NPDES Compliance Chronic Toxicity Testing of Chevron/Cawelo Water District 'Inlet to Reservoir B' Effluent

Sample collected August 31, 2009

Prepared For:

Chevron ETC
3901 Briarpark BRP 405
Houston, TX 77042

Prepared By:

Pacific EcoRisk
2250 Cordelia Road
Fairfield, CA 94534

October 2009

PACIFIC ECORISK
ENVIRONMENTAL CONSULTING & TESTING

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1. INTRODUCTION

Chevron ETC has contracted Pacific EcoRisk (PER) to perform NPDES compliance evaluations of the chronic toxicity of Chevron USA Inc. and Cawelo Water District (Chevron/Cawelo) effluent. These evaluations consist of performing the following US EPA freshwater chronic toxicity tests:

- 3-brood (6-8-day) survival and reproduction test with the crustacean *Ceriodaphnia dubia;* and
- 7-day survival and growth test with larval fathead minnows (*Pimephales promelas*).

The current round of testing was performed using an effluent sample that was collected on August 31, 2009. In order to assess the sensitivity of the test organisms to chronic toxic stress, reference toxicant tests were also performed. This report describes the performance and results of these effluent and reference toxicant tests.

2. TOXICITY TEST PROCEDURES

The methods used in conducting these tests followed EPA testing manual "Short-Term Methods for Estimating the Chronic Effects of Effluents and Receiving Waters to Freshwater Organisms, Fourth Edition" (EPA-821-R-02-013).

2.1 Sample Receipt and Handling

On August 31, Precision Analytical staff collected a sample of effluent into an appropriately cleaned sample containers. This sample was transported, on ice and under chain-of-custody, to the PER laboratory in Fairfield. Upon receipt at the testing laboratory, aliquots of the sample were collected for analysis of initial water quality characteristics (Table 1), with the remainder of the sample being stored at 0-6°C except when being used to prepare test solutions. The chain-of-custody records for the collection and delivery of this sample are provided in Appendix A.

Table 1. Initial water quality characteristics of the 'Inlet to Reservoir B' effluent sample.

Sample Receipt Date	Sample ID	Temp (°C)	pH	D.O. (mg/L)	Alkalinity (mg/L)	Hardness (mg/L)	Conductivity (µS/cm)	Total Ammonia (mg/L N)
8/31/09	Inlet to Reservoir B	25.3[a]	7.20	4.9	197	86	830	<1.0

a - This sample was transported and delivered on the day of sample collection.

2.2 Survival and Reproduction Toxicity Testing with *Ceriodaphnia dubia*

The short-term chronic *Ceriodaphnia* test consists of exposing individual females to effluent for the length of time it takes for the Lab Control treatment females to produce 3 broods (typically 6-8 days), after which effects on survival and reproduction are evaluated. The specific procedures used in this test are described below.

The Lab Water Control treatment for this test consisted of an 80:20 mixture of de-ionized water with commercial spring water (Perrier). The Lab Water and the effluent sample were used to prepare test solutions at the 12.5, 25, 50, 75, and 100% effluent concentrations. For each treatment, 200 mL of test solution was amended with the alga *Selenastrum capricornutum* and Yeast-Cerophyll®-Trout Food (YCT) to provide food for the test organisms. "New" water quality characteristics (pH, D.O., and conductivity) were measured on these food-amended test solutions prior to use in this test. Each day of the test, fresh test solutions and a "new" set of replicate cups were prepared and characterized, as before.

There were 10 replicates for each test treatment, each replicate consisting of 15 mL of test solution in a 30-mL plastic cup. This "3-brood" test was initiated by allocating one neonate (<24 hrs old) *Ceriodaphnia*, obtained from ongoing laboratory cultures, into each replicate. The replicate cups were placed into a temperature-controlled room at 25°C, under cool-white fluorescent lighting on a 16L:8D photoperiod.

Each test replicate cup was examined daily, with surviving "original" individual organisms being transferred to the corresponding new cup containing fresh test solution. The contents of each remaining "old" replicate cup were carefully examined, and the number of neonate offspring produced by each original organism was determined, after which "old" water quality characteristics (pH, D.O., and conductivity) were measured for the old media from one randomly-selected replicate at each treatment.

After it was determined that ≥ 60% of the *Ceriodaphnia* in the Lab Water Control treatment had produced their third brood of offspring, the test was terminated. The resulting survival and reproduction (number of offspring) data were analyzed to evaluate any impairment(s) caused by the effluent; all statistical analyses were performed using the CETIS® statistical software.

2.2.1 Reference Toxicant Testing of the *Ceriodaphnia dubia*

In order to assess the sensitivity of the *Ceriodaphnia* test organisms to toxic stress, a reference toxicant test was performed. The reference toxicant test was performed similarly to the effluent test except that test solutions consisted of Lab Control water spiked with NaCl at test concentrations of 250, 500, 1000, 1500 and 2000 mg/L. The resulting test response data were statistically analyzed to determine key dose-response point estimates (e.g., EC_{50}); all statistical analyses were made using the CETIS® software. These response endpoints were then compared

to the typical response range established by the mean ± 2 SD of the point estimates generated by the most recent previous reference toxicant tests performed by this lab.

2.3 Survival and Growth Toxicity Testing with Larval Fathead Minnows

The chronic fathead minnow test consists of exposing larval fish to effluent for 7 days, after which effects on survival and growth are evaluated. The specific procedures used in this test are described below.

The Lab Water Control treatment for this test consisted of US EPA synthetic moderately-hard water. The Lab Water and the effluent sample were used to prepare daily test solutions at the 12.5, 25, 50, 75, and 100% effluent concentrations. "New" water quality characteristics (pH, D.O., and conductivity) were measured on these test solutions prior to use in the test.

There were 4 replicates at each test treatment, each replicate consisting of 400 mL of test media in a 600-mL glass beaker. This test was initiated by randomly allocating 10 larval fathead minnows (<48 hrs old) into each replicate. The replicate beakers were placed in a temperature-controlled room at 25°C, under cool-white fluorescent lighting on a 16L:8D photoperiod. The test fish were fed brine shrimp nauplii thrice daily.

Each replicate was examined daily, with any dead animals, uneaten food, wastes, and other detritus being removed. The number of live fish in each replicate was determined and then approximately 80% of the test media in each beaker was carefully poured out and replaced with fresh test solution. "Old" water quality characteristics (pH, D.O., and conductivity) were measured on the old test water that had been discarded from one randomly-selected replicate at each treatment.

After 7 days exposure, the number of live fish in each replicate beaker was recorded. The fish from each replicate were then carefully euthanized in methanol, rinsed in de-ionized water, and transferred to a pre-dried and pre-tared weighing pan. These fish were then dried at 100°C for >24 hrs and re-weighed to determine the total weight of fish in each replicate; the total weight was then divided by the initial number of fish per replicate (n=10) to determine the "biomass value". The resulting survival and growth ("biomass value") data were analyzed to evaluate any impairment(s) caused by the effluent; all statistical analyses were performed using the CETIS® statistical software.

2.3.1 Reference Toxicant Testing of the Larval Fathead Minnows

In order to assess the sensitivity of the fish to toxic stress, a reference toxicant test was performed. The reference toxicant test was performed similarly to the effluent test, except that test solutions consisted of Lab Control water spiked with NaCl at test concentrations of 0.75, 1.5, 3, 6, and 9 gm/L. The resulting test response data were analyzed to determine key dose-response point estimates (e.g., EC_{50}); all statistical analyses were made using the CETIS® software. These

response endpoints were then compared to the 'typical response' range established by the mean ± 2 SD of the point estimates generated by the 20 most recent previous reference toxicant tests performed by this lab.

3. RESULTS

3.1 Chronic Effects of the Chevron/Cawelo Effluent on *Ceriodaphnia dubia*

The results of this test are summarized below in Table 2. There was 80% survival at the Lab Water Control treatment. There were no significant reductions in survival in the effluent; the survival NOEC was 100% effluent, resulting in 1.0 TUc (where TUc =100/NOEC).

There was a mean of 15.1 offspring per female at the Lab Water Control treatment. There were significant reductions in reproduction; the reproduction NOEC was 75% effluent, resulting in 1.3 TUc (where TUc =100/NOEC).

The test data and summary of statistical analyses for this test are presented in Appendix B.

Table 2. Effects of the effluent on *Ceriodaphnia dubia* survival and reproduction.

Effluent Treatment	% Survival	Reproduction (# neonates /female)
Lab Water Control	80	15.1
12.5%	100	24.1
25%	80	21.3
50%	100	19.7
75%	90	10.0
100%	90	**1.8***
Summary of Statistics		
No Observable Effect Concentration (NOEC) =	100% effluent	75% effluent
TUc (where TUc = 100/NOEC) =	1.0	1.3
Survival EC_{25} or Reproduction IC_{25} =	>100% effluent[a]	60.6% effluent
Survival EC_{50} or Reproduction IC_{50} =	>100% effluent[a]	74.7% effluent

* Significantly less than the Lab Control treatment response at $p < 0.05$.

a - Due to the absence of significant mortalities, the EC point estimates could not be calculated, but can be assumed to be >100% effluent a - Due to the absence of significant mortalities, the EC point estimates could not be calculated, but can be assumed to be >100% effluent.

3.1.1 Reference Toxicant Toxicity to *Ceriodaphnia dubia*

Results of this test are summarized below in Table 3. There was 90% survival and a mean of 15.4 neonates per female at the Lab Control treatment. The survival EC_{50} was 2190 mg/L NaCl, and the reproduction IC_{50} was 1340 mg/L NaCl.

These reference toxicant test results were consistent with the "typical response" ranges established by the reference toxicant test database, indicating that these test organisms were responding to toxic stress in a typical fashion.

The test data and the summary of statistical analyses for this test are presented in Appendix C.

Table 3. Reference toxicant testing: Effects of NaCl on *Ceriodaphnia dubia*

Treatment (mg/L NaCl)	% Survival	Reproduction (# neonates/female)
Lab Control	90	15.4
250	90	18.1
500	90	16.2
1000	60	12.6
1500	80	**6.7***
2000	40	**0***
Summary of Key Statistics		
Survival EC_{50} or Reproduction IC_{50} =	2190 mg/L NaCl	1340 mg/L NaCl

* Significantly less than the Lab Control treatment response at $p < 0.05$.

3.2 Chronic Effects of the Chevron/Cawelo Effluent on Fathead Minnows

The results of this test are summarized below in Table 4. There was 97.5% survival at the Lab Water Control treatment. There were significant reductions in survival in the effluent; the NOEC was 12.5% effluent, resulting in 8.0 TUc (where TUc =100/NOEC).

There was a mean 'biomass value' of 0.45 mg at the Lab Water Control treatment. The growth NOEC was <12.5% effluent, resulting in >8.0 TUc (where TUc =100/NOEC).

The test data and the summary of statistical analyses for this test are presented in Appendix D.

Table 4. Effects of the effluent on fathead minnow survival and growth.

Effluent Treatment	% Survival	Mean Fish Biomass Value (mg)
Lab Water Control	97.5	0.45
12.5%	90	**0.34***
25%	**80***	0.27
50%	**40***	0.09
75%	**0***	-
100%	**0***	-
Summary of Statistics		
No Observable Effect Concentration (NOEC) =	12.5% effluent	<12.5% effluent
TUc (where TUc = 100/NOEC) =	8.0	>8.0
Survival EC_{25} or Growth IC_{25} =	Could not be calculated	12.7% effluent
Survival EC_{50} or Growth IC_{50} =	39.8% effluent	29.5% effluent

* Significantly less than the Lab Water Control treatment response at $p < 0.05$.

3.2.1 Reference Toxicant Toxicity to Fathead Minnows

The results of this test are summarized below in Table 5. There was 97.5% survival and a mean biomass value of 0.36 mg at the Lab Control treatment. The survival EC_{50} was 5.1 gm/L NaCl and the growth IC_{50} was 5.8 gm/L NaCl.

The survival results of this reference toxicant test were consistent with the "typical response" range established by the reference toxicant test database, indicating that the survival response of these test organisms was typical and consistent. The growth IC_{50} of 5.8 gm/L NaCl was just above the upper threshold of 5.0 gm/L NaCl of the "typical response" range; this indicates that the growth of these test organisms may have been slightly less sensitive to toxicant stress than is typical. EPA guidelines state that at the $p<0.05$ level, it is to be expected that 1 out of 20 reference toxicant tests will fall outside of the "typical response" range due to statistical probability, so our observation of this "outlier" is not unexpected nor cause for undue concern.

The test data and summary of statistical analyses for this test are presented in Appendix E.

Table 5. Reference toxicant testing: effects of NaCl on fathead minnows.

NaCl Treatment (gm/L)	% Survival	Mean Fish Biomass Value (mg)
Lab Control	97.5	0.36
0.75	97.5	0.33
1.5	95	0.37
3	85	**0.29***
6	**52.5***	**0.18***
9	**0***	**0***
Summary of Statistics		
Survival EC_{50} or Growth IC_{50} =	5.1 gm/L NaCl	5.8 gm/L NaCl

* Significantly less than the Lab Control treatment response at $p < 0.05$.

4. SUMMARY AND CONCLUSIONS

Chronic Effects of Chevron/Cawelo Effluent on *Ceriodaphnia dubia*
There were no significant reductions in survival in the effluent; the survival NOEC was 100% effluent. There were significant reductions in reproduction; the reproduction NOEC was 75% effluent, resulting in 1.3 TUc (where TUc = 100/NOEC).

Chronic Effects of Chevron/Cawelo Effluent on Fathead Minnows
There were significant reductions in survival in the effluent; the NOEC was 12.5% effluent, resulting in 8.0 TUc (where TUc =100/NOEC). The reproduction NOEC was <12.5% effluent, resulting in >8.0 TUc (where TUc =100/NOEC).

4.1 QA/QC Summary

Test Conditions – Test conditions (pH, D.O., temperature, etc.) were all within acceptable limits for these tests. All analyses were performed according to laboratory Standard Operating Procedures.

Negative Lab Control – The biological responses in the Lab Water Control treatments for these tests were within acceptable limits.

Positive Control – The fathead minnow growth IC_{50} was just above the upper threshold of the "typical response" range, indicating that the growth of these test organisms may have been slightly less sensitive to toxicant stress than is typical. EPA guidelines state that at the $p < 0.05$ level, it is to be expected that 1 out of 20 reference toxicant tests will fall outside of the "typical response" range due to statistical probability, so our observation of this 'outlier' is not unexpected nor cause for undue concern.

All other reference toxicant responses were consistent with the "typical response" ranges established by the reference toxicant test databases.

Concentration Response Relationships – There were valid concentration-response relationships for the reference toxicant tests, which were determined to be acceptable for this testing.

Appendix A

Chain-of-Custody Record for the Collection and Delivery of the Chevron/Cawelo 'Inlet to Reservoir B' Effluent Sample

CHAIN OF CUSTODY RECORD

PACIFIC ECORISK
2250 Cordelia Rd
Fairfield, CA 94534
Ph: (707) 207-7760
Fax: (707) 207-7916
www.pacificecorisk.com

RESULTS TO:
CYNTHIA GULDE
CHEVRON CORP
Attn:
Phone:
Email:

BILL TO:
CYNTHIA GULDE
CHEVRON CORP
Attn:
Phone:
Email:

PROJECT:

ANALYSES REQUESTED

SAMPLE IDENTIFICATION	DATE	TIME	SAMPLE MATRIX	GRAB/ COMP.	# CONTAINERS/TYPE	ORGANIC + ACUTE TOXICITY								REMARKS
INLET TO RESERVOIR B	8/31/09	1:00	WATER	GRAB	2 / PLASTIC	X								
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METHOD OF SHIPMENT: FedEx:_____ UPS:_____ HAND:_____ OTHER:_____

COMMENTS: Temp: 33.6 °C
BLK: 28.8 °C

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[signature]	8/31/09	1950	[signature]	8/31/09	1950	

WHITE - RETURN W/ SAMPLE YELLOW - KEEP FOR YOUR RECORDS

CHAIN OF CUSTODY RECORD

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PROJECT:

ANALYSES REQUESTED

SAMPLE IDENTIFICATION	DATE	TIME	SAMPLE MATRIX	GRAB/ COMP.	# CONTAINERS/TYPE	CHRONIC + ACUTE TOXICITY								REMARKS
INLET TO RESERVOIR B	8/31/09	1:00	WATER	GRAB	2 / PLASTIC	X								
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Blk: 26.4 °C

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PROJECT:

ANALYSES REQUESTED

SAMPLE IDENTIFICATION	DATE	TIME	SAMPLE MATRIX	GRAB/ COMP.	# CONTAINERS/TYPE	Chemical + Acute Toxicity								REMARKS
INLET TO RESERVOIR B	8/31/09	1:00	WATER	GRAB	2 / PLASTIC	X								
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PROJECT:

ANALYSES REQUESTED

SAMPLE IDENTIFICATION	DATE	TIME	SAMPLE MATRIX	GRAB/ COMP.	# CONTAINERS/TYPE	CHRONIC + ACUTE ~~TOXICITY~~ TOXICITY							REMARKS
INLET TO RESERVOIR B	8/31/09	1:00	WATER	GRAB	2 / PLASTIC	X							
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METHOD OF SHIPMENT: FedEx:_____ UPS:_____ HAND:_____ OTHER:_____

COMMENTS: Temp: 30.7°C
BCC: 24.1°C

CODES:

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PROJECT:

ANALYSES REQUESTED

SAMPLE IDENTIFICATION	DATE	TIME	SAMPLE MATRIX	GRAB/ COMP.	# CONTAINERS/TYPE	CHRONIC TOXICITY								REMARKS
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METHOD OF SHIPMENT: FedEx:____ UPS:____ HAND:____ OTHER:____

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[signature]	8-31-09	1950	[signature]	8/31/09	1950	

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PROJECT:

ANALYSES REQUESTED

SAMPLE IDENTIFICATION	DATE	TIME	SAMPLE MATRIX	GRAB/ COMP.	# CONTAINERS/TYPE	CHLORINE FACILITY TOXICITY								REMARKS
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COMMENTS: Temp: 35.4 °C
Blue: 24.5 °C

CODES:

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[signature]	8-31-09	1950	[signature]	8/31/09	1950	

WHITE - RETURN W/ SAMPLE YELLOW - KEEP FOR YOUR RECORDS

Appendix B

Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of Chevron/Cawelo 'Inlet to Reservoir B' Effluent to *Ceriodaphnia dubia*

CETIS Summary Report

Report Date: 14 Sep-09 15:29 (p 1 of 2)
Test Code: 16-2498-9360/36009

Ceriodaphnia Survival and Reproduction Test — Pacific EcoRisk

Batch ID:	01-2928-7791	**Test Type:**	Reproduction-Survival (7d)	**Analyst:**	John Jirasritumrong
Start Date:	01 Sep-09 18:30	**Protocol:**	EPA-821-R-02-013 (2002)	**Diluent:**	Laboratory Water
Ending Date:	08 Sep-09 11:30	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Duration:	6d 17h	**Source:**	In-House Culture	**Age:**	1

Sample ID:	10-3556-7062	**Code:**	Eff	**Client:**	Precision Analytical
Sample Date:	31 Aug-09 13:00	**Material:**	Effluent	**Project:**	15164
Receive Date:	31 Aug-09 19:50	**Source:**	Precision Analytical		
Sample Age:	30h (25.3 °C)	**Station:**	Inlet Resv B		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
20-0972-2881	Reproduction	75	100	86.6	36.5%	1.33	Steel Many-One Rank Test
12-8581-4574	Survival	100	>100	N/A	N/A	1	Fisher Exact/Bonferroni-Holm Test

Point Estimate Summary

Analysis ID	Endpoint	Level	%	95% LCL	95% UCL	TU	Method
03-4573-7930	Reproduction	IC5	51.1	16.4	52.8	1.96	Linear Interpolation (ICPIN)
		IC10	53.4	21.5	55.9	1.87	
		IC15	55.7	29.7	59.2	1.8	
		IC20	58.1	50.6	62.6	1.72	
		IC25	60.6	53.3	66.2	1.65	
		IC40	68.7	61.7	77.4	1.46	
		IC50	74.7	66.7	81.2	1.34	

Reproduction Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	10	15.1	12.6	17.6	0	23	1.23	6.74	44.6%	0.0%
12.5		10	24.1	23	25.2	19	29	0.561	3.07	12.7%	-59.6%
25		10	21.3	18.3	24.3	0	29	1.45	7.96	37.4%	-41.1%
50		10	19.7	18	21.4	11	25	0.826	4.52	23.0%	-30.5%
75		10	10	7.97	12	0	16	0.993	5.44	54.4%	33.8%
100		10	1.8	0.889	2.71	0	6	0.446	2.44	136.0%	88.1%

Survival Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	10	0.8	0.643	0.957	0	1	0.077	0.422	52.7%	0.0%
12.5		10	1	1	1	1	1	0	0	0.0%	-25.0%
25		10	0.8	0.643	0.957	0	1	0.077	0.422	52.7%	0.0%
50		10	1	1	1	1	1	0	0	0.0%	-25.0%
75		10	0.9	0.782	1	0	1	0.0577	0.316	35.1%	-12.5%
100		10	0.9	0.782	1	0	1	0.0577	0.316	35.1%	-12.5%

Ceriodaphnia Survival and Reproduction Test — Pacific EcoRisk

Reproduction Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Lab Water Contr	0	21	23	13	18	20	15	14	9	18
12.5		23	25	29	28	19	24	20	24	24	25
25		21	25	25	29	0	25	24	19	22	23
50		22	11	25	18	20	23	13	20	23	22
75		15	12	9	13	15	0	6	3	11	16
100		5	0	0	6	0	0	0	3	0	4

Survival Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Lab Water Contr	0	1	1	1	1	1	1	1	0	1
12.5		1	1	1	1	1	1	1	1	1	1
25		1	1	1	1	0	1	1	0	1	1
50		1	1	1	1	1	1	1	1	1	1
75		1	1	1	1	1	0	1	1	1	1
100		1	1	0	1	1	1	1	1	1	1

CETIS Analytical Report

Report Date: 14 Sep-09 15:29 (p 1 of 1)
Test Code: 16-2498-9360/36009

Ceriodaphnia Survival and Reproduction Test — Pacific EcoRisk

Analysis ID:	12-8581-4574	Endpoint:	Survival	CETIS Version:	CETISv1.7.0
Analyzed:	14 Sep-09 15:17	Analysis:	STP 2x2 Contingency Tables	Official Results:	Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Untransformed		C > T	Not Run	100	>100	N/A	1	N/A

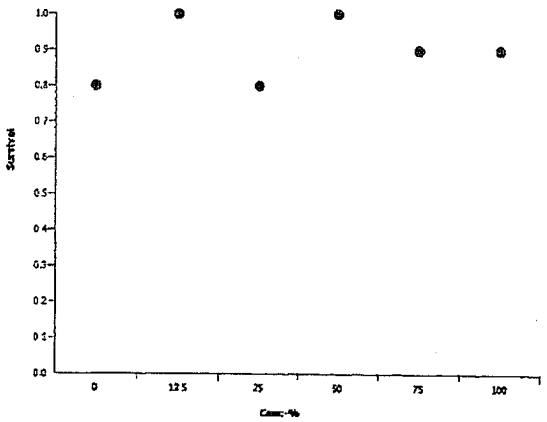
Fisher Exact/Bonferroni-Holm Test

Control	vs	Conc-%	Test Stat	P-Value	Decision(0.05)
Lab Water Control		12.5	1	1.0000	Non-Significant Effect
		25	0.709	1.0000	Non-Significant Effect
		50	1	1.0000	Non-Significant Effect
		75	1	1.0000	Non-Significant Effect
		100	1	1.0000	Non-Significant Effect

Data Summary

Conc-%	Control Type	No-Resp	Resp	Total
0	Lab Water Cont	8	2	10
12.5		10	0	10
25		8	2	10
50		10	0	10
75		9	1	10
100		9	1	10

Graphics



CETIS Analytical Report

Report Date: 14 Sep-09 15:29 (p 1 of 1)
Test Code: 16-2498-9360/36009

Ceriodaphnia Survival and Reproduction Test — Pacific EcoRisk

Analysis ID:	20-0972-2881	**Endpoint:**	Reproduction	**CETIS Version:**	CETISv1.7.0
Analyzed:	09 Sep-09 16:23	**Analysis:**	Nonparametric-Control vs Treatments	**Official Results:**	Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	0	C > T	Not Run	75	100	86.6	1.33	36.5%

Steel Many-One Rank Test

Control vs	Conc-%	Test Stat	Critical	Ties	P-Value	Decision(5%)
Lab Water Control	12.5	149	75	2	1.0000	Non-Significant Effect
	25	140	75	3	1.0000	Non-Significant Effect
	50	128	75	4	0.9987	Non-Significant Effect
	75	79.5	75	4	0.0977	Non-Significant Effect
	100*	62	75	1	0.0027	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	3431.733	686.3467	5	23.6	<0.0001	Significant Effect
Error	1567.6	29.02963	54			
Total	4999.333	715.3763	59			

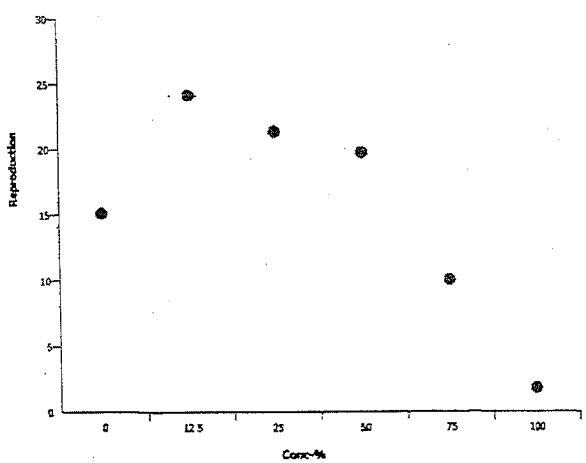
ANOVA Assumptions

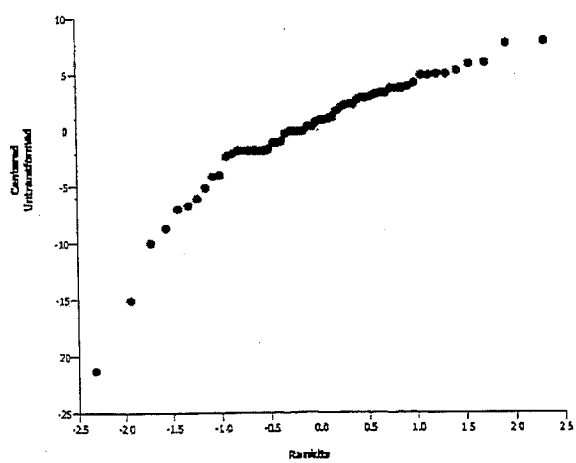
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	15.6	15.1	0.0080	Unequal Variances
Distribution	Shapiro-Wilk Normality	0.875		<0.0001	Non-normal Distribution

Reproduction Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	10	15.1	12.5	17.7	0	23	1.25	6.74	44.6%	0.0%
12.5		10	24.1	22.9	25.3	19	29	0.57	3.07	12.7%	-59.6%
25		10	21.3	18.3	24.3	0	29	1.48	7.96	37.4%	-41.1%
50		10	19.7	18	21.4	11	25	0.84	4.52	23.0%	-30.5%
75		10	10	7.93	12.1	0	16	1.01	5.44	54.4%	33.8%
100		10	1.8	0.872	2.73	0	6	0.453	2.44	136.0%	88.1%

Graphics





CETIS Analytical Report

Report Date: 14 Sep-09 15:29 (p 1 of 1)
Test Code: 16-2498-9360/36009

Ceriodaphnia Survival and Reproduction Test — Pacific EcoRisk

Analysis ID:	03-4573-7930	Endpoint:	Reproduction	CETIS Version:	CETISv1.7.0
Analyzed:	09 Sep-09 16:23	Analysis:	Linear Interpolation (ICPIN)	Official Results:	Yes

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	57951	200	Yes	Two-Point Interpolation

Point Estimates

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC5	51.1	16.4	52.8	1.96	1.89	6.08
IC10	53.4	21.5	55.9	1.87	1.79	4.65
IC15	55.7	29.7	59.2	1.8	1.69	3.37
IC20	58.1	50.6	62.6	1.72	1.6	1.97
IC25	60.6	53.3	66.2	1.65	1.51	1.88
IC40	68.7	61.7	77.4	1.46	1.29	1.62
IC50	74.7	66.7	81.2	1.34	1.23	1.5

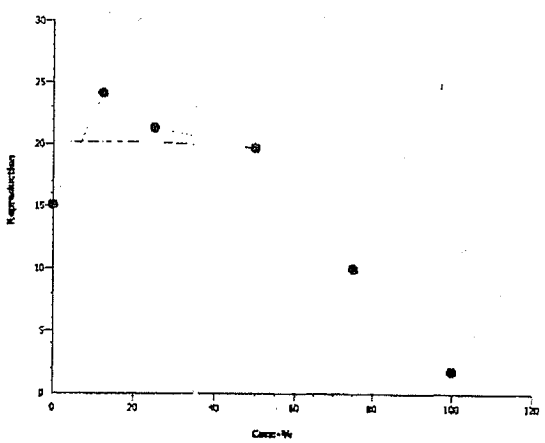
Reproduction Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contro	10	15.1	0	23	1.23	6.74	44.6%	0.0%
12.5		10	24.1	19	29	0.561	3.07	12.7%	-59.6%
25		10	21.3	0	29	1.45	7.96	37.4%	-41.1%
50		10	19.7	11	25	0.826	4.52	23.0%	-30.5%
75		10	10	0	16	0.993	5.44	54.4%	33.8%
100		10	1.8	0	6	0.446	2.44	136.0%	88.1%

Reproduction Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Lab Water Control	0	21	23	13	18	20	15	14	9	18
12.5		23	25	29	28	19	24	20	24	24	25
25		21	25	25	29	0	25	24	19	22	23
50		22	11	25	18	20	23	13	20	23	22
75		15	12	9	13	15	0	6	3	11	16
100		5	0	0	6	0	0	0	3	0	4

Graphics



Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: **Precision Analytical - Chevron Cawelo** Material: Inlet to Reserevoir B Test Date: 9/1/09

Project #: 15164 Test ID: 36009 Randomization: C.6.2 Control Water: Lab Water (80:20)

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Temp (°C)	Survival / Reproduction A	B	C	D	E	F	G	H	I	J	SIGN-OFF
Lab Water Control	0	8.09		8.8		235	25.6	0	0	0	0	0	0	0	0	0	0	Date: 9/1/09 New WQ: Sm Test Init.: DC Sol'n Prep: SH Time: 1030
	1	7.97	8.12	8.8	6.5	228	25.7	0	0	0	0	0	0	0	0	0	0	Date: 9/2/09 New WQ: Counts: Sol'n Prep: SH Old WQ: Time: 1315
	2	8.15	8.13	8.8	8.3	228	25.7	0	0	0	0	0	0	0	0	0	0	Date: 9/3/09 New WQ: Counts: Sol'n Prep: SH Old WQ: HV Time: 1010
	3	8.11	8.14	8.6	7.7	230	25.8	0	0	0	0	0	0	0	0	0	0	Date: 9/4/09 New WQ: Counts: Sol'n Prep: Old WQ: MB Time: 1430
	4	7.96	8.43	8.0	7.5	1991.9	25.8	X/0	3	4	0	3	3	0	3	3	4	Date: 9/5/09 New WQ: MB Counts: Sol'n Prep: Old WQ: MB Time: 1215
	5	7.89	8.25	8.3	7.8	263	25.7	-	0	0	5	6	5	3	5	0	0	Date: 9/6/09 New WQ: Counts: Sol'n Prep: Old WQ: Time: 1300
	6	8.31	8.05	8.2	8.1	222	25.7	-	8	8	8	9	0	0	6	X/6	6	Date: 9/7/09 New WQ: Counts: PA Sol'n Prep: SH Old WQ: Time: 1350
	7	—	8.11	—	7.5	220	25.6	-	10	11	0	0	12	12	0	-	8	Date: 9/8/09 New WQ: — Counts: Sol'n Prep: PA Old WQ: KR Time: 1120
	8							-								-		Date: New WQ: Counts: Sol'n Prep: Old WQ: Time:
							Total=	X/0	21	23	13	18	20	15	14	X/9	18	Mean Neonates/Female = 15.1

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Survival / Reproduction A	B	C	D	E	F	G	H	I	J	SAMPLE ID
12.5%	0	7.77		8.2		310	0	0	●	●	0	0	0	0	0	0	22637
	1	7.81	8.26	8.6	6.3	315	0	0	0	0	0	0	0	0	0	0	22637
	2	7.66	8.33	8.6	8.1	314	0	0	0	0	0	0	0	0	0	0	22637
	3	7.80	8.29	8.5	7.9	311	0	0	0	0	0	0	0	0	0	0	22637
	4	7.62	8.45	8.5	7.8	296	4	5	7	4	2	3	3	3	0	3	22637
	5	7.46	8.33	7.8	7.5	286	7	7	0	7	4	6	6	5	5	0	22637
	6	7.85	8.18	8.4	8.0	291	0	0	8	0	0	0	1	0	9	9	22637
	7	—	8.20	—	7.4	315	12	13	14	17	13	15	10	16	10	13	
	8																
							Total=	23	25	29	28	19	24	20	24	24	25	Mean Neonates/Female = ~~19.3~~ 24.1

Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: **Precision Analytical - Chevron Cawelo** Material: Inlet to Reserevoir B Test Date: 9/1/09

Project #: 15164 Test ID: 36009 Randomization: C.4.2 Control Water: Lab Water (80:20)

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Survival / Reproduction A	B	C	D	E	F	G	H	I	J	SIGN-OFF
25%	0	7.55		7.8		380	0	0	0	0	0	0	0	0	0	0	
	1	7.56	8.36	8.3	6.2	388	0	0	0	0	0	0	0	0	0	0	
	2	7.44	8.39	8.2	8.0	390	0	0	0	0	0	0	0	0	0	0	
	3	7.56	8.34	8.4	7.7	388	0	0	0	0	0	0	0	0	0	0	
	4	7.42	8.54	8.6	7.6	369	4	3	~~24~~	4	0	4	3	3	0	3	
	5	7.37	8.38	8.0	7.4	366	4	6	0	8	~~10~~	5	7	0	4	0	
	6	7.69	8.27	8.4	7.9	372	0	0	7	0	–	0	0	6	6	8	
	7	—	8.31	—	7.4	402	13	16	14	17	–	16	14	~~10~~	12	12	
	8										–			–			
						Total=	21	25	25	29	~~10~~	25	24	~~19~~	22	23	Mean Neonates/Female = 21.3

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Survival / Reproduction A	B	C	D	E	F	G	H	I	J	
50%	0	7.34		8.0		529	0	0	0	0	0	0	0	0	0	0	
	1	7.31	8.50	8.3	6.5	525	0	0	0	0	0	0	0	0	0	0	
	2	7.19	8.47	8.1	8.2	529	0	0	0	0	0	0	0	0	0	0	
	3	7.24	8.45	8.3	7.8	534	0	0	0	0	0	0	0	0	0	0	
	4	7.27	~~8.54~~ 8.59	8.0	7.5	521	0	0	0	4	3	3	3	3	0	3	
	5	7.21	8.46	8.0	7.4	513	7	5	5	3	4	7	4	4	5	6	
	6	7.50	8.36	8.5	7.9	516	5	6	7	0	0	0	6	0	8	7	
	7	—	8.46	—	7.5	551	10	0	13	11	13	13	0	13	10	12	
	8																
						Total=	22	11	25	18	~~20~~	23	13	20	23	22	Mean Neonates/Female = 19.7

Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: Precision Analytical - Chevron Cawelo Material: Inlet to Reserevoir B Test Date: 9/1/09

Project #: 15164 Test ID: 36009 Randomization: C.6.2 Control Water: Lab Water (80:20)

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Survival / Reproduction A	B	C	D	E	F	G	H	I	J	SIGN-OFF
75%	0	7.22		7.8		679	0	0	0	0	0	0	0	0	0	0	
	1	~~7.68~~ 7.16	8.54	8.2	6.6	667	0	0	0	0	0	0	0	0	0	0	
	2	7.06	8.56	7.7	8.2	672	0	0	0	0	0	0	0	0	0	0	
	3	7.16	8.52	8.3	7.7	689	0	0	0	0	0	x/0	0	0	0	0	
	4	7.18	8.62	7.2	7.7	672	0	0	0	0	0	-	0	0	0	0	
	5	7.06	8.59	7.9	7.3	665	5	0	0	2	0	-	4	3	4	5	
	6	7.40	8.46	8.8	7.9	663	0	3	0	0	5	-	2	0	7	0	
	7	—	8.51	—	7.3	700	10	9	9	11	10	-	0	0	0	11	
	8																
						Total=	15	12	9	13	15	0/0	6	3	11	16	Mean Neonates/Female = 10.0

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Survival / Reproduction A	B	C	D	E	F	G	H	I	J	SIGN-OFF
100%	0	7.10		8.0		824	0	0	0	0	0	0	0	0	0	0	
	1	7.08	8.54	8.1	6.5	825	0	0	0	0	0	0	0	0	0	0	
	2	6.94	8.62	7.3	8.2	819	0	0	0	0	0	0	0	0	0	0	
	3	7.03	8.61	7.9	7.8	832	0	0	0	0	0	0	0	0	0	0	
	4	7.07	8.68	8.1	7.1	833	0	0	0/0	0	0	0	0	0	0	0	
	5	6.99	8.63	7.7	7.4	825	0	0	-	0	0	0	0	0	0	0	
	6	7.28	8.57	9.3	7.9	828	0	0	-	0	0	0	0	2	0	0	
	7	—	KB ~~8.54~~ 8.61	—	7.3	KB ~~700~~ 866	5	0	-	6	0	0	0	1	0	4	
	8																
						Total=	5	0	0/0	6	0	0	0	3	0	4	Mean Neonates/Female = 1.8

Appendix C

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Ceriodaphnia dubia*

Ceriodaphnia Survival and Reproduction Test — **Pacific EcoRisk**

Batch ID:	10-0762-3779	**Test Type:**	Reproduction-Survival (7d)	**Analyst:**	Drew Gantner
Start Date:	01 Sep-09 18:55	**Protocol:**	EPA-821-R-02-013 (2002)	**Diluent:**	Laboratory Water
Ending Date:	09 Sep-09 15:30	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Duration:	7d 21h	**Source:**	In-House Culture	**Age:**	1

Sample ID:	03-2052-8953	**Code:**	NaCl	**Client:**	Pacific Ecorisk
Sample Date:	01 Sep-09 18:55	**Material:**	Sodium chloride	**Project:**	15165
Receive Date:	01 Sep-09 18:55	**Source:**	Reference Toxicant		
Sample Age:	N/A (25.9 °C)	**Station:**	In House		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
09-9425-2700	Reproduction	1000	1500	1220	28.5%		Dunnett's Multiple Comparison Test
08-4957-9737	Survival	2000	>2000	N/A	N/A		Fisher Exact/Bonferroni-Holm Test

Point Estimate Summary

Analysis ID	Endpoint	Level	mg/L	95% LCL	95% UCL	TU	Method
09-4578-7315	Reproduction	IC5	528	39.9	657		Linear Interpolation (ICPIN)
		IC10	621	312	862		
		IC15	730	370	1020		
		IC20	857	448	1070		
		IC25	1000	569	1130		
		IC40	1190	1010	1330		
		IC50	1340	1170	1520		
13-5714-9286	Survival	EC5	505	N/A	N/A		Linear Regression (MLE)
		EC10	698	N/A	N/A		
		EC15	869	N/A	N/A		
		EC20	1030	N/A	N/A		
		EC25	1200	N/A	N/A		
		EC40	1750	N/A	N/A		
		EC50	2190	N/A	N/A		

Reproduction Summary

Conc-mg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	10	15.4	13.1	17.7	4	27	1.14	6.26	40.6%	0.0%
250		10	18.1	16.3	19.9	9	23	0.884	4.84	26.7%	-17.5%
500		10	16.2	14.6	17.8	9	22	0.769	4.21	26.0%	-5.19%
1000		10	12.6	11.6	13.6	8	17	0.488	2.67	21.2%	18.2%
1500		10	6.7	5.53	7.87	0	11	0.571	3.13	46.7%	56.5%
2000		10	0	0	0	0	0	0	0		100.0%

Survival Summary

Conc-mg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	10	0.9	0.782	1	0	1	0.0577	0.316	35.1%	0.0%
250		10	0.9	0.782	1	0	1	0.0577	0.316	35.1%	0.0%
500		10	0.9	0.782	1	0	1	0.0577	0.316	35.1%	0.0%
1000		10	0.6	0.407	0.793	0	1	0.0943	0.516	86.1%	33.3%
1500		10	0.8	0.643	0.957	0	1	0.077	0.422	52.7%	11.1%
2000		10	0.4	0.207	0.593	0	1	0.0943	0.516	129.0%	55.6%

Ceriodaphnia Survival and Reproduction Test — Pacific EcoRisk

Reproduction Detail

Conc-mg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Lab Water Contr	13	17	22	13	18	27	12	16	12	4
250		9	11	22	22	20	23	16	18	18	22
500		16	9	19	16	9	18	22	18	16	19
1000		9	14	11	13	13	17	13	15	13	8
1500		10	0	6	11	8	4	8	8	6	6
2000		0	0	0	0	0	0	0	0	0	0

Survival Detail

Conc-mg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Lab Water Contr	1	1	1	1	1	0	1	1	1	1
250		1	1	1	0	1	1	1	1	1	1
500		1	1	1	1	0	1	1	1	1	1
1000		0	1	1	0	0	1	1	1	1	0
1500		1	1	1	1	1	0	1	1	0	1
2000		0	0	1	0	1	1	0	0	1	0

Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: **Reference Toxicant** Material: **Sodium Chloride** Test Date: 9/1/09

Project #: 15165 Test ID: 34097 Randomization: Board 17 Control Water: Lab Water (80:20)

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Temp (°C)	A	B	C	D	E	F	G	H	I	J
Lab Water Control	0	8.16		8.5		[illegible]	25.9	0	0	0	0	0	0	0	0	0	0
	1	8.06	8.53	9.8	8.2	234	25.7	0	0	0	0	0	0	0	0	0	0
	2	8.14	8.12	8.8	7.9	227	24.8	0	0	0	0	0	0	0	0	0	0
	3	8.05	8.20	8.9	8.1	225	25.8	0	0	0	0	0	[illegible]	0	0	0	0
	4	7.79	8.12	9.0	7.2	203	25.6	3	0	4	3	3	0	4	5	4	4
	5	8.00	8.21	8.4	8.1	205	25.7	0	4	0	0	0	5	0	0	0	0
	6	8.29	8.18	8.3	7.9	223	25.7	1	[illegible]	7	2	5	9	0	0	[illegible]	[illegible]
	7	7.86	8.09	8.0	7.0	202	25.3	9	8	11	8	10	0	0	4	0	0
	8		NM	.	NM	NM	25.8	0	0	8*	9*	7*	X/13	8	7	8	0
							Total=	13	17	[illegible]	[illegible]	[illegible]	X/27	12	16	12	4

SIGN-OFF

Date: 9/1/09 New WQ: KR Test Init.: KO
Sol'n Prep: [illegible] Time: [illegible]
Date: 9/2/09 New WQ: [illegible] Counts: [illegible]
Sol'n Prep: SH Old WQ: KR Time: 1400
Date: 9/3/09 New WQ: SH Counts: [illegible]
Sol'n Prep: [illegible] Old WQ: SH Time: 1400
Date: 9/4/09 New WQ: [illegible] Counts: [illegible]
Sol'n Prep: [illegible] Old WQ: [illegible] Time: [illegible]
Date: 9/5/09 New WQ: [illegible] Counts: [illegible]
Sol'n Prep: [illegible] Old WQ: MB Time: [illegible]
Date: 9/6/09 New WQ: [illegible] Counts: [illegible]
Sol'n Prep: SH Old WQ: [illegible] Time: 1330
Date: 9/7/09 New WQ: SH Counts: [illegible]
Sol'n Prep: [illegible] Old WQ: [illegible] Time: [illegible]
Date: 9/8/09 New WQ: [illegible] Counts: [illegible]
Sol'n Prep: [illegible] Old WQ: [illegible] Time: 1600
Date: 9/9/09 Old WQ: – Counts: [illegible]
Time: 1530

Mean Neonates/Female = 13.4 [illegible]

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	A	B	C	D	E	F	G	H	I	J
250 mg/L	0	8.19		9.1		720	0	0	0	0	0	0	0	0	0	0
	1	8.12	8.45	9.9	8.3	726	0	0	0	0	0	0	0	0	0	0
	2	8.04	8.12	8.5	8.0	785	0	0	0	0	0	0	0	0	0	0
	3	8.08	8.19	8.8	8.4	773	0	0	0	0	0	0	0	0	0	0
	4	[illegible]	8.11	8.8	6.5	735	0	0	3	3	3	3	3	3	4	4
	5	8.04	8.19	8.6	8	691	4	4	0	0	0	0	3	5	0	0
	6	8.22	8.16	8.4	7.7	715	5	7	6	0	7	1	1	0	6	7
	7	7.89	8.05	8.3	7.1	719	0	0	11	10	10	10	9	10	8	11
	8		NM		NM	NM	0	0	2	X/9	8*	9	10*	9*	6*	0
							Total=	9	11	22	X/22	[illegible]	23	[illegible]	[illegible]	[illegible]	22

RT BATCH NUMBER

0	19
1	20
2	20
3	20
4	20
5	20
6	20
7	20
8	–

Mean Neonates/Female = [illegible] 18.1

* fourth brood

Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: **Reference Toxicant** Material: **Sodium Chloride** Test Date: 9/1/07

Project #: 15165 Test ID: 34097 Randomization: Board 17 Control Water: Lab Water (80:20)

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Temp (°C)	A	B	C	D	E	F	G	H	I	J	SIGN-OFF
500 mg/L	0	8.17		9.1		1158		0	0	0	0	0	0	0	0	0	0	
	1	8.16	8.40	9.8	8.3	1244		0	0	0	0	0	0	0	0	0	0	
	2	8.19	8.11	8.7	7.9	1220		0	0	0	0	0	0	0	0	0	0	
	3	8.13	8.16	8.9	8.2	1248		0	0	0	0	0	0	0	0	0	0	
	4	8.05	8.10	8.9	6.5	1206		3	0	4	3	3	3	0	4	3	3	
	5	8.04	8.17	8.5	7.9	1246		0	4	0	4	0	0	5	0	0	0	
	6	8.19	8.14	8.6	7.9	1264		4	5	4	0	X/6	7	6	6	7	8	
	7	7.90	8.04	8.4	7.1	1208		9	0	9	9	–	8	11	8	6	8	
	8		NM		NM	NM		0	0	6*	7*	–	6*	0	9*	11*	0	
							Total=	16	9	~~25~~ 19	~~23~~ 16	X/9	~~24~~ 18	22	~~27~~ 18	~~27~~ 16	19	Mean Neonates/Female = ~~20.1~~ 16.2

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)		A	B	C	D	E	F	G	H	I	J	
1000 mg/L	0	8.19		9.4		2044		0	0	0	0	0	0	0	0	0	0	
	1	8.18	8.35	9.9	8.2	2138		0	0	0	0	0	0	0	0	0	0	
	2	8.17	8.09	9.0	8.0	2198		0	0	0	0	0	0	0	0	0	0	
	3	8.12	8.13	9.2	8.0	2171		0	0	0	0	0	0	0	0	0	0	
	4	~~8.05~~	~~8.10~~ 8.07	~~10.4~~ 8.5	~~6.4~~ 6.4	2203		0	0	2	0	0	0	2	3	2	0	
	5	8.02	8.15	8.8	7.8	2109		4	4	0	3	0	0	0	0	0	0	
	6	8.16	8.12	8.9	7.9	2172		5	5	6	X/8	6	5	6	5	4	5	
	7	7.91	8.04	8.7	7.1	2130		0	5	3	–	X/7	9	5	7	7	X/3	
	8		NM		NM	NM		X/0	0	2*	–	–	3	2*	2*	5*	–	
							Total=	X/9	14	~~15~~ 11	X/13	X/13	17	~~15~~ 13	~~17~~ 15	~~18~~ 13	X/8	Mean Neonates/Female = ~~14.3~~ 12.6

Short-Term Chronic 3-Brood *Ceriodaphnia dubia* Survival & Reproduction Test Data

Client: **Reference Toxicant** Material: **Sodium Chloride** Test Date: 9/1/09

Project #: 15165 Test ID: 34097 Randomization: Bond 17 Control Water: Lab Water (80:20)

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)	Temp (°C)	A	B	C	D	E	F	G	H	I	J	SIGN-OFF
1500 mg/L	0	8.05		9.8		2929		0	0	0	0	0	0	0	0	0	0	
	1	8.18	8.33	10.0	8.3	3120		0	0	0	0	0	0	0	0	0	0	
	2	8.14	8.08	9.3	8.0	3150		0	0	0	0	0	0	0	0	0	0	
	3	8.11	8.12	9.3	8.2	3120		0	0	0	0	0	0	0	0	0	0	
	4	8.04	8.11	8.9	6.6	3140		2	0	0	0	0	0	0	0	2	0	
	5	8.01	8.17	9.3	7.8	3160		0	0	0	0	0	0	0	0	0	0	
	6	8.15	8.12	9.4	7.9	3140		5	0	4	5	4	4	5	4	4	3	
	7	7.92	8.05	9.2	7.1	3060		3	0	2	6	4	0	3	4	0	3	
	8		NM		NM	NM		0	0	0	0	0	X/0	0	0	X/0	0	
							Total=	~~8~~ 10	0	6	11	8	X/4	8	8	X/6	6	Mean Neonates/Female = ~~6.5~~ 6.7

	Day	pH New	pH Old	D.O. New	D.O. Old	Cond. (µS/cm)		A	B	C	D	E	F	G	H	I	J	
2000 mg/L	0	7.97		10.3		3730		0	0	0	0	0	0	0	0	0	0	
	1	8.18	8.35	10.3	8.4	4080		0	0	0	0	0	0	0	0	0	0	
	2	8.11	8.11	9.5	8.2	4120		X/0	X/0	0	X/0	0	0	X/0	X/0	0	0	
	3	8.10	8.15	9.6	8.3	4060		–	–	0	–	0	0	–	–	0	X/0	
	4	8.02	8.16	9.9	7.0	4090		–	–	0	–	0	0	–	–	0	–	
	5	8.01	8.17	9.6	8.0	4090		–	–	0	–	0	0	–	–	0	–	
	6	8.12	8.13	9.5	7.8	4130		–	–	0	–	0	0	–	–	0	–	
	7	7.93	~~8.09~~	9.8	7.4	3970		–	–	0	–	0	0	–	–	0	–	
	8		NM		NM	NM		–	–	0	–	0	0	–	–	0	–	
							Total=	X/0	X/0	0	X/0	0	0	X/0	X/0	0	X/0	Mean Neonates/Female = 0

Appendix D

Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of Chevron/Cawelo 'Inlet to Reservoir B' Effluent to Fathead Minnows

CETIS Summary Report

Report Date: 09 Sep-09 16:13 (p 1 of 2)
Test Code: 08-1872-4281/36008

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Batch ID:	10-8732-2753	Test Type:	Growth-Survival (7d)	Analyst:	John Jirasritumrong
Start Date:	01 Sep-09 18:40	Protocol:	EPA/821/R-02-013 (2002)	Diluent:	Laboratory Water
Ending Date:	08 Sep-09 08:30	Species:	Pimephales promelas	Brine:	Not Applicable
Duration:	6d 14h	Source:	Enviro Sciences, Inc.	Age:	1

Sample ID:	10-3556-7062	Code:	Eff	Client:	Precision Analytical
Sample Date:	31 Aug-09 13:00	Material:	Effluent	Project:	15164
Receive Date:	31 Aug-09 19:50	Source:	Precision Analytical		
Sample Age:	30h (25.3 °C)	Station:	Inlet Resv B		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
13-2878-4765	7d Survival Rate	12.5	25	17.7	12.4%	8	Dunnett's Multiple Comparison Test
08-1008-1226	Mean Dry Biomass-mg	<12.5	12.5	N/A	16.3%	>8	Dunnett's Multiple Comparison Test
14-1524-3393	Mean Dry Weight-mg	<12.5	12.5	N/A	16.7%	>8	Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis ID	Endpoint	Level	%	95% LCL	95% UCL	TU	Method
02-2949-0593	7d Survival Rate	EC50	39.8	34.4	46	2.52	Trimmed Spearman-Kärber
05-2530-0471	Mean Dry Biomass-mg	IC5	0.697	0.326	2.43	143	Linear Interpolation (ICPIN)
		IC10	1.88	0.706	9.65	53.2	
		IC15	3.89	1.1	19.7	25.7	
		IC20	7.3	1.39	22.3	13.7	
		IC25	12.7	1.56	25.1	7.86	
		IC40	24.5	12.7	32.5	4.08	
		IC50	29.5	22.9	37.1	3.39	

7d Survival Rate Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	0.0%
12.5		4	0.9	0.87	0.93	0.8	1	0.0149	0.0816	9.07%	7.69%
25		4	0.8	0.77	0.83	0.7	0.9	0.0149	0.0816	10.2%	17.9%
50		4	0.4	0.347	0.453	0.2	0.5	0.0258	0.141	35.4%	59.0%
75		4	0	0	0	0	0	0	0		100.0%
100		4	0	0	0	0	0	0	0		100.0%

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.452	0.424	0.48	0.364	0.524	0.0137	0.075	16.6%	0.0%
12.5		4	0.341	0.332	0.35	0.309	0.359	0.00418	0.0229	6.72%	24.6%
25		4	0.269	0.255	0.283	0.236	0.319	0.00685	0.0375	13.9%	40.5%
50		4	0.086	0.0756	0.0964	0.045	0.104	0.00507	0.0277	32.3%	81.0%
75		4	0	0	0	0	0	0	0		100.0%
100		4	0	0	0	0	0	0	0		100.0%

Mean Dry Weight-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.462	0.44	0.485	0.404	0.524	0.011	0.0604	13.1%	0.0%
12.5		4	0.382	0.363	0.401	0.309	0.425	0.00922	0.0505	13.2%	17.4%
25		4	0.339	0.319	0.358	0.272	0.399	0.00948	0.0519	15.3%	26.8%
50		4	0.217	0.212	0.222	0.204	0.233	0.00248	0.0136	6.25%	53.0%
75		4	0	0	0	0	0	0	0		100.0%
100		4	0	0	0	0	0	0	0		100.0%

Chronic Larval Fish Survival and Growth Test

Pacific EcoRisk

7d Survival Rate Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	1	1	0.9	1
12.5		1	0.9	0.9	0.8
25		0.8	0.8	0.9	0.7
50		0.5	0.5	0.2	0.4
75		0	0	0	0
100		0	0	0	0

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	0.524	0.504	0.364	0.417
12.5		0.309	0.356	0.359	0.34
25		0.319	0.277	0.245	0.236
50		0.102	0.104	0.045	0.093
75		0	0	0	0
100		0	0	0	0

Mean Dry Weight-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	0.524	0.504	0.404	0.417
12.5		0.309	0.396	0.399	0.425
25		0.399	0.346	0.272	0.337
50		0.204	0.208	0.225	0.233
75		0	0	0	0
100		0	0	0	0

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis ID:	13-2878-4765	Endpoint:	7d Survival Rale	CETIS Version:	CETISv1.7.0
Analyzed:	09 Sep-09 16:12	Analysis:	Parametric-Control vs Treatments	Official Results:	Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)	0	C > T	Not Run	12.5	25	17.7	8	12.4%

Dunnett's Multiple Comparison Test

Control vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Lab Water Control	12.5	1.39	2.29	0.192	0.2033	Non-Significant Effect
	25*	3.07	2.29	0.192	0.0125	Significant Effect
	50*	8.24	2.29	0.192	<0.0001	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	1.096293	0.3654311	3	26	<0.0001	Significant Effect
Error	0.1689718	0.01408098	12			
Total	1.265265	0.379512	15			

ANOVA Assumptions

Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	1.05	11.3	0.7903	Equal Variances
Distribution	Shapiro-Wilk Normality	0.938		0.3275	Normal Distribution

7d Survival Rate Summary

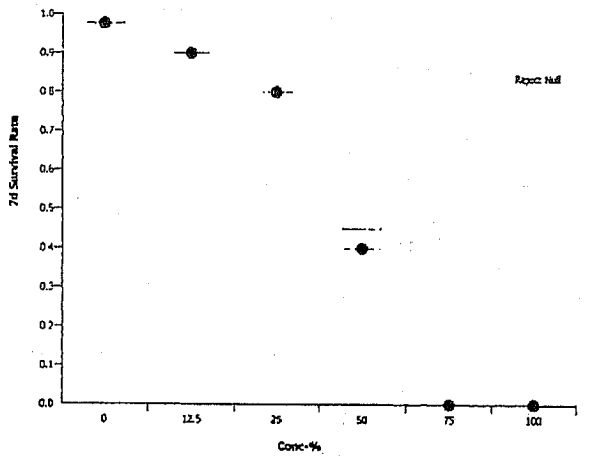
Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.975	0.956	0.994	0.9	1	0.00928	0.05	5.13%	0.0%
12.5		4	0.9	0.869	0.931	0.8	1	0.0152	0.0816	9.07%	7.69%
25		4	0.8	0.769	0.831	0.7	0.9	0.0152	0.0816	10.2%	17.9%
50		4	0.4	0.346	0.454	0.2	0.5	0.0263	0.141	35.4%	59.0%
75		4	0	0	0	0	0	0	0		100.0%
100		4	0	0	0	0	0	0	0		100.0%

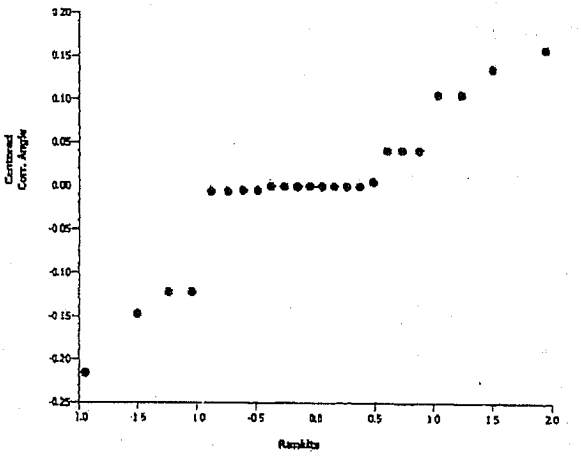
Angular (Corrected) Transformed Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Cont	4	1.37	1.34	1.4	1.25	1.41	0.0151	0.0815	5.94%	0.0%
12.5		4	1.25	1.21	1.3	1.11	1.41	0.0231	0.125	9.93%	8.53%
25		4	1.11	1.07	1.15	0.991	1.25	0.0196	0.106	9.48%	18.8%
50		4	0.68	0.622	0.737	0.464	0.785	0.0282	0.152	22.3%	50.4%
75		4	0.159	0.159	0.159	0.159	0.159	0	0	0.0%	88.4%
100		4	0.159	0.159	0.159	0.159	0.159	0	0	0.0%	88.4%

Chronic Larval Fish Survival and Growth Test					Pacific EcoRisk
Analysis ID:	13-2878-4765	Endpoint:	7d Survival Rate	CETIS Version:	CETISv1.7.0
Analyzed:	09 Sep-09 16:12	Analysis:	Parametric-Control vs Treatments	Official Results:	Yes

Graphics





Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis ID:	08-1008-1226	Endpoint:	Mean Dry Biomass-mg	CETIS Version:	CETISv1.7.0
Analyzed:	09 Sep-09 16:12	Analysis:	Parametric-Control vs Treatments	Official Results:	Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	0	C > T	Not Run	<12.5	12.5	N/A	>8	16.3%

Dunnett's Multiple Comparison Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Lab Water Control		12.5*	3.45	2.29	0.0738	0.0063	Significant Effect
		25*	5.67	2.29	0.0738	0.0001	Significant Effect
		50*	11.4	2.29	0.0738	<0.0001	Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.2837575	0.09458584	3	45.5	<0.0001	Significant Effect
Error	0.02496921	0.002080768	12			
Total	0.3087267	0.0966666	15			

ANOVA Assumptions

Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	4.66	11.3	0.1987	Equal Variances
Distribution	Shapiro-Wilk Normality	0.962		0.7057	Normal Distribution

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.452	0.424	0.481	0.364	0.524	0.0139	0.075	16.6%	0.0%
12.5		4	0.341	0.332	0.35	0.309	0.359	0.00425	0.0229	6.72%	24.6%
25		4	0.269	0.255	0.284	0.236	0.319	0.00697	0.0375	13.9%	40.5%
50		4	0.086	0.0754	0.0966	0.045	0.104	0.00515	0.0277	32.3%	81.0%
75		4	0	0	0	0	0	0	0		100.0%
100		4	0	0	0	0	0	0	0		100.0%

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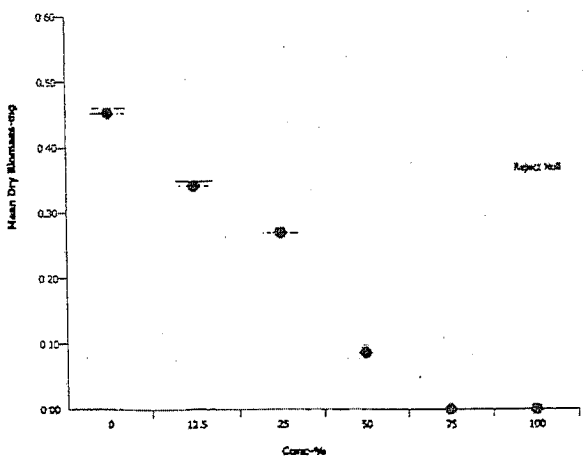

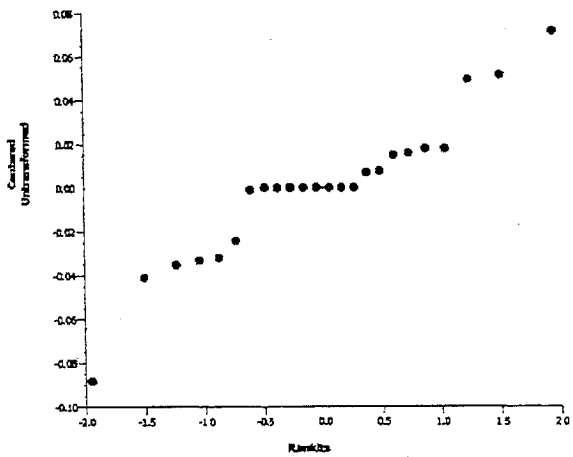

CETIS Analytical Report

Report Date: 09 Sep-09 16:13 (p 1 of 1)
Test Code: 08-1872-4281/36008

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis ID:	02-2949-0593	Endpoint:	7d Survival Rate	CETIS Version:	CETISv1.7.0
Analyzed:	09 Sep-09 16:12	Analysis:	Trimmed Spearman-Kärber	Official Results:	Yes

Trimmed Spearman-Kärber Estimates

Threshold Option	Threshold	Trim	Mu	Sigma	EC50	95% LCL	95% UCL
Control Threshold	0.025	7.69%	1.6	0.0315	39.8	34.4	46

7d Survival Rate Summary

			Calculated Variate(A/B)								
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%	A	B
0	Lab Water Contro	4	0.975	0.9	1	0.00913	0.05	5.13%	0.0%	39	40
12.5		4	0.9	0.8	1	0.0149	0.0816	9.07%	7.69%	36	40
25		4	0.8	0.7	0.9	0.0149	0.0816	10.2%	17.9%	32	40
50		4	0.4	0.2	0.5	0.0258	0.141	35.4%	59.0%	16	40
75		4	0	0	0	0	0		100.0%	0	40
100		4	0	0	0	0	0		100.0%	0	40

7d Survival Rate Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Control	1	1	0.9	1
12.5		1	0.9	0.9	0.8
25		0.8	0.8	0.9	0.7
50		0.5	0.5	0.2	0.4
75		0	0	0	0
100		0	0	0	0

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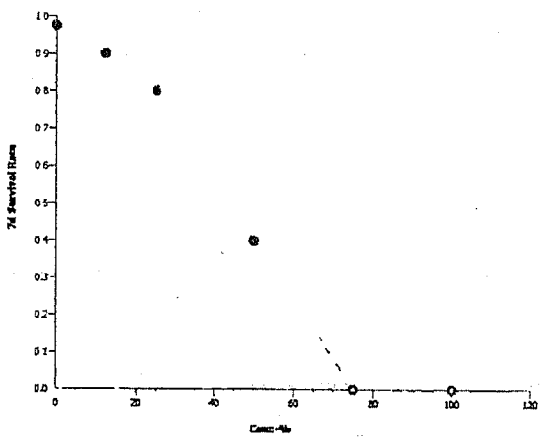

CETIS Analytical Report

Report Date: 09 Sep-09 16:13 (p 1 of 1)
Test Code: 08-1872-4281/36008

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis ID:	05-2530-0471	Endpoint:	Mean Dry Biomass-mg	CETIS Version:	CETISv1.7.0
Analyzed:	09 Sep-09 16:12	Analysis:	Linear Interpolation (ICPIN)	Official Results:	Yes

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	57951	200	Yes	Two-Point Interpolation

Point Estimates

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC5	0.697	0.326	2.43	143	41.1	307
IC10	1.88	0.706	9.65	53.2	10.4	142
IC15	3.89	1.1	19.7	25.7	5.09	91.1
IC20	7.3	1.39	22.3	13.7	4.48	71.9
IC25	12.7	1.56	25.1	7.86	3.98	64
IC40	24.5	12.7	32.5	4.08	3.08	7.9
IC50	29.5	22.9	37.1	3.39	2.69	4.36

Mean Dry Biomass-mg Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contro	4	0.452	0.364	0.524	0.0137	0.075	16.6%	0.0%
12.5		4	0.341	0.309	0.359	0.00418	0.0229	6.72%	24.6%
25		4	0.269	0.236	0.319	0.00685	0.0375	13.9%	40.5%
50		4	0.086	0.045	0.104	0.00507	0.0277	32.3%	81.0%
75		4	0	0	0	0	0		100.0%
100		4	0	0	0	0	0		100.0%

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Control	0.524	0.504	0.364	0.417
12.5		0.309	0.356	0.359	0.34
25		0.319	0.277	0.245	0.236
50		0.102	0.104	0.045	0.093
75		0	0	0	0
100		0	0	0	0

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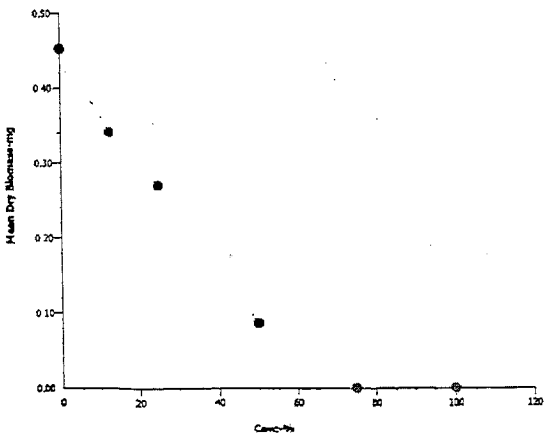

7 Day Chronic Fathead Minnow Toxicity Test Data

Client: Precision Analytical - Chevron Cawelo
Test Material: Inlet to Reservoir B
Test ID#: 36008 Project #: 15164
Test Date: 9/1/09 Randomization: 4.6.10

Organism Log#: 4740 Age: 048hrs
Organism Supplier: Envirosciences
Control/Diluent: EPAMH
Control Water Batch: 1237

Treatment (% Effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Conductivity (µs/cm)	# Live Organisms A	B	C	D	SIGN-OFF
Lab Water Control	25.8	7.98		8.7		345	10	10	10	10	Date: 9/01/09
12.5%	25.8	7.64		8.6		399	10	10	10	10	Sample ID 22637
25%	25.8	7.14		8.4		449	10	10	10	10	Test Solution Prep KO
50%	25.8	7.00		8.0		578	10	10	10	10	New WQ SA
75%	25.8	6.90		7.9		705	10	10	10	10	Initiation Time 1840
100%	25.8	6.84		7.0		829	10	10	10	10	Initiation Signoff [illegible]
Meter ID	22A	PH11		DO12		EC03					
Lab Water Control	25.6	8.05	8.09	9.9	6.3	341	10	10	10	10	Date: 9/2/09
12.5%	25.6	7.91	8.20	9.5	6.4	402	10	10	10	10	Sample ID 22637
25%	25.6	7.76	8.27	9.7	6.3	462	10	10	10	10	Test Solution Prep [illegible]
50%	25.6	7.59	8.40	9.4	6.3	569	10	10	10	10	New WQ [illegible]
75%	25.6	7.37	8.53	9.5	6.2	671	10	10	10	10	Renewal Time 10:05
100%	25.6	7.13	8.54	9.6	6.3	832	10	9	9	10	Renewal Signoff [illegible]
Meter ID	22A	PH12	PH14	DO12	DO14	EC03					Old WQ [illegible]
Lab Water Control	24.8	7.94	8.26	9.0	8.4	343	10	10	10	10	Date: 9/3/09
12.5%	24.8	7.55	8.24	9.0	8.1	424	10	10	10	10	Sample ID 22637
25%	24.8	7.42	8.30	8.7	8.0	464	10	10	10	10	Test Solution Prep [illegible]
50%	24.8	7.23	8.44	9.1	7.8	587	10	10	10	10	New WQ SH
75%	24.8	7.05	8.53	8.5	7.9	713	10	5	10	8	Renewal Time 1200
100%	24.8	6.98	8.53	8.2	7.8	835	5	9	6	5	Renewal Signoff SH
Meter ID	22A	PH11	PH14	DO12	DO13	EC05					Old WQ SH
Lab Water Control	25.8	7.89	8.08	9.0	[illegible]	332	10	10	10	10	Date: 9/4/09
12.5%	25.8	7.69	8.18	8.5	8.[illegible]	395	10	10	10	10	Sample ID 22637
25%	25.8	7.43	8.21	8.4	7.9	455	10	10	10	10	Test Solution Prep JL
50%	25.8	7.19	8.36	8.4	7.8	580	10	10	9	8	New WQ [illegible]
75%	25.8	7.05	8.50	8.3	7.9	703	6	0	9	5	Renewal Time 1130
100%	25.8	6.93	8.51	8.1	7.9	824	1	6	1	0	Renewal Signoff [illegible]
Meter ID	22A	PH14	PH14	DO13	DO13	EC03					Old WQ [illegible]

7 Day Chronic Fathead Minnow Toxicity Test Data

Client: Precision Analytical - Chevron Cawelo
Test Material: Inlet to Reservoir B
Test ID#: 36008 Project #: 15164
Test Date: 9/1/09 Randomization: 4.6.10

Organism Log#: 4740 Age: <48hrs
Organism Supplier: Envirosciences
Control/Diluent: EPAMH
Control Water Batch: 1237

Treatment (% Effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Conductivity (µs/cm)	# Live Organisms A	B	C	D	SIGN-OFF
Lab Water Control	25.6	7.07	7.98	8.0	8.4	331	10	10	10	10	Date: 9/05/09
12.5%	25.6	7.65	8.14	8.2	8.1	400	10	9	9	10	Sample ID 22637
25%	25.6	7.50	8.21	8.3	8.1	449	10	10	10	8	Test Solution Prep DC
50%	25.6	7.08	8.34	7.8	8.0	587	10	8	7	8	New WQ MB
75%	25.6	6.98	8.44	7.8	7.8	699	4	–	2	2	Renewal Time 1100
100%	25.6	6.90	8.48	7.3	7.8	836	0	2	0	–	Renewal Signoff JPC
Meter ID	22A	pH14	pH11	DO14	DO12	EC04					Old WQ DC
Lab Water Control	25.6	7.99	8.03	8.4	8.2	335	10	10	9	10	Date: 9/6/09
12.5%	25.6	7.72	8.13	8.4	8.1	397	10	9	9	9	Sample ID 22637
25%	25.6	7.46	8.22	8.3	8.0	458	9	8	9	8	Test Solution Prep DC
50%	25.6	7.16	8.38	8.4	7.9	588	9	6	6	7	New WQ EKK
75%	25.6	7.08	8.54	8.7	8.0	703	3	–	2	0	Renewal Time 1140
100%	25.6	6.98	8.61	8.8	8.0	836	–	0	–	–	Renewal Signoff SH
Meter ID	22A	pH11	pH11	DO13	DO13	EC05					Old WQ EKK
Lab Water Control	25.7	8.77 SH	8.10	8.4	8.2	340	10	10	9	10	Date: 9/7/09
12.5%	25.7	7.83	8.16	8.4	8.1	399	10	9	9	8	Sample ID 22637
25%	25.7	7.51	8.23	8.4	8.0	464	9	8	9	7	Test Solution Prep PA
50%	25.7	7.30	8.41	8.4	8.0	583	7	6	4	6	New WQ SH
75%	25.7	7.16	8.58	8.4	7.9	704	0	–	0	–	Renewal Time 1050
100%	25.7	7.02	–	8.0	–	833	–	–	–	–	Renewal Signoff PA
Meter ID	22A	pH14	pH14	DO13	DO13	EC05					Old WQ DC
Lab Water Control	25.3		7.98		7.7	358	10	10	9	10	Date 9/8/09
12.5%	25.3		8.11		7.9	417	10	9	9	8	Termination Time 0830
25%	25.3		8.17		7.8	489	9	8	9	7	Termination Signoff JKW
50%	25.3		8.36		7.8	614	5	5	2	4	Old WQ KR
75%	–		–		–	–	–	–	–	–	
100%	–		–		–	–	–	–	–	–	
Meter ID	22A		pH14		DO13	EC04					

Fathead Minnow Dry Weight Data Sheet

Client: Precision Analytical - Chevron Cawelo Test ID #: 36008 Project # 15164
Sample: Inlet to Reservoir B Tare Weight Date: 9/4/09 Sign-off: KR
Test Date: 9/1/09 Final Weight Date: 9/9/09 Sign-off: DRD

Pan ID	Concentration	Replicate	Initial Pan Weight (mg)	Final Pan Weight (mg)	Initial # of Organisms	Biomass Value (mg)
1	Lab Water	A	170.27	175.51	10	0.5240
2		B	151.64	156.68	10	0.5040
3		C	171.51	175.15	10	0.3640
4		D	170.84	~~150.87~~ 175.01	10	0.4170
5	12.5	A	147.84	~~184.73~~ 150.93	10	0.3090
6		B	181.17	184.73	10	~~0.335~~ 0.3560
7		C	152.66	156.25	10	0.3590
8		D	166.84	170.24	10	0.3400
9	25	A	180.63	183.82	10	0.3190
10		B	175.30	178.07	10	0.2770
11		C	185.30	187.75	10	0.2450
12		D	170.67	173.03	10	0.2360
13	50	A	172.32	173.34	10	0.1020
14		B	171.27	172.31	10	0.1040
15		C	188.68	189.13	10	0.0450
16		D	176.84	177.77	10	0.0930
17	75	A	174.59	-	~~0~~	0
18		B	167.80	-	~~0~~	0
19		C	176.55	-	~~0~~	0
20		D	178.32	-	~~0~~	0
21	100	A	161.68	-	~~0~~	0
22		B	171.38	-	~~0~~	0
23		C	167.57	-	~~0~~	0
24		D	161.58	-	~~0~~	0
QA 1			166.16	166.11		
QA 2			175.90	175.85		
Balance ID			1	1		

Appendix E

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the Fathead Minnows

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Batch ID:	19-3042-6112	Test Type:	Growth-Survival (7d)	Analyst:	Padrick Anderson
Start Date:	01 Sep-09 18:45	Protocol:	EPA-821-R-02-013 (2002)	Diluent:	Laboratory Water
Ending Date:	08 Sep-09 18:45	Species:	Pimephales promelas	Brine:	Not Applicable
Duration:	7d 0h	Source:	Enviro Sciences, Inc.	Age:	1

Sample ID:	07-2692-6773	Code:	NaCl	Client:	Reference Toxicant
Sample Date:	01 Sep-09 18:35	Material:	Sodium chloride	Project:	15170
Receive Date:	01 Sep-09 18:35	Source:	Reference Toxicant		
Sample Age:	10m (25.9 °C)	Station:	In House		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
16-5973-0337	7d Survival Rate	3	6	4.24	14.5%		Dunnett's Multiple Comparison Test
18-7014-3832	Mean Dry Biomass-mg	1.5	3	2.12	14.5%		Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis ID	Endpoint	Level	g/L	95% LCL	95% UCL	TU	Method
06-1534-1943	7d Survival Rate	EC50	5.13	4.55	5.77		Spearman-Kärber
17-7101-7464	Mean Dry Biomass-mg	IC5	1.61	N/A	2.15		Linear Interpolation (ICPIN)
		IC10	2.02	N/A	2.75		
		IC15	2.49	1.66	3.45		
		IC20	3.02	2.02	3.73		
		IC25	3.4	2.42	4.22		
		IC40	4.73	3.65	6.38		
		IC50	5.84	4.46	6.72		
05-2254-4356	Mean Dry Weight-mg	IC5	2.33	N/A	8.31		Linear Interpolation (ICPIN)
		IC10	6.06	0.00617	6.28		
		IC15	6.2	4.31	6.41		
		IC20	6.34	5.9	6.54		
		IC25	6.48	6.07	6.68		
		IC40	6.93	6.57	7.09		
		IC50	7.24	6.93	7.38		

7d Survival Rate Summary

Conc-g/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	0.0%
0.75		4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	0.0%
1.5		4	0.95	0.913	0.987	0.8	1	0.0183	0.1	10.5%	2.56%
3		4	0.85	0.802	0.898	0.7	1	0.0236	0.129	15.2%	12.8%
6		4	0.525	0.461	0.589	0.3	0.7	0.0312	0.171	32.5%	46.2%
9		4	0	0	0	0	0	0	0		100.0%

Mean Dry Biomass-mg Summary

Conc-g/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.359	0.352	0.365	0.336	0.376	0.00329	0.018	5.02%	0.0%
0.75		4	0.326	0.315	0.338	0.294	0.364	0.00582	0.0319	9.77%	8.99%
1.5		4	0.365	0.356	0.374	0.343	0.4	0.00451	0.0247	6.76%	-1.81%
3		4	0.288	0.278	0.298	0.255	0.319	0.00494	0.0271	9.39%	19.7%
6		4	0.175	0.157	0.192	0.112	0.225	0.00852	0.0467	26.7%	51.3%
9		4	0	0	0	0	0	0	0		100.0%

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Mean Dry Weight-mg Summary

Conc-g/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contr	4	0.369	0.36	0.377	0.336	0.392	0.00432	0.0236	6.42%	0.0%
0.75		4	0.337	0.318	0.355	0.294	0.404	0.00901	0.0494	14.7%	8.67%
1.5		4	0.387	0.371	0.404	0.343	0.443	0.00797	0.0436	11.3%	-5.11%
3		4	0.343	0.328	0.357	0.319	0.401	0.00724	0.0397	11.6%	7.05%
6		4	0.34	0.327	0.352	0.302	0.373	0.00615	0.0337	9.93%	7.85%
9		4	0	0	0	0	0	0	0		100.0%

7d Survival Rate Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	1	1	1	0.9
0.75		0.9	1	1	1
1.5		0.8	1	1	1
3		0.8	0.7	0.9	1
6		0.7	0.3	0.5	0.6
9		0	0	0	0

Mean Dry Biomass-mg Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	0.376	0.37	0.336	0.353
0.75		0.364	0.294	0.307	0.341
1.5		0.354	0.364	0.343	0.4
3		0.255	0.281	0.298	0.319
6		0.225	0.112	0.181	0.181
9		0	0	0	0

Mean Dry Weight-mg Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	0.376	0.37	0.336	0.392
0.75		0.404	0.294	0.307	0.341
1.5		0.443	0.364	0.343	0.4
3		0.319	0.401	0.331	0.319
6		0.321	0.373	0.362	0.302
9		0	0	0	0

7 Day Chronic Fathead Minnow Reference Toxicant Test Data

Client: Reference Toxicant
Test Material: Sodium Chloride
Test ID#: 36010 Project #: 15170
Test Date: 9/1/09 Randomization: 4.68

Organism Log#: 4740 Age: <48hrs
Organism Supplier: Enviro Sciences
Control/Diluent: EPAMH
Control Water Batch: 1236

Treatment (g/L)	Temp (°C)	pH New	pH Old	DO (mg/L) New	DO (mg/L) Old	Conductivity (µs/cm)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.9	8.52		8.6		328	10	10	10	10	Date: 9/1/09
0.75	25.9	8.35		8.6		1827	10	10	10	10	Test Solution Prep:
1.5	25.9	8.23		8.9		3238	10	10	10	10	New WQ:
3	25.9	8.13		9.3		6270	10	10	10	10	Initiation Time: 1845
6	25.9	8.05		9.3		11650	10	10	10	10	Initiation Signoff: JPC
9	25.9	7.98		9.6		16640	10	10	10	10	RT Stock Batch #: 27
Meter ID	22A	pH11		DO12		EC03					
Control	25.3	8.19	8.07	8.5	6.3	344	10	10	10	10	Date: 9/2/09
0.75	25.3	8.24	7.99	8.7	6.2	1801	10	10	10	10	Test Solution Prep:
1.5	25.3	8.16	7.95	8.7	6.2	3260	10	10	10	10	New WQ: KR
3	25.3	8.03	7.93	9.1	6.2	6320	10	10	10	10	Renewal Time: 930
6	25.3	7.94	7.83	9.7	6.1	11590	10	10	10	10	Renewal Signoff:
9	25.3	7.85	7.82	10.3	6.3	16790	10	10	10	10	Old WQ: KR
Meter ID	22A	pH14	pH14	DO14	DO14	EC04					RT Stock Batch #: 27
Control	25.5	8.09	7.83	8.7	7.7	341	10	10	10	10	Date: 9/3/09
0.75	25.5	8.12	7.73	8.8	7.4	1898	10	10	10	10	Test Solution Prep: JPC
1.5	25.5	8.11	7.74	9.2	7.3	3240	10	10	10	10	New WQ: SH
3	25.5	8.08	7.68	9.3	7.4	6010	10	9	10	10	Renewal Time: 1015
6	25.5	8.01	7.61	9.8	7.6	11220	8	9	7	9	Renewal Signoff: JPC
9	25.5	7.92	7.60	10.2	7.5	16540	0	0	0	0	Old WQ: HV
Meter ID	22A	pH11	pH14	DO12	DO13	EC05					RT Stock Batch #: 27
Control	25.8	8.00	7.86	8.4	7.5	343	10	10	10	10	Date: 9/4/09
0.75	25.8	7.99	7.84	8.2	7.3	1864	10	10	10	10	Test Solution Prep: JPC
1.5	25.8	7.98	7.71	8.3	7.2	3190	10	10	10	10	New WQ:
3	25.8	7.96	7.72	8.7	7.6	6140	10	9	10	10	Renewal Time: 1115
6	25.8	7.89	7.66	8.9	7.1	11320	7	8	7	9	Renewal Signoff:
9	—	—	—	—	—	—	—	—	—	—	Old WQ: HV
Meter ID	22A	pH14	pH14	DO13	DO13	EC03					RT Stock Batch #: 27

7 Day Chronic Fathead Minnow Reference Toxicant Test Data

Client: Reference Toxicant
Test Material: Sodium Chloride
Test ID#: 36010 Project #: 15170
Test Date: 9/1/09 Randomization: 4·68

Organism Log#: 4740 Age: <48 hrs
Organism Supplier: Enviro Sciences
Control/Diluent: EPAMH
Control Water Batch: 1236

Treatment (g/L)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Conductivity (µs/cm)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.6	8.14	7.90	8.8	7.7	327	10	10	10	10	Date 9/5/09
0.75	25.6	8.09	7.92	8.6	8.1	1735	10	10	10	10	Test Solution Prep JPC
1.5	25.6	8.06	7.86	8.6	7.9	3190	10	10	10	10	New WQ [illegible]
3	25.6	8.01	7.80	8.8	8.0	6060	9	9	10	10	Renewal Time 1030
6	25.6	7.92	7.76	9.0	7.6	11390	7	8	6	8	Renewal Signoff JPC
9	—	—	—	—	—	—	—	—	—	—	Old WQ MB
Meter ID	22A	pH 11	pH 14	DO12	DO14	EC03					RT Stock Batch #: 27
Control	25.6	7.98	7.95	8.3	7.7	336	10	10	10	10	Date: 9/6/09
0.75	25.6	7.93	7.92	8.5	7.7	1883	10	10	10	10	Test Solution Prep PA
1.5	25.6	7.91	7.87	8.5	7.3	3310	9	10	10	10	New WQ SH
3	25.6	7.86	7.81	8.6	7.4	6410	8	8	10	10	Renewal Time: 1210
6	25.6	7.81	7.78	8.6	7.5	11530	7	5	6	8	Renewal Signoff SH
9	—	—	—	—	—	—	—	—	—	—	Old WQ: EML
Meter ID	22A	pH 11	pH 11	DO13	DO13	EC05					RT Stock Batch #: 27+28
Control	25.6	8.40	8.19	8.3	7.4	350	10	10	10	9	Date: 9/7/09
0.75	25.6	8.28	7.95	8.4	7.4	1775	9	10	10	10	Test Solution Prep [illegible]
1.5	25.6	8.21	7.84	8.4	7.4	3160	8	10	10	10	New WQ: SH
3	25.6	8.15	7.77	8.5	7.4	6000	8	7	10	10	Renewal Time: 1015
6	25.6	8.05	7.72	8.9	7.4	11550	7	4	5	7	Renewal Signoff: [illegible]
9	—	—	—	—	—	—	—	—	—	—	Old WQ: SL
Meter ID	22A	pH 14	pH 14	DO13	DO13	EC05					RT Stock Batch #: 28
Control	25.3		8.20		7.4	351	10	10	10	9	Date: 9/8/09
0.75	25.3		8.12		7.5	1859	9	10	10	10	Termination Time: 0930
1.5	25.3		8.03		7.7	3260	8	10	10	10	Termination Signoff: [illegible]
3	25.3		7.98		7.8	6140	8	7	9	10	Old WQ: DBD
6	25.3		7.93		7.7	11750	2	3	5	6	
9	—		—		—	—	—	—	—	—	
Meter ID	22A		pH 11		DO12	EC03					

Fathead Minnow Dry Weight Data Sheet

Client: **Reference Toxicant** Test ID #: 36010 Project # 15170

Sample: Sodium Chloride Tare Weight Date: 9/4/09 Sign-off: KR

Test Date: 9/1/09 Final Weight Date: 9/9/09 Sign-off: JCD

Pan ID	Concentration	Replicate	Initial Pan Weight (mg)	Final Pan Weight (mg)	Initial # of Organisms	Biomass Value (mg)
1	Control	A	158.76	162.52	10	~~3.56~~ 0.376
2		B	160.06	163.76	10	~~3.7~~ 0.370
3		C	154.39	157.75	10	~~3.36~~ 0.336
4		D	157.05	160.58	10	~~3.53~~ 0.353
5	0.75	A	162.71	166.35	10	~~3.64~~ 0.364
6		B	162.81	165.75	10	~~2.94~~ 0.294
7		C	154.73	157.80	10	~~3.07~~ 0.307
8		D	172.13	175.54	10	~~3.41~~ 0.341
9	1.5	A	187.42	191.96	10	~~3.54~~ 0.354
10		B	160.21	163.85	10	~~3.64~~ 0.364
11		C	154.33	157.76	10	~~3.43~~ 0.343
12		D	155.49	159.49	10	~~4.00~~ 0.400
13	3	A	162.80	165.35	10	~~2.55~~ 0.255
14		B	173.08	175.89	10	~~2.81~~ 0.281
15		C	144.29	147.27	10	~~2.98~~ 0.298
16		D	173.72	176.91	10	~~3.19~~ 0.319
17	6	A	183.61	185.86	10	~~2.25~~ 0.225
18		B	168.66	169.78	10	~~1.12~~ 0.112
19		C	186.97	188.78	10	~~1.81~~ 0.181
20		D	175.19	177.00	10	~~1.81~~ 0.181
21	9	A	188.58	—	—	0
22		B	169.51	—	—	0
23		C	175.68	—	—	0
24		D	167.43	—	—	0
QA1			171.43	171.59		—
QA2			175.35	175.34		—
QA3			180.44	180.44		—
Balance ID:			1	1		