372	10	3.780	2.287	3.872	
100	23.000	0.9544	23.000	17.000	30.000	18.785	10	0.650	2.287	3.872	

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.50146	1.035	-0.22898	-0.17884
Bartlett's Test indicates equal variances (p = 0.56)	3.92402	15.0863		
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Dunnett's Test	<32	32		
	MSDu	MSDp	MSB	MSE
	3.87186	0.16066	71.2167	14.3352
			F-Prob	df
			8.2E-04	5, 54

Dose-Response Plot



CHRONIC TEST DATA SHEET
Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 101716 Time: 1320 Test Species: C. dubia
Dilution H₂O: MHAT Ending Date: 102416 Time: 1330 Age: <24 hours

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

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
Control	1	0	0	0	7	8	0	11	24
	2	↓	↓	↓	6	8	0	10	25 24 p
	3	↓	↓	↓	5	6	2	9	22
	4	↓	↓	↓	6	0	10	12	28
	5	↓	↓	↓	6	5	0	13	24
	6	↓	↓	↓	6	8	0	12	30 26 p
	7	↓	↓	↓	6	6	0	14	26
	8	↓	↓	↓	3	0	6	13	22
	9	↓	↓	↓	6	6	0	5	17
	10	↓	↓	↓	4	8	7	15	26
32%	1	0	0	0	7	11	0	0	18
	2	↓	↓	↓	6	6	0	8 8	20
	3	↓	↓	↓	6	6	0	5	17
	4	↓	↓	↓	5	0	3	6	14
	5	↓	↓	↓	7	8	0	5	20
	6	↓	↓	↓	7	8	0	6	21
	7	↓	↓	↓	5	9	0	10	24
	8	↓	↓	↓	6	0	3	12	21
	9	↓	↓	↓	5	9	0	5	19
	10	↓	↓	↓	6	6	0	7	19
Date		101816	101916	102016	102116	102216	102316	102416	102416
Initials		ps	den	ps	den	ps	ps	ps	den

CHRONIC TEST DATA SHEET
Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 10/1/16 Time: 1326 Test Species: C. dubia
Dilution H₂O: MH977 Ending Date: 10/24/16 Time: 1330 Age: 424 hours

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

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
42%	1	0	0	0	5	7	0	8	20
	2	↓	↓	↓	7	6	0	4	17
	3	↓	↓	↓	6	12	0	5	23
	4	↓	↓	↓	6	9	0	1	14
	5	↓	↓	↓	6	8	5	16	27
	6	↓	↓	↓	6	7	0	6	19
	7	↓	↓	↓	6	6	0	7	19
	8	↓	↓	↓	6	5	0	10	21
	9	↓	↓	↓	5	0	0	12	17
	10	↓	↓	↓	6	8	0	4	218
56%	1	0	0	0	6	5	0	6	17
	2	↓	↓	↓	5	0	3	10	18
	3	↓	↓	↓	7	11	0	2	20
	4	↓	↓	↓	6	8	0	5	19
	5	↓	↓	↓	5	9	0	7	21
	6	↓	↓	↓	5	2	0	4	11
	7	↓	↓	↓	6	5	0	0	11
	8	↓	↓	↓	5	7	0	5	17
	9	↓	↓	↓	3	9	0	12	24
	10	↓	↓	↓	6	8	0	7	21
Date		10/1/16	10/19/16	10/20/16	10/21/16	10/22/16	10/23/16	10/24/16	10/24/16
Initials		OP	Ben	OP	Ben	OP	OP	OP	Ben

CHRONIC TEST DATA SHEET

Ceriodaphnia dubia

Project: Walnut Ridge Beginning Date: 101716 Time: 1326 Test Species: C. dubia
 Dilution H₂O: MH977 Ending Date: 102416 Time: 1330 Age: 224 hours

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

Conc.	Rep	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Neonates
80%	1	0	0	0	5	0	5	0	10
	2	↓	↓	↓	6	0	2	8	16
	3	↓	↓	↓	5	0	5	10	20
	4	↓	↓	↓	6	7	0	7	20
	5	↓	↓	↓	4	6	0	5	15
	6	↓	↓	↓	7	11	0	7	25
	7	↓	↓	↓	5	6	0	0	11
	8	↓	↓	↓	7	2	0	11	20
	9	↓	↓	↓	3	0	6	10	19
	10	↓	↓	↓	6	9	0	6	21
100%	1	0	0	0	4	0	5	9	18
	2	↓	↓	↓	7	7	0	13	27
	3	↓	↓	↓	5	9	2	6	22
	4	↓	↓	↓	7	7	0	5	19
	5	↓	↓	↓	7	11	0	12	30
	6	↓	↓	↓	7	12	0	0	19
	7	↓	↓	↓	7	10	0	7	24
	8	↓	↓	↓	6	9	0	10	25
	9	↓	↓	↓	6	7	2	6	19 21
	10	↓	↓	↓	6	0	5	16	27
Date		101816	101916	102016	102116	102216	102316	102416	102416
Initials		op	den	op	den	op	op	op	den

Larval Fish Growth and Survival Test-7 Day Survival

Start Date: 10/17/2016 13:40	Test ID: W. Ridge	Sample ID: AR0046566-NPDES Permit #
End Date: 10/24/2016 14:00	Lab ID: ASU ERF	Sample Type: EFF1-POTW
Sample Date:	Protocol: EPAF 02-EPA Freshwater	Test Species: PP-Pimephales promelas

Comments:

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	0.8750	1.0000
42	0.6250	1.0000	1.0000	0.6250	0.7500
56	0.7500	1.0000	1.0000	0.8750	1.0000
80	0.8750	1.0000	0.8750	0.6250	1.0000
100	1.0000	1.0000	1.0000	0.6250	1.0000

Conc-%	Mean	N-Mean	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical
			Mean	Min	Max	CV%	N		
D-Control	1.0000	1.0000	1.3931	1.3931	1.3931	0.000	5		
32	0.9500	0.9500	1.3196	1.2094	1.3931	7.623	5	22.50	16.00
42	0.8000	0.8000	1.1314	0.9117	1.3931	21.676	5	20.00	16.00
56	0.9250	0.9250	1.2872	1.0472	1.3931	12.116	5	22.50	16.00
80	0.8750	0.8750	1.2234	0.9117	1.3931	16.097	5	20.00	16.00
100	0.9250	0.9250	1.2968	0.9117	1.3931	16.600	5	25.00	16.00

Auxiliary Tests

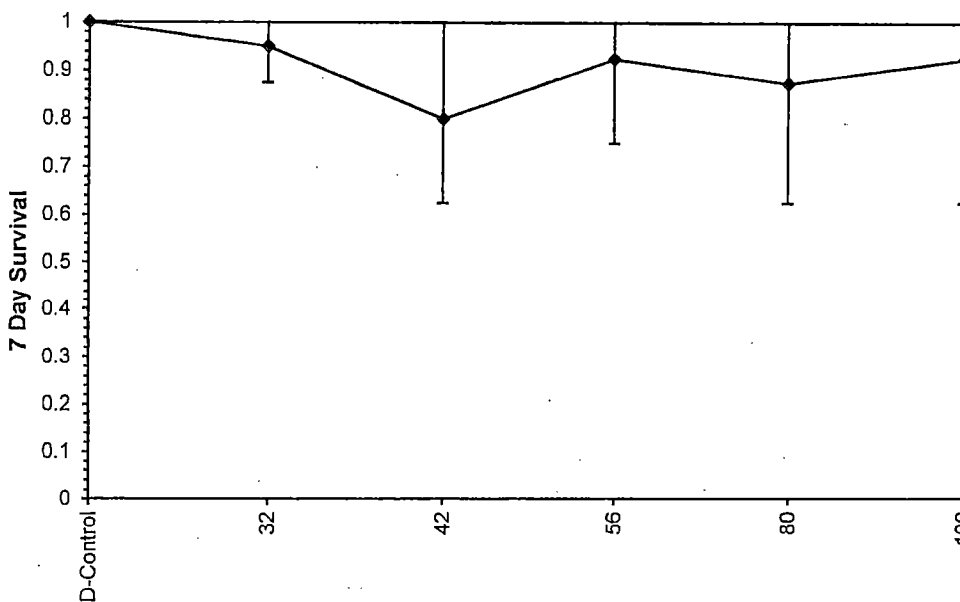
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	Statistic	Critical	Skew	Kurt
	0.9395	0.9	-0.68326	0.2272

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

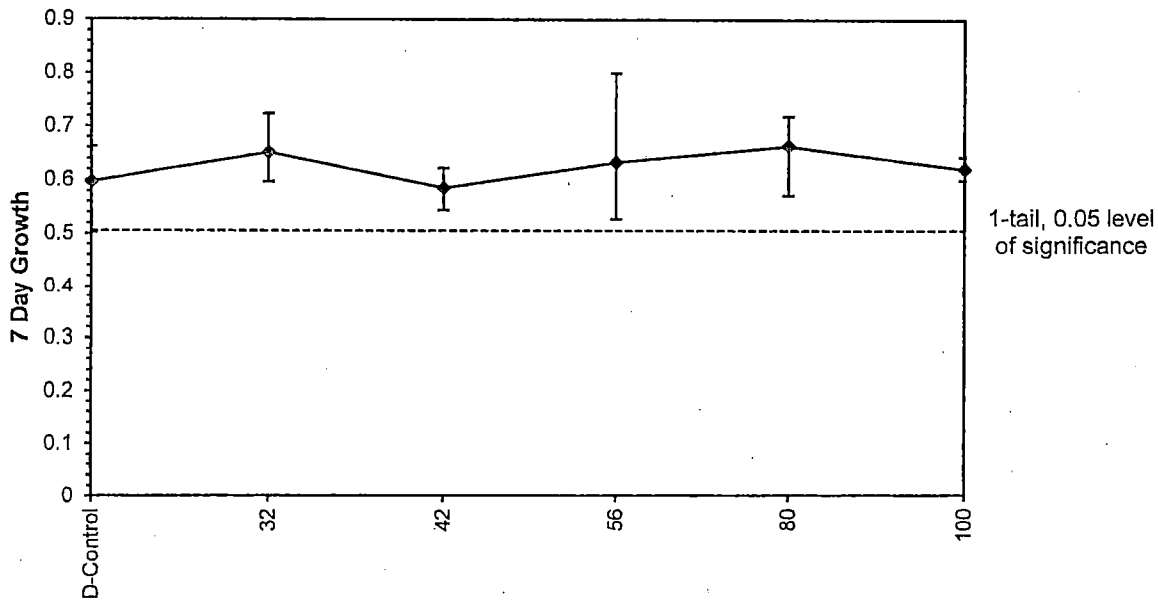
Start Date: 10/17/2016 13:40 Test ID: W. Ridge Sample ID: AR0046566-NPDES Permit #
 End Date: 10/24/2016 14:00 Lab ID: ASU ERF Sample Type: EFF1-POTW
 Sample Date: Protocol: EPAF 02-EPA Freshwater Test Species: PP-Pimephales promelas
 Comments:

Conc-%	1	2	3	4	5
D-Control	0.6125	0.5600	0.6475	0.5025	0.6625
32	0.7229	0.6112	0.6888	0.6357	0.5962
42	0.6220	0.5913	0.5775	0.5900	0.5433
56	0.5267	0.6238	0.8000	0.5829	0.6300
80	0.7186	0.6850	0.6257	0.7120	0.5712
100	0.6137	0.6075	0.5987	0.6360	0.6425

Conc-%	Mean	N-Mean	Transform: Untransformed					N	t-Stat	1-Tailed Critical	MSD
			Mean	Min	Max	CV%					
D-Control	0.5970	1.0000	0.5970	0.5025	0.6625	11.041	5				
32	0.6510	1.0904	0.6510	0.5962	0.7229	8.201	5	-1.386	2.360	0.0919	
42	0.5848	0.9796	0.5848	0.5433	0.6220	4.855	5	0.313	2.360	0.0919	
56	0.6327	1.0597	0.6327	0.5267	0.8000	16.159	5	-0.916	2.360	0.0919	
80	0.6625	1.1097	0.6625	0.5712	0.7186	9.481	5	-1.682	2.360	0.0919	
100	0.6197	1.0380	0.6197	0.5987	0.6425	3.028	5	-0.583	2.360	0.0919	

Auxiliary Tests	Statistic	Critical	Skew	Kurt						
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.96371	0.9	0.54022	1.64383						
Bartlett's Test indicates equal variances (p = 0.05)	11.0101	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	0.09191	0.15395	0.00457	0.00379	0.33719	5, 24

Dose-Response Plot



CHRONIC TEST DATA SHEET
Pimephales promelas

Project: Walnut Ridge Beginning Date: 10/7/16 Time: 1340 Test Species: P. promelas

Dilution H₂O: M4977 Ending Date: 10/24/16 Time: 1400 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	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	8/0	8/1	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	8/1	9
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	10
42%	1	8/0	8/1	7/0	7/0	7/1	6/1	5/0	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	8/1	7/0	5/0	5/0	5/0	14
	5	8/0	8/0	8/0	7/1	7/0	7/1	6/0	15
56%	1	8/0	8/0	8/0	8/1	7/0	7/1	6/0	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	8/1	7/0	7/0	7/0	7/0	19
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	20
Date		10/8/16	10/19/16	10/20/16	10/21/16	10/22/16	10/23/16	10/24/16	10/24/16
Initials		jen	jm	lsb	mm	lsb	lsb	jm	jm

CHRONIC TEST DATA SHEET

Pimephales promelas

Project: Walnut Ridge Beginning Date: 10/7/16 Time: 1340 Test Species: *P. promelas*

Dilution H₂O: M4977 Ending Date: 10/24/16 Time: 1400 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	Pan #
80%	1	8/0	8/0	8/0	8/0	8/0	8/0	8/1	21
	2	8/0	8/0	8/0	8/0	8/0	8/0	8/0	22
	3	8/0	8/0	8/0	8/1	7/0	7/0	7/0	23
	4	8/0	8/1	7/1	6/1	5/0	5/0	5/0	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	8/1	7/1	6/1	5/0 5/0	5/0	29
	5	8/0	8/0	8/0	8/0	8/0	8/0	8/0	30
Date		10/8/16	10/19/16	10/20/16	10/21/16	10/22/16	10/23/16	10/24/16	10/24/16
Initials		ALW	fm	ISO	MAN	ISO	ISO	fm	fm

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

Test Day		0	1	2	3	4	5	6
Date		10/17/16	10/18/16	10/19/16	10/20/16	10/21/16	10/22/16	10/23/16
H ₂ O Batch #		MH977	MH977	MH977	MH977	MH977	MH977	MH977
Temp. (°C)	Control	23.8	23.7	23.0	22.9	23.0	23.0	22.8
	32%	24.0	23.9	23.0	23.0	23.1	23.1	22.7
	42%	24.2	23.8	23.0	22.8	23.0	23.4	22.9
	56%	24.0	23.8	23.0	23.2	23.5	23.0	23.2
	80%	24.0	23.9	23.5	23.0	23.2	23.3	23.1
	100%	24.2	24.0	23.5	23.2	23.5	23.5	23.1
pH	Control	7.99	7.97	7.66	8.09	8.16	8.10	8.04
	32%	8.16	8.03	7.90	8.13	8.24	8.19	8.11
	42%	8.16	8.06	7.93	8.16	8.25	8.20	8.13
	56%	8.16	8.07	7.96	8.15	8.25	8.19	8.13
	80%	8.13	8.09	7.96	8.14	8.24	8.14	8.17
	100%	8.10	8.04	7.93	8.07	8.20	8.12	8.15
DO (mg/L)	Control	8.4	8.4	8.5	8.8	8.3	8.5	8.9
	32%	8.4	8.4	8.5	8.8	8.5	8.4	8.8
	42%	8.4	8.3	8.3	8.8	8.5	8.4	8.8
	56%	8.5	8.3	8.3	8.7	8.4	8.4	8.7
	80%	8.6	8.3	8.2	8.7	8.9	8.3	8.6
	100%	8.6	8.3	8.1	8.7	8.5	8.3	8.5
Cond. (µS/cm)	Control	310	306	301	305	312	309	311
	32%	380	382	422	412	421	419	416
	42%	400	409	448	449	457	461	450
	56%	432	448	497	500	507	503	499
	80%	485	506	588	587	596	576	588
	100%	535	562	621	651	674	664	660
Alk. (mg/L)	Control	62		62		62		
	100%	158		150		162		
Hard. (mg/L)	Control	90		90		90		
	100%	210		220		230		
Initials:		den/ps	den/ps	gaston	den/ps	den	den/ps	den/ps

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

Test Day		1	2	3	4	5	6	7
Date		10/8/16	10/9/16	10/20/16	10/21/16	10/22/16	10/25/16	10/24/16
H ₂ O Batch #		MH977	MH977	MH977	MH977	MH977	MH977	MH977
Temp. (°C)	Control	23.8	23.9	23.1	22.0	22.9	23.0	24.4
	32%	24.0	23.7	23.1	22.0	22.7	23.2	24.5
	42%	23.9	23.2	23.0	22.3	22.6	22.9	24.4
	56%	23.9	23.0	23.2	22.5	22.9	22.9	24.8
	80%	24.0	23.1	23.0	22.1	23.0	23.0	24.6
	100%	24.0	23.0	23.4	22.3	23.2	23.1	24.6
pH	Control	8.29	8.16	8.46	8.66	8.41	8.42	8.29
	32%	8.47	8.55	8.44	8.74	8.43	8.53	8.19
	42%	8.44	8.62	8.52	8.74	8.41	8.49	8.50
	56%	8.51	8.59	8.50	8.76	8.46	8.50	8.25
	80%	8.53	8.68	8.56	8.75	8.49	8.57	8.33
	100%	8.54	8.69	8.55	8.75	8.51	8.53	8.41
DO (mg/L)	Control	8.8	8.7	8.9	9.2	8.6	8.9	9.1
	32%	9.0	9.0	9.1	9.5	8.7	9.2	8.8
	42%	9.1	9.4	9.0	9.5	8.6	9.2	8.8
	56%	8.9	9.5	9.1	9.4	8.6	9.1	8.8
	80%	9.0	9.4	9.0	9.4	8.5	9.1	8.5
	100%	9.0	9.5	9.0	9.3	8.5	9.1	8.5
Initials		JS	JS	JS	JS	JS	JS	JS

Final Water Chemistry for Chronic Tests
Project: Walnut Ridge - *P. promelas*

Test Day		1	2	3	4	5	6	7
Date		10/8/16	10/9/16	10/20/16	10/21/16	10/22/16	10/23/16	10/24/16
H ₂ O Batch #		MH977	MH977	MH977	MH977	MH977	MH977	MH977
Temp. (°C)	Control	23.0	22.2	22.0	22.0	22.1	22.0	22.0
	32%	23.0	22.0	22.1	22.2	22.2	22.6	22.3
	42%	23.5	22.0	22.1	22.0	22.6	22.2	23.3
	56%	23.0	22.0	22.0	22.2	22.5	21.6	22.3
	80%	23.5	22.7	22.1	22.2	22.4	22.4	22.6
	100%	23.5	21.9	21.9	21.8	22.5	22.1	22.5
pH	Control	7.67	7.11	7.65	7.71	7.61	7.55	7.42
	32%	7.86	7.35	7.78	7.83	7.75	7.70	7.61
	42%	7.90	7.42	7.88	7.84	7.89	7.74	7.65
	56%	7.95	7.47	7.89	7.94	7.91	7.76	7.67
	80%	8.03	7.64	8.00	8.03	8.04	7.82	7.76
	100%	8.09	7.71	8.12	8.02	8.04	7.85	7.81
DO (mg/L)	Control	8.3	6.6	7.3	6.7	7.5	6.6	6.2
	32%	7.9	6.4	6.7	6.7	6.7	6.2	6.0
	42%	7.6	6.5	6.6	5.7	6.6	6.1	6.0
	56%	7.6	6.5	6.6	5.7	6.6	5.9	6.0
	80%	7.6	6.3	6.5	5.8	6.6	5.9	5.7
	100%	7.5	6.4	6.4	5.7	6.3	5.37	5.7
Initials		AM	AM	AM	AM	AM	AM	AM

AM AM AM AM AM AM AM AM



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: 101716 Sampling Date: 1016/1017 Arrival Time: 1133

Field Identification Number: EFF 001 Description: Composite

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

Drop-Off Location: ASU-ERF

Storage While Shipped: Cooler - Ice

Analysis Requested: Chronic NET Testing

Initial Water Chemistry Analysis:

Sample Received by: Jen

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

Date: 101716

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	Jen	101716	X	
Refrigerated at 4°C	Jen	↓	X	
Field Record Received	Jen			X
Sample Label Affixed Properly	Jen		X	
Project Leader Informed	Jen		X	

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: 10/19/16 Sampling Date: 10/18/10/19 Arrival Time: 12:15

Field Identification Number: EFF 002 Description: Composite

Shipped by: Federal Express UPS Hand delivered by: W. Ridge

Drop-Off Location: ASU-ERF

Storage While Shipped: Cooler - Ice

Analysis Requested: Chronic WET Testing

Initial Water Chemistry Analysis:

Sample Received by: Jen

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

Date: 10/19/16

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	Jen	10/19/16	X	
Refrigerated at 4°C	Jen	↓	X	
Field Record Received	Jen			X
Sample Label Affixed Properly	Jen		X	
Project Leader Informed	Jen		X	

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: 102116 Sampling Date: 1020/1021 Arrival Time: 1140

Field Identification Number: EFF 3 Description: Composite

Shipped by: Federal Express _____ UPS _____ Hand delivered by: Carlos Vidar

Drop-Off Location: ASU-ERF

Storage While Shipped: Cooler w/ ice

Analysis Requested: Chronic WET Testing

Initial Water Chemistry Analysis:

Sample Received by: Jen

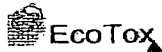
Temperature (°C): .5°C

Ice Present upon delivery: YES NO

Date: 102116

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	Jen	102116	X	
Refrigerated at 4°C	Jen		X	
Field Record Received	Jen	↓		X
Sample Label Affixed Properly	Jen		X	
Project Leader Informed	Jen	↓	X	

Comments: _____



Ecotoxicology Research Facility

Ecotoxicology Research Facility

Arkansas State University

2645 Caddo Drive

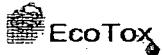
State University, AR 72467

(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 #:				Chronic C. dubia			
Remarks:			Contact: Jonathan Kopp							
Cont.#	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix			
					Comp	Grab	Aqueous	Soil	Other	
1	—	EFF 001	10-16-16	9am.	X					
			10-17-16	9am						
Ice present at delivery:			<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No							
Temp:			1.0 °C <i>ben</i> Initials							
1. Relinquished By (sign) <i>[Signature]</i>			Date	Time	1. Received By (sign) <i>[Signature]</i>			Date	Time	
2. Relinquished By (sign)			Date	Time	2. Received By (sign)			Date	Time	



Ecotoxicology Research Facility

Ecotoxicology Research Facility

Arkansas State University
2645 Gaddo 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) 			PO #:				Chronic <i>C. dubia</i>	Chronic <i>P. promelas</i>		
Remarks:			Contact: Jonathan Kopp							
Cont.#	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix			
					Comp	Grab	Aqueous	Soil	Other	
			10-18	10-19	9am - 9am	✓				
Ice present at delivery:			<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No							
Temp:			1.0 °C <i>ALH</i> Initials							
1. Relinquished By (sign) 			Date	Time	1. Received By (sign) 			Date	Time	
2. Relinquished By (sign)			Date	Time	2. Received By (sign)			Date	Time	



Ecotoxicology Research Facility
 Arkansas State University
 2645 Gaddo 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 #:				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	
			10-20	10-21	9am - 9am	<input checked="" type="checkbox"/>				
Ice present at delivery:			<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No							
Temp:			.5 °C <i>Sen</i> Initials							
1. Relinquished By (sign) <i>Caitlin [Signature]</i>			Date	Time	1. Received By (sign) <i>Ashley [Signature]</i>			Date	Time	
			10-21-16	1140				102116	1140	
2. Relinquished By (sign)			Date	Time	2. Received By (sign)			Date	Time	



Ecotoxicology Research Facility



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College of Sciences & Mathematics
www.astate.edu

November 18, 2016

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 November 2, 2016. No lethal or sublethal effects were measured in *Ceriodaphnia dubia* exposed to effluent dilutions.

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

Arkansas State University Ecotoxicology Research Facility

Laboratory Report

Facility Director / Lab Contact: Jennifer Bouldin
 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	11/01/16 0900 hrs to 11/02/16 0900 hrs	11/02/16 1158 hrs	2.1 °C
2	11/03/16 0900 hrs to 11/04/16 0900 hrs	11/04/16 1155 hrs	0.8 °C
3	11/05/16 0900 hrs to 11/06/16 0900 hrs	11/07/16 1134 hrs	0.0 °C

Test Methods:

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

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

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

Culture Source: ASU Ecotox

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

Critical Dilution: 100%

Statistical Method: Toxcalc 5.0.25

C. dubia

whole effluent toxicity

	lethality	sublethality
DMR Code	22414 10	22414 P0
Result	100%	100%

	lethality	sublethality
DMR Code	TGP3B	TLP3B
Result	0	0

	NOEC lethality	NOEC sublethal
DMR Code	TOP3B	TPP3B
Result	100%	100%

	CV%
DMR Code	TQP3B
Result	15.4%

control survival	control mean reproduction
100%	26.5
critical dil. survival	critical mean reproduction
100%	29.2

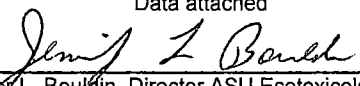
Results Summary:

No lethal or sublethal effects were measured to *C. dubia* exposed to effluent dilutions.

QA/Reference Testing:

Data attached

Reviewed By:


 Jennifer L. Bouldin, 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: 11/2/16 *C. dubia*
 Time Test Started: 1200 *C. dubia*
 Date Test Terminated: 11/9/16 *C. dubia*
 Time Test Terminated: 1105 *C. dubia*
 Laboratory Analyst: Sweeney/Nicholson/Cooper

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
Fathead Minnow Survival and Growth	Section 11

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: 24.8
 Range: 24.5-25.0
 Light Cycle: 16 hours light/ 8 hours dark
 Light Intensity: 100 footcandles
 Control Water: Moderately Hard Synthetic Water (#MH 978)

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 101016) and yeast/cereal/trout chow mix (#YCT 110216) 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: 11/14/16

Terminated: 11/21/16

Time of Reference Toxicant Test

Start: 1520

Terminated: 1025

Laboratory Analyst: Cooper

Dilution Water Used: Moderately Hard Synthetic Water #979

Results: Survival and Reproduction within control limits

Survival

LOEC: 2.60 g/L NaCl

EC50: 2.21 g/L NaCl

Reproduction

LOEC: 1.82 g/L NaCl

IC25: 1.29 g/L NaCl

V. Physical and Chemical Data - See Attached

VI. Survival and Growth Data - See Attached

VII. Statistical Methods - See Attached

VIII. NPDES Permit Results - 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	11/01/16		0900	11/02/16
Composite 2:	Collected from	0900	11/03/16		0900	11/04/16
Composite 3:	Collected from	0900	11/05/16		0900	11/06/16

Test Initiated: 1200

Date: 11/02/16

Time Terminated: 1105

Date: 11/09/16

Dilution H₂O: MH 978

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	100	100	100	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	28	28	29	35	30
B	29	28	20	34	28	32
C	27	35	26	23	36	31
D	29	27	26	31	28	29
E	23	27	20	27	23	32
F	25	30	25	24	30	17
G	24	25	24	26	31	31
H	29	36	25	28	36	30
I	26	28	21	33	36	28
J	27	35	33	29	33	32
Mean	26.5	29.9	24.8	28.4	31.6	29.2
CV%*	8.0	13.2	16.0	12.7	13.9	15.4

*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 15.4 % (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%

WET Testing Summary Form
***Ceriodaphnia dubia* (Cladoceran)**

Chemical Parameters Chart

Permittee: Walnut Ridge Wastewater Plant

Sample No. 1 Collected Ending Date: 11/02/16 Time: 0900

NPDES No.: AR0046566

Sample No. 2 Collected Ending Date: 11/04/16 Time: 0900

Contact: John Kopp

Sample No. 3 Collected Ending Date: 11/06/16 Time: 0900

Analyst: Sweeney/Cooper

Test Begin: Date: 11/02/16 Time: 1200 Test End: Date: 11/09/16 Time: 1105

Initial Water Chemistry for Chronic Tests

Project: Walnut Ridge – *C. dubia*

Test day		0	1	2	3	4	5	6
Date		11/2/2016	11/3/2016	11/4/2016	11/5/2016	11/6/2016	11/7/2016	11/8/2016
H ₂ O #		MH978	MH978	MH978	MH978	MH978	MH978	MH978
Temp (°C)	Control	23.4	23.2	22.0	23.0	22.2	23.1	23.2
	32%	23.6	23.2	22.1	23.3	22.8	23.2	23.4
	42%	23.3	23.5	22.0	23.2	22.9	23.4	23.5
	56%	23.5	23.7	22.6	23.2	23.1	23.4	23.8
	80%	23.7	23.9	22.8	23.5	23.2	23.6	23.9
	100%	24.0	23.9	23.0	23.5	23.0	23.9	24.0
pH (Standard Units)	Control	8.11	7.93	7.78	7.70	8.07	8.12	8.18
	32%	8.38	8.25	8.10	7.78	8.33	8.36	8.42
	42%	8.37	8.29	8.14	7.91	8.34	8.39	8.45
	56%	8.40	8.32	8.17	7.96	8.37	8.42	8.47
	80%	8.41	8.34	8.20	7.98	8.38	8.43	8.50
	100%	8.41	8.35	8.21	7.98	8.38	8.44	8.51
DO (mg/L)	Control	8.9	9.0	8.7	9.1	8.8	8.7	8.7
	32%	8.7	8.7	8.9	9.0	8.9	8.6	8.6
	42%	8.9	8.7	8.8	8.8	8.8	8.6	8.6
	56%	8.7	8.6	8.7	8.7	8.8	8.5	8.6
	80%	8.6	8.4	8.7	8.7	8.8	8.4	8.5
	100%	8.6	8.4	8.6	8.7	8.9	8.5	8.5
Cond (µS/cm)	Control	309	309	309	306	306	312	318
	32%	319	317	321	320	321	317	312
	42%	318	324	327	324	327	319	318
	56%	329	330	33	331	335	322	324
	80%	341	341	343	344	350	327	327
	100%	352	350	354	356	363	332	332
Alk (mg/L)	Control	60		60			60	
	100%	164		196			176	
Hard (mg/L)	Control	100		100			100	
	100%	230		220			200	

WET Testing Summary Form
***Ceriodaphnia dubia* (Cladoceran)**
Chemical Parameters Chart

Permittee: Walnut Ridge Wastewater Plant
 NPDES No.: AR0046566
 Contact: John Kopp
 Analyst: Sweeney/Cooper

Sample No. 1 Collected Ending Date: 11/02/16 Time: 0900
 Sample No. 2 Collected Ending Date: 11/04/16 Time: 0900
 Sample No. 3 Collected Ending Date: 11/06/16 Time: 0900
 Test Begin: Date: 11/02/16 Time: 1200 Test End: Date: 11/09/16 Time: 1105

Final Water Chemistry for Chronic Tests								
Project: Walnut Ridge - <i>C. dubia</i>								
Test day		1	2	3	4	5	6	7
Date:		11/3/2016	11/4/2016	11/5/2016	11/6/2016	11/7/2016	11/8/2016	11/9/2016
H ₂ O #		MH978	MH978	MH978	MH978	MH978	MH978	MH978
Temp (°C)	Control	23.7	22.7	23.0	22.7	23.4	23.5	24.7
	32%	23.9	22.5	23.4	23.2	23.3	23.9	24.8
	42%	23.6	22.8	22.5	23.1	23.0	23.9	24.6
	56%	23.7	23.0	23.0	23.2	23.3	23.8	24.7
	80%	23.6	23.0	22.9	23.0	23.3	23.8	24.7
	100%	23.5	22.9	23.0	23.2	23.4	23.9	24.6
pH (Standard Units)	Control	8.38	8.38	8.25	8.90	8.39	8.78	8.41
	32%	8.55	8.40	8.40	9.04	8.61	8.81	8.63
	42%	8.57	8.46	8.41	9.08	8.64	8.91	8.64
	56%	8.69	8.50	8.49	9.10	8.71	8.89	8.72
	80%	8.72	8.54	8.50	9.13	8.75	8.92	8.70
	100%	8.74	8.54	8.56	9.15	8.80	8.92	8.76
DO (mg/L)	Control	9.2	9.3	9.3	9.0	8.9	9.0	8.8
	32%	9.1	9.4	9.3	9.3	8.8	9.1	8.8
	42%	9.2	9.4	9.3	9.3	8.8	9.1	8.8
	56%	9.3	9.4	9.3	9.4	8.9	9.2	8.8
	80%	9.4	9.4	9.4	9.4	8.9	9.3	8.7
	100%	9.4	9.5	9.5	9.6	8.9	9.3	8.9

Ceriodaphnia Survival and Reproduction Test-7 Day Survival

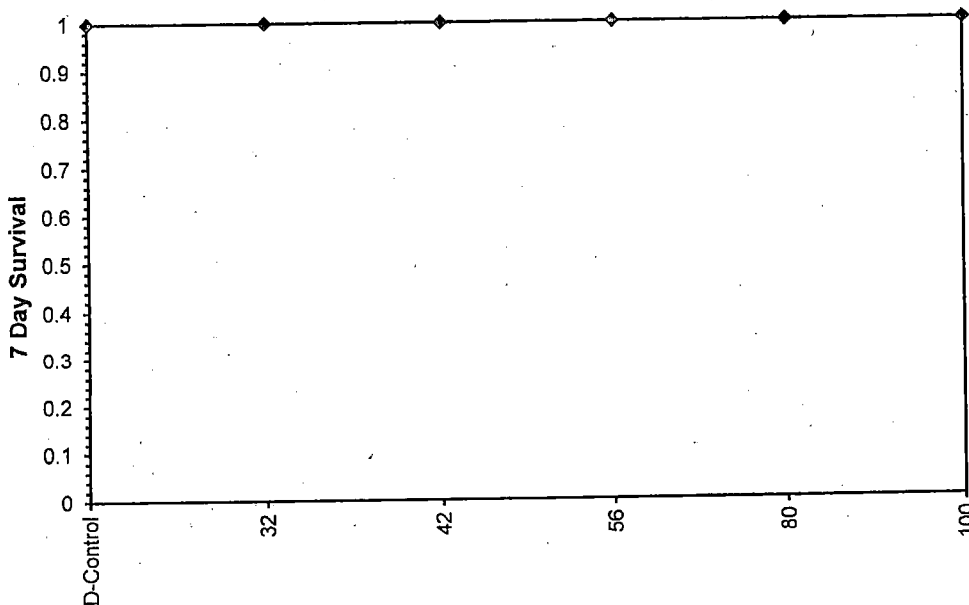
Start Date: 11/2/2016 12:00	Test ID: W. Ridge	Sample ID: AR0046566 -NPDES Permit #
End Date: 11/9/2016 11:05	Lab ID: ASU ERF	Sample Type: EFF1-POTW
Sample Date:	Protocol: EPAF 02-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia
Comments:		

Conc-%	1	2	3	4	5	6	7	8	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	1.0000	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	1.0000
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	1.0000	1.0000	0	10	10	10	1.0000	0.0500
42	1.0000	1.0000	0	10	10	10	1.0000	0.0500
56	1.0000	1.0000	0	10	10	10	1.0000	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: 11/2/2016 12:00	Test ID: W. Ridge	Sample ID: AR0046566-NPDES Permit #
End Date: 11/9/2016 11:05	Lab ID: ASU ERF	Sample Type: EFF1-POTW
Sample Date:	Protocol: EPAF 02-EPA Freshwater	Test Species: CD-Ceriodaphnia dubia

Comments:

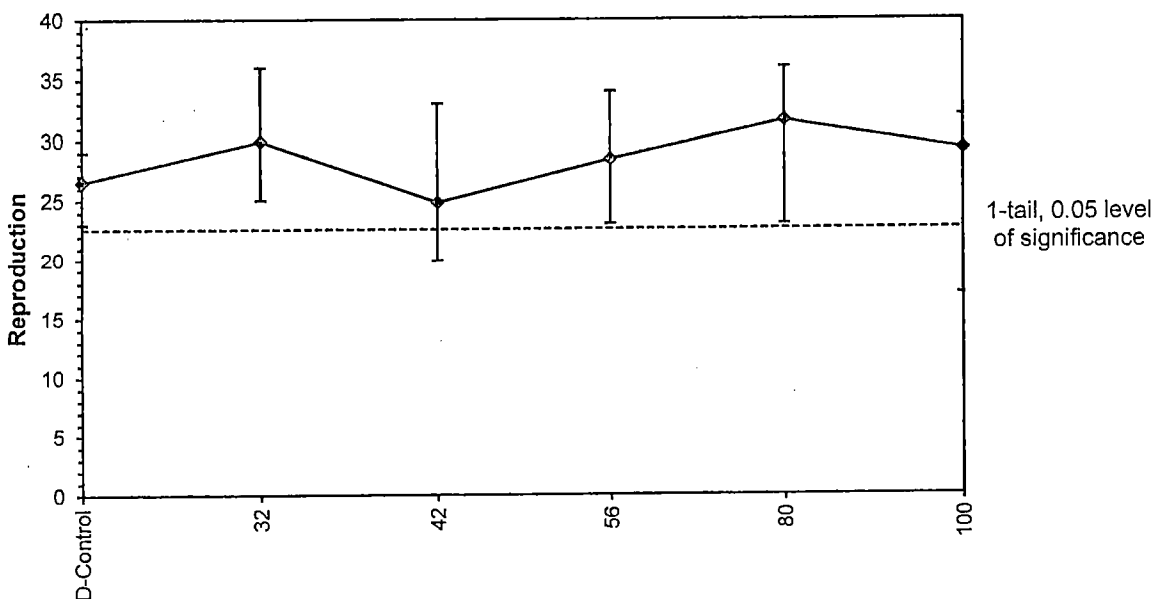
Conc-%	1	2	3	4	5	6	7	8	9	10
D-Control	26.000	29.000	27.000	29.000	23.000	25.000	24.000	29.000	26.000	27.000
32	28.000	28.000	35.000	27.000	27.000	30.000	25.000	36.000	28.000	35.000
42	28.000	20.000	26.000	26.000	20.000	25.000	24.000	25.000	21.000	33.000
56	29.000	34.000	23.000	31.000	27.000	24.000	26.000	28.000	33.000	29.000
80	35.000	28.000	36.000	28.000	23.000	30.000	31.000	36.000	36.000	33.000
100	30.000	32.000	31.000	29.000	32.000	17.000	31.000	30.000	28.000	32.000

Conc-%	Mean	N-Mean	Transform: Untransformed					N	t-Stat	1-Tailed Critical	MSD
			Mean	Min	Max	CV%					
D-Control	26.500	1.0000	26.500	23.000	29.000	8.005	10				
32	29.900	1.1283	29.900	25.000	36.000	13.233	10	-1.981	2.287	3.925	
42	24.800	0.9358	24.800	20.000	33.000	15.994	10	0.990	2.287	3.925	
56	28.400	1.0717	28.400	23.000	34.000	12.663	10	-1.107	2.287	3.925	
80	31.600	1.1925	31.600	23.000	36.000	13.930	10	-2.971	2.287	3.925	
100	29.200	1.1019	29.200	17.000	32.000	15.383	10	-1.573	2.287	3.925	

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Kolmogorov D Test indicates normal distribution (p > 0.01)	0.41122	1.035	-0.54625	1.16527
Bartlett's Test indicates equal variances (p = 0.39)	5.17603	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	3.92477	0.1481	59.4	14.7296	0.0035	5, 54

Dose-Response Plot



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

Test Day:		0	1	2	3	4	5	6
Date:		11/02/16	11/03/16	11/04/16	11/05/16	11/06/16	11/07/16	11/08/16
H ₂ O Batch #:		MH978	MH978	MH978	MH978	MH978	MH978	MH978
Temp. (°C)	Control	23.4	23.2	22.0	23.0	22.2	23.1	23.2
	32%	23.6	23.2	22.1	23.3	22.8	23.2	23.4
	42%	23.3	23.5	22.0	23.2	22.9	23.4	23.5
	56%	23.5	23.7	22.6	23.2	23.1	23.4	23.8
	80%	23.7	23.9	22.8	23.5	23.2	23.6	23.9
	100%	24.0	23.9	23.0	23.5	23.0	23.9	24.0
pH	Control	8.11	7.93	7.78	7.70	8.07	8.12	8.18
	32%	8.38	8.25	8.10	7.78	8.33	8.30	8.42
	42%	8.37	8.29	8.14	7.91	8.34	8.39	8.45
	56%	8.40	8.32	8.17	7.96	8.37	8.42	8.47
	80%	8.41	8.34	8.20	7.98	8.38	8.43	8.50
	100%	8.41	8.35	8.21	7.98	8.38	8.44	8.51
DO (mg/L)	Control	8.9	9.0	8.7	8.85	8.8	8.7	8.7
	32%	8.7	8.7	8.9	9.0	8.9	8.7	8.6
	42%	8.9	8.7	8.8	8.8	8.8	8.6	8.6
	56%	8.7	8.6	8.7	8.7	8.8	8.6	8.6
	80%	8.6	8.5	8.7	8.7	8.8	8.5	8.5
	100%	8.6	8.4	8.6	8.7	8.9	8.4	8.5
Cond. (µS/cm)	Control	309	309	309	306	306	312	318
	32%	319	317	321	320	321	317	312
	42%	318	324	327	324	327	319	318
	56%	329	330	333	331	335	322	324
	80%	341	341	343	344	350	327	327
	100%	352	350	354	356	363	332	332
Alk. (mg/L)	Control	60		60			60	
	100%	164		196			176	
Hard. (mg/L)	Control	100		100			100	
	100%	230		220			200	
Initials		jen/ruc	jen/aps	ruc	ruc	ruc	jen/ruc	jen/ruc

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

Test Day:		1	2	3	4	5	6	7
Date:		110316	110416	110516	110616	110716	110816	110916
H ₂ O Batch #:		MH978	MH978	MH978	MH978	MH978	MH978	MH978
Temp. (°C)	Control	23.7	22.7	23.0	22.7	23.4	23.5	24.7
	32%	23.9	22.5	23.4	23.2	23.3	23.9	24.8
	42%	23.6	22.8	22.5	23.1	23.0	23.9	24.6
	56%	23.7	23.0	23.0	23.2	23.3	23.8	24.7
	80%	23.6	23.0	22.9	23.0	23.3	23.8	24.7
	100%	23.5	22.9	23.0	23.2	23.4	23.9	24.6
pH	Control	8.35	8.38	8.25	8.90	8.39	8.78	8.41
	32%	8.55	8.40	8.40	9.04	8.61	8.81	8.63
	42%	8.57	8.46	8.41	9.08	8.64	8.91	8.64
	56%	8.49	8.50	8.49	9.10	8.71	8.89	8.72
	80%	8.72	8.54	8.50	9.13	8.75	8.92	8.70
	100%	8.74	8.54	8.56	9.15	8.80	8.92	8.76
DO (mg/L)	Control	9.2	9.3	9.3	9.0	8.9	9.0	8.8
	32%	9.1	9.4	9.3	9.3	8.8	9.1	8.8
	42%	9.2	9.4	9.3	9.3	8.8	9.1	8.8
	56%	9.3	9.4	9.3	9.4	8.9	9.2	8.8
	80%	9.4	9.4	9.4	9.4	8.9	9.3	8.7
	100%	9.4	9.5	9.5	9.6	8.9	9.3	8.9
Initials		DP/len	RIC	RIC	RIC	RIC	RIC	RIC



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: 11 02 16 Sampling Date: 11 01 16 / 11 02 16 Arrival Time: 11:58

Field Identification Number: WR 001 Description: Composite

Shipped by: Federal Express _____ UPS _____ Hand delivered by: Jon Kopp

Drop-Off Location: ASU-ERF

Storage While Shipped: Cooler w/ Ice

Analysis Requested: Chronic C. dubia

Initial Water Chemistry Analysis:

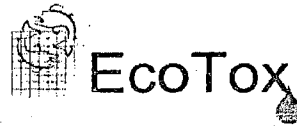
Sample Received by: Jen

Temperature (°C): 2.1°C Ice Present upon delivery: YES NO

Date: 11 02 16

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	Jen	11 02 16	X	
Refrigerated at 4°C	Jen		X	
Field Record Received	Jen	↓		X
Sample Label Affixed Properly	Jen		X	
Project Leader Informed	Jen	↓	X	

Comments: _____



Ecotoxicology Research Facility

SAMPLE CHECK INSample 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: 110416 Sampling Date: 110316/110416 Arrival Time: 1155Field Identification Number: WR002 Description: CompositeShipped by: Federal Express _____ UPS _____ Hand delivered by: Jon KoppDrop-Off Location: ASU-ERFStorage While Shipped: Cooler w/ iceAnalysis Requested: Chronic C. dubia

Initial Water Chemistry Analysis:

Sample Received by: RICTemperature (°C): 0.8 Ice Present upon delivery: YES NODate: 110416

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	RIC	110416	X	
Refrigerated at 4°C	RIC	↓	X	
Field Record Received	RIC			X
Sample Label Affixed Properly	RIC		X	
Project Leader Informed	RIC		X	

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: 11/07/16 Sampling Date: 11/06 - 11/07 Arrival Time: 1134

Field Identification Number: EFF 001 Description: Composite Aquatics

Shipped by: Federal Express _____ UPS _____ Hand delivered by: Jon Kopp

Drop-Off Location: ASU-ERF

Storage While Shipped: Ice/cooler

Analysis Requested: Chronic C. dubia

Initial Water Chemistry Analysis:

Sample Received by: Jen/RIC

Temperature (°C): 0.0 Ice Present upon delivery: YES NO

Date: 11/07/16

Quality Assurance	Initial	Date	Yes	No
Chain of Custody	Jen	110716	X	
Refrigerated at 4°C	Jen	↓	X	
Field Record Received	Jen			X
Sample Label Affixed Properly	Jen		X	
Project Leader Informed	Jen		X	

Comments: _____

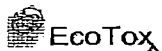


Ecotoxicology Research Facility
 Arkansas State University
 2645 Gaddo Drive
 State University, AR 72467
 (870) 972-2570 Fax (870) 972-2577

CHAIN OF CUSTODY RECORD



Client Name			Phone: (870) 886-2312				Analyses (List Below)					
Walnut Ridge Wastewater Treatment			Fax:									
Project #			PO #:				Chronic C. dubia	Chronic P. promelas				
Sampler (sign)			Remarks:									
[Signature]			Contact: Jonathan Kopp									
Cont.#	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix					
					Comp	Grab	Aqueous	Soil	Other			
			11-1/11-2	9am - 9am	✓							
Ice present at delivery:		<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No										
Temp:		2.0 °C		AEN		Initials						
1. Relinquished By (sign)			Date	Time	1. Received By (sign)			Date	Time			
[Signature]			11-2-16	11:58	[Signature]			110216	1158			
2. Relinquished By (sign)			Date	Time	2. Received By (sign)			Date	Time			



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			Phone: (870) 886-2312				Analyses (List Below)							
Walnut Ridge Wastewater Treatment			Fax:											
Project #			PO #:				Chronic C. dubia	Chronic P. promelas						
Sampler (sign) <i>[Signature]</i>			Remarks: Contact: Jonathan Kopp											
Cont.#	Sample ID Number	Location	Sample Date	Sample Time	Sample Type		Matrix							
					Comp	Grab	Aqueous	Soil	Other					
			11-3	11-4	9am - 9am	✓								
Ice present at delivery:			<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No											
Temp:			0.8 °C <i>RLC</i> Initials											
1. Relinquished By (sign) <i>[Signature]</i>			Date 11-4-16		Time 11:55		1. Received By (sign) <i>Rebecca d. Corp</i>			Date 110416		Time 1155		
2. Relinquished By (sign)			Date		Time		2. Received By (sign)			Date		Time		



Ecotoxicology Research Facility

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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 #:					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	W.R.	EFF 001	10/6/16	9am-9am	X								
Ice present at delivery:			<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No										
Temp:			0.0°C		Jen Initials								
1. Relinquished By (sign) <i>[Signature]</i>			Date 10/7/16		Time 1134		1. Received By (sign) <i>[Signature]</i>			Date 11/07/16		Time 1134	
2. Relinquished By (sign)			Date		Time		2. Received By (sign)			Date		Time	

City Water Works
216 South West 4th St.
Walnut Ridge, AR 72476



**RETURN RECEIPT
REQUESTED**

ADEQ/Water Division
ATTN: Mary Barrett
5301 North Shore Dr.
North Little Rock, AR, 72118-5317

