APPENDIX G

Concentrate Toxicity Testing

TECHNICAL MEMORANDUM

Bay Area Regional Desalination Project
Pilot Study

Subject:	**Brine Toxicity Testing Results**		
Prepared by:	BEB/JAJ/DDH; Applied Marine Sciences, Inc.	**Reference:**	1481449
Reviewed by:	JAJ-AMS; COB-MWH	**Date:**	November 2009

The Bay Area's four largest water agencies, the Contra Costa Water District (CCWD), the East Bay Municipal Utility District (EBMUD), the San Francisco Public Utilities Commission (SFPUC), and the Santa Clara Valley Water District (SCVWD), are jointly exploring a regional desalination project that could provide the region an additional water source, diversify the area's water supply, and foster long-term regional sustainability. The Bay Area Regional Desalination project (RDP) could consist of one or more desalination facilities, with an ultimate total capacity of up to 65 million gallons per day.

The RDP tested the operation and maintenance of a joint desalination facility on a pilot scale. The Pilot Plant Study (PPS) was located at CCWD's Mallard Slough Pumping Plant site near Pittsburg, CA, adjacent to the San Francisco Bay Estuary in Suisun Bay. The PPS operated between October 2008 and March 2009, in order to capture both wet and dry season conditions. Water from Mallard Slough underwent microfiltration pretreatment and then reverse osmosis (RO) treatment to produce potable water at the PPS. The PPS then blended the permeate with the backwash and brine/reject streams for subsequent discharge into the CCWD water treatment facility.

One of the major potential issues associated with RO operations in the Delta is the discharge of the RO brine, backwash, and reject streams back into the Delta. The potential toxic effects of brine on local organisms involve both increased concentrations of ions (e.g., salinity or total dissolved solids) as well as more concentrated contaminants from the source water (e.g., pesticides or heavy metals). Identifying whether any toxicity effects of the brine are due to salinity or contaminants is necessary to determine the operational solutions needed to minimize them.

This Technical Memorandum describes the results of tests that were conducted to: (1) assess the potential toxicity of brine produced by the PPS desalination plant, and (2) differentiate toxicity sources (i.e., salinity or contaminants).

1.0 Brine Toxicity Testing Approach

The desalination process concentrates both salts and contaminants in the brine effluent, which are potentially toxic to aquatic organisms living in the Delta. Thus, brine toxicity at the PPS location is susceptible to two potential influences that affect the source water: (1) seawater

salinity, due to the PPS's proximity to Suisun Bay, and (2) the concentration of contaminants washed off the surrounding land by storm runoff and transported via Delta outflow.

Testing the PPS brine focused on evaluating its potential toxicity to several test organisms during extremes in salinity and contaminant input for the source water, which typically occur during the dry and wet seasons of the year. Dry season conditions represent the period of highest ambient salinities, whereas wet season conditions represent highest contaminant concentrations associated with storm runoff. Assessing the toxicity of brine produced during both extremes was intended to differentiate the separate effects.

As a QA measure, a positive control test (i.e., reference toxicant test) was conducted concurrently with the salinity and brine toxicity testing to assess the sensitivity of each test organism to toxic stress.

2.0 Objectives

The objectives of the brine toxicity testing were to:

- Sample brine from the PPS during a dry period (high salinity, low contaminant concentration) and a wet period (low salinity, high contaminant concentration).
- Assess algal growth toxicity with the diatom *Thalassiosira pseudonana*
- Assess survival and growth toxicity with the crustacean *Americamysis bahia*
- Assess survival and growth toxicity with the fish *Menidia beryllina*

3.0 Methods

3.1 Sampling Methods

Samples of brine were collected from the PPS facility on November 14, 2008 and February 25, 2009. The November sample was expected to be representative of "dry" conditions, with elevated salinity and reduced contaminants. It was collected during the high tide (estimated +4.8 ft at Mallard Island Ferry Wharf, Suisun Bay), when saltwater incursion within the Estuary, and salinity at the intake for the site, was expected to be maximized. The February sample was expected to be representative of "wet" conditions, with reduced salinity and elevated contaminants from freshwater runoff during recent storms. This sample was collected during the low tide (estimated +1.0 ft), when salinity at the intake for the site was expected to be minimized.

Brine water was collected from the PPS in plastic cubitainers and transported in coolers on ice to Pacific EcoRisk for toxicity testing. Samples were homogenized at the laboratory before analysis.

3.2 Analytical Methods

Pacific EcoRisk performed toxicity evaluations of the samples (Appendix A, B). These evaluations consisted of the following USEPA short-term chronic toxicity tests:

- Chronic growth test with the diatom *Thalassiosira pseudonana*
- Chronic survival and growth test with the crustacean *Americamysis bahia*

- Chronic survival and growth test with larvae of the fish *Menidia beryllina*.

These specific test organisms were selected because they represented a "best fit" of standard effluent test organisms approved by EPA for testing under 40 CFR Part 136, are representative of taxa that inhabit the San Francisco Bay-Delta region, and are known to be less tolerant of high salinity concentrations than other potential test organisms.

All tests involving concentration treatments involved diluting brine or a reference toxicant (KCl or $K_2Cr_2O_7$) in a Lab Control water (reverse-osmosis, deionized water). During dilutions, the brine and reference toxicant were adjusted to a test salinity of 25 ppt.

3.2.1 Growth Toxicity Testing with Thalassiosira pseudonana

The diatom *Thalassiosira pseudonana* was exposed to varied treatment concentrations of brine for 96 hours (2.5%, 5%, 10%, 25%, 50%, and 100%), and then the effects on cell growth (i.e., cell density) were assessed to identify any toxicity caused by the brine (Pacific EcoRisk, 2008, 2009). A reference toxicity test was performed to determine the sensitivity of the diatoms to toxic stress, by exposing *Thalassiosira* to varied concentrations of KCl (0.625, 1.25, 2.5, 5, and 10 g/L) for 96 hours. The data were analyzed to determine key dose-response point estimates, including No Effect Concentration (NOEC) and Inhibition Concentration 50% (IC_{50}) and 25% (IC_{25}).

3.2.2 Survival and Growth Toxicity Testing with Americamysis bahia

The crustacean *Americamysis bahia* was exposed to a series of brine dilutions for seven days, and fed brine shrimp nauplii twice daily. After the test, the effects on survival (number of live mysids) and growth ("biomass value" = dry weight of live mysids divided by initial number) were assessed to determine any impairments caused by the brine (Pacific EcoRisk, 2008, 2009). A reference toxicity test was performed to determine the sensitivity of the mysids to toxic stress, by exposing *Americamysis* to serial dilutions of a toxicant for seven days. The data were analyzed to determine key dose-response point estimates, including NOEC, IC_{50}, and IC_{25}, as well as Effect Concentration 50% (EC_{50}) and 25% (EC_{25}).

Between the two sample collections, the testing laboratory transitioned from using chromium to KCl as the toxicant in these tests, for safety reasons (Pacific EcoRisk, 2008, 2009). For the "dry" sample, Lab Control water was spiked with chromium (as $K_2Cr_2O_7$) at concentrations of 0.88, 1.75, 3.5, 7, and 14 mg/L. For the "wet" sample, KCl was used at concentrations of 0.125, 0.25, 0.5, 1, and 2 g/L. Using different toxicants in the reference tests has no effect on sample tests.

3.2.3 Survival and Growth Toxicity Testing with Menidia beryllina

Larvae of the fish *Menidia beryllina* were exposed to a series of brine dilutions for seven days, and fed brine shrimp nauplii twice daily. After the test, the effects on survival (number of live fish) and growth ("biomass value" = dry weight of live fish divided by initial number) were evaluated (Pacific EcoRisk, 2008, 2009). A reference toxicity test was performed to determine the sensitivity of the fish to toxic stress, by exposing *Menidia* to serial dilutions of KCl at concentrations of 0.5, 1.0, 1.25, 1.5, and 2 mg/L for seven days. The test response data were

analyzed to determine key dose-response point estimates, including NOEC, IC_{50}, IC_{25}, EC_{50}, and EC_{25}.

4.0 Results

4.1 Water Quality Characteristics of Brine Samples

The brine samples differed between seasons in several water quality characteristics (Table 4-1). Brine collected from the PPS during the "dry" season (November 2008) at high tide had a salinity of 17.7 ppt and conductivity of 29.35 mS/cm, whereas the sample collected during the "wet" season (February 2009) at low tide had lower values of 7.0 ppt and 12.42 mS/cm, respectively (Pacific EcoRisk, 2008, 2009). Although not directly relevant to this study, the "dry" season sample had a higher dissolved oxygen content (7.3 mg/L) than the "wet" season sample (5.5 mg/L). pH and total ammonia concentrations in both samples were the same (i.e., <1.0 mg/L N).

Table 4-1. Collection information and water quality characteristics for brine samples collected during "dry" (11/14/08) and "wet" (2/25/09) periods from the PPS desalination plant (Pacific EcoRisk, 2008, 2009).

Season[1]	Target Salinity	Target Contam.	Date	Time	Tidal Height[2]	pH	DO (mg/L)	Salinity (ppt)	Cond. (mS/cm)	Total Ammonia (mg/L N)
"Dry"	High	Low	11/14/08	1444-1451	High (+4.8 ft)	7.55	7.3	17.7	29.35	<1.0
"Wet"	Low	High	2/25/09	0915-0935	Low (+1.0 ft)	7.55	5.5	7.0	12.42	<1.0

[1] Representative, based on precipitation and river discharge

[2] Estimated at Mallard Island Ferry Wharf, Suisun Bay (38° 02.6' N, 121° 55.1' W)

4.2 Toxicity Test Results

Copies of the bioassay test result reports are contained in Appendices A and B for the November 2008 and February 2009 PPS brine discharge sample events, respectively.

4.2.1 Growth Toxicity on Thalassiosira pseudonana

There were no significant reductions in algal growth in the desalination brine for either the "dry" or "wet" sample (Table 4-2) (Pacific EcoRisk, 2008, 2009). NOEC estimates were 100% brine, and IC_{50} and IC_{25} estimates were >100% brine.

4.2.2 Survival and Growth Toxicity on Americamysis bahia

There were no significant reductions in invertebrate survival or growth in the desalination brine for either the "dry" or "wet" sample (Table 4-2) (Pacific EcoRisk, 2008, 2009). NOEC, IC_{50}, IC_{25}, EC_{50}, and EC_{25} estimates were all >100% brine.

4.2.3 Survival and Growth Toxicity on Menidia beryllina

There were no significant reductions in fish survival or growth in the desalination brine for either the "dry" or "wet" sample (Table 4-2) (Pacific EcoRisk, 2008, 2009). NOEC, IC_{50}, IC_{25}, EC_{50}, and EC_{25} estimates were all >100% brine.

5.0 Discussion

5.1 Toxicity Test Results

No significant growth toxicity of the desalination brine was found for the algae, and no significant survival or growth toxicity was found for the invertebrate or fish test organisms for either the "dry" sample (salinity-dominant scenario) or the "wet" sample (contaminant-dominant scenario) (Table 4-2). Because neither salinity- nor contaminant-related toxicity was found, it was not possible to distinguish the relative effects of each. The toxicity results suggest that, if the source water used at the PPS desalination plant and the brine samples tested are representative of those at an operational desalination plant at the Mallard Slough location, then there would be no expected toxic effects of the effluent on biota.

5.2 Representative Source Water Extremes

One concern with these results is whether the brine samples were collected during the extremes in salinity and contaminant input at Mallard Slough, as intended. As discussed previously, the "dry" season sample was collected during expected conditions of low freshwater flow through the Delta and high tidal seawater incursion, when salinity would be maximized and contaminant input from precipitation-driven surface runoff would be minimal (Table 4-1). The "wet" season sample was collected during expected conditions of high freshwater flow and low seawater influence, when salinity would be minimized and contaminant concentrations from surface runoff maximized. Measurements of the collected brine confirm the expected higher brine salinity and conductivity during "dry" conditions and lower values during "wet" conditions (Table 4-1).

Hydrographic conditions and water quality data measured near the PPS desalination plant from January 2008 to March 2009 also substantiate that the samples were collected during extremes in source water composition during the study period. Freshwater input to the region varied significantly throughout 2008 and early 2009, when the PPS was operating. Precipitation data for Concord Airport show that the 2008 dry season extended from March through mid October (Figure 5-1a). In early November, a storm delivered two inches of rain to the region, but for nearly two weeks before the "dry" sampling event on November 14, 2008, and during the sampling event itself, conditions were dry (National Weather Service, 2009). A series of storms in late November of 2008 through February of 2009 delivered 8.3 inches of rain, with nearly 6

inches of that falling during the three weeks immediately before the "wet" sampling event on February 25, 2009.

The effects of precipitation runoff during the PPS operation can be seen in records related to contaminant input near Mallard Island. Sacramento River discharge at Rio Vista (USGS water quality station "SRV") (California Data Exchange Center, 2009) varied sinusoidally around a

Table 4-2. Summary of toxicity testing results for brine samples collected from the PPS desalination plant during "dry" (11/14/08) and "wet" (2/25/09) periods, as well as results for reference toxicant tests (Pacific EcoRisk, 2008, 2009).

Test	Test	Test	Algae (*Thalassiosira pseudonana*)		Invertebrate (*Americamysis bahia*)		Fish (*Menidia beryllina*)	
Parameter	**Treatment**	**Statistic**	**"Dry"**	**"Wet"**	**"Dry"**	**"Wet"**	**"Dry"**	**"Wet"**
Survival	Brine	NOEC	-	-	>100% brine	>100% brine	>100% brine	>100% brine
Survival	Brine	EC_{25}	-	-	>100% brine	>100% brine	>100% brine	>100% brine
Survival	Brine	EC_{50}	-	-	>100% brine	>100% brine	>100% brine	>100% brine
Survival	Ref Tox	EC_{50}	-	-	6.1 mg/L Cr	0.60 g/L KCl	1.2 g/L KCl	1.2 g/L KCl
Growth	Brine	NOEC	100% brine	100% brine	>100% brine	>100% brine	>100% brine	>100% brine
Growth	Brine	IC_{25}	>100% brine	>100% brine	>100% brine	>100% brine	>100% brine	>100% brine
Growth	Brine	IC_{50}	>100% brine	>100% brine	>100% brine	>100% brine	>100% brine	>100% brine
Growth	Ref Tox	IC_{50}	1.72 g/L KCl	4.25 g/L KCl	4.6 mg/L Cr	0.64 g/L KCl	1.2 g/L KCl	1.3 g/L KCl

median value of approximately 11,800 cfs from March through October of 2008 (Figure 5-1b). Discharge during the "dry" sample collection was on the lower end of this variation, at 440 cfs. In contrast, discharge peaked at over 91,000 cfs on the day the "wet" sample was collected, which indicates a significant increase in the fresh water flow (and presumably, contaminant input) from the Delta to Mallard Slough. Water turbidity in the Sacramento River at Mallard Island (California Department of Water Resources station "MAL") (California Data Exchange Center, 2009), which we use here as a proxy for suspended sediment and contaminant input to the Sacramento River, also increased between the "dry" and "wet" sample collections, from 14.8 to 38.1 ntu (Figure 5-1c).

The effect of freshwater runoff on source water salinity is also apparent in electrical conductivity records from Sacramento River at Mallard Island (California Data Exchange Center, 2009). Conductivity increased throughout the 2008 dry season, but decreased between the "dry" and "wet" events, from 11.6 to 0.5 mS/cm (Figure 5-1d). Conductivity of the feed water tank for the PPS desalination plant closely tracked that of the nearby MAL station (Figure 5-1d). The conductivity of the feed water on 11/14/08 is unknown, due to an instrument error (Stefani Harrison, Pers. Comm., 2009), but based on values immediately before and after that date, the value was probably about 12 mS/cm. This value dropped to 4.2 mS/cm on the 2/25/09 event.

The logistics of collecting a field sample under target weather and hydrographic conditions is difficult, and much more so when coordinating with a complex operation such as the PPS desalination plant. Yet, the brine samples appear to have been collected during conditions that represent the seasonal extremes of source water quality for Mallard Slough reasonably well. Although the "dry" brine sample ideally would have been collected several weeks earlier (i.e., before the first significant storm in early November of 2008), unexpected start-up delays prevented sampling earlier in the year. However, the "wet" sample was collected during peak freshwater runoff of the 2008-2009 winter storm season.

5.3 Representative Water Year

Another concern with the results of the toxicity tests is that they may not be representative of conditions during other years. California is in its third year of drought. The samples were collected when the Sacramento River was in a Moderate to Severe Drought, based on 8 Station Index (8SI) percentiles for the current water year (October 1, 2008 to September 30, 2009) (California Department of Water Resources, 2009). It is possible that if the source water for the desalination plant had higher salinity and/or contaminant concentration than those tested, then the desalination process could potentially concentrate the salts and contaminants enough to produce brine with toxic effects on biota. However, because the survival and growth NOEC values for all tests were $\geq$ 100% brine, it is unclear how much the salinity or contaminant concentration would need to increase in the brine to see a toxic effect on the test organisms.

6.0 Conclusions

Toxicity tests of desalination brine samples collected during the "dry" season (salinity-dominant scenario) and "wet" season (contaminant-dominant scenario) from the PPS in Mallard Slough

showed no significant effects on the survival or growth of algal (*Thalassiosira pseudonana*), invertebrate (*Americamysis bahia*), or fish (*Menidia beryllina*) test organisms.

Assuming that brine samples tested in this study are representative of those produced by an operational desalination plant at Mallard Slough, there would be no expected toxic effects of the brine on biota were the brine to be discharged into the Delta.

7.0 References

California Data Exchange Center, 2009. http://cdec.water.ca.gov/.

California Department of Water Resources, April 6, 2009. Sacramento River Drought Status. http://www.cnrfc.noaa.gov/products/Drought_8si_product.pdf.

National Weather Service, 2009. Observed Weather Reports. http://www.weather.gov/climate/index.php?wfo=mtr.

Pacific EcoRisk, December, 2008. A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent, Sample Collected November 14, 2008. 63 pp.

Pacific EcoRisk, March, 2009. Supplemental Report: A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent, Sample Collected February 25, 2009.

Stefani Harrison, Pers. Comm., 2009. Electrical conductivity data from raw water tank at PPS desalination plant.

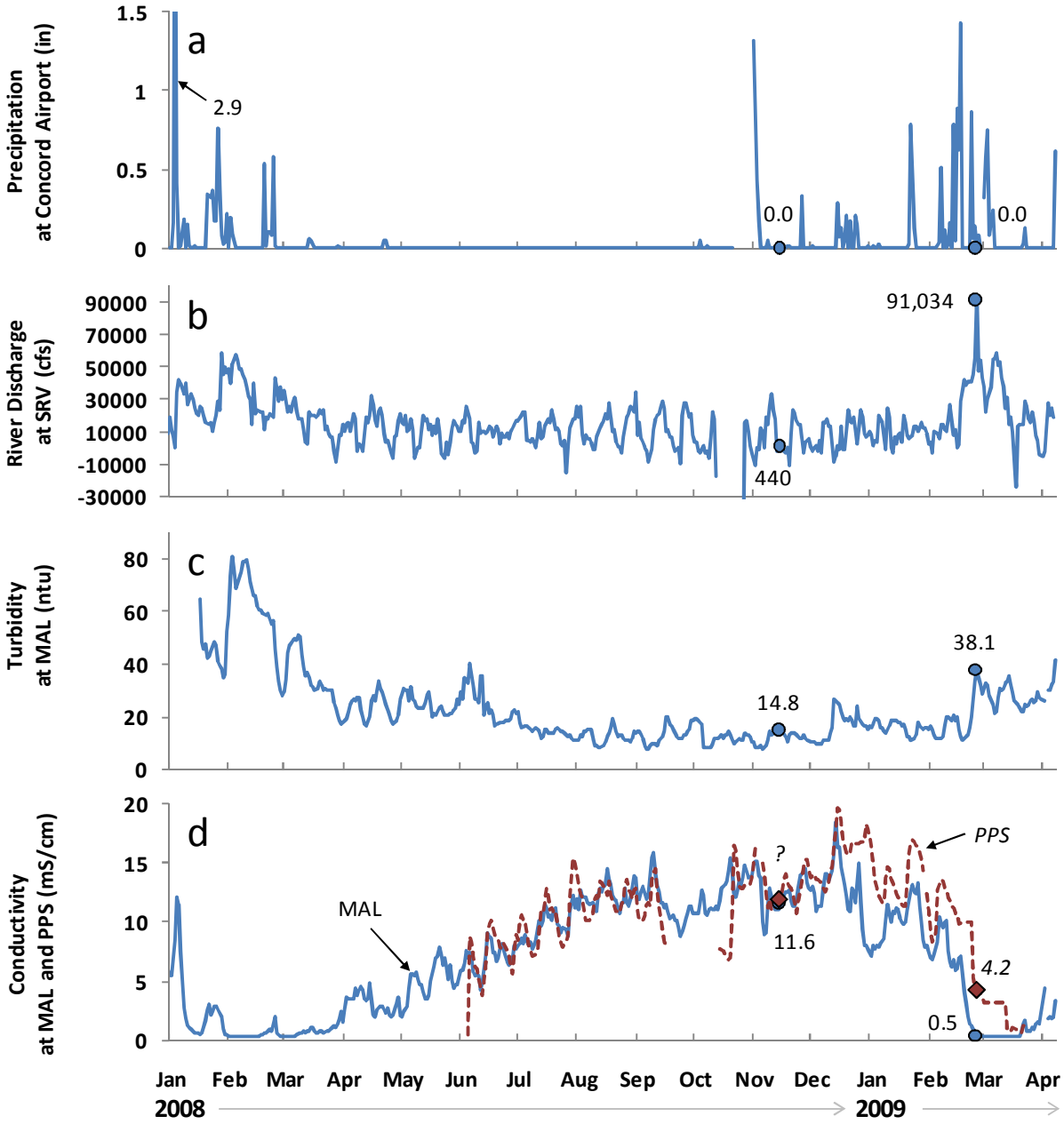


Figure 5-1. Measured precipitation (a), hydrographic (b), and water quality (c, d) parameters near the Pilot Plant Study (PPS) site. River discharge, water turbidity, and water electrical conductivity are plotted as median daily values. SRV = Sacramento River at Rio Vista station (USGS); MAL = Sacramento River at Mallard Slough station (California DWR). Symbols with labels indicate parameter values during brine sampling events on 11/14/08 ("dry") and 2/25/09 ("wet"). The dashed line in plot d indicates measurements within the feed water tank of the desalination plant. The "?" label for 11/14/08 feed water at PPS denotes an instrument error (symbol position interpolated from surrounding values).

8.0 Appendix A

See separate attachment for detailed toxicity testing results for the "dry" season brine sample, collected on November 14, 2008 (AppendixA_111408Results.pdf) (Pacific EcoRisk, December, 2008).

Jay Johnson
Applied Marine Sciences
4749 Bennett Dr., Suite L
Livermore, CA 94550

December 16, 2007

Dear Jay:

I have enclosed two copies of our report "A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent" for the sample collected November 14, 2008. The results of this testing are summarized below:

Chronic Effects of the Desalination Effluent on *Thalassiosira pseudonana*
There were ***no*** significant reductions in *Thalassiosira pseudonana* growth.

Chronic Effects of the Desalination Effluent on *Americamysis bahia*
There were ***no*** significant reductions in *Americamysis bahia* survival or growth.

Chronic Effects of the Desalination Effluent on *Menidia beryllina*
There were ***no*** significant reductions in *Menidia beryllina* survival or growth.

If you have any questions regarding the performance and interpretation of these tests, please give me a call at (707) 207-7760.

Sincerely,

R. Scott Ogle, Ph.D.
Principal & Special Projects Director

This testing was performed under Lab Order 14150. The test results reported herein conform to the most current NELAC standards, where applicable, unless otherwise narrated in the body of the report, and only relate to the sample(s) tested. This report shall not be reproduced, except in full, without the written consent of Pacific EcoRisk.

A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent

Sample collected November 14, 2008

Prepared For:

Applied Marine Sciences
4749 Bennett Dr., Suite L
Livermore, CA 94550

Prepared By:

Pacific EcoRisk, Inc.
2250 Cordelia Rd.
Fairfield, CA 94534

December 2008

PACIFIC ECORISK
ENVIRONMENTAL CONSULTING & TESTING

A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent

Sample collected November 14, 2008

Prepared For:

Applied Marine Sciences
4749 Bennett Dr., Suite L
Livermore, CA 94550

Prepared By:

Pacific EcoRisk, Inc.
2250 Cordelia Rd.
Fairfield, CA 94534

December 2008

A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent

Sample collected November 14, 2008

Table of Contents

Appendices

1. INTRODUCTION

Under contract to the Applied Marine Sciences, Pacific EcoRisk (PER) performed chronic toxicity evaluations of effluent sample collected from the Bay Area Regional Desalination Project. The toxicity evaluations consist of performing the following chronic toxicity tests:

- chronic (96-hr) growth test with the diatom *Thalassiosira pseudonana*;
- chronic survival and growth test with the crustacean *Americamysis bahia*; and
- chronic survival and growth test with larval *Menidia beryllina*.

These toxicity tests were performed on water the sample collected on November 14, 2008. This report describes the performance and results of these tests.

2. COLLECTION AND DELIVERY OF THE EFFLUENT SAMPLE

On November 14, an effluent sample was collected from the Bay Area Regional Desalination plant. The sample was transported, on ice and under chain-of-custody, to the PER laboratory facility in Fairfield. Upon receipt at the testing laboratory, aliquots of sample were collected for analysis of initial water quality characteristics (Table 1). The remainder of the water sample was stored at 0-6°C except when being used to prepare test solutions. The chain-of-custody record for the collection and delivery of this sample is provided in Appendix A.

Table 1. Initial water quality characteristics of the Bay Area Regional Desalination Project effluent.

Date Sample Collected	Date Sample Received	Sample ID	Temp (°C)	pH	D.O (mg/L)	Salinity	Conductivity (μS/cm)	Total Ammonia (mg/L N)
11/14/08	11/14/08	MWHA-Dry	11.7*	7.55	7.3	17.7	29350	<1.0

* Sample was delivered on the day of collection, and was transported at ≤6.0°C.

3. CHRONIC TOXICITY TEST PROCEDURES

The Bay Area Desalination Project effluent sample was tested for toxicity using the following US EPA short-term chronic toxicity tests:

- chronic (96-hr) growth test with the diatom *Thalassiosira pseudonana*;
- chronic survival and growth test with the crustacean *Americamysis bahia*; and
- chronic survival and growth test with larval *Menidia beryllina*.

The methods used in conducting these toxicity tests followed the guidelines established by the following manuals:

- "Standard Guide for Conducting Static 96-h Toxicity Tests with Microalgae" (ASTM E 1218-97a); and
- "Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Marine and Estuarine Organisms, Third Edition" (EPA-821-R-02-014).

3.1 Algal Growth Toxicity Testing with *Thalassiosira pseudonana*

The short-term chronic diatom toxicity test consists of exposing *Thalassiosira pseudonana* to the effluent for ~96-hrs, after which the effects on cell growth are evaluated. The specific procedures used in these tests are described below.

The Lab Control water for this test consisted of reverse osmosis, de-ionized (RO/DI) water adjusted up to the test salinity of 25 ppt using an artificial sea salt (Crystal Seas®-bioassay grade). For use in this test, an aliquot of the effluent was similarly adjusted to 25 ppt using the same sea salt. The Lab Control water and ambient waters were filtered (0.45 μm) and then spiked with nutrients, as per ASTM guidelines. The salinity-adjusted Lab Control water and effluent were used to prepare test solutions at test treatment concentrations of 2.5%, 5%, 10%, 25%, 50% and 100% effluent. Water quality characteristics were measured on these test solutions prior to use in this test.

There were 4 replicates at each test treatment, each replicate consisting of a 250-mL glass Erlenmeyer flask containing 100 mL of test solution; an additional replicate was established at each test treatment for the measurement of test solution water quality characteristics during the test and at test termination. Each treatment was inoculated to an initial diatom cell density of 20,000 cells/mL from a from a laboratory culture of *Thalassiosira* that is maintained in log growth phase. These flasks were loosely capped and randomly positioned within a temperature-controlled room at 20°C, under continuous illumination from cool-white fluorescent bulbs.

Each day, the temperature and pH were determined for the designated "water quality" replicate at each treatment; each replicate flask was gently shaken in the morning and randomly re-positioned within the temperature-controlled room.

After 96 ($\pm$2) hrs exposure, the cell density in each replicate flask was determined by microscopic analysis. The resulting cell density data were analyzed to determine any growth impairment, or toxicity, caused by the ambient water; all statistical analyses were performed using CETIS® statistical software (Tidepool Scientific, McKinleyville, CA).

3.1.1 Reference Toxicant Testing of the *Thalassiosira pseudonana*

In order to assess the sensitivity of the *Thalassiosira* 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 KCl at concentrations of 0.625, 1.25, 2.5, 5, and 10 gm/L. The resulting test response data were statistically analyzed to determine key

dose-response point estimates (e.g., IC_{50}); all statistical analyses were performed 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.

3.2 Survival and Growth Toxicity Testing with *Americamysis bahia*

The short-term chronic *Americamysis bahia* test consists of exposing the organisms to a series of effluent dilutions for 7 days, after which effects on survival and growth are evaluated. The specific procedures used in this test are described below.

The *Americamysis bahia* used in this test were obtained from Aquatic BioSystems (Ft. Collins, CO); upon receipt at the lab, the mysids were transferred into aerated tanks containing saltwater at 25 ppt, and were fed brine shrimp nauplii during the pre-test holding period.

The Lab Control/dilution water for this test was prepared by salting up reverse-osmosis, de-ionized water to a salinity of 25 ppt using a commercial artificial sea salt (Crystal Sea Salt®-bioassay grade). Each day, an aliquot of the final effluent sample was similarly adjusted to a salinity of 25 ppt using the same artificial sea salt. The salinity-adjusted Lab Control/dilution water and effluent sample were used to prepare daily test solutions at concentrations of 2.5%, 5%, 10%, 25%, 50% and 100% effluent. "New" water quality characteristics (pH, D.O., and salinity) were measured on these test solutions prior to use in the test.

There were 8 replicates at each test treatment, each replicate consisting of 200 mL of test solution in a 400-mL glass beaker. The test was initiated by randomly allocating five 7-day old mysids into each replicate beaker. The beakers were randomly positioned in a temperature-controlled room at 26°C (with temperature being monitored daily) under a 16L:8D photoperiod. The mysids were fed freshly-hatched brine shrimp nauplii twice daily.

Each day of the test, fresh test solutions were prepared and characterized as before. The test replicate beakers were examined, with any dead animals, uneaten food, wastes, and other detritus being removed. The number of live mysids in each replicate was determined and ~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 salinity) were measured on the old test water that had been discarded from one randomly-selected replicate at each treatment.

After 7 days exposure, the test was terminated and the number of live mysids in each replicate beaker was recorded. The mysids 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. The mysids were then dried at 100°C for >24 hrs and re-weighed to determine the total weight of mysids in each replicate; the total weight was divided by the initial number of mysids per replicate (n=5) 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 CETIS® statistical software.

3.2.1 Reference Toxicant Testing of the *Americamysis bahia*

In order to assess the sensitivity of the mysid 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 media spiked with chromium (as $K_2Cr_2O_7$) at concentrations of 0.88, 1.75, 3.5, 7, and 14 mg/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 ± 3 SD of the point estimates generated by the 20 most recent previous reference toxicant tests performed by this lab.

3.3 Survival and Growth Toxicity Testing with *Menidia beryllina*

The short-term chronic *Menidia beryllina* test consists of exposing larval fish to a series of effluent dilutions for 7 days, after which effects on survival and growth are evaluated. The specific procedures used in this test are described below.

The larval fish used in this bioassay were obtained from a commercial supplier (Aquatic Biosystems, Fort Collins, CO). These fish were maintained at 25°C in aerated aquaria containing Lab Control water (described below) prior to their use in this test. During this pre-test period, the fish were fed brine shrimp nauplii *ad libitum*.

The Lab Control/dilution water for this bioassay was prepared by salting up reverse-osmosis, de-ionized water to a salinity of 25 ppt using a commercial artificial sea salt (Crystal Sea® -bioassay grade). Each day, an aliquot of the final effluent sample was similarly adjusted to a salinity of 25 ppt using the same artificial sea salt. The salinity-adjusted Lab Control/dilution water and effluent sample were used to prepare daily test solutions at concentrations of 2.5%, 5%, 10%, 25%, 50% and 100% effluent. "New" water quality characteristics (pH, D.O., and salinity) were measured on these test solutions prior to use in the test.

There were 4 replicates for the Lab Control and each effluent treatment, each replicate consisting of 400 mL of test media in a 600-mL glass beaker. This test was initiated by randomly allocating 10 nine-day old *Menidia beryllina* into each replicate. These 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 twice daily.

Each day of the test, fresh test solutions were prepared and characterized as before. The replicate beakers containing the larval fish were examined, 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 media. "Old" water quality characteristics (pH, D.O., and conductivity) were measured on the old test water collected from one randomly selected replicate at each treatment.

After 7 days exposure, the number of live fish in each replicate beaker was recorded. Then, the fish from each replicate were carefully euthanized in methanol, rinsed in de-ionized water, and transferred to a pre-dried and pre-tared weighing pan. These 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 "biomass value" data were analyzed to determine key dose-response point estimates (e.g., EC_{50}); all statistical analyses were performed using the CETIS® statistical software.

3.3.1 Reference Toxicant Testing of the *Menidia beryllina*

In order to assess the sensitivity of the fish test organisms to toxic stress, a reference toxicant test was performed concurrently with the effluent test. This reference toxicant test was performed similarly to the effluent toxicity test, except that test solutions consisted of Lab Control (25 ppt water) spiked with KCl at concentrations of 0.5, 1.0, 1.25, 1.5, and 2 gm/L. After 7 days exposure, the survival and weight data were evaluated as in the effluent test. 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.

4. RESULTS

4.1 Effects of the Desalination Effluent on *Thalassiosira pseudonana*

The results of this test are summarized below in Table 2. There were ***no*** significant reductions in algal growth in the Desalination effluent; the growth NOEC was 100% effluent. The IC_{25} and IC_{50} were both >100% effluent, resulting in <1.0 survival TUc (where survival TUc = $100/IC_{25}$ or $100/IC_{50}$) for both test endpoints..

The test data and the summary of statistical analyses for these tests are presented in Appendix B.

Table 2. Effect of the Desalination effluent on *Thalassiosira pseudonana*.

Effluent Treatment	Mean Diatom Cell Density (cells/mL x 10^6)
Lab Water Control	1.91
2.5%	2.28
5%	2.27
10%	2.51
25%	2.63
50%	3.02
100%	3.18
Summary of Key Statistics	
Growth NOEC =	100% effluent
TUc (TUc = 100/NOEC) =	1.0
Growth IC_{25} =	>100% effluent
TUc (TUc = $100/IC_{25}$) =	<1.0
Growth IC_{50} =	>100% effluent
TUc (TUc = $100/IC_{50}$) =	<1.0

4.1.1 Reference Toxicant Toxicity to *Thalassiosira pseudonana*

The results of this test are presented in Table 3. There was a mean of 833,000 cells/mL at the Lab Control treatment. The growth IC_{50} was 1.72 gm/L KCl.

The IC_{50} of 1.72 gm/L KCl is just outside of the lower threshold of 1.80 gm/L KCl of the "typical response" range established by the mean ± 2 SD from the 20 most recent previously performed reference toxicant tests; this indicates that these test organisms may have been slightly more sensitive to toxicant stress than is typical. The U.S. 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. Moreover, as there was no impairment of algal growth in the effluent, the observation of algal cells that may be more sensitive than is typical does not affect the interpretation of the effluent test results.

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

Table 3. Reference toxicant testing: effects of KCl on *Thalassiosira pseudonana*.

KCl Treatment (gm/L)	Mean Diatom Cell Density (cells/mL x 10^6)
Lab Water Control	0.83
0.625	0.66
1.25	**0.51***
2.5	**0.26***
5	**0.00***
10	**0.00***
Summary of Key Statistic	
IC_{50} =	1.72 gm/L KCl

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

4.2 Effects of the Desalination Effluent on *Americamysis bahia*

The results of this test are presented in Table 4. There was a mean of 97.5% survival at the Lab Control treatment. There were ***no*** significant reductions in survival in the Desalination effluent; the survival NOEC was 100% effluent. Due to the absence of significant mortalities, the EC_{25} and EC_{50} could not be calculated, but can both be assumed >100% effluent, resulting in <1.0 survival TUc (where TUc = $100/EC_{25}$ or $100/EC_{50}$) for both test endpoints.

The mean 'biomass value' was 0.31 mg at the Lab Control treatment. There were ***no*** significant reductions in growth in the Desalination effluent; the growth NOEC was 100% effluent. The IC_{25} and IC_{50} were both >100% effluent, resulting in <1.0 growth TUc (where TUc = $100/IC_{25}$ or $100/IC_{50}$) for both test endpoints.

The test data and summary of statistical analyses for this test are attached as Appendix D.

Table 4. Effects of the Desalination effluent on *Americamysis bahia*.

Effluent Treatment	Mean % Survival	Mean "Biomass Value" (mg)
Lab Control	97.5	0.31
2.5%	100	0.35
5%	100	0.34
10%	97.5	0.35
25%	100	0.34
50%	97.5	0.34
100%	95	0.36
Summary of Key Statistics		
NOEC	>100% effluent	>100% effluent
TUc (TUc = 100/NOEC) =	<1.0	<1.0
Survival EC_{25} or Growth IC_{25}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{25}$ or $100/IC_{25}$) =	<1.0	<1.0
Survival EC_{50} or Growth IC_{50}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{50}$ or $100/IC_{50}$) =	<1.0	<1.0

4.2.1 Reference Toxicant Toxicity to *Americamysis bahia*

The results of this test are presented in Table 5. There was a mean of 92.5% survival and a mean mysid biomass of 0.31 mg at the Lab Control treatment. The survival EC_{50} was 6.1 mg/L, and the growth IC_{50} was 4.6 mg/L.

The results of the concurrent reference toxicant test were consistent with 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 attached as Appendix E.

Table 5. Reference toxicant testing: effects of chromium on *Americamysis bahia*.

Chromium Treatment (mg/L)	Mean % Survival	Mean Biomass Value (mg)
Lab Control	92.5	0.31
0.88	92.5	0.31
1.75	95	**0.25***
3.5	91.4	**0.21***
7	**26.7***	0.04
14	**0***	0
Summary of Key Statistics		
Survival EC_{50} or Growth IC_{50} =	6.1 mg/L Cr	4.6 mg/L Cr

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

4.3 Effects of the Desalination Effluent on *Menidia beryllina*

The results of this test are presented in Table 6. There was a mean of 97.5% survival at the Lab Control treatment. There were ***no*** significant reductions in survival in the Desalination effluent; the survival NOEC was 100% effluent. Due to the absence of significant mortalities, the EC_{25} and EC_{50} could not be calculated, but can both be assumed >100% effluent, resulting in <1.0 survival TUc (where TUc = $100/EC_{25}$ or $100/EC_{50}$) for both test endpoints.

The mean fish biomass value was 1.29 mg at the Lab Control treatment. There were ***no*** significant reductions in growth in the Bay Area Regional Desalination effluent; the growth NOEC was 100% effluent. The IC_{25} and IC_{50} were both >100% effluent, resulting in <1.0 growth TUc (where TUc = $100/IC_{25}$ or $100/IC_{50}$) for both test endpoints.

The test data and the summary of statistical analyses for this test are attached as Appendix F.

Table 6. Effects the Desalination effluent on *Menidia beryllina*.

Effluent Treatment	Mean % Survival	Mean Biomass Value (mg)
Lab Control	97.5	1.29
2.5%	97.5	1.23
5%	95	1.24
10%	100	1.40
25%	100	1.23
50%	95	1.32
100%	100	1.44
Summary of Key Statistics		
NOEC	>100% effluent	>100% effluent
TUc (TUc = 100/NOEC) =	<1.0	<1.0
Survival EC_{25} or Growth IC_{25}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{25}$ or $100/IC_{25}$) =	<1.0	<1.0
Survival EC_{50} or Growth IC_{50}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{50}$ or $100/IC_{50}$) =	<1.0	<1.0

4.3.1 Reference Toxicant Toxicity to *Menidia beryllina*

The results of this test are summarized below in Table 7. There was a mean of 97.5% survival and a mean fish biomass value of 1.35 mg at the Lab Control treatment; the survival EC_{50} value was 1.2 gm/L KCl, and the growth IC_{50} was 1.2 gm/L KCl.

These reference toxicant test responses were consistent with previous performance of this test in our lab, indicating that these organisms were responding to toxic stress in a typical fashion.

The test data and summary of statistical analyses for this test are attached as Appendix G.

Table 7. Reference toxicant testing: effects of KCl on *Menidia beryllina*.

KCl Treatment (gm/L)	Mean % Survival	Mean Biomass Value (mg)
Lab Control	97.5	1.35
0.5	95	1.24
1	**77.5***	1.10
1.25	**40***	0.59
1.5	**7.5***	0.06
2	**0***	0.0
Summary of Key Statistics		
Survival EC_{50} or Growth IC_{50} =	1.2 gm/L KCl	1.2 gm/L KCl

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

5. SUMMARY AND CONCLUSIONS

Chronic Effects of the Desalination Effluent on *Thalassiosira pseudonana*
There were no significant reductions in algal growth in the effluent.

Chronic Effects of the Desalination Effluent on *Americamysis bahia*
There were no significant reductions in invertebrate survival or growth in the effluent.

Chronic Effects of the Desalination Effluent on *Menidia beryllina*
There were no significant reductions in fish survival or growth in the effluent.

6. AQUATIC TOXICITY DATA QUALITY CONTROL

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

Negative Control – The test organism responses at the Lab Control treatments were within acceptable limits.

Positive Control – The results for the *Americamysis bahia* and *Menidia beryllina* reference toxicant test were consistent with the reference toxicant test database, indicating that these test organisms were responding to toxic stress in a typical fashion. The *Thalassiosira pseudonana* cells were slightly more sensitive to toxic stress than is typical; however, as there was no impairment of algal growth in the effluent, the observation of algal cells that may be more sensitive than is typical does not affect the interpretation of the effluent test results.

Appendix A

Chain-of-Custody Record for the Collection and Delivery of the Bay Area Regional Desalination Project Effluent Sample

4749 Bennett Dr., Ste L
Livermore, CA 94551
925-373-7142

APPLIED amarine SCIENCES

Chain of Custody Record

White: Lab Copy
Yellow: AMS Copy

Project ID: MWHA Regional Desal	Client Contact:
PO #: 2279	Address: .
Results to: ☒ AMS ☐ Client	
AMS Contact: Jay Johnson	Phone:
Email: johnson@amarine.com	Email:

Sample ID	Site Name	Date	Time	Analyses: Most sens. species: Thalassiosira pseudonana (x3)	Mysid sens. species: Americamysis bahia (x3)	Most sens. species: Menidia beryllina (x3)	Contaminant toxicity	Reference toxicity	Salinity toxicity	Comments
MWHA-Dry		11/14/08	1442	X	X	X	X	X	X	60L water total (6 jugs)

bottles numbe 1 to 4

Relinquished by:	Date: 11/14/08	Time: 15:39	Shipping: ☒ Delivered by AMS
Received by:	Date: 11/14/08	Time: 1537	☐ Picked up by lab
# coolers:			☐ Fedex / Other Tracking# _____

Appendix B

Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of the Bay Area Regional Desalination Project Effluent to *Thalassiosira pseudonana*

CETIS Summary Report

Phytoplankton Growth Inhibition Test — **Pacific EcoRisk**

Test Run No:	02-1743-4374	**Test Type:**	Cell Growth	**Analyst:**	Jason Walker
Start Date:	15 Nov-08 11:20	**Protocol:**	EPA/821/R/02/014 (2002)	**Diluent:**	Laboratory Water
Ending Date:	19 Nov-08 15:00	**Species:**	Thalassiosira pseudonana	**Brine:**	Crystal Sea
Duration:	4d 4h	**Source:**	In-House Culture	**Age:**	5

Sample No:	11-2799-6109	**Code:**	EFF	**Client:**	AMS
Sample Date:	14 Nov-08 14:42	**Material:**	Effluent	**Project:**	14150
Receive Date:	14 Nov-08 15:39	**Source:**	Applied Marine Sciences		
Sample Age:	21h (11.7 °C)	**Station:**	MWHA		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
03-7220-8470	Cell Density	100	>100	N/A	7.93%	1	Equal Variance t Two-Sample Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-%	95% LCL	95% UCL	TU	Method
06-5571-6402	Cell Density	IC2.5	>100	N/A	N/A	<1	Linear Interpolation (ICPIN)
		IC5	>100	N/A	N/A	<1	
		IC10	>100	N/A	N/A	<1	
		IC15	>100	N/A	N/A	<1	
		IC20	>100	N/A	N/A	<1	
		IC25	>100	N/A	N/A	<1	
		IC40	>100	N/A	N/A	<1	
		IC50	>100	N/A	N/A	<1	

Cell Density Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.91E+6	1.87E+6	1.94E+6	1.83E+6	2.04E+6	1.69E+4	9.26E+4	4.86%	0.0%
2.5		4	2.28E+6	2.21E+6	2.34E+6	2.09E+6	2.43E+6	3.08E+4	1.69E+5	7.41%	-19.6%
5		4	2.27E+6	2.24E+6	2.29E+6	2.18E+6	2.35E+6	1.32E+4	7.23E+4	3.19%	-18.9%
10		4	2.51E+6	2.46E+6	2.55E+6	2.32E+6	2.60E+6	2.37E+4	1.30E+5	5.18%	-31.5%
25		4	2.63E+6	2.58E+6	2.68E+6	2.46E+6	2.76E+6	2.35E+4	1.29E+5	4.9%	-38.1%
50		4	3.02E+6	2.98E+6	3.05E+6	2.86E+6	3.09E+6	1.95E+4	1.07E+5	3.54%	-58.3%
100		4	3.18E+6	3.13E+6	3.22E+6	3.04E+6	3.34E+6	2.28E+4	1.25E+5	3.93%	-66.8%

Cell Density Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.88E+6	2.04E+6	1.87E+6	1.83E+6
2.5		2.18E+6	2.41E+6	2.09E+6	2.43E+6
5		2.35E+6	2.18E+6	2.24E+6	2.29E+6
10		2.60E+6	2.59E+6	2.51E+6	2.32E+6
25		2.46E+6	2.69E+6	2.76E+6	2.61E+6
50		2.86E+6	3.09E+6	3.08E+6	3.03E+6
100		3.19E+6	3.04E+6	3.14E+6	3.34E+6

CETIS Analytical Report

Phytoplankton Growth Inhibition Test — Pacific EcoRisk

Analysis No:	03-7220-8470	Endpoint:	Cell Density	CETIS Version:	CETISv1.6.5
Analyzed:	30 Nov-08 12:22	Analysis:	Parametric-Two Sample	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	7.93%

Equal Variance t Two-Sample Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Control		2.5	-3.87	1.94	187000	0.9960	Non-Significant Effect
		5	-6.13	1.94	114000	1.0000	Non-Significant Effect
		10	-7.53	1.94	155000	1.0000	Non-Significant Effect
		25	-9.14	1.94	154000	1.0000	Non-Significant Effect
		50	-15.7	1.94	137000	1.0000	Non-Significant Effect
		100	-16.4	1.94	151000	1.0000	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	4.756436E+12	7.927393E+11	6	54	0.0000	Significant Effect
Error	3.0815E+11	14673810000	21			
Total	5.064585642E+12	8.0741312512E+11	27			

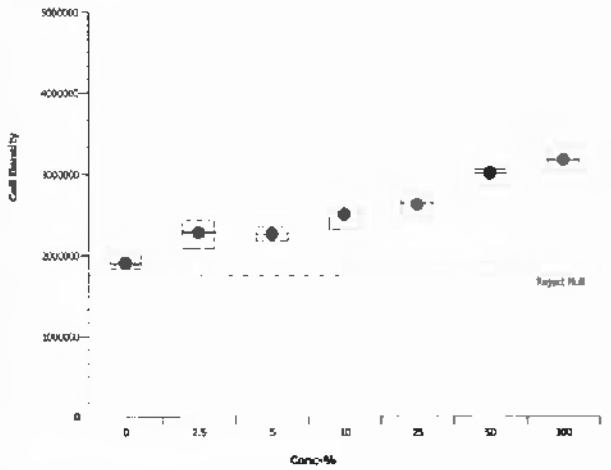
ANOVA Assumptions

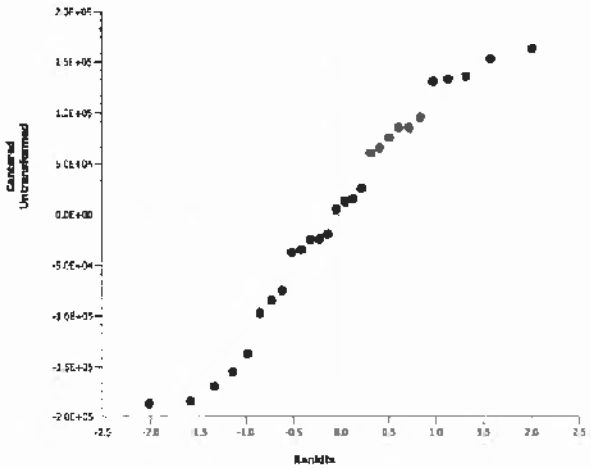
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	2.23	16.8	0.8970	Equal Variances
Distribution	Shapiro-Wilk Normality	0.95		0.2020	Normal Distribution

Cell Density Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.91E+6	1.87E+6	1.94E+6	1.83E+6	2.04E+6	1.72E+4	9.26E+4	4.86%	0.0%
2.5		4	2.28E+6	2.21E+6	2.34E+6	2.09E+6	2.43E+6	3.13E+4	1.69E+5	7.41%	-19.6%
5		4	2.27E+6	2.24E+6	2.29E+6	2.18E+6	2.35E+6	1.34E+4	7.23E+4	3.19%	-18.9%
10		4	2.51E+6	2.46E+6	2.55E+6	2.32E+6	2.60E+6	2.41E+4	1.30E+5	5.18%	-31.5%
25		4	2.63E+6	2.58E+6	2.68E+6	2.46E+6	2.76E+6	2.39E+4	1.29E+5	4.9%	-38.1%
50		4	3.02E+6	2.97E+6	3.06E+6	2.86E+6	3.09E+6	1.98E+4	1.07E+5	3.54%	-58.3%
100		4	3.18E+6	3.13E+6	3.23E+6	3.04E+6	3.34E+6	2.32E+4	1.25E+5	3.93%	-66.8%

Graphics





CETIS Analytical Report

Phytoplankton Growth Inhibition Test — Pacific EcoRisk

Analysis No:	06-5571-6402	**Endpoint:**	Cell Density	**CETIS Version:**	CETISv1.6.5
Analyzed:	30 Nov-08 12:23	**Analysis:**	Linear Interpolation (ICPIN)	**Official Results:**	Yes

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	5334240	280	Yes	Two-Point Interpolation

Point Estimates

Level	Conc-%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC2.5	>100	N/A	N/A	<1	N/A	N/A
IC5	>100	N/A	N/A	<1	N/A	N/A
IC10	>100	N/A	N/A	<1	N/A	N/A
IC15	>100	N/A	N/A	<1	N/A	N/A
IC20	>100	N/A	N/A	<1	N/A	N/A
IC25	>100	N/A	N/A	<1	N/A	N/A
IC40	>100	N/A	N/A	<1	N/A	N/A
IC50	>100	N/A	N/A	<1	N/A	N/A

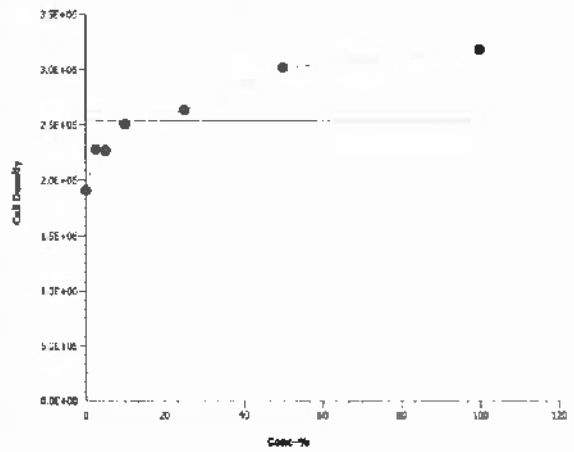
Cell Density Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.91E+6	1.83E+6	2.04E+6	1.69E+4	9.26E+4	4.86%	0.0%
2.5		4	2.28E+6	2.09E+6	2.43E+6	3.08E+4	1.69E+5	7.41%	-19.6%
5		4	2.27E+6	2.16E+6	2.35E+6	1.32E+4	7.23E+4	3.19%	-18.9%
10		4	2.51E+6	2.32E+6	2.60E+6	2.37E+4	1.30E+5	5.18%	-31.5%
25		4	2.63E+6	2.46E+6	2.76E+6	2.35E+4	1.29E+5	4.9%	-38.1%
50		4	3.02E+6	2.86E+6	3.09E+6	1.95E+4	1.07E+5	3.54%	-58.3%
100		4	3.18E+6	3.04E+6	3.34E+6	2.28E+4	1.25E+5	3.93%	-66.8%

Cell Density Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.88E+6	2.04E+6	1.87E+6	1.83E+6
2.5		2.18E+6	2.41E+6	2.09E+6	2.43E+6
5		2.35E+6	2.18E+6	2.24E+6	2.29E+6
10		2.60E+6	2.59E+6	2.51E+6	2.32E+6
25		2.46E+6	2.69E+6	2.75E+6	2.61E+6
50		2.86E+6	3.09E+6	3.08E+6	3.03E+6
100		3.19E+6	3.04E+6	3.14E+6	3.34E+6

Graphics



Thalassiosira pseudonana Cell Density Enumeration Data

Client: **Applied Marine Sciences** Initial Count: 20,000 cells/mL

Test Material: ~~effluent~~ MWHA - Dry Enumerating Scientist: SM

Test Start Date: 11/15/08 Start Time: 11:20 Project #: 31045

Test End Date: 11/19/08 End Time: 1500 Test ID #: 14150

Treatment %	Rep A	Rep B	Rep C	Rep D	Mean
Lab Water Control	1.88	2.04	1.87	1.83	1.90
2.5	2.18	2.41	2.09	2.43	2.28
5	2.35	2.18	2.24	2.29	2.26
10	2.60	2.59	2.51	2.32	2.50
25	~~2.59~~246	2.69	2.76	2.61	2.63
50	2.86	3.09	3.08	3.03	3.02
100	3.19	3.04	3.14	3.34	3.18

This datasheet has been reviewed for completeness and consistency with Test Acceptability Criteria and/or other issues of concern.	**Control Mean Density (cells/mL x 10^6)**	**Date:**	**Time:**	**Signoff:**
	1.90	11-19-08	1800	~~XS~~

Thalassiosira pseudonana Toxicity Test Water Quality Data

Client: Applied Marine Sciences Test ID #: ~~14158~~ 31045 Test Date: 11/15/08

Test Material: ~~Effluent~~ MWHA-Dry Project #: ~~31045~~ 14158 Control/Diluent: Algal Medium

Treatment %	Temp (°C)	pH	D.O. (mg/L)	Salinity (ppt)	Sign-Off
Lab Water Control	20.0	7.96	7.8	25.0	Test Solution Prep: JR
2.5	20.0	8.14	7.7	25.1	New WQ: MEC
5	20.0	8.15	7.7	25.1	Inoculation Date: 11/15/08
10	20.0	8.09	7.6	25.2	Inoculation Time: 1120
25	20.0	8.01	7.6	25.2	Inoculation Signoff: JR
50	20.0	7.93	7.7	25.2	
100	20.0	7.83	7.7	25.9	
Meter ID:	40	pH03	D014	EcO4	
Lab Water Control	20.0	8.02			Date: 11/16/08
2.5	20.0	7.96			WQ Time: 1220
5	20.0	7.93			WQ Signoff: SL
10	20.0	7.94			
25	20.0	7.96			
50	20.0	7.98			
100	20.0	8.03			
Meter ID:	40	ph11			
Lab Water Control	20.3	8.28			Date: 11/17/08
2.5	20.3	8.39			WQ Time: 1540
5	20.3	8.38			WQ Signoff: JNC
10	20.3	8.41			
25	20.3	8.42			
50	20.3	8.38			
100	20.3	8.31			
Meter ID:	40	pH11			
Lab Water Control	21.1	9.18			Date: 11/18/08
2.5	21.1	~~AR 9.3~~ 9.24			WQ Time: 1645
5	21.1	9.31			WQ Signoff: AR
10	21.1	9.36			
25	21.1	9.47			
50	21.1	9.38			
100	21.1	9.30			
Meter ID:	40	ph12			
Lab Water Control	21.0	9.08	9.9	25.2	Date: 11/19/08
2.5	21.0	9.15	10.1	25.2	Termination Time: SL 9:00 1500
5	21.0	9.17	10.2	25.3	Termination Signoff: SM
10	21.0	9.22	10.5	25.3	WQ Time: 9:00
25	21.0	9.29	9.6	25.3	WQ Signoff: SL
50	21.0	9.33	9.9	25.3	
100	21.0	9.36	11.1	25.4	
Meter ID:	40	ph 11	D010	EC01	

Initial Test Conditions			Light Intensity (lux)
			~~4345~~ 6347

Appendix C

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Thalassiosira pseudonana*

CETIS Summary Report

Phytoplankton Growth Inhibition Test — Pacific EcoRisk

Test Run No:	02-8530-0394	**Test Type:**	Cell Growth	**Analyst:**	Rivian Villanueva
Start Date:	15 Nov-08 15:00	**Protocol:**	ASTM E 1218-97a (Algae)	**Diluent:**	Laboratory Water
Ending Date:	19 Nov-08 16:00	**Species:**	Thalassiosira pseudonana	**Brine:**	Crystal Sea
Duration:	4d 1h	**Source:**	In-House Culture	**Age:**	5

Sample No:	15-7757-6135	**Code:**	KCl	**Client:**	Reference Toxicant
Sample Date:	15 Nov-08 15:00	**Material:**	Potassium chloride	**Project:**	14151
Receive Date:	15 Nov-08 15:00	**Source:**	Reference Toxicant		
Sample Age:	N/A (20 °C)	**Station:**	In House		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
14-8507-6429	Cell Density	0.625	1.25	0.884	31.5%		Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-g/L	95% LCL	95% UCL	TU	Method
00-3822-5947	Cell Density	IC2.5	0.0754	0.0166	1.03		Linear Interpolation (ICPIN)
		IC5	0.151	0.0372	1.05		
		IC10	0.302	0.0744	1.22		
		IC15	0.452	0.112	1.81		
		IC20	0.603	0.149	1.9		
		IC25	0.773	0.174	1.98		
		IC40	1.3	0.277	2.2		
		IC50	1.72	0.661	2.38		

Cell Density Summary

Conc-g/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	8.33E+5	7.92E+5	8.73E+5	7.10E+5	9.30E+5	2.00E+4	1.10E+5	13.2%	0.0%
0.625		4	6.60E+5	5.82E+5	7.38E+5	3.90E+5	9.00E+5	3.82E+4	2.09E+5	31.7%	20.7%
1.25		4	5.10E+5	4.29E+5	5.91E+5	3.40E+5	8.20E+5	3.94E+4	2.16E+5	42.3%	38.7%
2.5		4	2.60E+5	2.40E+5	2.80E+5	2.00E+5	3.20E+5	1.00E+4	5.48E+4	21.1%	68.8%
5		4	0.00E+0	0.00E+0	0.00E+0	0.00E+0	0.00E+0	0.00E+0	0.00E+0		100.0%
10		4	0.00E+0	0.00E+0	0.00E+0	0.00E+0	0.00E+0	0.00E+0	0.00E+0		100.0%

Cell Density Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	7.70E+5	7.10E+5	9.30E+5	9.20E+5
0.625		9.00E+5	6.90E+5	6.60E+5	3.90E+5
1.25		3.90E+5	3.40E+5	8.20E+5	4.90E+5
2.5		2.30E+5	2.90E+5	2.00E+5	3.20E+5
5		0.00E+0	0.00E+0	0.00E+0	0.00E+0
10		0.00E+0	0.00E+0	0.00E+0	0.00E+0

Thalassiosira pseudonana Reference Toxicant Test Cell Density Data

Client: **Reference Toxicant** Initial Count: 20,000 cells/mL

Test Material: Potassium chloride Enumerating Scientist: GM

Test Start Date: 11/5/08 Start Time: 15:00 Project #: 14151

Test End Date: 11/19/08 End Time: 1600 Test ID #: 31048

Treatment	Rep A	Rep B	Rep C	Rep D	Mean
Lab Water Control	0.77	0.71	0.93	0.92	0.83
0.625 g/L KCl	0.90	0.69	0.66	0.39	0.66
1.25 g/L KCl	0.39	0.34	0.82	0.49	0.51
2.5 g/L KCl	0.23	0.29	0.20	0.32	0.26
5 g/L KCl	0.0	0.0	0.0	0.0	0.0
10 g/L KCl	0.0	0.0	0.0	0.0	0.0

This datasheet has been reviewed for completeness and consistency with Test Acceptability Criteria and/or other issues of concern.	**Control Mean Density (cells/mL x 10^4)**	**Date:**	**Time:**	**Signoff:**
	0.83	11-19-08	1750	AB

Thalassiosira pseudonana Reference Toxicant Test Water Quality Data

Client: Reference Toxicant | Test ID #: 31048 | Test Date: JL 11/15/08

Test Material: Potassium chloride | Project #: 14151 | Control/Diluent: Algal Medium

Reference Toxicant Test Treatment (g/L KCl)	Temp (°C)	pH	D.O. (mg/L)	Salinity (ppt)	Sign-Off
Lab Water Control	20.0	7.96	7.3	25.7	Test Solution Prep: JL
0.625	20.0	8.18	7.4	25.7	New WQ: JL
1.25	20.0	8.21	6.9	26.4	Inoculation Date: 11/15/08
2.5	20.0	8.20	7.0	27.6	Inoculation Time: 1600
5	20.0	8.21	7.1	30.1	Inoculation Signoff: JL
10	20.0	8.20	7.1	34.7	
Meter ID:	40	PH12	DO12	ECO4	
Lab Water Control	20.0	8.07			Date: 11/16/08
0.625	20.0	8.04			WQ Time: 1230
1.25	20.0	8.03			WQ Signoff: SL
2.5	20.0	8.03			
5	20.0	8.01			
10	20.0	8.01			
Meter ID:	40	PH11			
Lab Water Control	20.3	7.93			Date: 11/17/08
0.625	20.3	7.96			WQ Time: 1540
1.25	20.3	7.95			WQ Signoff: JNC
2.5	20.3	7.94			
5	20.3	7.90			
10	20.3	7.88			
Meter ID:	40	PH11			
Lab Water Control	21.1	8.24			Date: 11/18/08
0.625	21.1	8.19			WQ Time: 1640
1.25	21.1	8.18			WQ Signoff: AR
2.5	21.1	8.13			
5	21.1	8.04			
10	21.1	8.01			
Meter ID:	40	PH12			
Lab Water Control	21.0	8.24	8.1	25.3	Date: 11/19/08
0.625	21.0	8.15	7.9	25.9	Termination Time: 1600
1.25	21.0	8.17	7.9	26.8	Termination Signoff: SM
2.5	21.0	8.10	7.6	27.9	WQ Time: 920
5	21.0	8.02	7.4	30.4	WQ Signoff: SL
10	21.0	7.97	6.5	34.8	
Meter ID:	40	ph03	DO10	ECO1	

Initial Test Conditions			Light Intensity (lux)
			~~6034.75~~ 6347.5

Appendix D

Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of the Bay Area Regional Desalination Project Effluent to *Americamysis bahia*

Chronic Mysid Survival, Growth and Fecundity Test — **Pacific EcoRisk**

Test Run No:	11-3743-9629	**Test Type:**	Growth-Survival-Fec (7d)	**Analyst:**	Jason Walker
Start Date:	15 Nov-08 15:50	**Protocol:**	EPA/821/R/02/014 (2002)	**Diluent:**	Laboratory Water
Ending Date:	22 Nov-08 08:00	**Species:**	Americamysis bahia	**Brine:**	Crystal Sea
Duration:	6d 16h	**Source:**	Aquatic Biosystems, CO	**Age:**	7

Sample No:	11-2799-6109	**Code:**	EFF	**Client:**	AMS
Sample Date:	14 Nov-08 14:42	**Material:**	Effluent	**Project:**	14150
Receive Date:	14 Nov-08 15:39	**Source:**	Applied Marine Sciences		
Sample Age:	25h (11.7 °C)	**Station:**	MWHA		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
18-5198-7028	7d Survival Rate	100	>100	N/A	8.64%	1	Steel Many-One Rank Test
04-7215-3166	Mean Dry Biomass-mg	100	>100	N/A	14.9%	1	Equal Variance t Two-Sample Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-%	95% LCL	95% UCL	TU	Method
00-9868-1767	Mean Dry Biomass-mg	IC2.5	>100	N/A	N/A	<1	Linear Interpolation (ICPIN)
		IC5	>100	N/A	N/A	<1	
		IC10	>100	N/A	N/A	<1	
		IC15	>100	N/A	N/A	<1	
		IC20	>100	N/A	N/A	<1	
		IC25	>100	N/A	N/A	<1	
		IC40	>100	N/A	N/A	<1	
		IC50	>100	N/A	N/A	<1	

7d Survival Rate Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	0.0%
2.5		8	1	1	1	1	1	0	0	0.0%	-2.56%
5		8	1	1	1	1	1	0	0	0.0%	-2.56%
10		8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	0.0%
25		8	1	1	1	1	1	0	0	0.0%	-2.56%
50		8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	0.0%
100		8	0.95	0.915	0.985	0.8	1	0.0169	0.0926	9.75%	2.56%

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.309	0.296	0.321	0.25	0.352	0.00592	0.0324	10.5%	0.0%
2.5		8	0.347	0.331	0.363	0.294	0.408	0.00759	0.0416	12.0%	-12.5%
5		8	0.34	0.326	0.354	0.274	0.388	0.00698	0.0382	11.2%	-10.2%
10		8	0.346	0.334	0.359	0.3	0.408	0.00595	0.0326	9.4%	-12.3%
25		8	0.34	0.325	0.355	0.266	0.398	0.0072	0.0394	11.6%	-10.1%
50		8	0.338	0.32	0.357	0.242	0.392	0.00909	0.0498	14.7%	-9.64%
100		8	0.355	0.33	0.38	0.222	0.438	0.0121	0.0665	18.7%	-15.1%

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

7d Survival Rate Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	1	1	1	1	0.8	1	1	1
2.5		1	1	1	1	1	1	1	1
5		1	1	1	1	1	1	1	1
10		0.8	1	1	1	1	1	1	1
25		1	1	1	1	1	1	1	1
50		0.8	1	1	1	1	1	1	1
100		0.8	1	1	0.8	1	1	1	1

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	0.3	0.302	0.288	0.352	0.25	0.324	0.344	0 308
2.5		0.308	0.31	0.368	0.394	0.344	0.35	0.408	0.294
5		0.33	0.38	0.388	0.34	0.37	0.312	0.326	0.274
10		0.342	0.408	0.314	0.348	0.34	0.36	0.3	0.36
25		0.266	0.33	0.376	0.32	0.344	0.332	0.352	0.398
50		0.242	0.382	0.298	0.366	0.318	0.354	0.354	0.392
100		0.222	0.374	0.34	0.438	0.392	0.31	0.404	0.36

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

Analysis No: 18-5198-7028 Endpoint: 7d Survival Rate CETIS Version: CETISv1.6.5
Analyzed: 30 Nov-08 12:37 Analysis: Nonparametric-Control vs Treatments Official Results: Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)		C > T	Not Run	100	>100	N/A	1	8.64%

Steel Many-One Rank Test

Control	vs	Conc-%	Test Stat	Critical	Ties	P-Value	Decision(5%)
Control		2.5	72	46	1	0.9430	Non-Significant Effect
		5	72	46	1	0.9430	Non-Significant Effect
		10	68	46	2	0.8570	Non-Significant Effect
		25	72	46	1	0.9430	Non-Significant Effect
		50	68	46	2	0.8570	Non-Significant Effect
		100	64	46	2	0.7130	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.02322722	0.003871203	6	0.809	0.5680	Non-Significant Effect
Error	0.2345659	0.004787059	49			
Total	0.25779308751226	0.00865826173685	55			

ANOVA Assumptions

Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Mod Levene Equality of Variance	0.809	3.19	0.5680	Equal Variances
Distribution	Shapiro-Wilk Normality	0.597		0.0000	Non-normal Distribution

7d Survival Rate Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.975	0.948	1	0.8	1	0.0131	0.0707	7.25%	0.0%
2.5		8	1	1	1	1	1	0	0	0.0%	-2.56%
5		8	1	1	1	1	1	0	0	0.0%	-2.56%
10		8	0.975	0.948	1	0.8	1	0.0131	0.0707	7.25%	0.0%
25		8	1	1	1	1	1	0	0	0.0%	-2.56%
50		8	0.975	0.948	1	0.8	1	0.0131	0.0707	7.25%	0.0%
100		8	0.95	0.915	0.985	0.8	1	0.0172	0.0926	9.75%	2.56%

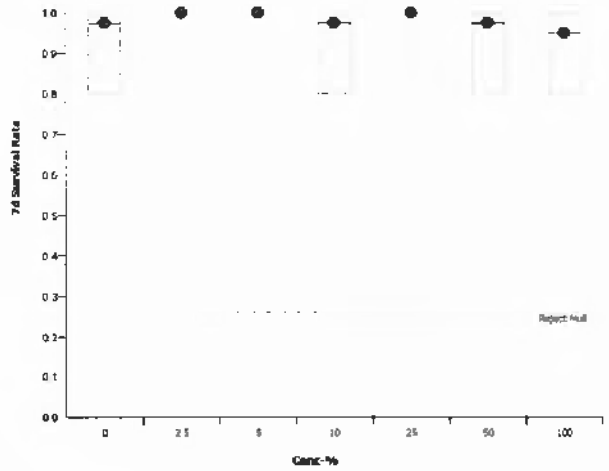
Angular (Corrected) Transformed Summary

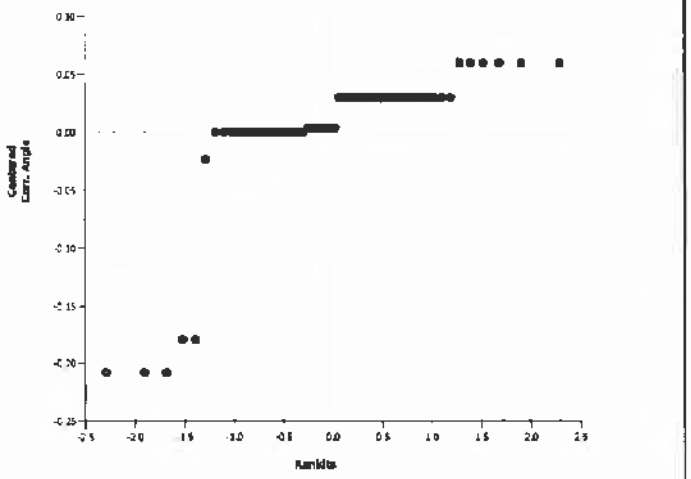
Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	1.32	1.28	1.35	1.11	1.35	0.0156	0.0842	6.4%	0.0%
2.5		8	1.35	1.35	1.35	1.35	1.35	0	0	0.0%	-2.26%
5		8	1.35	1.35	1.35	1.35	1.35	0	0	0.0%	-2.26%
10		8	1.32	1.28	1.35	1.11	1.35	0.0156	0.0842	6.4%	0.0%
25		8	1.34	1.34	1.35	1.32	1.35	0.00178	0.0096	0.72%	-2.0%
50		8	1.32	1.28	1.35	1.11	1.35	0.0156	0.0842	6.4%	0.0%
100		8	1.29	1.24	1.33	1.11	1.35	0.0205	0.11	8.57%	2.26%

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

Analysis No:	18-5198-7028	Endpoint:	7d Survival Rate	CETIS Version:	CETISv1.6.5
Analyzed:	30 Nov-08 12:37	Analysis:	Nonparametric-Control vs Treatments	Official Results:	Yes

Graphics





Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

Analysis No:	04-7215-3166	**Endpoint:**	Mean Dry Biomass-mg	**CETIS Version:**	CETISv1.6.5
Analyzed:	30 Nov-08 12:37	**Analysis:**	Parametric-Two Sample	**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	14.9%

Equal Variance t Two-Sample Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Control		2.5	-2.07	1.76	0.0328	0.9710	Non-Significant Effect
		5	-1.78	1.76	0.0312	0.9510	Non-Significant Effect
		10	-2.34	1.76	0.0286	0.9830	Non-Significant Effect
		25	-1.74	1.76	0.0316	0.9480	Non-Significant Effect
		50	-1.42	1.76	0.037	0.9110	Non-Significant Effect
		100	-1.78	1.76	0.0461	0.9510	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.01046493	0.001744155	6	0.887	0.5120	Non-Significant Effect
Error	0.09632742	0.001965865	49			
Total	0.10679234843701	0.00371002079919	55			

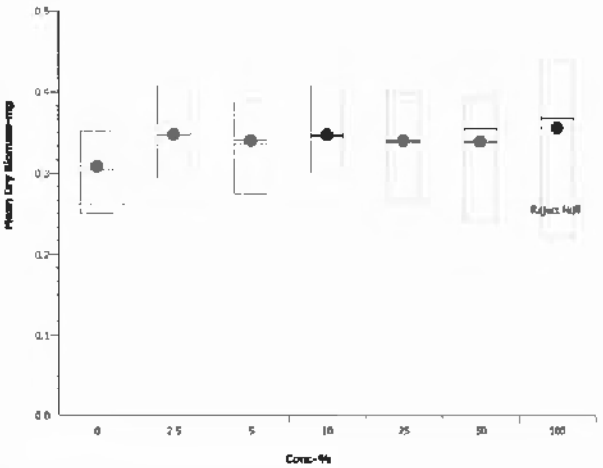
ANOVA Assumptions

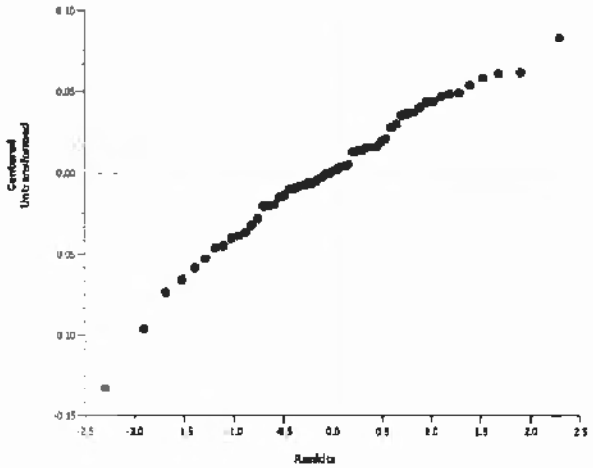
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	5.71	16.8	0.4560	Equal Variances
Distribution	Shapiro-Wilk Normality	0.973		0.2280	Normal Distribution

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.309	0.296	0.321	0.25	0.352	0.00602	0.0324	10.5%	0.0%
2.5		8	0.347	0.331	0.363	0.294	0.408	0.00772	0.0416	12.0%	-12.5%
5		8	0.34	0.325	0.355	0.274	0.388	0.0071	0.0382	11.2%	-10.2%
10		8	0.346	0.334	0.359	0.3	0.408	0.00605	0.0326	9.4%	-12.3%
25		8	0.34	0.325	0.355	0.266	0.398	0.00732	0.0394	11.6%	-10.1%
50		8	0.338	0.319	0.357	0.242	0.392	0.00925	0.0498	14.7%	-9.64%
100		8	0.355	0.33	0.38	0.222	0.438	0.0124	0.0665	18.7%	-15.1%

Graphics





Chronic Mysid Survival, Growth and Fecundity Test					Pacific EcoRisk
Analysis No:	00-9868-1767	**Endpoint:**	Mean Dry Biomass-mg	**CETIS Version:**	CETISv1.6.5
Analyzed:	30 Nov-08 12:37	**Analysis:**	Linear Interpolation (ICPIN)	**Official Results:**	Yes

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	2895625	280	Yes	Two-Point Interpolation

Point Estimates

Level	Conc-%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC2.5	>100	N/A	N/A	<1	N/A	N/A
IC5	>100	N/A	N/A	<1	N/A	N/A
IC10	>100	N/A	N/A	<1	N/A	N/A
IC15	>100	N/A	N/A	<1	N/A	N/A
IC20	>100	N/A	N/A	<1	N/A	N/A
IC25	>100	N/A	N/A	<1	N/A	N/A
IC40	>100	N/A	N/A	<1	N/A	N/A
IC50	>100	N/A	N/A	<1	N/A	N/A

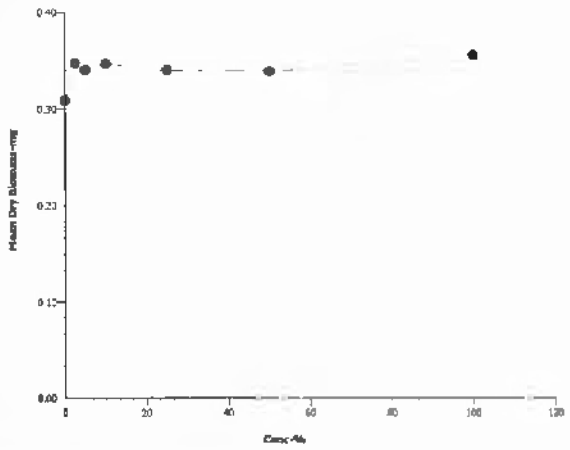
Mean Dry Biomass-mg Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.309	0.25	0.352	0.00592	0.0324	10.5%	0.0%
2.5		8	0.347	0.294	0.408	0.00759	0.0416	12.0%	-12.5%
5		8	0.34	0.274	0.388	0.00698	0.0382	11.2%	-10.2%
10		8	0.346	0.3	0.408	0.00595	0.0326	9.4%	-12.3%
25		8	0.34	0.266	0.398	0.0072	0.0394	11.6%	-10.1%
50		8	0.338	0.242	0.392	0.00909	0.0498	14.7%	-9.64%
100		8	0.355	0.222	0.438	0.0121	0.0665	18.7%	-15.1%

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	0.3	0.302	0.288	0.352	0.25	0.324	0.344	0.308
2.5		0.308	0.31	0.368	0.394	0.344	0.35	0.408	0.294
5		0.33	0.38	0.388	0.34	0.37	0.312	0.326	0.274
10		0.342	0.408	0.314	0.348	0.34	0.36	0.3	0.36
25		0.266	0.33	0.376	0.32	0.344	0.332	0.353	0.398
50		0.242	0.382	0.298	0.366	0.318	0.354	0.354	0.392
100		0.222	0.374	0.34	0.438	0.392	0.31	0.404	0.36

Graphics



7 Day Chronic *Americamysis bahia* Toxicity Test Test Data

Client: Applied Marine Sciences
Test Material: ~~effluent~~ AB mWHA-Dry
Test ID #: 31047 Project #: 14150
Test Date: 11-15-08

Organism Log#: 4255 Age: 7
Organism Supplier: ABS
Control/Diluent: DI & Crystal Sea @ 25 ppt
Control Water Batch: 696

Treatment (% effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	25.6	7.92		7.7		24.3		5	5	5	5	5	5	5	5	Date: 11.15.08
2.5	25.8	7.92		7.8		24.5		5	5	5	5	5	5	5	5	Sample ID: 20926
5	25.8	7.92		7.7		24.5		5	5	5	5	5	5	5	5	Test Solution Prep: EAK
10	25.8	7.91		7.7		24.5		5	5	5	5	5	5	5	5	New WQ: MEC
25	25.8	7.88		7.9		24.4		5	5	5	5	5	5	5	5	Initiation Time: 1550
50	25.8	7.85		8.0		24.2		5	5	5	5	5	5	5	5	Initiation Signed: RV
100	25.8	7.79		8.5		24.0		5	5	5	5	5	5	5	5	
Meter ID	11A	PH03		DO14		EC04										
Control	25.8	8.23	7.52	7.8	5.5	24.8	24.9	5	5	5	5	5	5	5	5	Date: 11/16/08
2.5	25.8	8.24	7.43	7.9	5.0	24.8	25.1	5	5	5	5	5	5	5	5	Sample ID: 20926
5	25.8	8.23	7.45	7.8	5.1	24.8	25.1	5	5	5	5	5	5	5	5	Test Solution Prep: EAK
10	25.8	8.19	7.46	7.8	4.8	24.7	25.1	5	5	5	5	5	5	5	5	New WQ: SL
25	25.8	8.09	7.59	8.1	4.9	24.6	25.0	5	5	5	5	5	5	5	5	Renewal Time: 1750
50	25.8	7.95	7.81	8.4	5.1	24.6	24.9	5	5	5	5	5	5	5	5	Renewal-Sign-off: SM
100	25.8	7.74	7.97	8.8	5.0	24.3	24.6	5	5	5	4	5	5	5	5	Old WQ: SM
Meter ID	11A	ph11	Ph11	DO12	DO12	EC04	EC04									
Control	25.7	8.05	7.70	7.8	5.8	24.8	25.3	5	5	5	5	5	5	5	5	Date: 11/17/08 1146
2.5	25.7	8.16	7.77	6.8	5.9	24.8	25.4	5	5	5	5	5	5	5	5	Sample ID: 20926
5	25.7	8.17	7.60	6.8	5.3	24.7	25.5	5	5	5	5	5	5	5	5	Test Solution Prep: PA
10	25.7	8.16	7.76	6.9	5.5	24.7	25.4	5	5	5	5	5	5	5	5	New WQ: AR
25	25.7	8.09	7.79	6.9	5.0	24.7	25.1	5	5	5	5	5	5	5	5	Renewal Time: 1640
50	25.7	7.96	7.90	7.4	5.2	24.6	25.2	5	5	5	5	5	5	5	5	Renewal-Sign-off: AB
100	25.7	7.79	8.04	7.2	5.3	24.1	25.5	5	5	5	4	5	5	5	5	Old WQ: JNC
Meter ID	11A	ph11	ph12	No10	DO14	EC04	EC05									
Control	25.8	8.23	7.70	7.0	5.4	24.6	25.4	5	5	5	5	5	5	5	5	Date: 11/18/08
2.5	25.8	8.22	7.61	6.9	4.7	24.9	25.3	5	5	5	5	5	5	5	5	Sample ID: 20926
5	25.8	8.20	7.65	7.0	5.4	24.9	25.6	5	5	5	5	5	5	5	5	Test Solution Prep: JL
10	25.8	8.16	7.63	7.0	5.1	24.8	25.5	5	5	5	5	5	5	5	5	New WQ: JNC
25	25.8	8.07	7.66	6.9	5.4	24.7	25.4	4	5	5	5	5	5	5	5	Renewal Time: 1615
50	25.9	7.90	7.75	7.6	5.2	24.5	25.4	4	5	5	5	5	5	5	5	Renewal-Sign-off: JL
100	25.8	7.66	7.94	8.5	5.2	24.3	25.5	4	5	5	4	5	5	5	5	Old WQ: JL
Meter ID	11A	pH11	PH11	DO10	DO10	EC04	EC04									

7 Day Chronic *Americamysis bahia* Toxicity Test Test Data

Client: Applied Marine Sciences
Test Material: ~~XS Effluent~~ MWHA-Dry
Test ID #: 31047 Project #: 14150
Test Date: 11-15-08

Organism Log#: 4258 Age: 7
Organism Supplier: ABS
Control/Diluent: DI & Crystal Sea @ 25 ppt
Control Water Batch: 696

Treatment (% effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	25.8	8.34	7.65	6.9	6.0	24.6	25.2	5	5	5	5	4	5	5	5	Date: 11/15/08
2.5	25.8	8.35	7.65	6.9	6.3	24.9	25.4	5	5	5	5	5	5	5	5	Sample ID: 20926
5	25.8	8.34	7.71	6.7	5.6	24.9	25.6	5	5	5	5	5	5	5	5	Test Solution Prep: JW
10	25.8	8.30	7.73	6.8	6.0	24.8	25.7	5	5	5	5	5	5	5	5	New WQ: JW
25	25.8	8.22	7.83	6.5	5.8	24.7	25.4	5	5	5	5	5	5	5	5	Renewal Time: 1510
50	25.8	8.10	7.89	7.0	6.3	24.6	25.5	4	5	5	5	5	5	5	5	Renewal-Sign-off: JPC
100	25.8	7.91	8.02	7.3	6.0	24.3	25.1	4	5	5	4	5	5	5	5	Old WQ: JW
Meter ID	11A	PH11	PH12	DO12	DO14	EC05	EC1									
Control	25.7	8.34	7.79	7.6	5.8	24.7	25.0	5	5	5	5	4	5	5	5	Date: 11/20/08
2.5	25.7	8.33	7.79	7.4	5.7	24.9	25.4	5	5	5	5	5	5	5	5	Sample ID: 20926
5	25.7	8.32	7.87	7.4	5.7	24.9	25.4	5	5	5	5	5	5	5	5	Test Solution Prep: JW
10	25.7	8.29	7.71	7.4	5.5	24.9	25.4	5	5	5	5	5	5	5	5	New WQ: OAP
25	25.7	8.17	7.90	7.6	5.5	24.9	25.3	5	5	5	5	5	5	5	5	Renewal Time: 1100
50	25.7	8.02	7.89	7.9	5.5	24.9	25.3	4	5	5	5	5	5	5	5	Renewal-Sign-off: JPC
100	25.7	7.80	8.05	8.3	5.3	24.7	25.2	4	5	5	4	5	5	5	5	Old WQ: MDM
Meter ID	11A	PH12	PH03	DO14	DO10	EC01	EC04									
Control	25.7	8.07	8.09	8.8	5.6	25.1	25.1	5	5	5	5	4	5	5	5	Date: 11-21-08
2.5	25.7	8.09	8.15	8.9	5.9	25.2	25.5	5	5	5	5	5	5	5	5	Sample ID: 20926
5	25.7	8.09	8.14	8.8	5.5	25.2	25.3	5	5	5	5	5	5	5	5	Test Solution Prep: JW
10	25.7	8.08	8.18	9.0	5.6	25.1	25.4	4	5	5	5	5	5	5	5	New WQ: MATM
25	25.7	8.07	8.24	9.0	5.5	24.6	25.3	5	5	5	5	5	5	4	5	Renewal Time: 1725
50	25.7	8.01	8.34	9.3	5.4	24.1	25.1	4	5	5	5	5	5	5	5	Renewal-Sign-off: KO
100	25.7	7.96	8.49	9.5	5.7	23.7	25.2	4	5	5	4	5	5	5	5	Old WQ: DMP
Meter ID	11A	PH11	PH03	DO14	DO14	EC04	EC									
Control	25.4		7.84		6.6		25.6	5	5	5	5	4	5	5	5	Date: 11-22-08
2.5	25.6		7.84		6.6		25.7	5	5	5	5	5	5	5	5	Termination-Time: 0800
5	25.6		7.82		6.5		25.6	5	5	5	5	5	5	5	5	Termination-Signoff: EKC
10	25.6		7.84		6.2		25.4	4	5	5	5	5	5	5	5	Old WQ: DMP
25	25.6		7.89		6.1		25.4	5	5	5	5	5	5	4	5	
50	25.6		8.01		6.2		25.1	4	5	5	5	5	5	5	5	
100	25.6		8.09		6.0		24.4	4	5	5	4	5	5	5	5	
Meter ID	11A		PH03		DO14		EC04									

Americamysis bahia Dry Weight and Biomass Value Data

Client: **Applied Marine Sciences** Test ID #: 31047 Project # 14150

Sample: MWHA-Dry Tare Weight Date: 11-17-08 Sign-off: MC

Test Date: 11-15-08 Final Weight Date: 11-24-08 Sign-off: MC

Pan ID	Concentration (%)	Replicate	Initial Pan Weight (mg)	Pan + Dry Mysid Weight (mg)	Initial # of Organisms	Mysid Biomass Value (mg)
1	Control	A	22.88	24.38	5	0.300
2		B	28.32	29.83	5	0.302
3		C	22.45	2389	5	0.288
4		D	26.87	28.63	5	0.352
5		E	23.56	24.81	5	0.250
6		F	2582	27.44	5	0.324
7		G	25.16	26.88	5	0.344
8		H	26.32	27.86	5	0.308
9	2.5	A	30.50	32.04	5	0.308
10		B	24.52	26.07	5	0.310
11		C	28.48	30.32	5	0.368
12		D	24.01	25.98	5	0.394
13		E	25.27	26.99	5	0.344
14		F	28.71	30.46	5	0.350
15		G	25.49	27.53	5	0.408
16		H	28.01	29.48	5	0.294
17	5	A	28.31	29.96	5	0.330
18		B	22.66	24.56	5	0.380
19		C	26.47	28.41	5	0.388
20		D	27.26	28.96	5	0.340
21		E	31.80	33.65	5	0.370
22		F	27.65	29.21	5	0.312
23		G	24.84	26.47	5	0.326
24		H	24.82	26.19	5	0.274
25	10	A	27.90	29.61	5	0.342
26		B	25.53	27.57	5	0.408
27		C	23.33	24.90	5	0.314
28		D	31.10	32.84	5	0.348
29		E	24.62	26.32	5	0.346
30		F	26.52	28.23	5	0.360
31		G	26.09	27.59	5	0.300
32		H	26.52	28.32	5	0.360

Americamysis bahia Dry Weight and Biomass Value Data

Client: **Applied Marine Sciences** Test ID #: 31047 Project # 14150

Sample: MWHA - DRY Tare Weight Date: 11-17-08 Sign-off: MC

Test Date: 11-15-08 Final Weight Date: 11-24-08 Sign-off: MC

Pan ID	Concentration (%)	Replicate	Initial Pan Weight (mg)	Pan + Dry Mysid Weight (mg)	Initial # of Organisms	Mysid Biomass Value (mg)
33	25	A	22.03	23.36	5	0.266
34		B	27.49	29.14	5	0.330
35		C	27.25	29.13	5	0.376
36		D	22.23	23.83	5	0.320
37		E	26.12	27.84	5	0.344
38		F	25.58	27.24	5	0.332
39		G	34.99	36.40	~~5~~ 4	0.3525
40		H	27.83	29.82	5	0.398
41	50	A	26.57	27.78	5	0.242
42		B	28.07	29.98	5	0.382
43		C	31.76	33.25	5	0.298
44		D	24.43	26.26	5	0.366
45		E	30.44	32.03	5	0.318
46		F	24.92	26.69	5	0.354
47		G	25.90	27.67	5	0.354
48		H	26.80	28.76	5	0.392
49	100	A	25.83	26.94	5	0.222
50		B	25.57	27.44	5	0.3748
51		C	24.18	25.88	5	0.340
52		D	25.15	27.34	5	0.4385
53		E	28.08	30.04	5	0.3928
54		F	27.15	28.70	5	0.310
55		G	28.94	30.96	5	0.404
56		H	27.59	29.39	5	0.360
QA 1			27.86	27.85		-0.010
QA 2			24.61	24.62		0.010
QA 3			27.32	27.31		0.010
QA 4			29.60	29.60		0.000
QA 5			21.87	21.87		0.000
QA 6			25.83	25.80		-0.030

Appendix E

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Americamysis bahia*

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

Test Run No:	02-6444-3409	Test Type:	Growth-Survival-Fec (7d)	Analyst:	Lisa Nugent
Start Date:	15 Nov-08 16:45	Protocol:	EPA/821/R/02/014 (2002)	Diluent:	Laboratory Water
Ending Date:	22 Nov-08 08:40	Species:	Americamysis bahia	Brine:	Crystal Sea
Duration:	6d 16h	Source:	Aquatic Biosystems, CO	Age:	7

Sample No:	18-8681-4098	Code:	K2CrO4	Client:	Reference Toxicant
Sample Date:	15 Nov-08 16:45	Material:	Potassium dichromate	Project:	14153
Receive Date:	15 Nov-08 16:45	Source:	Reference Toxicant		
Sample Age:	N/A (25.8 °C)	Station:	In House		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	Method
16-3540-2606	7d Survival Rate	3.5	7	4.95	16.6%	Wilcoxon/Bonferroni Adj Test
06-9574-8000	Mean Dry Biomass-mg	0.88	1.75	1.24	21.8%	Bonferroni Adj t Test

Point Estimate Summary

Analysis No	Endpoint	Effect-%	Conc-mg/L	95% LCL	95% UCL	Method
08-6753-1982	7d Survival Rate	2.5	3.76	1E-10	5.19	Linear Regression (MLE)
		10	4.45	1E-10	5.66	
		15	4.72	1E-10	5.85	
		20	4.95	1E-10	6.01	
		25	5.16	1E-10	6.15	
		40	5.72	1E-10	6.56	
		50	6.09	1E-10	6.88	
04-1344-8866	Mean Dry Biomass-mg	2.5	0.979	0.186	1.2	Linear Interpolation (ICPIN)
		5	1.08	0.372	1.58	
		10	1.28	0.744	2.09	
		15	1.48	1.03	2.49	
		20	1.68	1.21	2.86	
		25	2.24	1.37	3.32	
		40	3.95	2.3	4.31	
		50	4.6	4.08	4.96	

7d Survival Rate Summary

Conc-mg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.925	0.886	0.964	0.8	1	0.0189	0.104	11.2%	0.0%
0.88		8	0.925	0.886	0.964	0.8	1	0.0189	0.104	11.2%	0.0%
1.75		7	0.943	0.906	0.979	0.8	1	0.0178	0.0976	10.4%	-1.93%
3.5		7	0.914	0.874	0.954	0.8	1	0.0195	0.107	11.7%	1.16%
7		6	0.267	0.19	0.344	0	0.6	0.0377	0.207	77.5%	71.2%
14		8	0	0	0	0	0	0	0		100.0%

Mean Dry Biomass-mg Summary

Conc-mg/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.312	0.295	0.328	0.244	0.37	0.0081	0.0444	14.2%	0.0%
0.88		8	0.314	0.295	0.332	0.232	0.382	0.00899	0.0492	15.7%	-0.6%
1.75		7	0.244	0.213	0.276	0.068	0.314	0.0153	0.0841	34.4%	21.6%
3.5		7	0.209	0.198	0.22	0.166	0.264	0.00549	0.0301	14.4%	32.9%
7		6	0.0407	0.0243	0.057	0	0.112	0.00799	0.0438	108.0%	87.0%
14		8	0	0	0	0	0	0	0		100.0%

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

7d Survival Rate Detail

Conc-mg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	1	1	1	0.8	1	0.8	0.8	1
0.88		1	1	1	0.8	0.8	1	1	0.8
1.75		1	1	0.8	1	1	0.8	1	
3.5		1	0.8	0.8	1	1	1	0.8	
7		0	0.2	0.6	0.4	0.2	0.2		
14		0	0	0	0	0	0	0	0

Mean Dry Biomass-mg Detail

Conc-mg/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	0.316	0.342	0.346	0.28	0.37	0.244	0.264	0.332
0.88		0.324	0.324	0.325	0.232	0.278	0.382	0.366	0.278
1.75		0.314	0.28	0.242	0.068	0.312	0.23	0.264	
3.5		0.214	0.208	0.166	0.22	0.202	0.264	0.19	
7		0	0.014	0.112	0.076	0.028	0.014		
14		0	0	0	0	0	0	0	0

7 Day Chronic *Americamysis bahia* Reference Toxicant Test Data

Client: **Reference Toxicant** Organism Log#: 4255

Test Material: Potassium dichromate Organism Supplier: ABS

Test ID #: 31050 Project #: 14153 Control/Diluent: DI & Crystal Sea @ 25ppt

Test Date: 11-15-08 Randomization: 8 6 10 Control Water Batch: 696

Treatment (mg/L Cr)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	25.8	7.94		7.7		24.3		5	5	5	5	5	5	5	5	Date: 11-15-08
0.88	25.8	7.89		7.8		24.5		5	5	5	5	5	5	5	5	Test Solution Prep: RV
1.75	25.8	7.83		7.6		24.6		5	5	5	5	5	5	5	5	New WQ: MEC
3.5	25.8	7.71		7.7		24.5		5	5	5	5	5	5	5	5	Initiation Time: 1645
7	25.9	7.49		7.7		24.6		5	5	5	5	5	5	5	5	Initiation Signoff: RV
14	25.8	7.11		7.7		24.6		5	5	5	5	5	5	5	5	
Meter ID	11A	pH03		DO14		EC04										
Control	25.8	8.36	7.62	7.7	7.2	25.0	25.3	5	5	5	4	5	5	5	5	Date: 11/16/08
0.88	25.8	8.32	7.69	7.3	6.5	25.2	25.2	5	5	5	5	5	5	5	5	Test Solution Prep: RV
1.75	25.8	8.28	7.68	7.3	6.8	25.1	25.1	5	5	4	5	5	5	5	5	New WQ: JNC
3.5	25.8	8.19	7.63	7.3	6.8	25.1	25.0	5	5	4	5	5	5	5	4	Renewal Time: 1515
7	25.8	7.98	7.63	7.3	6.3	25.2	25.1	5	5	5	5	5	4	5	5	Renewal-Sign-off: RV
14	25.8	7.55	7.57	7.8	6.3	25.3	25.0	4	3	4	2	3	3	4	4	Old WQ: SL
Meter ID	11A	pH12	ph12	DO10	DO10	EC01	EC05									
Control	25.6	7.95	7.91	8.1	5.9	24.9	25.4	5	5	5	4	5	5	5	5	Date: 11/17/08
0.88	25.6	8.09	7.87	7.5	5.9	25.0	25.5	5	5	5	5	5	5	5	5	Test Solution Prep: QD
1.75	25.6	8.09	7.92	7.5	6.0	25.6	25.6	5	5	4	5	5	5	5	5	New WQ: AR
3.5	25.6	8.03	7.89	7.5	6.0	25.6	25.3	5	4	4	5	5	5	5	4	Renewal Time: 1430
7	25.6	7.88	7.83	7.5	5.9	25.6	25.6	3	4	2	2	3	2	4	3	Renewal-Sign-off: QD
14	25.6	7.52	7.79	7.8	6.0	24.8	25.5	1	1	3	2	2	0	2	1	Old WQ: SL
Meter ID	11A	pH11	ph12	DO10	DO14	EC04	EC05									
Control	25.8	8.30	7.81	6.7	6.3	24.7	25.4	5	5	5	4	5	5	5	5	Date: 11/18/08
0.88	25.8	8.27	7.80	6.7	6.1	25.0	25.6	5	5	5	5	5	5	5	5	Test Solution Prep: JT
1.75	25.8	8.24	7.80	6.6	6.2	25.0	25.6	5	5	4	5	5	5	4	5	New WQ: JNC
3.5	25.8	8.15	7.76	6.6	6.0	25.0	25.3	5	4	4	4	5	5	5	4	Renewal Time: 1100
7	25.9	7.96	7.76	6.6	6.0	24.9	25.4	2	4	2	1	3	2	3	2	Renewal-Sign-off: SLR
14	25.8	7.46	7.74	6.6	6.1	25.0	25.6	0	0	0	0	0	-	0	1	Old WQ: MEC
Meter ID	11A	pH03	pH11	DO14	DO10	EC05	EC04									

7 Day Chronic *Americamysis bahia* Reference Toxicant Test Data

Client: **Reference Toxicant** Organism Log#: 4255

Test Material: Potassium dichromate Organism Supplier: ABS

Test ID #: 31050 Project #: 14153 Control/Diluent: DI & Crystal Sea @ 25ppt

Test Date: 11/15/08 Randomization: 8.6.10 Control Water Batch: 696

Treatment (mg/L Cr)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	25.8	8.33	7.73	7.6	7.8	24.8	26.4	5	5	5	4	5	4	5	5	Date: 11/19/08
0.88	25.9	8.35	7.74	7.4	6.6	25.0	26.7	5	5	5	4	4	5	4	5	Test Solution Prep: SLR
1.75	25.9	8.32	7.74	7.3	6.2	25.0	26.3	5	5	1	4	5	5	4	5	New WQ: AR
3.5	25.9	8.27	7.71	7.3	5.9	25.0	26.0	5	4	—	4	5	5	5	4	Renewal Time: 1430 1230 TH
7	25.8	8.09	7.72	7.3	6.0	24.9	26.1	2	—	—	1	3	2	2	1	Renewal-Sign-off: SLR
14	25.8	7.66	NM	7.2	NM	25.0	NM	—	—	—	—	—	—	—	0	Old WQ: AR
Meter ID	11A	pH03	pH12	DO16	DO14	Eco1	Eco1									
Control	25.8	8.32	7.73	6.9	6.3	24.6	26.3	5	5	5	4	5	4	4	5	Date: 11/20/08
0.88	25.8	8.13	7.62	7.0	4.7	25.4	26.7	5	5	5	4	4	5	5	4	Test Solution Prep: JT
1.75	25.8	8.09	7.63	7.0	4.6	25.4	26.7	5	5	1	4	5	5	4	5	New WQ: DAP
3.5	25.8	8.02	7.69	7.0	4.7	25.3	25.8	5	4	—	4	5	5	5	4	Renewal Time: 1430
7	25.8	7.86	7.71	7.0	4.9	25.1	26.1	2	—	—	1	3	2	1	1	Renewal-Sign-off: SLR
14	25.8	7.5	—	6.8	—	24.9	—	—	—	—	—	—	—	—	—	Old WQ: AMM
Meter ID	11A	pH12	pH03	DO14	DO16	EC01	EC01									
Control	25.8	8.13	8.25	8.1	5.9	25.2	26.5	5	5	5	4	5	4	4	5	Date: 11/21/08
0.88	25.8	8.08	8.20	7.7	5.7	25.4	26.6	5	5	5	4	4	5	5	4	Test Solution Prep: JT
1.75	25.8	8.03	8.21	7.5	5.7	25.4	26.3	5	5	1	5	5	5	4	5	New WQ: AMM
3.5	25.8	8.02	8.22	7.5	5.7	25.3	26.2	5	4	—	4	5	5	5	4	Renewal Time: 1415
7	25.8	7.73	8.27	7.6	5.6	25.4	25.9	0	—	—	1	3	2	1	1	Renewal-Sign-off: SLR
14	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Old WQ: AM
Meter ID	11A	pH11	pH03	DO14	DO10	EC04	EC05									
Control	25.7		7.82		6.5		25.9	5	5	5	4	5	4	4	5	Date: 11/22/08
0.88	25.7		7.83		6.2		26.4	5	5	4	4	4	5	5	4	Termination Time: 0940
1.75	25.7		7.83		6.7		26.1	5	5	1	4	5	5	JPE 5	5	Final Sign-off: JPE
3.5	25.7		7.82		6.2		26.1	5	4	—	4	5	5	5	4	Old WQ: Bm
7	25.7		7.89		6.5		26.3	—	—	—	1	3	2	1	1	
14	—		—		—		—	—	—	—	—	—	—	—	—	
Meter ID	11A		pH11		DO12		EC01									

Americamysis bahia Dry Weight and Biomass Value Data

Client: **Reference Toxicant Test** Test ID #: 31050 Project #: 14153

Sample ID: Potassium dichromate Tare Weight Date: 11-17-08 Sign-off: MC

Test Date: 11-15-08 Final Weight Date: 11-24-08 Sign-off: MC

Pan	Concentration	Replicate	Initial Weight (mg)	Final Weight (mg)	# Organisms	Biomass (mg)
1	Control	A	26.78	28.36	5	0.316
2		B	26.44	28.15	5	0.342
3		C	25.67	27.40	5	0.346
4		D	29.17	30.57	5	0.280
5		E	30.02	31.87	5	0.370
6		F	28.15	29.37	5	0.244
7		G	25.35	26.67	5	0.264
8		H	24.46	26.12	5	0.332
9	0.88 mg/L	A	24.99	26.61	5	0.324
10		B	~~28.64~~ 28.35	29.97	5	0.324
11		C	25.06	26.36	5	~~0.260~~ 0.325
12		D	24.69	25.85	5	0.232
13		E	25.43	26.82	5	0.278
14		F	26.64	28.55	5	0.382
15		G	27.57	29.40	5	0.366
16		H	24.59	25.98	5	0.278
17	1.75 mg/L	A	28.66	30.23	5	0.314
18		B	29.28	30.78	5	0.280
19		C	24.63	24.91	~~5~~ 1 0	~~0.140~~ —
20		D	24.51	25.72	5	0.242
21		E	27.29	27.63	5	0.068
22		F	30.66	32.22	5	0.312
23		G	24.81	25.96	5	0.230
24		H	25.58	26.90	5	0.264
25	3.5 mg/L	A	24.78	25.85	5	0.214
26		B	30.01	31.05	5	0.208
27		C	23.82	—	~~5~~ 0	—
28		D	29.00	29.83	5	0.166
29		E	25.03	26.13	5	0.220
30		F	26.43	27.44	5	0.202
31		G	26.32	27.64	5	0.264
32		H	24.20	~~25.18~~ 25.18	5	0.190

Americamysis bahia Dry Weight and Biomass Value Data

Client: **Reference Toxicant Test** Test ID #: 31050 Project #: 14153

Sample ID: Potassium dichromate Tare Weight Date: 11-17-08 Sign-off: MC

Test Date: 11-15-08 Final Weight Date: 11-24-08 Sign-off: MC

Pan	Concentration	Replicate	Initial Weight (mg)	Final Weight (mg)	# Organisms	Biomass (mg)
33	7 mg/L	A	24.18	–	5	~~0.080~~
34		B	24.05	–	~~5~~ 0	~~0.080~~ –
35		C	26.32	–	~~5~~ 0	~~0.080~~ –
36		D	28.93	29.00	5	0.014
37		E	26.81	27.37	5	0.112
38		F	26.62	27.00	5	0.076
39		G	25.40	25.54	5	0.028
40		H	23.37	23.44	5	0.014
41	14 mg/L	A	–	–	5	~~0.000~~ 0
42		B	–	–	5	0
43		C	–	–	5	0
44		D	–	–	5	0
45		E	–	–	5	0
46		F	–	–	5	0
47		G	–	–	5	0
48		H	–	–	5	0
Q1			24.56	24.55	–	-0.010
Q2			25.91	25.90	–	-0.010
Q3			27.68	27.68	–	0.000
Q4			25.00	24.99	–	-0.010
Balance ID			1	1		

Appendix F

Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of the Bay Area Regional Desalination Project Effluent to *Menidia beryllina*

Chronic Larval Fish Survival and Growth Test — **Pacific EcoRisk**

Test Run No:	19-1593-6539	**Test Type:**	Growth-Survival (7d)	**Analyst:**	Jason Walker
Start Date:	15 Nov-08 11:30	**Protocol:**	EPA/821/R/02/014 (2002)	**Diluent:**	Laboratory Water
Ending Date:	22 Nov-08 09:15	**Species:**	Menidia beryllina	**Brine:**	Crystal Sea
Duration:	6d 22h	**Source:**	Aquatic Biosystems, CO	**Age:**	9

Sample No:	11-2799-6109	**Code:**	EFF	**Client:**	AMS
Sample Date:	14 Nov-08 14:42	**Material:**	Effluent	**Project:**	14150
Receive Date:	14 Nov-08 15:39	**Source:**	Applied Marine Sciences		
Sample Age:	21h (11.7 °C)	**Station:**	MWHA		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
20-5578-9613	7d Survival Rate	100	>100	N/A	5.2%	1	Equal Variance t Two-Sample Test
17-6240-2220	Mean Dry Biomass-mg	100	>100	N/A	17.5%	1	Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-%	95% LCL	95% UCL	TU	Method
13-5969-4097	Mean Dry Biomass-mg	IC2.5	>100	N/A	N/A	<1	Linear Interpolation (ICPIN)
		IC5	>100	N/A	N/A	<1	
		IC10	>100	N/A	N/A	<1	
		IC15	>100	N/A	N/A	<1	
		IC20	>100	N/A	N/A	<1	
		IC25	>100	N/A	N/A	<1	
		IC40	>100	N/A	N/A	<1	
		IC50	>100	N/A	N/A	<1	

7d Survival Rate Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	0.0%
2.5		4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	0.0%
5		4	0.95	0.928	0.972	0.9	1	0.0105	0.0577	6.08%	2.56%
10		4	1	1	1	1	1	0	0	0.0%	-2.56%
25		4	1	1	1	1	1	0	0	0.0%	-2.56%
50		4	0.95	0.928	0.972	0.9	1	0.0105	0.0577	6.08%	2.56%
100		4	1	1	1	1	1	0	0	0.0%	-2.56%

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.29	1.29	1.3	1.28	1.31	0.00253	0.0139	1.07%	0.0%
2.5		4	1.23	1.16	1.3	1	1.45	0.0337	0.185	15.0%	4.53%
5		4	1.24	1.2	1.27	1.13	1.33	0.0157	0.0859	6.95%	4.26%
10		4	1.4	1.36	1.43	1.27	1.47	0.0163	0.0892	6.4%	-8.03%
25		4	1.23	1.18	1.28	1.07	1.38	0.0232	0.127	10.3%	4.76%
50		4	1.32	1.27	1.38	1.16	1.48	0.0266	0.146	11.0%	-2.5%
100		4	1.44	1.37	1.51	1.26	1.65	0.0326	0.179	12.4%	-11.6%

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	Control	1	1	1	0.9
2.5		1	1	0.9	1
5		0.9	0.9	1	1
10		1	1	1	1
25		1	1	1	1
50		1	0.9	0.9	1
100		1	1	1	1

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.29	1.31	1.29	1.28
2.5		1.45	1.27	1	1.21
5		1.13	1.26	1.33	1.23
10		1.27	1.45	1.47	1.38
25		1.25	1.07	1.38	1.22
50		1.41	1.25	1.16	1.48
100		1.65	1.52	1.33	1.26

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis No: 20-5578-9613 Endpoint: 7d Survival Rate CETIS Version: CETISv1.6.5
Analyzed: 30 Nov-08 12:30 Analysis: Parametric-Two Sample Official Results: Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)		C > T	Not Run	100	>100	N/A	1	5.2%

Equal Variance t Two-Sample Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Control		2.5	0	1.94	0.112	0.5000	Non-Significant Effect
		5	0.655	1.94	0.121	0.2680	Non-Significant Effect
		10	-1	1.94	0.0792	0.8220	Non-Significant Effect
		25	-1	1.94	0.0792	0.8220	Non-Significant Effect
		50	0.655	1.94	0.121	0.2680	Non-Significant Effect
		100	-1	1.94	0.0792	0.8220	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.03225062	0.005375103	6	1.21	0.3380	Non-Significant Effect
Error	0.09295766	0.004426555	21			
Total	0.1252082772553	0.00980165787041	27			

ANOVA Assumptions

Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Mod Levene Equality of Variance	2.63	3.81	0.0351	Equal Variances
Distribution	Shapiro-Wilk Normality	0.875		0.0032	Non-normal Distribution

7d Survival Rate Summary

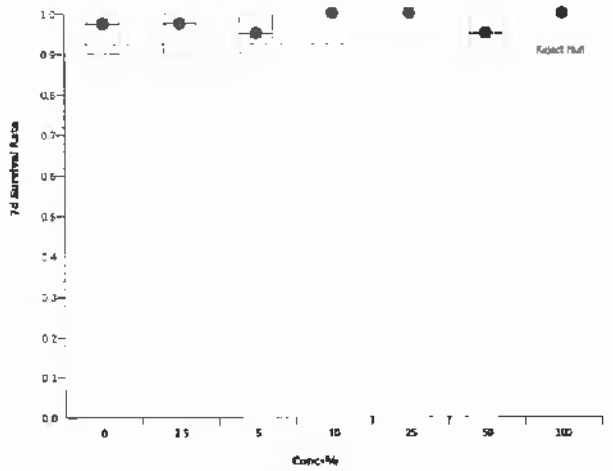
Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	0.975	0.956	0.994	0.9	1	0.00928	0.05	5.13%	0.0%
2.5		4	0.975	0.956	0.994	0.9	1	0.00928	0.05	5.13%	0.0%
5		4	0.95	0.928	0.972	0.9	1	0.0107	0.0577	6.08%	2.56%
10		4	1	1	1	1	1	0	0	0.0%	-2.56%
25		4	1	1	1	1	1	0	0	0.0%	-2.56%
50		4	0.95	0.928	0.972	0.9	1	0.0107	0.0577	6.08%	2.56%
100		4	1	1	1	1	1	0	0	0.0%	-2.56%

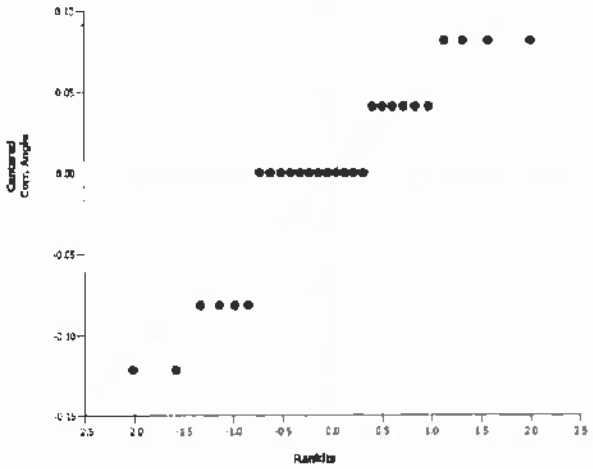
Angular (Corrected) Transformed Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.37	1.34	1.4	1.25	1.41	0.0151	0.0815	5.94%	0.0%
2.5		4	1.37	1.34	1.4	1.25	1.41	0.0151	0.0815	5.94%	0.0%
5		4	1.33	1.29	1.37	1.25	1.41	0.0175	0.0941	7.07%	2.97%
10		4	1.41	1.41	1.41	1.41	1.41	0	0	0.0%	-2.97%
25		4	1.41	1.41	1.41	1.41	1.41	0	0	0.0%	-2.97%
50		4	1.33	1.29	1.37	1.25	1.41	0.0175	0.0941	7.07%	2.97%
100		4	1.41	1.41	1.41	1.41	1.41	0	0	0.0%	-2.97%

Chronic Larval Fish Survival and Growth Test					Pacific EcoRisk
Analysis No:	20-5578-9613	Endpoint:	7d Survival Rate	CETIS Version:	CETISv1.6.5
Analyzed:	30 Nov-08 12:30	Analysis:	Parametric-Two Sample	Official Results:	Yes

Graphics





Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis No:	17-6240-2220	Endpoint:	Mean Dry Biomass-mg	CETIS Version:	CETISv1.6.5
Analyzed:	30 Nov-08 12:30	Analysis:	Parametric-Control vs Treatments	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	17.5%

Dunnett's Multiple Comparison Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Control		2.5	0.634	2.45	0.226	0.6220	Non-Significant Effect
		5	0.597	2.45	0.226	0.6390	Non-Significant Effect
		10	-1.13	2.45	0.226	0.9910	Non-Significant Effect
		25	0.667	2.45	0.226	0.6070	Non-Significant Effect
		50	-0.35	2.45	0.226	0.9310	Non-Significant Effect
		100	-1.62	2.45	0.226	0.9980	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.1702912	0.02838187	6	1.67	0.1780	Non-Significant Effect
Error	0.3570597	0.01700284	21			
Total	0.52735090255737	0.04536471437991	27			

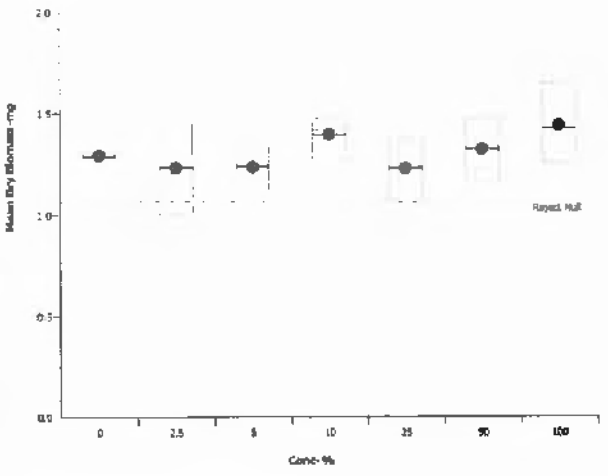
ANOVA Assumptions

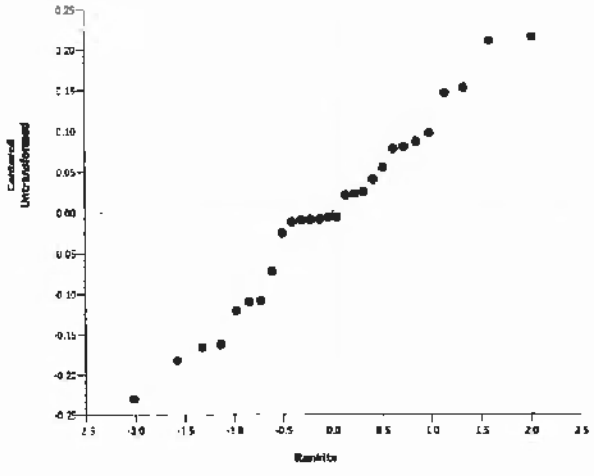
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	12.2	16.8	0.0581	Equal Variances
Distribution	Shapiro-Wilk Normality	0.974		0.6790	Normal Distribution

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.29	1.29	1.3	1.28	1.31	0.00257	0.0139	1.07%	0.0%
2.5		4	1.23	1.16	1.3	1	1.45	0.0343	0.185	15.0%	4.53%
5		4	1.24	1.2	1.27	1.13	1.33	0.0159	0.0859	6.95%	4.26%
10		4	1.4	1.36	1.43	1.27	1.47	0.0166	0.0892	6.4%	-8.03%
25		4	1.23	1.18	1.28	1.07	1.38	0.0236	0.127	10.3%	4.76%
50		4	1.32	1.27	1.38	1.16	1.48	0.0271	0.146	11.0%	-2.5%
100		4	1.44	1.37	1.51	1.26	1.65	0.0332	0.179	12.4%	-11.6%

Graphics





Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis No:	13-5969-4097	Endpoint:	Mean Dry Biomass-mg	CETIS Version:	CETISv1.6.5
Analyzed:	30 Nov-08 12:31	Analysis:	Linear Interpolation (ICPIN)	Official Results:	Yes

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	5795186	280	Yes	Two-Point Interpolation

Point Estimates

Level	Conc-%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC2.5	>100	N/A	N/A	<1	N/A	N/A
IC5	>100	N/A	N/A	<1	N/A	N/A
IC10	>100	N/A	N/A	<1	N/A	N/A
IC15	>100	N/A	N/A	<1	N/A	N/A
IC20	>100	N/A	N/A	<1	N/A	N/A
IC25	>100	N/A	N/A	<1	N/A	N/A
IC40	>100	N/A	N/A	<1	N/A	N/A
IC50	>100	N/A	N/A	<1	N/A	N/A

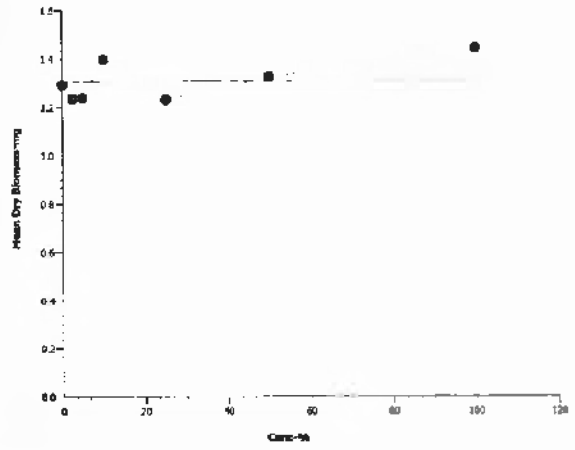
Mean Dry Biomass-mg Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.29	1.26	1.31	0.00253	0.0139	1.07%	0.0%
2.5		4	1.23	1	1.45	0.0337	0.185	15.0%	4.53%
5		4	1.24	1.13	1.33	0.0157	0.0859	6.95%	4.26%
10		4	1.4	1.27	1.47	0.0163	0.0892	6.4%	-8.03%
25		4	1.23	1.07	1.38	0.0232	0.127	10.3%	4.76%
50		4	1.32	1.16	1.48	0.0266	0.146	11.0%	-2.5%
100		4	1.44	1.26	1.65	0.0326	0.179	12.4%	-11.6%

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.29	1.31	1.29	1.28
2.5		1.45	1.27	1	1.21
5		1.13	1.26	1.33	1.23
10		1.27	1.45	1.47	1.38
25		1.25	1.07	1.38	1.22
50		1.41	1.25	1.16	1.48
100		1.65	1.52	1.33	1.26

Graphics



7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Applied Marine Sciences
Test Material: MWHA - Dry
Test ID#: 31046 Project #: 14150
Test Date: 11-15-08

Organism Log#: 4256 Age: 9d.0.
Organism Supplier: ABS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 695

Treatment (% Effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.9	7.92		7.7		24.3	10	10	10	10	Date: 11.15.08
2.5	25.9	7.92		7.8		24.5	10	10	10	10	Sample ID 20926
5	25.9	7.92		7.7		24.5	10	10	10	10	Test Solution Prep EMC
10	25.9	7.91		7.7		24.5	10	10	10	10	New WQ MEL
25	25.9	7.88		7.9		24.4	10	10	10	10	Initiation Time 1150
50	25.9	7.85		8.0		24.2	10	10	10	10	Initiation Signoff
100	25.9	7.79		8.5		24.0	10	10	10	10	
Meter ID	11A	pH03		DO14		EC04					
Control	25.6	8.33	7.35	7.7	5.8	24.9	10	10	10	10	Date 11/16/08
2.5	25.6	8.31	7.37	7.4	6.4	25.2	10	10	9	10	Sample ID 20926
5	25.6	8.29	7.44	7.4	6.1	25.2	9	9	10	10	Test Solution Prep SA
10	25.6	8.25	7.47	7.5	5.8	25.2	10	10	10	10	New WQ JNC
25	25.6	8.14	7.58	7.5	5.9	25.1	10	10	10	10	Renewal Time 1030
50	25.6	8.00	7.71	8.1	6.0	25.1	10	9	9	10	Renewal Signoff
100	25.6	7.78	7.82	8.1	5.8	25.0	10	10	10	10	Old WQ ARL
Meter ID	11A	~~pH02~~ pH12	pH11	D010	D012	EC01					
Control	25.8	8.45	7.94	7.3	5.9	24.8	10	10	10	9	Date 11/17/08
2.5	25.8	8.44	7.98	7.2	5.7	25.0	10	10	9	10	Sample ID 20926
5	25.8	8.44	8.01	7.3	5.8	25.1	9	9	10	10	Test Solution Prep PH
10	25.8	8.42	7.96	7.3	5.5	25.0	10	10	10	10	New WQ JNC
25	25.8	8.34	8.04	7.4	5.7	25.0	10	10	10	10	Renewal Time 1550
50	25.8	8.22	8.07	7.9	5.3	24.9	10	9	9	10	Renewal Signoff
100	25.8	8.00	8.14	8.6	5.3	24.9	10	10	10	10	Old WQ
Meter ID	11A	pH03	pH03	D012	D012	EC01					
Control	25.7	8.25	7.84	7.1	25 5.5	24.8	10	10	10	9	Date 11/18/08
2.5	25.7	8.23	7.85	7.0	5.8	24.9	10	10	9	10	Sample ID 20926
5	25.7	8.21	7.89	7.1	5.5	24.9	9	9	10	10	Test Solution Prep
10	25.7	8.17	7.88	7.1	5.7	24.9	10	10	10	10	New WQ JNC
25	25.7	8.06	7.95	7.2	5.5	24.8	10	10	10	10	Renewal Time 1645
50	25.7	7.91	8.02	7.6	5.5	24.5	10	9	9	10	Renewal Signoff
100	25.7	7.67	8.04	8.9	5.3	24.3	10	10	10	10	Old WQ RV
Meter ID	11A	pH11	PH 12	D010	D014	EC04					

7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Applied Marine Sciences
Test Material: MWHA-Dry
Test ID#: 31046 Project #: 14150
Test Date: 11-15-08

Organism Log#: 4256 Age: 9d 0h
Organism Supplier: ATS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 695

Treatment (% Effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.6	8.27	7.89	7.0	5.9	24.6	10	10	10	9	Date 11/14/08
2.5	25.6	8.29	7.87	7.3	5.7	24.7	10	10	9	10	Sample ID 20926
5	25.6	8.29	7.89	7.3	5.5	24.7	9	9	10	10	Test Solution Prep JPW
10	25.6	8.25	7.84	7.4	5.5	24.7	10	10	10	10	New WQ
25	25.6	8.16	7.90	7.5	5.4	24.6	10	10	10	10	Renewal Time 10:45
50	25.6	8.01	7.97	7.5	5.5	24.4	10	~~10~~ 9	9	10	Renewal Signoff
100	25.6	~~8.05~~ 8.12	8.01	~~7.8~~ 8.14	5.6	~~24.1~~ EC04	10	10	10	10	Old WQ
Meter ID	11A	Ph12	Ph03	D014	D010	EC04					
Control	25.7	8.32	7.99	7.5	5.0	24.8	10	10	10	9	Date 11/20/08
2.5	25.7	8.32	7.83	7.2	5.6	24.9	10	10	9	10	Sample ID 20926
5	25.7	8.31	7.82	7.3	5.4	24.9	9	9	10	10	Test Solution Prep
10	25.7	8.27	7.85	7.4	5.3	24.9	10	10	10	10	New WQ
25	25.7	8.17	7.87	7.5	5.3	24.8	10	10	10	10	Renewal Time 0930
50	25.7	8.01	7.87	7.8	5.0	24.9	10	9	9	10	Renewal Signoff
100	25.7	7.79	7.95	8.2	4.7	24.8	10	10	10	10	Old WQ
Meter ID	11A	PH12	PH12	D014	D014	EC01					
Control	25.9	8.11	7.83	7.16	6.0	25.2	10	10	10	9	Date 11-21-08
2.5	25.9	8.10	7.80	7.4	5.7	25.1	10	10	9	10	Sample ID 20926
5	25.9	8.10	7.77	7.6	5.6	25.1	9	9	10	10	Test Solution Prep
10	25.9	8.08	7.79	7.7	5.5	25.0	10	10	10	10	New WQ
25	25.9	8.05	7.90	7.8	5.8	24.8	10	10	10	10	Renewal Time 1100
50	25.9	8.00	7.90	7.9	6.0	24.3	10	9	9	10	Renewal Signoff
100	25.9	7.92	8.02	8.6	5.5	~~25.9~~ 25.8	10	10	10	10	Old WQ
Meter ID	11A	pH11	pH11	D014	D014	EC04					
Control	25.8		7.65		6.0	25.8	10	10	10	9	Date 11/22/08
2.5	25.8		7.64		5.8	25.7	10	10	9	10	Termination Time 0915
5	25.8		7.73		5.9	25.8	9	9	10	10	Termination Signoff SM
10	25.8		7.71		5.8	25.8	10	10	10	10	Old WQ
25	25.8		7.83		5.7	25.2	10	10	10	10	
50	25.8		7.83		5.3	25.1	10	9	9	10	
100	25.8		7.95		5.4	24.3	10	10	10	10	
Meter ID	11A		PH03		D014	EC04					

Chronic Inland Silverside Dry Weight and Biomass Data

Client: Applied Marine Sciences Test ID #: 31046 Project # 14150

Sample: MWA-Dry Tare Weight Date: 11-17-08 Sign-off: JMC

Test Date: 11-15-08 Final Weight Date: 11-24-08 Sign-off: JMC

Pan ID	Concentration	Replicate	Initial Pan Weight (mg)	Final Pan Weight (mg)	Initial # of Organisms	Biomass Value (mg)
1	Control	A	185.18	198.03	10	1.285
2		B	168.56	181.68	10	1.312
3		C	161.95	174.80	10	1.285
4		D	167.28	180.11	10	1.283
5	2.5	A	167.04	181.53	10	1.449
6		B	161.79	174.52	10	1.273
7		C	176.95	186.97	10	1.002
8		D	157.68	169.75	10	1.207
9	5	A	170.44	181.70	10	1.126
10		B	163.92	176.51	10	1.259
11		C	173.03	186.36	10	1.333
12		D	163.29	175.56	10	1.227
13	10	A	160.37	173.11	10	1.274
14		B	177.57	192.07	10	1.450
15		C	171.12	185.85	10	1.473
16		D	183.07	196.90	10	1.383
17	25	A	176.49	189.04	10	1.255
18		B	184.64	195.31	10	1.067
19		C	185.43	199.19	10	1.376
20		D	185.56	197.77	10	1.221
21	50	A	182.13	196.23	10	1.410
22		B	175.45	187.96	10	1.251
23		C	181.99	193.56	10	1.157
24		D	182.22	196.98	10	1.476
25	100	A	170.95	187.46	10	1.651
26		B	188.06	203.27	10	1.521
27		C	179.45	192.77	10	1.332
28		D	171.65	184.23	10	1.258
QA 1			167.97	168.00		
QA 2			175.69	175.70		
QA 3			119.48	119.46		
Balance ID			1	1		

Appendix G

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Menidia beryllina*

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Test Run No:	04-4336-0470	Test Type:	Growth-Survival (7d)	Analyst:	Rivian Villanueva
Start Date:	15 Nov-08 12:05	Protocol:	EPA/821/R/02/014 (2002)	Diluent:	Laboratory Water
Ending Date:	22 Nov-08 11:15	Species:	Menidia beryllina	Brine:	Crystal Sea
Duration:	6d 23h	Source:	Aquatic Biosystems, CO	Age:	9

Sample No:	17-0698-4462	Code:	KCl	Client:	Reference Toxicant
Sample Date:	15 Nov-08 12:05	Material:	Potassium chloride	Project:	14152
Receive Date:	15 Nov-08 12:05	Source:	Reference Toxicant		
Sample Age:	N/A (25.8 °C)	Station:	In House		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
11-8178-4333	7d Survival Rate	0.5	1	0.707	17.0%		Dunnett's Multiple Comparison Test
12-2977-0287	Mean Dry Biomass-mg	0.5	1	0.707	17.8%		Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-g/L	95% LCL	95% UCL	TU	Method
03-7103-6061	7d Survival Rate	EC2.5	0.83	0.689	0.92		Linear Regression (MLE)
		EC10	0.936	0.817	1.01		
		EC15	0.977	0.868	1.05		
		EC20	1.01	0.911	1.08		
		EC25	1.04	0.948	1.11		
		EC40	1.12	1.05	1.18		
		EC50	1.17	1.11	1.23		
16-3238-7402	Mean Dry Biomass-mg	IC2.5	0.15	0.0136	0.845		Linear Interpolation (ICPIN)
		IC5	0.301	0.0271	1.42		
		IC10	0.581	0.0668	1.29		
		IC15	0.819	0.131	1.18		
		IC20	1.01	0.321	1.11		
		IC25	1.04	0.708	1.12		
		IC40	1.13	1.03	1.24		
		IC50	1.19	1.11	1.3		

7d Survival Rate Summary

Conc-g/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	0.0%
0.5		4	0.95	0.928	0.972	0.9	1	0.0105	0.0577	6.08%	2.56%
1		4	0.775	0.704	0.846	0.5	0.9	0.0346	0.189	24.4%	20.5%
1.25		4	0.4	0.339	0.461	0.2	0.6	0.0298	0.163	40.8%	59.0%
1.5		4	0.075	0.0392	0.111	0	0.2	0.0175	0.0957	128.0%	92.3%
2		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	Control	4	1.35	1.32	1.39	1.24	1.45	0.018	0.0988	7.3%	0.0%
0.5		4	1.24	1.18	1.3	1.05	1.41	0.0287	0.157	12.7%	8.31%
1		4	1.1	1.03	1.17	0.908	1.35	0.034	0.186	16.9%	18.8%
1.25		4	0.591	0.527	0.655	0.443	0.827	0.0312	0.171	28.9%	56.4%
1.5		4	0.061	0.0308	0.0912	0	0.171	0.0148	0.081	133.0%	95.5%
2		4	0	0	0	0	0	0	0		100.0%

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

7d Survival Rate Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	0.9	1	1	1
0.5		0.9	1	0.9	1
1		0.9	0.9	0.5	0.8
1.25		0.2	0.6	0.4	0.4
1.5		0	0	0.2	0.1
2		0	0	0	0

Mean Dry Biomass-mg Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.42	1.24	1.31	1.45
0.5		1.05	1.32	1.18	1.41
1		1.09	1.35	0.908	1.04
1.25		0.443	0.827	0.601	0.492
1.5		0	0	0.171	0.073
2		0	0	0	0

7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Reference Toxicant
Test Material: Potassium Chloride
Test ID#: 31049 Project #: 14152
Test Date: 11-15-08

Organism Log#: 4256 Age: 9
Organism Supplier: ABS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 695

Treatment (g KCl/L)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.8	7.94		7.9		24.3	10	10	10	10	Date 11-15-08
0.5	25.8	7.95		7.7		25.0	10	10	10	10	Sample ID —
1	25.8	7.95		7.7		25.6	10	10	10	10	Test Solution Prep RV
1.25	25.8	7.95		7.6		25.7	10	10	10	10	New WQ MEC
1.5	25.8	7.96		7.6		26.1	10	10	10	10	Initiation Time 1205
2	25.8	7.94		7.7		26.6	10	10	10	10	Initiation Signoff RV
Meter ID	11A	pH03		DO14		EC04					
Control	25.6	8.34	7.53	8.0	6.5	24.9	10	10	10	10	Date 11/16/08
0.5	25.6	8.34	7.53	7.9	6.9	25.5	9	10	9	10	Sample ID —
1	25.6	8.34	7.55	7.9	6.5	26.0	9	9	8	10	Test Solution Prep RV
1.25	25.6	8.35	7.53	7.8	6.1	26.2	9	10	7	7	New WQ JNC
1.5	25.6	8.35	7.52	7.8	6.5	26.4	2	2	9	4	Renewal Time 1510
2	25.6	8.36	7.48	7.8	6.5	27.2	0	2	2	2	Renewal Signoff PG
											Old WQ SL
Meter ID	11A	pH12	pH12	DO10	DO10	EC01					
Control	25.8	8.08	7.86	7.7	5.6	24.8	10	10	10	10	Date 11-17-08
0.5	25.8	8.12	7.87	7.1	5.8	25.5	9	10	9	10	Sample ID —
1	25.8	8.13	7.88	7.0	5.7	26.0	9	9	6	10	Test Solution Prep RV
1.25	25.8	8.15	7.84	7.0	5.6	26.1	7	10	6	7	New WQ AR
1.5	25.8	8.16	7.94	6.9	5.7	26.5	1	1	7	3	Renewal Time 1215
2	25.8	8.16	7.99	6.9	5.7	27.0	—	0	0	0	Renewal Signoff RV
											Old WQ NW
Meter ID	11A	pH11	pH12	DO10	DO10	Eco4					
Control	25.8	8.14	7.70	7.4	5.7	24.8	10	10	10	10	Date 11/18/08
0.5	25.8	8.34	7.73	7.2	5.7	25.6	9	10	9	10	Sample ID —
1	25.8	8.35	7.72	7.1	5.5	26.1	9	9	5	10	Test Solution Prep JAW
1.25	25.8	8.35	7.72	7.2	5.2	26.3	3	8	5	7	New WQ AR
1.5	25.8	8.35	7.82	7.1	5.5	26.5	1	0	6	3	Renewal Time 1415
2	—	—	—	—	—	—	—	—	—	—	Renewal Signoff JCR
											Old WQ DCW
Meter ID	11A	pH12	pH11	DO12	DO10	Eco1					

7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Reference Toxicant
Test Material: Potassium Chloride
Test ID#: 31049 Project #: 14152
Test Date: 11-15-08

Organism Log#: 4256 Age: 9
Organism Supplier: ABS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 695

Treatment (g KCl/L)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.7	8.39	7.80	6.8	6.0	24.6	10	10	10	10	Date 11/19/08
0.5	25.7	8.40	7.82	6.9	6.5	25.4	9	10	9	10	Sample ID —
1	25.7	8.40	7.81	6.9	5.8	25.9	9	9	5	9	Test Solution Prep JW
1.25	25.7	8.39	7.82	6.7	5.6	26.1	3	7	5	6	New WQ JNC
1.5	25.7	8.39	7.96	6.8	5.5	26.4	0	—	3	2	Renewal Time 1130
2	—	—	—	—	—	—	—	—	—	—	Renewal Signoff JLR
											Old WQ AP
Meter ID	11A	pH11	DH03	D012	D010	EC05					
Control	25.8	8.33	7.68	6.8	4.9	24.6	10	10	10	10	Date 11/20/08
0.5	25.8	8.23	7.79	6.9	4.8	25.7	9	10	9	10	Sample ID —
1	25.8	8.31 8.31	7.66	6.9	4.8	~~26.4~~ 25.9	9	9	5	9	Test Solution Prep JT
1.25	25.8	8.29	7.73	6.9	4.8	26.4	2	7	5	5	New WQ DAP
1.5	25.8	8.31	7.84	6.9	5.3	26.6	—	—	2	1	Renewal Time 1400
2	—	8.36	—	6.8	—	27.0	—	—	—	—	Renewal Signoff JLR
											Old WQ
Meter ID	11A	PH12	pH03	D014	D010	EC01					
Control	25.9	8.05	7.79	7.7	6.0	25.2	10	10	10	10	Date 11/21/08
0.5	25.9	8.06	7.78	7.6	6.0	25.9	9	10	9	10	Sample ID —
1	25.9	8.08	7.79	7.7	5.5	26.4	9	9	5	8	Test Solution Prep JT
1.25	25.9	8.08	7.82	7.6	5.7	26.6	2	6	5	4	New WQ MTM
1.5	25.9	8.09	7.88	7.7	5.9	26.9	—	—	2	1	Renewal Time 1115
2	—	—	—	—	—	—	—	—	—	—	Renewal Signoff JLR
											Old WQ MTM
Meter ID	11A	pH11	pH11	D014	D014	EC04					
Control	25.9		7.67		5.9	26.0	9	10	10	10	Date 11/22/08
0.5	25.9		7.73		5.8	26.5	9	10	9	10	Termination Time 1115
1	25.9		7.77		5.7	27.2	9	9	5	8	Termination Signoff JT
1.25	25.9		7.80		5.7	27.0	2	6	4	4	Old WQ SM
1.5	25.9		7.84		5.9	27.7	—	—	2	1	
2	—		—		—	—	—	—	—	—	
Meter ID	11A		pH11		D012	EC01					

Chronic Inland Silverside (*M. beryllina*) Dry Weight and Biomass Data

Client: Reference Toxicant Test ID #: 31049 Project # 14152

Sample: Potassium Chloride Tare Weight Date: 11-17-08 Sign-off: TLC

Test Date: 11-15-08 Final Weight Date: 11-24-08 Sign-off: TLC

Pan ID	Concentration	Replicate	Initial Pan Weight (mg)	Final Pan Weight (mg)	Initial # of Organisms	Biomass Value (mg)
1	Control	A	177.32	191.49	10	1.417
2		B	177.53	189.92	10	1.239
3		C	186.83	199.89	10	1.306
4		D	179.50	194.03	10	1.453
5	0.5	A	168.46	179.00	10	1.054
6		B	165.33	178.49	10	1.316
7		C	176.48	188.29	10	1.181
8		D	168.97	183.11	10	1.414
9	1	A	190.91	201.83	10	1.092
10		B	174.63	188.16	10	~~1.85~~ 1.353
11		C	197.82	206.90	10	0.908
12		D	169.91	180.36	10	1.045
13	1.25	A	177.97	182.40	10	6.443
14		B	131.09	139.36	10	0.827
15		C	152.35	158.36	10	0.601
16		D	118.72	123.64	10	0.492
17	1.5	A	117.21	—	10	—
18		B	179.52	—	10	—
19		C	185.03	186.74	10	0.171
20		D	163.16	163.89	10	0.073
21	2	A	189.15	—	10	—
22		B	162.41	—	10	—
23		C	154.89	—	10	—
24		D	181.91	—	10	—
QA1			116.80	116.80		0.000
QA2			147.01	146.99		-0.020
QA3			117.92	117.92		0.000
Balance ID						

9.0 Appendix B

See separate attachment for detailed toxicity testing results for the "wet" season brine sample, collected on February 25, 2009 (AppendixB_022509Results.pdf) (Pacific EcoRisk, March, 2009).

Jay Johnson
Applied Marine Sciences
4749 Bennett Dr., Suite L
Livermore, CA 94550

March 30, 2009

Dear Jay:

I have enclosed two copies of our *Supplemental* report "A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent" for the sample collected February 25, 2009. This report was revised to reflect the correction of the sample collection date.

The results of this testing are summarized below:

Chronic Effects of the Desalination Effluent on *Thalassiosira pseudonana*
There were ***no*** significant reductions in *Thalassiosira pseudonana* growth.

Chronic Effects of the Desalination Effluent on *Americamysis bahia*
There were ***no*** significant reductions in *Americamysis bahia* survival or growth.

Chronic Effects of the Desalination Effluent on *Menidia beryllina*
There were ***no*** significant reductions in *Menidia beryllina* survival or growth.

If you have any questions regarding the performance and interpretation of these tests, feel free to call me at (707) 207-7760.

Sincerely,

R. Scott Ogle, Ph.D.
Principal & Special Projects Director

This testing was performed under Lab Order 14499. The test results reported herein conform to the most current NELAC standards, where applicable, unless otherwise narrated in the body of the report, and only relate to the sample(s) tested. This report shall not be reproduced, except in full, without the written consent of Pacific EcoRisk.

Supplemental Report

A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent

Sample collected February 25, 2009

Prepared For:

Applied Marine Sciences
4749 Bennett Dr., Suite L
Livermore, CA 94550

Prepared By:

Pacific EcoRisk, Inc.
2250 Cordelia Rd.
Fairfield, CA 94534

March 2009
Report Revised March 2009

PACIFIC ECORISK
ENVIRONMENTAL CONSULTING & TESTING

Supplemental Report

A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent

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March 2009
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A Toxicity Evaluation of the Bay Area Regional Desalination Project Effluent

Sample collected February 25, 2009

Table of Contents

Appendices

1. INTRODUCTION

Under contract to the Applied Marine Sciences, Pacific EcoRisk (PER) performed chronic toxicity evaluations of effluent sample collected as part of the Bay Area Regional Desalination Project. The toxicity evaluations consist of performing the following chronic toxicity tests:

- chronic (96-hr) growth test with the diatom *Thalassiosira pseudonana*;
- chronic survival and growth test with the crustacean *Americamysis bahia*; and
- chronic survival and growth test with larval *Menidia beryllina*.

These toxicity tests were performed on water the sample collected on February 25, 2009. This report describes the performance and results of these tests.

2. COLLECTION AND DELIVERY OF THE EFFLUENT SAMPLE

On February 25, an effluent sample was collected from the Bay Area Regional Desalination plant. The sample was transported, on ice and under chain-of-custody, to the PER laboratory facility in Fairfield. Upon receipt at the testing laboratory, aliquots of sample were collected for analysis of initial water quality characteristics (Table 1). The remainder of the water sample was stored at 0-6°C except when being used to prepare test solutions. The chain-of-custody record for the collection and delivery of this sample is provided in Appendix A.

Table 1. Initial water quality characteristics of the Bay Area Regional Desalination Project effluent.

Date Sample Collected	Date Sample Received	Sample ID	Temp (˚C)	pH	D.O (mg/L)	Salinity	Conductivity (μS/cm)	Total Ammonia (mg/L N)
02/25/09	02/25/09	Brine	13.7*	7.55	5.5	7.0	12420	<1.0

* Sample was delivered on the day of collection, and was transported at ≤6.0˚C.

3. CHRONIC TOXICITY TEST PROCEDURES

The Bay Area Desalination Project effluent sample was tested for toxicity using the following US EPA short-term chronic toxicity tests:

- chronic (96-hr) growth test with the diatom *Thalassiosira pseudonana*;
- chronic survival and growth test with the crustacean *Americamysis bahia*; and
- chronic survival and growth test with larval *Menidia beryllina*.

The methods used in conducting these toxicity tests followed the guidelines established by the following manuals:

- "Standard Guide for Conducting Static 96-h Toxicity Tests with Microalgae" (ASTM E 1218-97a); and
- "Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Marine and Estuarine Organisms, Third Edition" (EPA-821-R-02-014).

3.1 Algal Growth Toxicity Testing with *Thalassiosira pseudonana*

The short-term chronic diatom toxicity test consists of exposing *Thalassiosira pseudonana* to the effluent for ~96-hrs, after which the effects on cell growth are evaluated. The specific procedures used in these tests are described below.

The Lab Control water for this test consisted of reverse osmosis, de-ionized (RO/DI) water adjusted up to the test salinity of 25 ppt using an artificial sea salt (Crystal Seas®-bioassay grade). For use in this test, an aliquot of the effluent was similarly adjusted to 25 ppt using the same sea salt. The Lab Control water and ambient waters were filtered (0.45 μm) and then spiked with nutrients, as per ASTM guidelines. The salinity-adjusted Lab Control water and effluent were used to prepare test solutions at test treatment concentrations of 2.5%, 5%, 10%, 25%, 50% and 100% effluent. Water quality characteristics were measured on these test solutions prior to use in this test.

There were 4 replicates at each test treatment, each replicate consisting of a 250-mL glass Erlenmeyer flask containing 100 mL of test solution; an additional replicate was established at each test treatment for the measurement of test solution water quality characteristics during the test and at test termination. Each treatment was inoculated to an initial diatom cell density of 20,000 cells/mL from a from a laboratory culture of *Thalassiosira* that is maintained in log growth phase. These flasks were loosely capped and randomly positioned within a temperature-controlled room at 20°C, under continuous illumination from cool-white fluorescent bulbs.

Each day, the temperature and pH were determined for the designated “water quality” replicate at each treatment; each replicate flask was gently shaken in the morning and randomly re-positioned within the temperature-controlled room.

After 96 ($\pm$2) hrs exposure, the cell density in each replicate flask was determined by microscopic analysis. The resulting cell density data were analyzed to determine any growth impairment, or toxicity, caused by the ambient water; all statistical analyses were performed using CETIS® statistical software (Tidepool Scientific, McKinleyville, CA).

3.1.1 Reference Toxicant Testing of the *Thalassiosira pseudonana*

In order to assess the sensitivity of the *Thalassiosira* 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 KCl at concentrations of 0.625, 1.25, 2.5, 5, and 10 gm/L. The resulting test response data were statistically analyzed to determine key

dose-response point estimates (e.g., IC_{50}); all statistical analyses were performed 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.

3.2 Survival and Growth Toxicity Testing with *Americamysis bahia*

The short-term chronic *Americamysis bahia* test consists of exposing the organisms to a series of effluent dilutions for 7 days, after which effects on survival and growth are evaluated. The specific procedures used in this test are described below.

The *Americamysis bahia* used in this test were obtained from Aquatic BioSystems (Ft. Collins, CO); upon receipt at the lab, the mysids were transferred into aerated tanks containing saltwater at 25 ppt, and were fed brine shrimp nauplii during the pre-test holding period.

The Lab Control/dilution water for this test was prepared by salting up reverse-osmosis, de-ionized water to a salinity of 25 ppt using a commercial artificial sea salt (Crystal Sea Salt®-bioassay grade). Each day, an aliquot of the final effluent sample was similarly adjusted to a salinity of 25 ppt using the same artificial sea salt. The salinity-adjusted Lab Control/dilution water and effluent sample were used to prepare daily test solutions at concentrations of 2.5%, 5%, 10%, 25%, 50% and 100% effluent. "New" water quality characteristics (pH, D.O., and salinity) were measured on these test solutions prior to use in the test.

There were 8 replicates at each test treatment, each replicate consisting of 200 mL of test solution in a 400-mL glass beaker. The test was initiated by randomly allocating five 7-day old mysids into each replicate beaker. The beakers were randomly positioned in a temperature-controlled room at 26˚C (with temperature being monitored daily) under a 16L:8D photoperiod. The mysids were fed freshly-hatched brine shrimp nauplii twice daily.

Each day of the test, fresh test solutions were prepared and characterized as before. The test replicate beakers were examined, with any dead animals, uneaten food, wastes, and other detritus being removed. The number of live mysids in each replicate was determined and ~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 salinity) were measured on the old test water that had been discarded from one randomly-selected replicate at each treatment.

After 7 days exposure, the test was terminated and the number of live mysids in each replicate beaker was recorded. The mysids 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. The mysids were then dried at 100˚C for >24 hrs and re-weighed to determine the total weight of mysids in each replicate; the total weight was divided by the initial number of mysids per replicate (n=5) 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 CETIS® statistical software.

3.2.1 Reference Toxicant Testing of the *Americamysis bahia*

In order to assess the sensitivity of the mysid 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 media spiked with KCl at concentrations of 0.125, 0.25, 0.5, 1, and 2 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.

3.3 Survival and Growth Toxicity Testing with *Menidia beryllina*

The short-term chronic *Menidia beryllina* test consists of exposing larval fish to a series of effluent dilutions for 7 days, after which effects on survival and growth are evaluated. The specific procedures used in this test are described below.

The larval fish used in this bioassay were obtained from a commercial supplier (Aquatic Biosystems, Fort Collins, CO). These fish were maintained at 25˚C in aerated aquaria containing Lab Control water (described below) prior to their use in this test. During this pre-test period, the fish were fed brine shrimp nauplii *ad libitum*.

The Lab Control/dilution water for this bioassay was prepared by salting up reverse-osmosis, de-ionized water to a salinity of 25 ppt using a commercial artificial sea salt (Crystal Sea® -bioassay grade). Each day, an aliquot of the final effluent sample was similarly adjusted to a salinity of 25 ppt using the same artificial sea salt. The salinity-adjusted Lab Control/dilution water and effluent sample were used to prepare daily test solutions at concentrations of 2.5%, 5%, 10%, 25%, 50% and 100% effluent. “New” water quality characteristics (pH, D.O., and salinity) were measured on these test solutions prior to use in the test.

There were 4 replicates for the Lab Control and each effluent treatment, each replicate consisting of 400 mL of test media in a 600-mL glass beaker. This test was initiated by randomly allocating ten 11-day old *Menidia beryllina* into each replicate. These 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 twice daily.

Each day of the test, fresh test solutions were prepared and characterized as before. The replicate beakers containing the larval fish were examined, 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 media. “Old” water quality characteristics (pH, D.O., and conductivity) were measured on the old test water collected from one randomly selected replicate at each treatment.

After 7 days exposure, the number of live fish in each replicate beaker was recorded. Then, the fish from each replicate were carefully euthanized in methanol, rinsed in de-ionized water, and transferred to a pre-dried and pre-tared weighing pan. These 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 “biomass value” data were analyzed to determine key dose-response point estimates (e.g., EC_{50}); all statistical analyses were performed using the CETIS® statistical software.

3.3.1 Reference Toxicant Testing of the *Menidia beryllina*

In order to assess the sensitivity of the fish test organisms to toxic stress, a reference toxicant test was performed concurrently with the effluent test. This reference toxicant test was performed similarly to the effluent toxicity test, except that test solutions consisted of Lab Control (25 ppt water) spiked with KCl at concentrations of 0.5, 1.0, 1.25, 1.5, and 2 gm/L. After 7 days exposure, the survival and weight data were evaluated as in the effluent test. 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.

4. RESULTS

4.1 Effects of the Desalination Effluent on *Thalassiosira pseudonana*

The results of this test are summarized below in Table 2. There were ***no*** significant reductions in algal growth in the Desalination effluent; the growth NOEC was 100% effluent. The IC_{25} and IC_{50} were both >100% effluent, resulting in <1.0 survival TUc (where survival TUc = $100/IC_{25}$ or $100/IC_{50}$) for both test endpoints..

The test data and the summary of statistical analyses for these tests are presented in Appendix B.

Table 2. Effect of the Desalination effluent on *Thalassiosira pseudonana*.

Effluent Treatment	Mean Diatom Cell Density (cells/mL x 10^6)
Lab Water Control	1.91
2.5%	2.26
5%	2.53
10%	2.99
25%	3.70
50%	3.85
100%	5.58
Summary of Key Statistics	
Growth NOEC =	100% effluent
TUc (TUc = 100/NOEC) =	1.0
Growth IC_{25} =	>100% effluent
TUc (TUc = $100/IC_{25}$) =	<1.0
Growth IC_{50} =	>100% effluent
TUc (TUc = $100/IC_{50}$) =	<1.0

4.1.1 Reference Toxicant Toxicity to *Thalassiosira pseudonana*

The results of this test are presented in Table 3. There was a mean of 1,990,000 cells/mL at the Lab Control treatment. The growth IC_{50} was 4.25 gm/L KCl.

The results of the concurrent reference toxicant test were consistent with 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 these tests are presented in Appendix C.

Table 3. Reference toxicant testing: effects of KCl on *Thalassiosira pseudonana*.

KCl Treatment (gm/L)	Mean Diatom Cell Density (cells/mL x 10^6)
Lab Water Control	1.99
0.625	2.17
1.25	2.38
2.5	2.01
5	**0.70***
10	**0.19***
Summary of Key Statistic	
IC_{50} =	4.25 gm/L KCl

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

4.2 Effects of the Desalination Effluent on *Americamysis bahia*

The results of this test are presented in Table 4. There was a mean of 92.5% survival at the Lab Control treatment. There were ***no*** significant reductions in survival in the Desalination effluent; the survival NOEC was 100% effluent. Due to the absence of significant mortalities, the EC_{25} and EC_{50} could not be calculated, but can both be assumed >100% effluent, resulting in <1.0 survival TUc (where TUc = $100/EC_{25}$ or $100/EC_{50}$) for both test endpoints.

The mean 'biomass value' was 0.26 mg at the Lab Control treatment. There were ***no*** significant reductions in growth in the Desalination effluent; the growth NOEC was 100% effluent. The IC_{25} and IC_{50} were both >100% effluent, resulting in <1.0 growth TUc (where TUc = $100/IC_{25}$ or $100/IC_{50}$) for both test endpoints.

The test data and summary of statistical analyses for this test are attached as Appendix D.

Table 4. Effects of the Desalination effluent on *Americamysis bahia*.

Effluent Treatment	Mean % Survival	Mean "Biomass Value" (mg)
Lab Control	92.5	0.26
2.5%	97.5	0.31
5%	97.5	0.29
10%	100	0.31
25%	95.0	0.31
50%	95.0	0.29
100%	97.5	0.31
Summary of Key Statistics		
NOEC	>100% effluent	>100% effluent
TUc (TUc = 100/NOEC) =	<1.0	<1.0
Survival EC_{25} or Growth IC_{25}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{25}$ or $100/IC_{25}$) =	<1.0	<1.0
Survival EC_{50} or Growth IC_{50}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{50}$ or $100/IC_{50}$) =	<1.0	<1.0

4.2.1 Reference Toxicant Toxicity to *Americamysis bahia*

The results of this test are presented in Table 5. There was a mean of 97.5% survival and a mean mysid biomass of 0.28 mg at the Lab Control treatment. The survival EC_{50} was 0.60 gm/L, and the growth IC_{50} was 0.64 gm/L.

The results of the concurrent reference toxicant test were consistent with 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 attached as Appendix E.

Table 5. Reference toxicant testing: effects of potassium chloride on *Americamysis bahia*.

KCl Treatment (gm/L)	Mean % Survival	Mean Biomass Value (mg)
Lab Control	97.5	0.28
0.125	97.5	0.28
0.25	100	0.26
0.5	75	**0.19***
1	**0***	**0***
2	**0***	**0***
Summary of Key Statistics		
Survival EC_{50} or Growth IC_{50} =	0.60 gm/L KCl	0.64 gm/L KCl

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

4.3 Effects of the Desalination Effluent on *Menidia beryllina*

The results of this test are presented in Table 6. There was 100% survival at the Lab Control treatment. There were ***no*** significant reductions in survival in the Desalination effluent; the survival NOEC was 100% effluent. Due to the absence of significant mortalities, the EC_{25} and EC_{50} could not be calculated, but can both be assumed >100% effluent, resulting in <1.0 survival TUc (where TUc = $100/EC_{25}$ or $100/EC_{50}$) for both test endpoints.

The mean fish biomass value was 0.94 mg at the Lab Control treatment. There were ***no*** toxicologically significant reductions in growth in the Desalination effluent; the growth NOEC was 100% effluent. The IC_{25} and IC_{50} were both >100% effluent, resulting in <1.0 growth TUc (where TUc = $100/IC_{25}$ or $100/IC_{50}$) for both test endpoints.

The test data and the summary of statistical analyses for this test are attached as Appendix F.

Table 6. Effects the Desalination effluent on *Menidia beryllina*.

Effluent Treatment	Mean % Survival	Mean Biomass Value (mg)
Lab Control	100	0.94
2.5%	97.5	0.92
5%	97.5	0.90
10%	100	0.89
25%	97.5	**0.82***
50%	100	0.94
100%	100	0.97
Summary of Key Statistics		
NOEC	>100% effluent	>100% effluent
TUc (TUc = 100/NOEC) =	<1.0	<1.0
Survival EC_{25} or Growth IC_{25}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{25}$ or $100/IC_{25}$) =	<1.0	<1.0
Survival EC_{50} or Growth IC_{50}	>100% effluent	>100% effluent
TUc (TUc = $100/EC_{50}$ or $100/IC_{50}$) =	<1.0	<1.0

* There was an interrupted dose-response with a biomass of 0.82 mg at the 25% effluent treatment, which was indicated as being statistically less than the Lab Control treatment. However, the biomass values at the remaining 50% and 100% effluent treatments were not significantly less than the Lab Control, indicating that the apparent reduction in biomass in the 25% effluent was not toxicologically significant.

4.3.1 Reference Toxicant Toxicity to *Menidia beryllina*

The results of this test are summarized below in Table 7. There was a mean of 100% survival and a mean fish biomass value of 1.03 mg at the Lab Control treatment; the survival EC_{50} value was 1.2 gm/L KCl, and the growth IC_{50} was 1.3 gm/L KCl.

These reference toxicant test responses were consistent with previous performance of this test in our lab, indicating that these organisms were responding to toxic stress in a typical fashion.

The test data and summary of statistical analyses for this test are attached as Appendix G.

Table 7. Reference toxicant testing: effects of KCl on *Menidia beryllina*.

KCl Treatment (gm/L)	Mean % Survival	Mean Biomass Value (mg)
Lab Control	100	1.03
0.5	100	1.03
1	85	0.91
1.25	**40***	**0.54***
1.5	**7.5***	**0.13***
2	**0***	**0***
Summary of Key Statistics		
Survival EC_{50} or Growth IC_{50} =	1.2 gm/L KCl	1.3 gm/L KCl

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

5. SUMMARY AND CONCLUSIONS

Chronic Effects of the Desalination Effluent on *Thalassiosira pseudonana*
There were no significant reductions in algal growth in the effluent.

Chronic Effects of the Desalination Effluent on *Americamysis bahia*
There were no significant reductions in invertebrate survival or growth in the effluent.

Chronic Effects of the Desalination Effluent on *Menidia beryllina*
There were no significant reductions in fish survival or growth in the effluent.

6. AQUATIC TOXICITY DATA QUALITY CONTROL

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

Negative Control – The test organism responses at the Lab Control treatments were within acceptable limits.

Positive Control – The results for the *Americamysis bahia, Menidia beryllina,* and *Thalassiosira pseudonana* reference toxicant tests were consistent with the reference toxicant test databases, indicating that these test organisms were responding to toxic stress in a typical fashion.

Concentration Response Relationships – There was an interrupted dose-response in the Menidia beryllina growth response, with an indication of a statistically significant reduction at the 25% effluent concentration. However, the biomass value at the remaining 50% and 100% effluent treatments were not significantly less than the Lab Control, indicating that the apparent reduction in biomass in the 25% effluent was not toxicologically significant.

There were valid concentration-response relationships for the remaining effluent and reference toxicant tests (EPA821-B-00-004), which were determined to be acceptable for this testing.

Appendix A

Chain-of-Custody Record for the Collection and Delivery of the Bay Area Regional Desalination Project 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: Applied Marine Sciences, Inc.

Attn: Jay Johnson
Phone: (925) 373-7142
Email: jjohnson@amarine.com

BILL TO:

Attn:
Phone:
Email:

PROJECT: MWHA Regional Desal Brine Sampling (post-storm event)

ANALYSES REQUESTED — Per Scott Ogle

SAMPLE IDENTIFICATION	DATE	TIME	SAMPLE MATRIX	GRAB/ COMP.	# CONTAINERS/TYPE	REMARKS
cubitainers Brine	2/25/09	0915	water		2x 20L } cubitainer 2x 10L }	
					/	
					/	
					/	
					/	
					/	
					/	
					/	
					/	

METHOD OF SHIPMENT: FedEx:_____ UPS:_____ HAND:_____ OTHER:_____

COMMENTS:

CODES:

RELINQUISHED BY: (SIGNATURE)	DATE	TIME	RECEIVED BY: (SIGNATURE)	DATE	TIME	PAGE #
	2/25/09	10:33		2/25/09	10:35	1 OF 1

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 the Bay Area Regional Desalination Project Effluent to *Thalassiosira pseudonana*

Phytoplankton Growth Inhibition Test — **Pacific EcoRisk**

Test Run No:	18-5118-4444	**Test Type:**	Cell Growth	**Analyst:**	Jason Walker
Start Date:	25 Feb-09 14:30	**Protocol:**	EPA/821/R/02/014 (2002)	**Diluent:**	Laboratory Water
Ending Date:	01 Mar-09 13:30	**Species:**	Thalassiosira pseudonana	**Brine:**	Crystal Sea
Duration:	95h	**Source:**	In-House Culture	**Age:**	7

Sample No:	08-7787-0147	**Code:**	Brine	**Client:**	AMS
Sample Date:	25 Feb-09 09:15	**Material:**	Effluent	**Project:**	14499
Receive Date:	25 Feb-09 10:35	**Source:**	Applied Marine Sciences		
Sample Age:	5h (13.7 °C)	**Station:**	MWHA		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
03-5037-7206	Cell Density	0	>0		6.26%		Equal Variance t Two-Sample Test
04-4861-5439		100	>100	N/A	19.5%	1	Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-%	95% LCL	95% UCL	TU	Method
15-8463-6205	Cell Density	IC2.5	>100	N/A	N/A	<1	Linear Interpolation (ICPIN)
		IC5	>100	N/A	N/A	<1	
		IC10	>100	N/A	N/A	<1	
		IC15	>100	N/A	N/A	<1	
		IC20	>100	N/A	N/A	<1	
		IC25	>100	N/A	N/A	<1	
		IC40	>100	N/A	N/A	<1	
		IC50	>100	N/A	N/A	<1	

Cell Density Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.91E+6	1.88E+6	1.95E+6	1.82E+6	2.01E+6	1.60E+4	8.77E+4	4.59%	0.0%
0	Seawater	4	2.33E+6	2.29E+6	2.36E+6	2.21E+6	2.42E+6	1.58E+4	8.66E+4	3.72%	-21.6%
2.5		4	2.26E+6	2.25E+6	2.27E+6	2.23E+6	2.28E+6	3.76E+3	2.06E+4	0.91%	-18.0%
5		4	2.53E+6	2.47E+6	2.58E+6	2.35E+6	2.72E+6	2.93E+4	1.61E+5	6.36%	-32.0%
10		4	2.99E+6	2.90E+6	3.07E+6	2.68E+6	3.19E+6	3.98E+4	2.18E+5	7.3%	-56.1%
25		4	3.70E+6	3.58E+6	3.82E+6	3.35E+6	4.06E+6	5.81E+4	3.18E+5	8.6%	-93.3%
50		4	3.85E+6	3.74E+6	3.96E+6	3.53E+6	4.22E+6	5.17E+4	2.83E+5	7.36%	-101.0%
100		4	5.58E+6	5.49E+6	5.68E+6	5.33E+6	5.81E+6	4.52E+4	2.48E+5	4.44%	-192.0%

Cell Density Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.82E+6	2.01E+6	1.86E+6	1.96E+6
0	Seawater	2.34E+6	2.33E+6	2.42E+6	2.21E+6
2.5		2.26E+6	2.23E+6	2.26E+6	2.28E+6
5		2.58E+6	2.72E+6	2.45E+6	2.35E+6
10		3.19E+6	3.07E+6	3.00E+6	2.68E+6
25		3.35E+6	4.06E+6	3.53E+6	3.85E+6
50		3.53E+6	4.22E+6	3.83E+6	3.82E+6
100		5.81E+6	5.78E+6	5.33E+6	5.41E+6

Phytoplankton Growth Inhibition Test — **Pacific EcoRisk**

Analysis No:	04-4861-5439	**Endpoint:**	Cell Density	**CETIS Version:**	CETISv1.6.5
Analyzed:	08 Mar-09 15:57	**Analysis:**	Parametric-Control vs Treatments	**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	19.5%

Dunnett's Multiple Comparison Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Control		2.5	-2.27	2.45	372000	1.0000	Non-Significant Effect
		5	-4.03	2.45	372000	1.0000	Non-Significant Effect
		10	-7.05	2.45	372000	1.0000	Non-Significant Effect
		25	-11.7	2.45	372000	1.0000	Non-Significant Effect
		50	-12.7	2.45	372000	1.0000	Non-Significant Effect
		100	-24.1	2.45	372000	1.0000	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	3.748044E+13	6.246741E+12	6	135	0.0000	Significant Effect
Error	9.725E+11	46309520000	21			
Total	3.8452943127E+13	6.2930500485E+12	27			

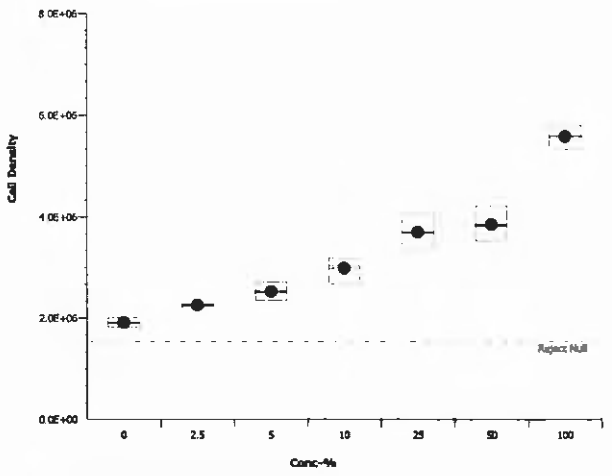
ANOVA Assumptions

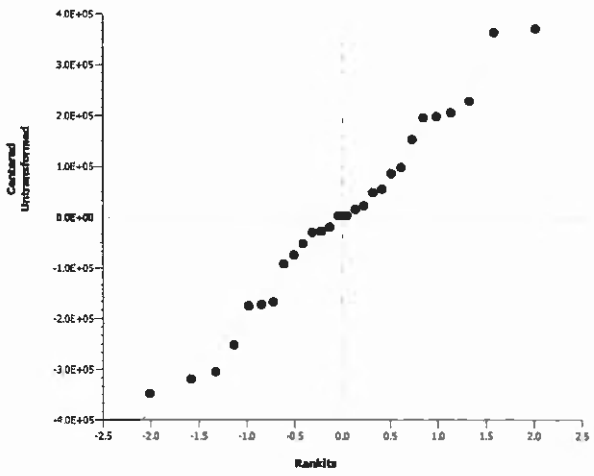
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	14.5	16.8	0.0248	Equal Variances
Distribution	Shapiro-Wilk Normality	0.975		0.7130	Normal Distribution

Cell Density Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.91E+6	1.88E+6	1.95E+6	1.82E+6	2.01E+6	1.63E+4	8.77E+4	4.59%	0.0%
2.5		4	2.26E+6	2.25E+6	2.27E+6	2.23E+6	2.28E+6	3.83E+3	2.06E+4	0.91%	-18.0%
5		4	2.53E+6	2.46E+6	2.59E+6	2.35E+6	2.72E+6	2.98E+4	1.61E+5	6.36%	-32.0%
10		4	2.99E+6	2.90E+6	3.07E+6	2.68E+6	3.19E+6	4.05E+4	2.18E+5	7.3%	-56.1%
25		4	3.70E+6	3.58E+6	3.82E+6	3.35E+6	4.06E+6	5.91E+4	3.18E+5	8.6%	-93.3%
50		4	3.85E+6	3.74E+6	3.96E+6	3.53E+6	4.22E+6	5.26E+4	2.83E+5	7.36%	-101.0%
100		4	5.58E+6	5.49E+6	5.68E+6	5.33E+6	5.81E+6	4.60E+4	2.48E+5	4.44%	-192.0%

Graphics





Phytoplankton Growth Inhibition Test — Pacific EcoRisk

Analysis No:	15-8463-6205	**Endpoint:**	Cell Density	**CETIS Version:**	CETISv1.6.5
Analyzed:	08 Mar-09 15:57	**Analysis:**	Linear Interpolation (ICPIN)	**Official Results:**	Yes

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	7055475	280	Yes	Two-Point Interpolation

Point Estimates

Level	Conc-%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC2.5	>100	N/A	N/A	<1	N/A	N/A
IC5	>100	N/A	N/A	<1	N/A	N/A
IC10	>100	N/A	N/A	<1	N/A	N/A
IC15	>100	N/A	N/A	<1	N/A	N/A
IC20	>100	N/A	N/A	<1	N/A	N/A
IC25	>100	N/A	N/A	<1	N/A	N/A
IC40	>100	N/A	N/A	<1	N/A	N/A
IC50	>100	N/A	N/A	<1	N/A	N/A

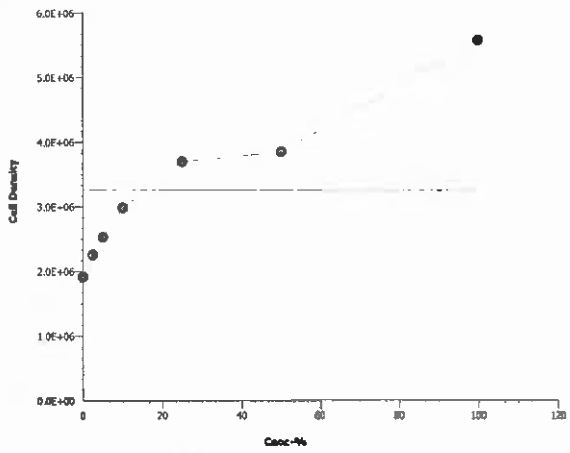
Cell Density Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.91E+6	1.82E+6	2.01E+6	1.60E+4	8.77E+4	4.59%	0.0%
2.5		4	2.26E+6	2.23E+6	2.28E+6	3.76E+3	2.06E+4	0.91%	-18.0%
5		4	2.53E+6	2.35E+6	2.72E+6	2.93E+4	1.61E+5	6.36%	-32.0%
10		4	2.99E+6	2.68E+6	3.19E+6	3.98E+4	2.18E+5	7.3%	-56.1%
25		4	3.70E+6	3.35E+6	4.06E+6	5.81E+4	3.18E+5	8.6%	-93.3%
50		4	3.85E+6	3.53E+6	4.22E+6	5.17E+4	2.83E+5	7.36%	-101.0%
100		4	5.58E+6	5.33E+6	5.81E+6	4.52E+4	2.48E+5	4.44%	-192.0%

Cel' Density Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.82E+6	2.01E+6	1.86E+6	1.96E+6
2.5		2.26E+6	2.23E+6	2.26E+6	2.28E+6
5		2.58E+6	2.72E+6	2.45E+6	2.35E+6
10		3.19E+6	3.07E+6	3.00E+6	2.68E+6
25		3.35E+6	4.06E+6	3.53E+6	3.85E+6
50		3.53E+6	4.22E+6	3.83E+6	3.82E+6
100		5.81E+6	5.78E+6	5.33E+6	5.41E+6

Graphics



Phytoplankton Growth Inhibition Test — Pacific EcoRisk

Analysis No:	03-5037-7206	**Endpoint:**	Cell Density	**CETIS Version:**	CETISv1.6.5
Analyzed:	08 Mar-09 15:57	**Analysis:**	Parametric-Two Sample	**Official Results:**	Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Untransformed		C > T	Not Run	0	>0			6.26%

Equal Variance t Two-Sample Test

Control	vs	Control	Test Stat	Critical	MSD	P-Value	Decision(5%)
Control		Seawater	-6.69	1.94	120000	1.0000	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	3.403125E+11	3.403125E+11	1	44.8	0.0005	Significant Effect
Error	45575000000	7595833000	6			
Total	3.8588749005E+11	3.4790832333E+11	7			

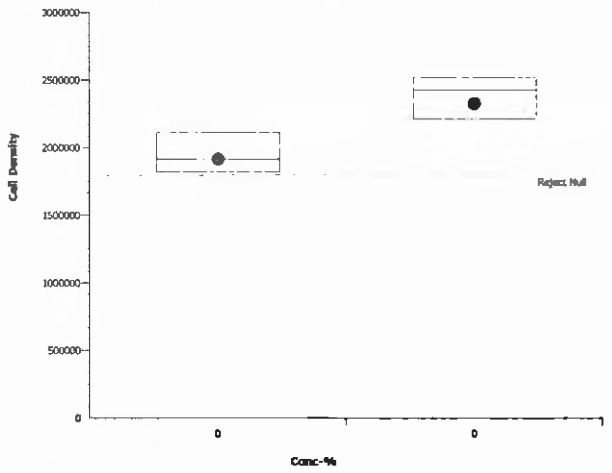
ANOVA Assumptions

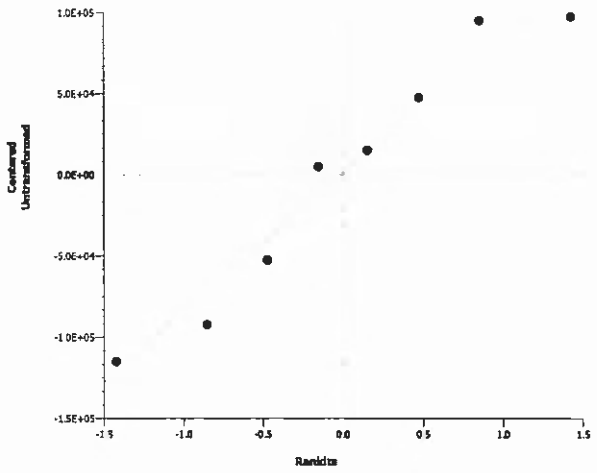
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Variance Ratio F	1.03	47.5	0.9840	Equal Variances
Distribution	Shapiro-Wilk Normality	0.931		0.5230	Normal Distribution

Cell Density Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Seawater	4	2.33E+6	2.29E+6	2.36E+6	2.21E+6	2.42E+6	1.61E+4	8.66E+4	3.72%	0.0%
0	Control	4	1.91E+6	1.88E+6	1.95E+6	1.82E+6	2.01E+6	1.63E+4	8.77E+4	4.59%	17.7%

Graphics





Thalassiosira pseudonana Cell Density Enumeration Data

Client: Applied Marine Sciences Initial Count: N/A

Test Material: Brine Effluent Enumerating Scientist: CG

Test Start Date: 2/25/09 Start Time: 1430 Project #: 14499

Test End Date: ~~2/27/09~~ 3/1/09 End Time: 1330 Test ID #: 32189

Treatment %	Rep A	Rep B	Rep C	Rep D	Mean
Lab Water Control	1.82	2.01	1.86	1.96	1.91
2.5	2.26	2.23	2.26	2.28	2.26
5	2.58	2.72	2.45	2.35	2.52
10	3.19	3.07	3.00	2.68	2.98
25	3.35	4.06	3.53	3.85	3.70
50	3.53	4.22	3.83	3.82	3.85
100	5.81	5.78	5.33	5.41	5.58

This datasheet has been reviewed for completeness and consistency with Test Acceptability Criteria and/or other issues of concern.	**Control Mean Density (cells/mL x 10^6)**	**Date:**	**Time:**	**Signoff:**
	1.91	3/1/09	14:00	

Thalassiosira pseudonana Toxicity Test Water Quality Data

Client: Applied Marine Sciences Test ID #: 32189 Test Date: 2/25/09

Test Material: BrINE EFFLUENT Project #: 14499 Control/Diluent: Algal Medium

Treatment %	Temp (°C)	pH	D.O. (mg/L)	Salinity (ppt)	Sign-Off
Lab Water Control	20.7	8.00	8.4	25.2	Test Solution Prep: JL
2.5	20.7	8.01	8.3	25.3	New WQ: JNC
5	20.7	7.99	8.0	25.5	Inoculation Date: 2/25/09
10	20.7	7.97	8.2	25.6	Inoculation Time: 1430
25	20.7	7.92	8.4	25.5	Inoculation Signoff: JL
50	20.7	7.88	8.1	25.5	
100	20.7	7.81	7.8	25.5	
Meter ID:	40	pH12	DO12	EC05	
Lab Water Control	21.0	8.10			Date: 2/26/09
2.5	21.0	8.13			WQ Time: 0915
5	21.0	8.14			WQ Signoff: DGV
10	21.0	8.15			
25	21.0	8.19			
50	21.0	8.18			
100	21.0	8.18			
Meter ID:	40	pH11			
Lab Water Control	20.5	8.39			Date: 2/27/09
2.5	20.5	~~8.38~~ 8.38			WQ Time: 12:00
5	20.5	8.46			WQ Signoff: MH
10	20.5	8.48			
25	20.5	8.58			
50	20.5	8.47			
100	20.5	8.42			
Meter ID:	40	pH 12			
Lab Water Control	20.9	9.48			Date: 2/28/09
2.5	20.9	9.41			WQ Time: 12:10
5	20.9	9.38			WQ Signoff: MEL
10	20.9	9.57			
25	20.9	9.64			
50	20.9	9.47			
100	20.9	9.19			
Meter ID:	40	pH12			
Lab Water Control	20.5	9.20	11.6	25.0	Date: 3/1/09
2.5	20.5	9.34	13.1	25.5	Termination Time: 1330
5	20.5	9.41	12.1	25.4	Termination Signoff: MH
10	20.5	9.46	13.1	25.5	WQ Time: 0930
25	20.5	9.38	12.3	25.5	WQ Signoff: JNC
50	20.5	9.55	14.1	25.5	
100	20.5	9.55	> 20.0	25.3	
Meter ID:	40	pH03	DO14	EC05	

Initial Test Conditions			Light Intensity (lux)
			6167

Thalassiosira pseudonana Cell Density Enumeration Data Sheet

Client: **Applied Marine Sciences** Sample ID: **Sea Water Control**

Test Start Date: 2/25/09 Test ID #: 32189 Project #: 14499

Test End Date: 2/29/09 Control/Diluent: Algal Medium

Test Treatment	Temp (C)	pH	D.O. (mg/L)	Salinity (ppt)	Sign-Off
Lab Water Control	20.7	8.00	8.4	25.4	Date: 2/25/09
Sea Water Control	20.7	7.90	8.8	24.5	Sample ID #: 21508
					Test Solution Prep: JC
					New WQ: JNC
					Inoculation Time: 1430
Meter ID	40	PH12	DO12	ECO5	Innoculation Signoff: JC
Lab Water Control	21.0	8.10			Date: 2/26/09
Sea Water Control	21.0	8.04			WQ Time: 0915
					WQ Signoff: DGO
Meter ID	40	PH11			
Lab Water Control	20.5	8.39			Date: 2/27/09
Sea Water Control	20.5	8.43			WQ Time: 12:00
					WQ Signoff: MC
Meter ID	40	PH 12			
Lab Water Control	20.9	9.48			Date: 2/28/09
Sea Water Control	20.9	9.52			WQ Time: 12:10
					WQ Signoff: MEC
Meter ID	40	PH12			
Lab Water Control	20.5	9.20	11.6	25.0	Date: 3/1/09
Sea Water Control	20.5	9.23	12.4	24.8	WQ Time: 0930
					WQ Signoff: JNC
Meter ID	40	PH03	DO14	ECO5	

Initial Count: 20.000 cells/mL Termination Time: 1330 Enumerating Scientist: SA

Treatment	Cell Density (cells/mL x 10^6)				Mean Cell Density (cells/mL x 10^6)
	Rep A	Rep B	Rep C	Rep D	
Lab Water Control	1.82	2.01	1.86	1.96	1.91
Sea Water Control	2.34	2.33	2.42	2.21	2.32

This datasheet has been reviewed for completeness and consistency with Test Acceptability Criteria and/or other issues of concern.	Control Mean Density (cells/mL x 10^6)	Date:	Time:	Signoff:
	1.91	3/1/09	14:00	

Initial Test Conditions	Light Intensity (lux)
	6167

Appendix C

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Thalassiosira pseudonana*

Phytoplankton Growth Inhibition Test — Pacific EcoRisk

Test Run No:	02-3978-3152	**Test Type:**	Cell Growth	**Analyst:**	Rivian Villanueva
Start Date:	25 Feb-09 14:30	**Protocol:**	ASTM E 1218-97a (Algae)	**Diluent:**	Laboratory Water
Ending Date:	01 Mar-09 13:10	**Species:**	Thalassiosira pseudonana	**Brine:**	Not Applicable
Duration:	95h	**Source:**	In-House Culture	**Age:**	7

Sample No:	13-7905-2419	**Code:**	KCl	**Client:**	Reference Toxicant
Sample Date:	25 Feb-09 14:30	**Material:**	Potassium chloride	**Project:**	14502
Receive Date:	25 Feb-09 14:30	**Source:**	Reference Toxicant		
Sample Age:	N/A (20.7 °C)	**Station:**	In House		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
03-5814-1385	Cell Density	2.5	5	3.54	7.2%		Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-g/L	95% LCL	95% UCL	TU	Method
20-1499-7251	Cell Density	IC2.5	1.66	1.49	1.95		Linear Interpolation (ICPIN)
		IC5	2.06	1.72	2.64		
		IC10	2.6	2.36	2.74		
		IC15	2.8	2.63	2.94		
		IC20	3.01	2.85	3.14		
		IC25	3.22	3.07	3.34		
		IC40	3.84	3.71	3.94		
		IC50	4.25	4.14	4.34		

Cell Density Summary

Conc-g/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	4	1.99E+6	1.95E+6	2.02E+6	1.87E+6	2.08E+6	1.66E+4	9.11E+4	4.59%	0.0%
0.625		4	2.17E+6	2.10E+6	2.23E+6	2.02E+6	2.40E+6	3.00E+4	1.64E+5	7.59%	-9.07%
1.25		4	2.38E+6	2.35E+6	2.40E+6	2.32E+6	2.45E+6	1.20E+4	6.56E+4	2.76%	-19.6%
2.5		4	2.01E+6	1.99E+6	2.02E+6	1.97E+6	2.06E+6	6.89E+3	3.77E+4	1.88%	-1.13%
5		4	6.95E+5	6.84E+5	7.06E+5	6.60E+5	7.30E+5	5.27E+3	2.89E+4	4.15%	65.0%
10		4	1.90E+5	1.82E+5	1.98E+5	1.60E+5	2.10E+5	3.94E+3	2.16E+4	11.4%	90.4%

Cell Density Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Control	1.87E+6	1.96E+6	2.08E+6	2.03E+6
0.625		2.40E+6	2.02E+6	2.10E+6	2.14E+6
1.25		2.41E+6	2.32E+6	2.32E+6	2.45E+6
2.5		2.00E+6	1.97E+6	2.06E+6	2.00E+6
5		7.00E+5	6.90E+5	7.30E+5	6.60E+5
10		1.90E+5	2.10E+5	1.60E+5	2.00E+5

Thalassiosira pseudonana Reference Toxicant Test Cell Density Data

Client: **Reference Toxicant** Initial Count: N/A

Test Material: Potassium chloride Enumerating Scientist: SM

Test Start Date: 2/25/09 Start Time: 1430 Project #: 14502

Test End Date: 3/1/09 End Time: 1310 Test ID #: 32194

Treatment	Rep A	Rep B	Rep C	Rep D	Mean
Lab Water Control	1.87	1.96	2.08	2.03	1.98
0.625 g/L KCl	2.40	2.02	2.10	2.14	2.16
1.25 g/L KCl	2.41	2.32	2.32	2.45	2.38
2.5 g/L KCl	2.00	1.97	2.06	2.00	2.01
5 g/L KCl	0.70	0.69	0.73	0.66	0.70
10 g/L KCl	0.19	0.21	0.16	0.20	0.19

This datasheet has been reviewed for completeness and consistency with Test Acceptability Criteria and/or other issues of concern.	**Control Mean Density (cells/mL x 10^6)**	**Date:**	**Time:**	**Signoff:**
	1.98	3/1/09	14:00	

Thalassiosira pseudonana Reference Toxicant Test Water Quality Data

Client: **Reference Toxicant** Test ID #: 32194 Test Date: 2/25/09

Test Material: Potassium chloride Project #: 14502 Control/Diluent: Algal Medium

Reference Toxicant Test Treatment (g/L KCl)	Temp (°C)	pH	D.O. (mg/L)	Salinity (ppt)	Sign-Off
Lab Water Control	20.7	8.00	8.4	25.4	Test Solution Prep JC
0.625	20.7	8.03	8.1	26.3	New WQ MDM
1.25	20.7	8.04	7.9	27.0	Innoculated Date 2/25/09
2.5	20.7	8.05	7.8	28.2	Innoculation Time 1430
5	20.7	8.05	7.7	30.8	Innoculation Signoff JC
10	20.7	8.05	7.9	35.8	
Meter ID:	58	pH03	D012	EC05	
Lab Water Control	20.7	8.07			Date: 2/26/09
0.625	20.7	8.11			WQ Time: 0930
1.25	20.7	8.10			WQ Signoff: DG
2.5	20.7	8.11			
5	20.7	8.08			
10	20.7	8.07			
Meter ID:	58	pH11			
Lab Water Control	20.5	8.31			Date: 2/27/09
0.625	20.5	8.34			WQ Time: 12:00
1.25	20.5	8.35			WQ Signoff: YK
2.5	20.5	8.31			
5	20.5	8.09			
10	20.5	8.00			
Meter ID:	58	pH 12			
Lab Water Control	20.8	8.78			Date: 2/28/09
0.625	20.8	9.08			WQ Time: 12:00
1.25	20.8	9.01			WQ Signoff: MEL
2.5	20.8	8.87			
5	20.8	8.40			
10	20.8	8.14			
Meter ID:	58	pH12			
Lab Water Control	20.5	8.82	11.2	25.2	Date: 3/1/09
0.625	20.5	9.16	14.6	26.0	Termination Time 1310
1.25	20.5	9.17	14.2	26.8	Termination Signoff SH
2.5	20.5	9.04	12.9	28.0	WQ Time: 0910
5	20.5	8.40	9.9	30.2	WQ Signoff: JNC
10	20.5	7.95	7.3	35.5	
Meter ID:	58	pH03	D014	EC05	

Initial Test Conditions			Light Intensity (lux)
			6200

Appendix D

Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of the Bay Area Regional Desalination Project Effluent to *Americamysis bahia*

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

Test Run No:	21-2258-0498	Test Type:	Growth-Survival (7d)	Analyst:	Jason Walker
Start Date:	25 Feb-09 15:00	Protocol:	EPA/821/R/02/014 (2002)	Diluent:	Laboratory Water
Ending Date:	04 Mar-09 08:35	Species:	Americamysis bahia	Brine:	Crystal Sea
Duration:	6d 18h	Source:	Aquatic Biosystems, CO	Age:	7

Sample No:	08-7787-0147	Code:	Brine	Client:	AMS
Sample Date:	25 Feb-09 09:15	Material:	Effluent	Project:	14499
Receive Date:	25 Feb-09 10:35	Source:	Applied Marine Sciences		
Sample Age:	6h (13.7 °C)	Station:	MWHA		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
18-0763-0142	7d Survival Rate	100	>100	N/A	11.2%	1	Steel Many-One Rank Test
16-9066-4752	Mean Dry Biomass-mg	100	>100	N/A	23.9%	1	Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-%	95% LCL	95% UCL	TU	Method
01-1773-8098	Mean Dry Biomass-mg	IC2.5	>100	N/A	N/A	<1	Linear Interpolation (ICPIN)
		IC5	>100	N/A	N/A	<1	
		IC10	>100	N/A	N/A	<1	
		IC15	>100	N/A	N/A	<1	
		IC20	>100	N/A	N/A	<1	
		IC25	>100	N/A	N/A	<1	
		IC40	>100	N/A	N/A	<1	
		IC50	>100	N/A	N/A	<1	

7d Survival Rate Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.925	0.886	0.964	0.8	1	0.0189	0.104	11.2%	0.0%
2.5		8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	-5.41%
5		8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	-5.41%
10		8	1	1	1	1	1	0	0	0.0%	-8.11%
25		8	0.95	0.897	1	0.6	1	0.0258	0.141	14.9%	-2.7%
50		8	0.95	0.915	0.985	0.8	1	0.0169	0.0926	9.75%	-2.7%
100		8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	-5.41%

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.262	0.243	0.28	0.176	0.316	0.00925	0.0507	19.4%	0.0%
2.5		8	0.306	0.293	0.319	0.236	0.346	0.00629	0.0345	11.3%	-16.9%
5		8	0.294	0.275	0.312	0.204	0.372	0.00904	0.0495	16.9%	-12.2%
10		8	0.309	0.293	0.326	0.23	0.378	0.00798	0.0437	14.1%	-18.3%
25		8	0.31	0.288	0.332	0.19	0.398	0.011	0.0602	19.4%	-18.5%
50		8	0.293	0.268	0.318	0.23	0.446	0.0121	0.0665	22.7%	-12.0%
100		8	0.307	0.285	0.329	0.216	0.384	0.0109	0.0595	19.4%	-17.3%

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

7d Survival Rate Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	1	1	0.8	1	1	0.8	1	0.8
2.5		1	1	1	1	0.8	1	1	1
5		0.8	1	1	1	1	1	1	1
10		1	1	1	1	1	1	1	1
25		1	1	1	0.6	1	1	1	1
50		1	0.8	1	0.8	1	1	1	1
100		1	1	1	1	0.8	1	1	1

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	0.316	0.316	0.2	0.27	0.284	0.252	0.278	0.176
2.5		0.328	0.284	0.346	0.236	0.312	0.332	0.312	0.296
5		0.204	0.308	0.302	0.262	0.296	0.372	0.33	0.274
10		0.304	0.378	0.306	0.322	0.274	0.336	0.324	0.23
25		0.308	0.398	0.284	0.19	0.36	0.316	0.308	0.316
50		0.306	0.23	0.446	0.246	0.266	0.292	0.286	0.272
100		0.374	0.29	0.322	0.216	0.234	0.384	0.314	0.32

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

Analysis No:	18-0763-0142	**Endpoint:**	7d Survival Rate	**CETIS Version:**	CETISv1.6.5
Analyzed:	08 Mar-09 16:12	**Analysis:**	Nonparametric-Control vs Treatments	**Official Results:**	Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)		C > T	Not Run	100	>100	N/A	1	11.2%

Steel Many-One Rank Test

Control	vs	Conc-%	Test Stat	Critical	Ties	P-Value	Decision(5%)
Control		2.5	76	46	2	0.9820	Non-Significant Effect
		5	76	46	2	0.9820	Non-Significant Effect
		10	80	46	1	0.9950	Non-Significant Effect
		25	74.5	46	1	0.9710	Non-Significant Effect
		50	72	46	2	0.9430	Non-Significant Effect
		100	76	46	2	0.9820	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.03935887	0.006559811	6	0.614	0.7180	Non-Significant Effect
Error	0.5237871	0.01068953	49			
Total	0.56314594671130	0.01724934205413	55			

ANOVA Assumptions

Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Mod Levene Equality of Variance	0.614	3.19	0.7180	Equal Variances
Distribution	Shapiro-Wilk Normality	0.694		0.0000	Non-normal Distribution

7d Survival Rate Summary

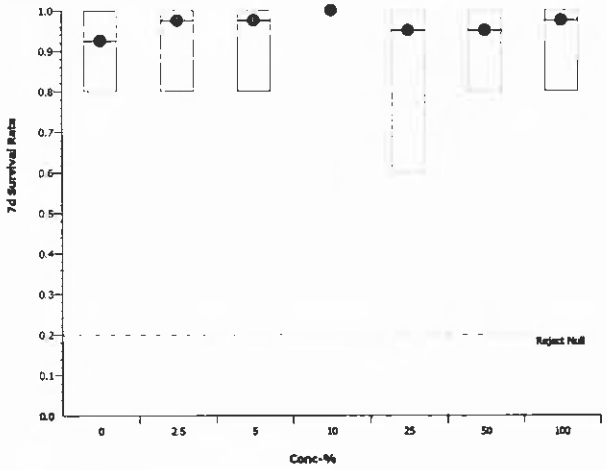
Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.925	0.886	0.964	0.8	1	0.0192	0.104	11.2%	0.0%
2.5		8	0.975	0.948	1	0.8	1	0.0131	0.0707	7.25%	-5.41%
5		8	0.975	0.948	1	0.8	1	0.0131	0.0707	7.25%	-5.41%
10		8	1	1	1	1	1	0	0	0.0%	-8.11%
25		8	0.95	0.896	1	0.6	1	0.0263	0.141	14.9%	-2.7%
50		8	0.95	0.915	0.985	0.8	1	0.0172	0.0926	9.75%	-2.7%
100		8	0.975	0.948	1	0.8	1	0.0131	0.0707	7.25%	-5.41%

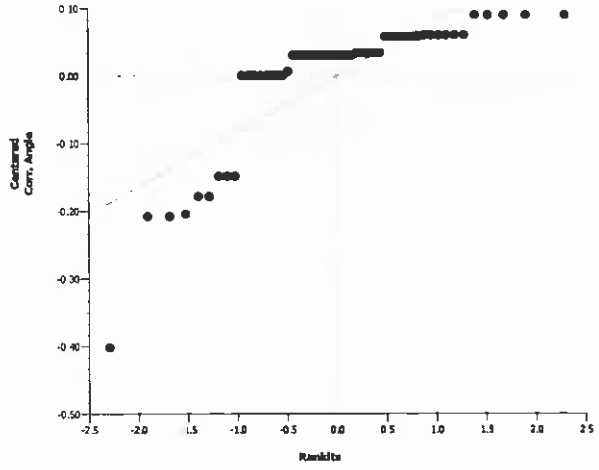
Angular (Corrected) Transformed Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	1.26	1.21	1.3	1.11	1.35	0.0229	0.123	9.81%	0.0%
2.5		8	1.32	1.28	1.35	1.11	1.35	0.0156	0.0842	6.4%	-4.74%
5		8	1.32	1.28	1.35	1.11	1.35	0.0156	0.0842	6.4%	-4.74%
10		8	1.35	1.35	1.35	1.35	1.35	0	0	0.0%	-7.11%
25		8	1.29	1.23	1.35	0.886	1.35	0.0301	0.162	12.6%	-2.54%
50		8	1.29	1.24	1.33	1.11	1.35	0.0205	0.11	8.57%	-2.37%
100		8	1.31	1.28	1.34	1.11	1.35	0.0155	0.0834	6.35%	-4.47%

Chronic Mysid Survival, Growth and Fecundity Test					Pacific EcoRisk
Analysis No:	18-0763-0142	**Endpoint:**	7d Survival Rate	**CETIS Version:**	CETISv1.6.5
Analyzed:	08 Mar-09 16:12	**Analysis:**	Nonparametric-Control vs Treatments	**Official Results:**	Yes

Graphics





Chronic Mysid Survival, Growth and Fecundity Test — **Pacific EcoRisk**

Analysis No:	16-9066-4752	**Endpoint:**	Mean Dry Biomass-mg	**CETIS Version:**	CETISv1.6.5
Analyzed:	08 Mar-09 16:16	**Analysis:**	Parametric-Control vs Treatments	**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	23.9%

Dunnett's Multiple Comparison Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Control		2.5	-1.67	2.36	0.0625	0.9990	Non-Significant Effect
		5	-1.21	2.36	0.0625	0.9940	Non-Significant Effect
		10	-1.8	2.36	0.0625	0.9990	Non-Significant Effect
		25	-1.83	2.36	0.0625	0.9990	Non-Significant Effect
		50	-1.19	2.36	0.0625	0.9930	Non-Significant Effect
		100	-1.71	2.36	0.0625	0.9990	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.01424249	0.002373748	6	0.843	0.5430	Non-Significant Effect
Error	0.1379901	0.002816125	49			
Total	0.15223260316998	0.00518987257965	55			

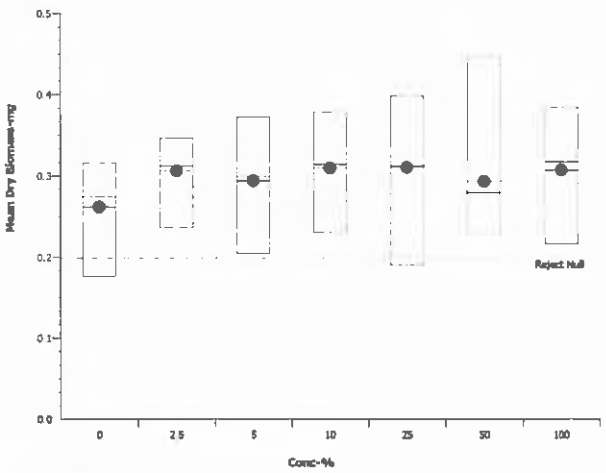
ANOVA Assumptions

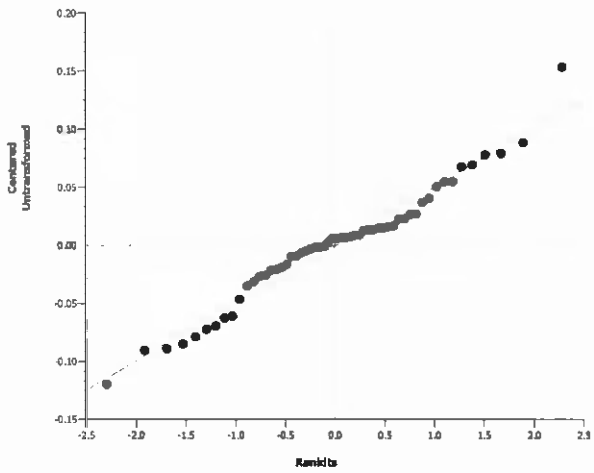
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	3.63	16.8	0.7260	Equal Variances
Distribution	Shapiro-Wilk Normality	0.97		0.1800	Normal Distribution

Mean Dry Biomass-mg Summary

Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.262	0.242	0.281	0.176	0.316	0.00941	0.0507	19.4%	0.0%
2.5		8	0.306	0.293	0.319	0.236	0.346	0.0064	0.0345	11.3%	-16.9%
5		8	0.294	0.275	0.312	0.204	0.372	0.0092	0.0495	16.9%	-12.2%
10		8	0.309	0.293	0.326	0.23	0.378	0.00812	0.0437	14.1%	-18.3%
25		8	0.31	0.287	0.333	0.19	0.398	0.0112	0.0602	19.4%	-18.5%
50		8	0.293	0.268	0.318	0.23	0.446	0.0124	0.0665	22.7%	-12.0%
100		8	0.307	0.284	0.329	0.216	0.384	0.011	0.0595	19.4%	-17.3%

Graphics





CETIS Analytical Report

Chronic Mysid Survival, Growth and Fecundity Test — **Pacific EcoRisk**

Analysis No:	01-1773-8098	**Endpoint:**	Mean Dry Biomass-mg	**CETIS Version:**	CETISv1.6.5
Analyzed:	08 Mar-09 16:16	**Analysis:**	Linear Interpolation (ICPIN)	**Official Results:**	Yes

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	5795186	280	Yes	Two-Point Interpolation

Point Estimates

Level	Conc-%	95% LCL	95% UCL	TU	95% LCL	95% UCL
IC2.5	>100	N/A	N/A	<1	N/A	N/A
IC5	>100	N/A	N/A	<1	N/A	N/A
IC10	>100	N/A	N/A	<1	N/A	N/A
IC15	>100	N/A	N/A	<1	N/A	N/A
IC20	>100	N/A	N/A	<1	N/A	N/A
IC25	>100	N/A	N/A	<1	N/A	N/A
IC40	>100	N/A	N/A	<1	N/A	N/A
IC50	>100	N/A	N/A	<1	N/A	N/A

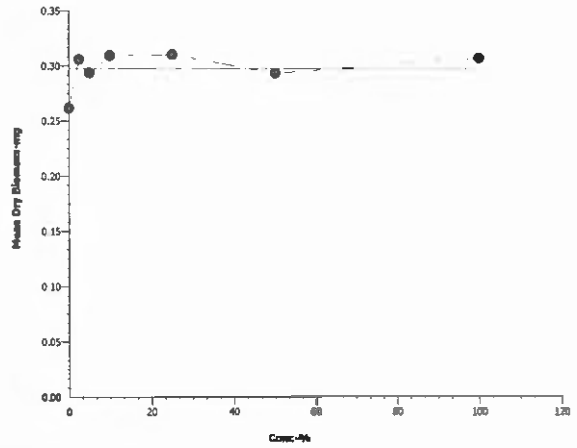
Mean Dry Biomass-mg Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Control	8	0.262	0.176	0.316	0.00925	0.0507	19.4%	0.0%
2.5		8	0.306	0.236	0.346	0.00629	0.0345	11.3%	-16.9%
5		8	0.294	0.204	0.372	0.00904	0.0495	16.9%	-12.2%
10		8	0.309	0.23	0.378	0.00798	0.0437	14.1%	-18.3%
25		8	0.31	0.19	0.398	0.011	0.0602	19.4%	-18.5%
50		8	0.293	0.23	0.446	0.0121	0.0665	22.7%	-12.0%
100		8	0.307	0.216	0.384	0.0109	0.0595	19.4%	-17.3%

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Control	0.316	0.316	0.2	0.27	0.284	0.252	0.278	0.176
2.5		0.328	0.284	0.346	0.236	0.312	0.332	0.312	0.296
5		0.372	0.33	0.274	0.204	0.308	0.302	0.262	0.296
10		0.304	0.378	0.306	0.322	0.274	0.336	0.324	0.23
25		0.308	0.398	0.284	0.19	0.36	0.316	0.308	0.316
50		0.306	0.23	0.446	0.246	0.266	0.292	0.286	0.272
100		0.374	0.29	0.322	0.216	0.234	0.384	0.314	0.32

Graphics



7 Day Chronic *Americamysis bahia* Toxicity Test Test Data

Client: Applied Marine Sciences
Test Material: ~~Storm water~~ Brine
Test ID #: 32190 Project #: 14499
Test Date: 2/25/09

Organism Log#: 4408 Age: 4 days
Organism Supplier: ABS
Control/Diluent: DI & Crystal Sea @ 25 ppt
Control Water Batch: 708

Treatment (% effluent)	Temp (C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	26.9	7.90		8.1		25.0		5	5	5	5	5	5	5	5	Date: 2/25/09
2.5	26.9	7.91		8.2		25.2		5	5	5	5	5	5	5	5	Sample ID: 21508
5	26.9	7.90		8.5		25.2		5	5	5	5	5	5	5	5	Test Solution Prep: JPW
10	26.9	7.88		8.6		25.1		5	5	5	5	5	5	5	5	New WQ: JNC
25	26.9	7.84		8.1		24.9		5	5	5	5	5	5	5	5	Initiation Time: 1500
50	26.9	7.79		8.2		24.5		5	5	5	5	5	5	5	5	Initiation Signoff: JPW
100	26.9	7.73		7.3		24.2		5	5	5	5	5	5	5	5	
Meter ID	30A	pH12		DO12	8.0	EC05										
Control	26.3	8.05	7.64	8.9	~~8.0~~ 6.0	25.1	25.7	5	5	5	5	5	5	5	5	Date: 2/26/09
2.5	26.3	8.05	7.70	8.3	5.2	25.2	25.8	5	5	5	5	5	5	5	5	Sample ID: 21508
5	26.3	8.03	7.77	8.4	4.9	25.2	25.8	5	5	5	5	5	5	5	5	Test Solution Prep: JPW
10	26.3	8.01	7.80	7.9	5.3	25.1	25.7	5	5	5	5	5	5	5	5	New WQ: SL
25	26.3	7.96	7.93	8.0	5.3	24.9	25.7	5	5	5	4	5	5	5	5	Renewal Time: 1330
50	26.3	7.90	8.02	8.2	5.0	24.5	25.5	5	5	5	5	5	5	5	5	Renewal-Sign-off: EKK
100	26.3	7.82	8.14	8.3	4.5	24.0	24.9	5	5	5	5	5	5	5	5	Old WQ: JDN
Meter ID	30A	pH11	pH12	DO14	DO10	EC04	EC01									
Control	26.6	8.04	7.74	7.1	5.1	25.2	25.8	5	5	5	5	5	4	5	5	Date: 2/27/09
2.5	26.6	8.05	7.79	7.4	5.1	25.3	26.6	5	5	5	5	4	5	5	5	Sample ID: 21508
5	26.6	8.04	7.79	7.3	4.7	25.3	26.5	5	5	5	5	5	5	5	5	Test Solution Prep: KO
10	26.6	8.03	7.84	7.4	4.9	25.3	26.4	5	5	5	5	5	5	5	5	New WQ: EKK
25	26.6	7.99	8.07	7.6	5.4	25.2	25.5	5	5	5	4	5	5	5	5	Renewal Time: 1205
50	26.6	7.95	8.17	7.8	5.3	25.4	25.5	5	5	5	4	5	5	5	5	Renewal-Sign-off: KO
100	26.6	7.89	8.30	7.7	5.4	25.3	24.8	5	5	5	5	5	5	5	5	Old WQ: MJM
Meter ID	~~30A~~	pH03	pH03	DO10	DO10	EC05	EC05									
Control	25.7	8.22	7.87	7.3	5.8	25.2	26.1	5	5	5	5	5	4	5	5	Date: 2/28/09
2.5	25.7	8.26	7.92	7.4	5.5	25.2	26.2	5	5	5	5	4	5	5	5	Sample ID: 21508
5	25.7	8.28	7.97	7.4	5.7	25.2	26.1	5	5	5	5	5	5	5	5	Test Solution Prep: EK
10	25.7	8.28	8.02	7.4	5.5	25.0	26.2	5	5	5	5	5	5	5	5	New WQ: JPC
25	25.7	8.25	8.13	7.5	5.4	24.7	26.4	5	5	5	4	5	5	5	5	Renewal Time: 1030
50	25.7	8.22	8.24	7.5	5.3	24.3	26.3	5	5	5	4	5	5	5	5	Renewal-Sign-off: EKK
100	25.7	8.20	8.38	7.3	5.2	24.4	26.5	5	5	5	5	5	5	5	5	Old WQ: DMM
Meter ID	24A	pH03	PH11	DO10	DO12	EC05	EC01									

7 Day Chronic *Americamysis bahia* Toxicity Test Test Data

Client: Applied Marine Sciences
Organism Log#: 4408 Age: 1 days
Test Material: ~~AB Storm water~~ Brine
Organism Supplier: ABS
Test ID #: 32190 Project #: 14499
Control/Diluent: DI & Crystal Sea @ 25 ppt
Test Date: 2/27/09
Control Water Batch: 708

Treatment (% effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	25.4	7.86	7.59	8.5	5.9	24.5	25.9	5	5	5	5	5	4	5	5	Date: 3/1/09
2.5	25.4	7.88	7.58	8.3	5.6	24.6	26.0	5	5	5	5	4	5	5	5	Sample ID: 21508
5	25.4	7.89	7.61	8.4	5.4	24.6	25.9	5	5	5	5	5	5	5	5	Test Solution Prep: JM
10	25.4	7.89	7.68	8.4	5.3	24.6	25.9	5	5	5	5	5	5	5	5	New WQ: SL
25	25.4	7.90	7.78	8.6	5.0	24.5	26.0	5	5	5	4	5	5	5	5	Renewal Time: 1310
50	25.4	7.91	7.96	8.3	5.1	24.4	25.3	5	5	5	4	5	5	5	5	Renewal-Sign-off: PA
100	25.4	7.91	8.12	8.7	5.4	24.0	25.1	5	5	5	5	5	5	5	5	Old WQ: NW
Meter ID	24A	pH11	pH12	DO10	DO12	EC05	EC04									
Control	25.6	7.80	7.48	8.4	6.7	24.7	25.1	5	5	4	5	5	4	5	4	Date: 3/2/09
2.5	25.6	7.82	7.47	8.3	5.8	24.7	25.6	5	5	5	5	4	5	5	5	Sample ID: 21508
5	25.6	7.83	7.49	8.2	5.6	24.7	25.4	5	5	5	5	5	5	5	5	Test Solution Prep: JM
10	25.6	7.84	7.59	8.1	5.7	24.6	25.2	5	5	5	5	5	5	5	5	New WQ: MDM
25	25.6	7.84	7.75	8.2	5.8	24.5	25.4	5	5	5	4	5	5	5	5	Renewal Time: 1330
50	25.6	7.85	7.81	8.3	5.6	24.3	25.3	5	5	5	4	5	5	5	5	Renewal-Sign-off: JM
100	25.6	7.85	8.04	8.1	5.5	24.0	24.9	5	5	5	5	5	5	5	5	Old WQ: AR
Meter ID	24A	pH12	pH12	DO14	DO19	EC05	EC03									
Control	25.7	7.86	7.58	8.9	6.3	24.7	25.4	5	5	4	5	5	4	5	4	Date: 3/3/09
2.5	25.7	7.87	7.70	8.6	5.8	24.9	25.7	5	5	5	5	4	5	5	5	Sample ID: 21508
5	25.7	7.85	7.75	8.3	6.1	24.8	25.4	4	5	5	5	5	5	5	5	Test Solution Prep: JM
10	25.7	7.84	7.83	8.6	5.8	24.8	25.6	5	5	5	5	5	5	5	5	New WQ: SL
25	25.7	7.82	7.99	8.6	5.7	24.7	25.4	5	5	5	3	5	5	5	5	Renewal Time: 1435
50	25.7	7.80	8.01	8.5	5.3	24.6	25.3	5	5	5	4	5	5	5	5	Renewal-Sign-off: JR
100	25.7	7.77	8.26	8.3	5.8	24.1	25.0	5	5	5	5	4	5	5	5	Old WQ: DGV
Meter ID	24A	pH12	pH11	DO14	DO10	EC01	EC04									
Control	25.0		7.84		7.1		25.4	5	5	4	5	5	4	5	4	Date: 3/4/09
2.5	25.0		7.85		6.8		25.4	5	5	5	5	4	5	5	5	Termination-Time: 835
5	25.0		7.90		6.9		25.4	4	5	5	5	5	5	5	5	Termination-Signoff: PA
10	25.0		7.93		6.7		25.4	5	5	5	5	5	5	5	5	Old WQ: SL
25	25.0		8.05		6.6		25.4	5	5	5	3	5	5	5	5	
50	25.0		8.16		6.5		25.2	5	4	5	4	5	5	5	5	
100	25.0		8.27		6.3		25.0	5	5	4	5	4	5	5	6	
Meter ID	24A		pH12		DO14		EC05									

Americamysis bahia Dry Weight and Biomass Value Data

Client: **Applied Marine Sciences** Test ID #: 32190 Project # 14499

Sample: _____ Tare Weight Date: 2-26-09 Sign-off: MC

Test Date: 2/25/09 Final Weight Date: 3-5-09 Sign-off: MC

Pan ID	Concentration (%)	Replicate	Initial Pan Weight (mg)	Pan + Dry Mysid Weight (mg)	Initial # of Organisms	Mysid Biomass Value (mg)
1	Control	A	27.42	29.00	5	0.316
2		B	25.74	27.32	5	0.316
3		C	27.94	28.94	5	0.200
4		D	30.32	31.67	5	0.270
5		E	25.91	27.33	5	0.284
6		F	24.80	26.06	5	0.252
7		G	22.90	24.29	5	0.278
8		H	28.74	29.62	5	0.176
9	2.5	A	23.73	25.37	5	0.328
10		B	26.81	28.23	5	0.284
11		C	27.97	29.70	5	0.346
12		D	24.22	25.40	5	0.236
13		E	25.87	27.43	5	0.312
14		F	30.45	32.11	5	0.332
15		G	27.83	29.39	5	0.312
16		H	31.08	32.56	5	0.296
17	5	A	27.57	28.59	5	0.204
18		B	31.33	32.87	5	0.308
19		C	23.88	25.39	5	0.302
20		D	26.40	27.71	5	0.262
21		E	25.30	26.78	5	0.296
22		F	28.82	30.68	5	0.372
23		G	26.82	28.47	5	0.330
24		H	28.15	29.52	5	0.274
25	10	A	26.51	28.03	5	0.304
26		B	25.19	27.08	5	0.378
27		C	30.13	31.66	5	0.306
28		D	24.88	26.49	5	0.322
29		E	27.26	28.63	5	0.274
30		F	24.89	26.57	5	0.336
31		G	26.88	28.50	5	0.324
32		H	28.48	29.63	5	0.230

Americamysis bahia Dry Weight and Biomass Value Data

Client: **Applied Marine Sciences** Test ID #: 32190 Project # 14499

Sample: _____ Tare Weight Date: 2-26-09 Sign-off: mc

Test Date: 2/25/09 Final Weight Date: 3-5-09 Sign-off: mc

Pan ID	Concentration (%)	Replicate	Initial Pan Weight (mg)	Pan + Dry Mysid Weight (mg)	Initial # of Organisms	Mysid Biomass Value (mg)
33	25	A	26.00	27.54	5	0.308
34		B	24.86	26.85	5	0.398
35		C	28.45	29.87	5	0.284
36		D	25.36	26.31	5	0.190
37		E	26.33	28.13	5	0.360
38		F	26.33	27.91	5	0.316
39		G	25.98	27.52	5	0.308
40		H	25.72	27.30	5	0.316
41	50	A	27.73	29.26	5	0.306
42		B	25.59	29.74	5	0.230
43		C	25.47	27.70	5	0.446
44		D	27.70	28.93	5	0.246
45		E	22.95	24.28	5	0.266
46		F	28.21	29.67	5	0.292
47		G	25.72	27.15	5	0.286
48		H	27.18	28.54	5	0.272
49	100	A	30.53	32.40	5	0.374
50		B	23.94	25.39	5	0.290
51		C	28.43	29.72	~~5~~ 4	0.322
52		D	29.45	30.53	5	0.216
53		E	27.12	28.29	5	0.234
54		F	29.13	31.05	5	0.384
55		G	29.41	30.98	5	0.314
56		H	30.24	31.84	5	0.320
QA 1			26.99	26.99		
QA 2			25.52	25.51		
QA 3			28.65	28.63		
QA 4			26.03	26.03		
QA 5			27.33	27.33		
QA 6			27.20	27.19		

Appendix E

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Americamysis bahia*

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

Test Run No:	13-2398-9421	**Test Type:**	Growth-Survival-Fec (7d)	**Analyst:**	Rivian Villanueva
Start Date:	25 Feb-09 15:45	**Protocol:**	EPA/821/R/02/014 (2002)	**Diluent:**	Laboratory Water
Ending Date:	04 Mar-09 08:45	**Species:**	Americamysis bahia	**Brine:**	Crystal Sea
Duration:	6d 17h	**Source:**	Aquatic Biosystems, CO	**Age:**	7

Sample No:	12-4853-2547	**Code:**	NaCl	**Client:**	Reference Toxicant
Sample Date:	25 Feb-09 15:45	**Material:**	Sodium chloride	**Project:**	14500
Receive Date:	25 Feb-09 15:45	**Source:**	Reference Toxicant		
Sample Age:	N/A (26.7 °C)	**Station:**	In House		

Comparison Summary

Analysis No	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
16-3010-1391	7d Survival Rate	0.5	1	0.707	12.1%		Steel Many-One Rank Test
00-3908-7142	Mean Dry Biomass-mg	0.25	0.5	0.354	17.4%		Steel Many-One Rank Test

Point Estimate Summary

Analysis No	Endpoint	Level	Conc-g/L	95% LCL	95% UCL	TU	Method
15-1823-5578	7d Survival Rate	EC50	0.6	0.546	0.658		Spearman-Kärber
03-4110-6648	Mean Dry Biomass-mg	IC2.5	0.181	0.0306	0.276		Linear Interpolation (ICPIN)
		IC5	0.236	0.0612	0.305		
		IC10	0.293	0.122	0.373		
		IC15	0.343	0.219	0.474		
		IC20	0.392	0.283	0.523		
		IC25	0.441	0.336	0.552		
		IC40	0.566	0.45	0.642		
		IC50	0.638	0.515	0.702		

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	8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	0.0%
0.125		8	0.975	0.949	1	0.8	1	0.0129	0.0707	7.25%	0.0%
0.25		8	1	1	1	1	1	0	0	0.0%	-2.56%
0.5		8	0.75	0.663	0.837	0.4	1	0.0425	0.233	31.1%	23.1%
1		8	0	0	0	0	0	0	0		100.0%
2		8	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	8	0.278	0.263	0.293	0.212	0.328	0.00734	0.0402	14.4%	0.0%
0.125		8	0.279	0.261	0.296	0.212	0.352	0.00859	0.047	16.9%	-0.09%
0.25		8	0.263	0.245	0.28	0.23	0.37	0.00863	0.0473	18.0%	5.57%
0.5		8	0.192	0.167	0.217	0.066	0.262	0.0122	0.0671	34.9%	30.9%
1		8	0	0	0	0	0	0	0		100.0%
2		8	0	0	0	0	0	0	0		100.0%

Chronic Mysid Survival, Growth and Fecundity Test — Pacific EcoRisk

7d Survival Rate Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Lab Water Contr	1	1	0.8	1	1	1	1	1
0.125		1	1	1	0.8	1	1	1	1
0.25		1	1	1	1	1	1	1	1
0.5		1	0.8	0.4	0.8	0.8	1	0.4	0.8
1		0	0	0	0	0	0	0	0
2		0	0	0	0	0	0	0	0

Mean Dry Biomass-mg Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8
0	Lab Water Contr	0.286	0.328	0.228	0.312	0.212	0.288	0.27	0.302
0.125		0.212	0.272	0.282	0.252	0.262	0.352	0.342	0.254
0.25		0.248	0.29	0.236	0.25	0.246	0.232	0.37	0.23
0.5		0.262	0.204	0.126	0.228	0.198	0.262	0.066	0.192
1		0	0	0	0	0	0	0	0
2		0	0	0	0	0	0	0	0

7 Day Chronic *Americamysis bahia* Reference Toxicant Test Data

Client: **Reference Toxicant**
Test Material: Potassium chloride
Test ID #: 32192 Project #: 14500
Test Date: 2/25/09 Randomization: 8.6.1

Organism Log#: 4408
Organism Supplier: ABS
Control/Diluent: DI & Crystal Sea @ 25ppt
Control Water Batch: 708

Treatment (g/L KCl)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	26.7	7.75		7.7		25.2		5	5	5	5	5	5	5	5	Date: 2/25/09
0.125	26.7	7.83		7.9		25.4		5	5	5	5	5	5	5	5	Test Solution Prep: SB
0.25	26.7	7.84		7.8		25.5		5	5	5	5	5	5	5	5	New WQ: AR
0.5	26.7	7.85		8.2		25.5		5	5	5	5	5	5	5	5	Initiation Time: 1545
1	26.7	7.85		8.2		25.6		5	5	5	5	5	5	5	5	Initiation Signoff: PA
2	26.7	7.84		9.8		26.2		5	5	5	5	5	5	5	5	
Meter ID	30A	pH11		DO14		Eco1										
Control	26.7	7.94	7.65	7.3	5.4	25.0	25.8	5	5	4	5	5	5	5	5	Date: 2/26/09
0.125	26.7	8.00	7.69	7.0	4.7	25.3	26.1	5	5	5	4	5	5	5	5	Test Solution Prep: PA
0.25	26.7	8.01	7.70	7.1	5.5	25.1	26.1	5	5	5	5	5	5	5	5	New WQ: VK
0.5	26.7	8.02	7.70	7.2	5.6	25.5	26.1	5	4	5	5	4	5	4	5	Renewal Time: 1410
1	26.7	8.02	7.71	7.0	5.7	26.1	26.4	0	1	0	0	0	0	1	0	Renewal-Sign-off: PA
2	26.7	8.02	7.70	6.9	5.7	27.2	26.8	0	0	0	0	0	0	0	0	Old WQ: MJM
Meter ID	30A	pH03	pH11	DO12	DO14	EC05	EC04									
Control	26.7	8.08	7.67	7.2	5.8	25.3	25.8	5	5	4	5	5	5	5	5	Date: 2/27/09
0.125	26.7	8.10	7.80	7.6	5.7	25.4	26.0	5	5	5	4	5	5	5	5	Test Solution Prep: KO
0.25	26.7	8.11	7.78	7.7	5.7	25.5	26.2	5	5	5	5	5	5	5	5	New WQ: EKK
0.5	26.7	8.11	7.77	7.7	5.4	25.7	26.2	5	4	4	4	4	5	4	5	Renewal Time: 1040
1	26.7	8.13	7.85	8.0	5.9	25.9	26.9	–	0	–	–	–	–	0	–	Renewal-Sign-off: KO
2	26.7	–	–	–	–	–	–	–	–	–	–	–	–	–	–	Old WQ: MJM
Meter ID	30A	pH 03	pH03	DO 10	DO 10	EC 05	EC05									
Control	25.6	8.16	7.94	7.3	6.7	25.2	26.1	5	5	4	5	5	5	5	5	Date: 2/28/09
0.125	25.6	8.17	7.95	7.2	5.7	25.5	26.1	5	5	5	4	5	5	5	5	Test Solution Prep: SB
0.25	25.6	8.18	7.97	7.3	5.6	25.6	26.2	5	5	5	5	5	5	5	5	New WQ: QMP
0.5	25.6	8.19	7.99	7.5	5.7	25.8	26.3	5	4	2	4	4	5	4	5	Renewal Time: 0940
1	25.6	8.20	–	7.7	–	26.3	–	–	–	–	–	–	–	–	–	Renewal-Sign-off: EKK
2	25.6	–	–	–	–	–	–	–	–	–	–	–	–	–	–	Old WQ: JPL
Meter ID	24A	pH11	pH12	DO12	DO10	EC01	EC05									

7 Day Chronic *Americamysis bahia* Reference Toxicant Test Data

Client: **Reference Toxicant** Organism Log#: 4408

Test Material: Potassium chloride Organism Supplier: ABS

Test ID #: 32192 Project #: 14500 Control/Dilluent: DI & Crystal Sea @ 25ppt

Test Date: 2/25/09 Randomization: 8.6.1 Control Water Batch: 708

Treatment (g/L KCl)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt) new	Salinity (ppt) old	# Live Organisms A	B	C	D	E	F	G	H	SIGN-OFF
Control	25.5	~~8.9~~ 7.88	7.70	8.3	6.8	24.2	25.8	5	5	4	5	5	5	5	5	Date: 3/1/09
0.125	25.5	7.89	7.62	7.9	6.2	24.7	27.0	5	5	5	4	5	5	5	5	Test Solution Prep: PA
0.25	25.5	7.90	7.61	8.4	5.9	24.8	27.2	5	5	5	5	5	5	5	5	New WQ: SL
0.5	25.5	7.94	7.68	8.5	6.0	25.1	26.6	5	4	2	4	4	5	4	4	Renewal Time: 1030
1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Renewal-Sign-off: JPL
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Old WQ: SL
Meter ID	24A	pH11	pH12	DO10	DO12	EC01	EC04									
Control	25.7	7.75	7.54	8.2	6.7	24.5	24.9	5	6	4	5	5	5	5	5	Date: 3/2/09
0.125	25.7	7.81	7.55	8.0	6.0	24.8	25.6	5	5	5	4	5	5	5	5	Test Solution Prep: RV
0.25	25.7	7.82	7.54	7.9	5.7	25.0	25.9	5	5	5	5	5	5	5	5	New WQ: MJM
0.5	25.7	7.84	7.54	8.3	5.9	25.2	25.8	5	4	2	4	4	5	4	4	Renewal Time: 1030
1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Renewal-Sign-off: JW
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Old WQ: MJM
Meter ID	24A	pH12	pH03	DO14	DO16	EC05	EC01									
Control	~~26.8~~	7.86	7.76	7.7	6.7	24.6	24.9	5	5	4	5	5	5	5	5	Date: 3/2/09
0.125	25.8	7.87	7.72	7.8	6.4	24.9	25.7	5	5	5	4	5	5	5	5	Test Solution Prep: JW
0.25	25.8	7.90	7.72	8.0	6.3	25.1	25.9	5	5	5	5	5	5	5	5	New WQ: DBJ
0.5	25.8	7.93	7.71	8.1	6.3	25.4	26.0	5	4	2	4	4	5	4	4	Renewal Time: 1015
1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Renewal-Sign-off: JLR
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	Old WQ: MEL
Meter ID	24A	pH11	pH03	DO10	DO12	EC04	EC05									
Control	25.0		7.77		6.6		25.0	5	5	4	5	5	5	5	5	Date: 3/4/09
0.125	25.0		7.76		6.7		25.6	5	5	5	4	5	5	5	5	Termination Time: 0845
0.25	25.0		7.75		6.8		25.8	5	5	5	5	5	5	5	5	Final Sign-off: JLR
0.5	25.0		7.75		6.9		26.1	5	4	2	4	4	5	2	4	Old WQ: SL
1	—		—		—		—	—	—	—	—	—	—	—	—	
2	—		—		—		—	—	—	—	—	—	—	—	—	
Meter ID	24A		pH12		DO14		EC04									

Appendix F

Test Data and Summary of Statistics for the Evaluation of the Chronic Toxicity of the Bay Area Regional Desalination Project Effluent to *Menidia beryllina*

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Batch ID:	16-3894-6560	**Test Type:**	Growth-Survival (7d)	**Analyst:**	Jason Walker
Start Date:	26 Feb-09 11:00	**Protocol:**	EPA/821/R/02/014 (2002)	**Diluent:**	Laboratory Water
Ending Date:	05 Mar-09 08:30	**Species:**	Menidia beryllina	**Brine:**	Crystal Sea
Duration:	6d 21h	**Source:**	Aquatic Biosystems, CO	**Age:**	11

Sample ID:	08-7787-0147	**Code:**	Brine	**Client:**	AMS
Sample Date:	25 Feb-09 09:15	**Material:**	Effluent	**Project:**	14499
Receive Date:	25 Feb-09 10:35	**Source:**	Applied Marine Sciences		
Sample Age:	26h (13.7 °C)	**Station:**	MWHA		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
08-8190-6186	7d Survival Rate	100	>100	N/A	6.17%	1	Steel Many-One Rank Test
04-8325-9227	Mean Dry Biomass-mg	100	>100	N/A	10.1%	1	Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis ID	Endpoint	Level	%	95% LCL	95% UCL	TU	Method
09-7075-4299	Mean Dry Biomass-mg	IC5	>100	N/A	N/A	<1	Linear Interpolation (ICPIN)
		IC10	>100	N/A	N/A	<1	
		IC15	>100	N/A	N/A	<1	
		IC20	>100	N/A	N/A	<1	
		IC25	>100	N/A	N/A	<1	
		IC40	>100	N/A	N/A	<1	
		IC50	>100	N/A	N/A	<1	

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	1	1	1	1	1	0	0	0.0%	0.0%
2.5		4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	2.5%
5		4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	2.5%
10		4	1	1	1	1	1	0	0	0.0%	0.0%
25		4	0.975	0.956	0.994	0.9	1	0.00913	0.05	5.13%	2.5%
50		4	1	1	1	1	1	0	0	0.0%	0.0%
100		4	1	1	1	1	1	0	0	0.0%	0.0%

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	0.94	0.931	0.949	0.911	0.971	0.00463	0.0254	2.7%	0.0%
2.5		4	0.915	0.901	0.929	0.866	0.944	0.00663	0.0363	3.97%	2.66%
5		4	0.899	0.87	0.929	0.788	0.956	0.0144	0.079	8.78%	4.34%
10		4	0.893	0.87	0.916	0.84	0.966	0.011	0.0604	6.76%	5.0%
25		4	0.824	0.8	0.848	0.741	0.895	0.0118	0.0645	7.83%	12.3%
50		4	0.935	0.91	0.96	0.867	1.02	0.0124	0.068	7.27%	0.53%
100		4	0.966	0.96	0.973	0.952	0.988	0.00303	0.0166	1.72%	-2.82%

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	1	1
2.5		1	1	0.9	1
5		1	0.9	1	1
10		1	1	1	1
25		0.9	1	1	1
50		1	1	1	1
100		1	1	1	1

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	0.971	0.911	0.947	0.931
2.5		0.909	0.941	0.866	0.944
5		0.955	0.788	0.898	0.956
10		0.966	0.847	0.919	0.84
25		0.741	0.815	0.895	0.846
50		0.867	0.957	0.896	1.02
100		0.955	0.952	0.971	0.988

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis ID: 08-8190-6186 Endpoint: 7d Survival Rate CETIS Version: CETISv1.7.0
Analyzed: 11 Mar-09 15:37 Analysis: Nonparametric-Control vs Treatments Official Results: Yes

Data Transform	Zeta	Alt Hyp	Monte Carlo	NOEL	LOEL	TOEL	TU	PMSD
Angular (Corrected)		C > T	Not Run	100	>100	N/A	1	6.17%

Steel Many-One Rank Test

Control	vs	Conc-%	Test Stat	Critical	Ties	P-Value	Decision(5%)
Lab Water Control		2.5	16	10	1	0.6450	Non-Significant Effect
		5	16	10	1	0.6450	Non-Significant Effect
		10	18	10	1	0.8571	Non-Significant Effect
		25	16	10	1	0.6450	Non-Significant Effect
		50	18	10	1	0.8571	Non-Significant Effect
		100	18	10	1	0.8571	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.01138257	0.001897095	6	0.667	0.6774	Non-Significant Effect
Error	0.0597585	0.002845643	21			
Total	0.07114106	0.004742738	27			

ANOVA Assumptions

Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Mod Levene Equality of Variance	0.667	3.81	0.6774	Equal Variances
Distribution	Shapiro-Wilk Normality	0.653		<0.0001	Non-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	1	1	1	1	1	0	0	0.0%	0.0%
2.5		4	0.975	0.956	0.994	0.9	1	0.00928	0.05	5.13%	2.5%
5		4	0.975	0.956	0.994	0.9	1	0.00928	0.05	5.13%	2.5%
10		4	1	1	1	1	1	0	0	0.0%	0.0%
25		4	0.975	0.956	0.994	0.9	1	0.00928	0.05	5.13%	2.5%
50		4	1	1	1	1	1	0	0	0.0%	0.0%
100		4	1	1	1	1	1	0	0	0.0%	0.0%

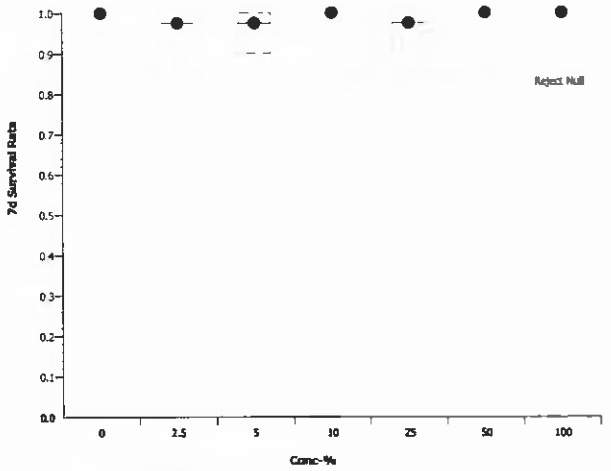
Angular (Corrected) Transformed Summary

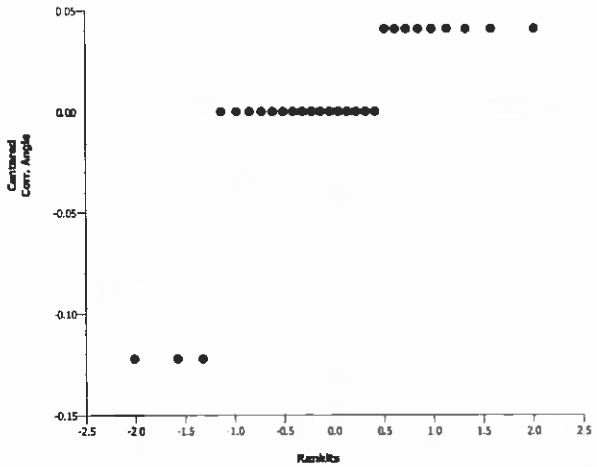
Conc-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Cont	4	1.41	1.41	1.41	1.41	1.41	0	0	0.0%	0.0%
2.5		4	1.37	1.34	1.4	1.25	1.41	0.0151	0.0815	5.94%	2.89%
5		4	1.37	1.34	1.4	1.25	1.41	0.0151	0.0815	5.94%	2.89%
10		4	1.41	1.41	1.41	1.41	1.41	0	0	0.0%	0.0%
25		4	1.37	1.34	1.4	1.25	1.41	0.0151	0.0815	5.94%	2.89%
50		4	1.41	1.41	1.41	1.41	1.41	0	0	0.0%	0.0%
100		4	1.41	1.41	1.41	1.41	1.41	0	0	0.0%	0.0%

Chronic Larval Fish Survival and Growth Test — **Pacific EcoRisk**

Analysis ID:	08-8190-6186	**Endpoint:**	7d Survival Rate	**CETIS Version:**	CETISv1.7.0
Analyzed:	11 Mar-09 15:37	**Analysis:**	Nonparametric-Control vs Treatments	**Official Results:**	Yes

Graphics





Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis ID:	04-8325-9227	**Endpoint:**	Mean Dry Biomass-mg	**CETIS Version:**	CETISv1.7.0
Analyzed:	11 Mar-09 15:40	**Analysis:**	Parametric-Control vs Treatments	**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	10.1%

Dunnett's Multiple Comparison Test

Control	vs	Conc-%	Test Stat	Critical	MSD	P-Value	Decision(5%)
Lab Water Control		2.5	0.647	2.45	0.0946	0.6163	Non-Significant Effect
		5	1.05	2.45	0.0946	0.4290	Non-Significant Effect
		10	1.22	2.45	0.0946	0.3584	Non-Significant Effect
		25*	3	2.45	0.0946	0.0161	Significant Effect
		50	0.129	2.45	0.0946	0.8193	Non-Significant Effect
		100	-0.686	2.45	0.0946	0.9697	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(5%)
Between	0.04999426	0.008332376	6	2.79	0.0372	Significant Effect
Error	0.06269658	0.002985551	21			
Total	0.1126908	0.01131793	27			

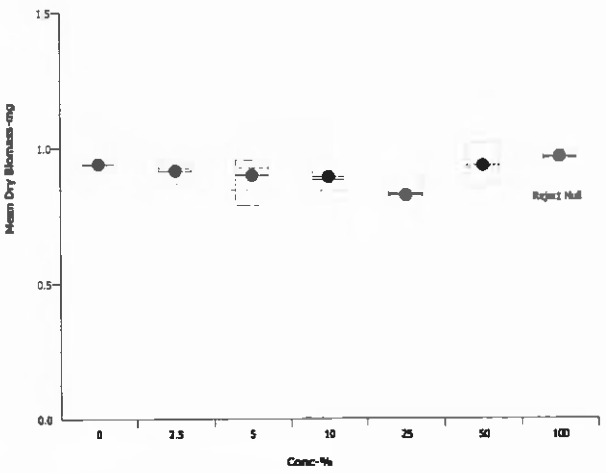
ANOVA Assumptions

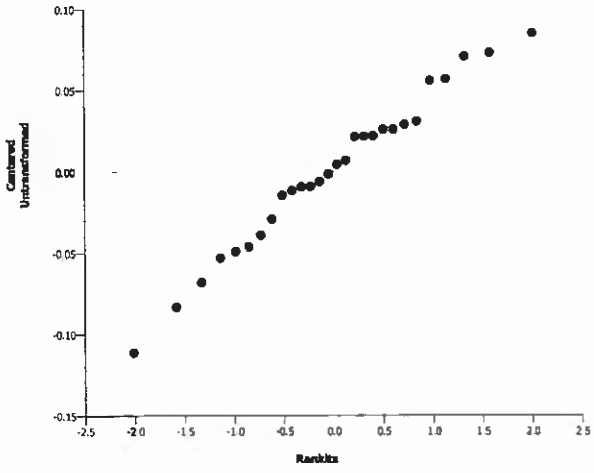
Attribute	Test	Test Stat	Critical	P-Value	Decision(1%)
Variances	Bartlett Equality of Variance	8.07	16.8	0.2330	Equal Variances
Distribution	Shapiro-Wilk Normality	0.98		0.8420	Normal Distribution

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	0.94	0.93	0.95	0.911	0.971	0.00471	0.0254	2.7%	0.0%
2.5		4	0.915	0.901	0.929	0.866	0.944	0.00674	0.0363	3.97%	2.66%
5		4	0.899	0.869	0.929	0.788	0.956	0.0147	0.079	8.78%	4.34%
10		4	0.893	0.87	0.916	0.84	0.966	0.0112	0.0604	6.76%	5.0%
25		4	0.824	0.8	0.849	0.741	0.895	0.012	0.0645	7.83%	12.3%
50		4	0.935	0.909	0.961	0.867	1.02	0.0126	0.068	7.27%	0.53%
100		4	0.966	0.96	0.973	0.952	0.988	0.00308	0.0166	1.72%	-2.82%

Graphics





Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

Analysis ID:	09-7075-4299	**Endpoint:**	Mean Dry Biomass-mg	**CETIS Version:**	CETISv1.7.0
Analyzed:	11 Mar-09 15:40	**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	>100	N/A	N/A	<1	N/A	N/A
IC10	>100	N/A	N/A	<1	N/A	N/A
IC15	>100	N/A	N/A	<1	N/A	N/A
IC20	>100	N/A	N/A	<1	N/A	N/A
IC25	>100	N/A	N/A	<1	N/A	N/A
IC40	>100	N/A	N/A	<1	N/A	N/A
IC50	>100	N/A	N/A	<1	N/A	N/A

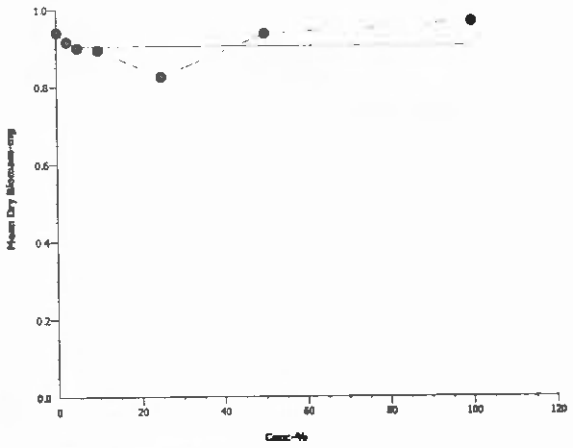
Mean Dry Biomass-mg Summary

			Calculated Variate						
Conc-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	Diff%
0	Lab Water Contro	4	0.94	0.911	0.971	0.00463	0.0254	2.7%	0.0%
2.5		4	0.915	0.866	0.944	0.00663	0.0363	3.97%	2.66%
5		4	0.899	0.788	0.956	0.0144	0.079	8.78%	4.34%
10		4	0.893	0.84	0.966	0.011	0.0604	6.76%	5.0%
25		4	0.824	0.741	0.895	0.0118	0.0645	7.83%	12.3%
50		4	0.935	0.867	1.02	0.0124	0.068	7.27%	0.53%
100		4	0.966	0.952	0.988	0.00303	0.0166	1.72%	-2.82%

Mean Dry Biomass-mg Detail

Conc-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Control	0.971	0.911	0.947	0.931
2.5		0.909	0.941	0.866	0.944
5		0.955	0.788	0.898	0.956
10		0.966	0.847	0.919	0.84
25		0.741	0.815	0.895	0.846
50		0.867	0.957	0.896	1.02
100		0.955	0.952	0.971	0.988

Graphics



7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Applied Marine Sciences
Test Material: AB effluent Brine
Test ID#: 32191 Project #: 14499
Test Date: 2/26/09

Organism Log#: 4409 Age: 11 days
Organism Supplier: ABS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 108

Treatment (% Effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.7	8.05		8.9		25.1	10	10	10	10	Date: 2/26/09
2.5	25.7	8.05		8.3		25.2	10	10	10	10	Sample ID 21508
5	25.7	8.03		8.4		25.2	10	10	10	10	Test Solution Prep JPL
10	25.7	8.01		7.9		25.1	10	10	10	10	New WQ SL
25	25.7	7.96		8.0		24.9	10	10	10	10	Initiation Time 1100
50	25.7	7.90		8.2		24.5	10	10	10	10	Initiation Signoff JPL
100	25.7	7.82		8.3		24.0	10	10	10	10	
Meter ID	24A	pH11		DO14		EC04					
Control	25.5	8.04	7.76	7.1	5.9	25.2	10	10	10	10	Date 2/27/09
2.5	25.5	8.05	7.82	7.4	5.8	25.3	10	10	10	10	Sample ID 21508
5	25.6	8.04	7.84	7.3	5.6	25.3	10	10	10	10	Test Solution Prep KO
10	25.5	8.03	7.92	7.4	5.8	25.3	10	10	10	10	New WQ EKK
25	25.5	7.91	8.02	7.6	5.7	25.2	10	10	10	10	Renewal Time 1115
50	25.5	7.95	8.10	7.8	5.7	25.4	10	10	10	10	Renewal Signoff KO
100	25.5	7.89	8.19	7.7	5.7	25.3	10	10	10	10	Old WQ MJM
Meter ID	24A	pH03	pH03	DO10	DO10	EC05					
Control	25.5	8.22	8.02	7.3	5.7	25.2	10	10	10	10	Date 2/28/09
2.5	25.5	8.26	8.10	7.4	5.9	25.2	10	10	9	10	Sample ID 21508
5	25.5	8.29	8.09	7.4	5.7	25.2	10	10	10	10	Test Solution Prep KO
10	25.5	8.28	8.18	7.4	6.0	25.0	10	10	10	10	New WQ JPL
25	25.5	8.25	8.28	7.5	6.0	24.7 JPL	9	10	10	10	Renewal Time 1035
50	25.5	8.22	8.36	7.5	6.0	24.3	10	10	10	10	Renewal Signoff KO
100	25.5	8.20	8.45	7.3	5.9	24.4	10	10	10	10	Old WQ JPL
Meter ID	24A	pH03	pH12	DO10	DO10	EC05					
Control	25.5	7.86	7.61	8.5	6.3	24.5	10	10	10	10	Date 3/1/09
2.5	25.5	7.88	7.69	8.3	6.2	24.6	10	10	9	10	Sample ID 21508
5	25.5	7.89	7.67	8.4	6.1	24.6	10	10	10	10	Test Solution Prep JPL
10	25.5	7.89	7.78	8.4	6.2	24.6	10	10	10	10	New WQ SL
25	25.5	7.90	7.90	8.6	6.2	24.5	9	10	10	10	Renewal Time 1000
50	25.5	7.91	7.99	8.3	6.2	24.4	10	10	10	10	Renewal Signoff PA
100	25.5	7.91	8.10	8.7	6.0	24.0	10	10	10	10	Old WQ SL
Meter ID	24A	pH11	pH12	DO10	DO12	EC05					

7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Applied Marine Sciences
Test Material: ~~effluent~~ Brine
Test ID#: 32191 Project #: 14499
Test Date: 2/26/09

Organism Log#: 2/4/09 Age: 11 days
Organism Supplier: ABS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 705

Treatment (% Effluent)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.5	7.80	7.36	8.4	5.9	24.7	10	10	10	10	Date 3/2/09
2.5	25.5	7.82	7.45	8.3	5.8	24.7	10	10	9	10	Sample ID 2508
5	25.5	7.83	7.50	8.2	5.8	24.7	10	10	10	10	Test Solution Prep JW
10	25.5	7.84	7.57	8.1	5.8	24.6	10	10	10	10	New WQ AGM
25	25.5	7.84	7.70	8.2	5.8	24.5	9	10	10	10	Renewal Time 0930
50	25.5	7.85	7.86	8.3	5.8	24.3	10	10	10	10	Renewal Signoff PA
100	25.5	7.85	8.02	8.1	5.4	24.0	10	10	10	10	Old WQ MGM
Meter ID	24A	pH12	pH12	DO14	DO14	EC05					
Control	25.5	7.86	7.63	8.8	6.1	24.7	10	10	10	10	Date 3/3/09
2.5	25.5	7.87	7.70	8.6	6.0	24.9	10	10	9	10	Sample ID 2508
5	25.5	7.85	7.73	8.3	6.0	24.8	10	10	10	10	Test Solution Prep JW
10	25.5	7.84	7.81	8.6	6.1	24.8	10	10	10	10	New WQ SL
25	25.5	7.82	7.94	8.6	6.1	24.7	~~10~~ 9	10	10	10	Renewal Time 1030
50	25.5	7.90	8.04	8.5	5.9	24.6	10	10	~~10~~ 9	10	Renewal Signoff JR
100	25.5	7.77	8.17	8.3	6.0	24.1	10	10	10	10	Old WQ MEL
Meter ID	24A	pH12	pH03	DO14	DO12	EC01					
Control	25.2	8.08	7.77	7.9	6.7	24.6	10	10	10	10	Date 3/4/09
2.5	25.2	8.06	7.79	7.9	6.4	24.8	10	10	9	10	Sample ID 2508
5	25.2	8.04	7.82	7.8	6.4	24.7	10	10	10	10	Test Solution Prep JW
10	25.2	8.01	7.90	8.0	6.4	24.7	10	10	10	10	New WQ SL
25	25.2	7.95	8.01	8.0	6.3	24.6	9	10	10	10	Renewal Time 1035
50	25.2	7.89	8.12	8.2	6.3	24.4	10	10	10	10	Renewal Signoff PA
100	25.2	7.83	8.23	8.6	6.3	24.1	10	10	10	10	Old WQ SL
Meter ID	24A	pH12	pH12	DO14	DO14	EC04					
Control	25.6		7.63		6.9	25.0	10	10	10	10	Date 3/5/09
2.5	25.6		7.72		6.9	25.1	10	10	9	10	Termination Time 0830
5	25.6		7.78		6.8	25.1	10	9	10	10	Termination Signoff ~~08~~ JR
10	25.6		7.85		6.8	25.2	10	10	10	10	Old WQ MC
25	25.6		7.95		6.8	~~24.9~~ 24.9	9	10	10	10	
50	25.6		8.09		6.3	24.9	10	10	10	10	
100	25.6		8.21		5.8	24.7	10	10	10	10	
Meter ID	24A		pH11		DO12	EC04					

Chronic Inland Silverside Dry Weight and Biomass Data

Client: Applied Marine Sciences Test ID #: 32191 Project #: 14499

Sample: Effluent Tare Weight Date: 2-26-09 Sign-off: MC

Test Date: 2/~~27~~28/09 Final Weight Date: 3-9-09 Sign-off: MC

Pan ID	Concentration	Replicate	Initial Pan Weight (mg)	Final Pan Weight (mg)	Initial # of Organisms	Biomass Value (mg)
1	Control	A	169.82	179.53	10	0.971
2		B	165.71	~~174.2~~ 174.82	10	0.911
3		C	152.55	162.02	10	0.947
4		D	167.61	176.92	10	~~0.957~~ 0.931
5	2.5	A	163.28	172.37	10	0.909
6		B	162.27	171.68	10	0.941
7		C	152.27	160.93	10	0.866
8		D	152.96	162.40	10	0.944
9	5	A	150.73	160.28	10	0.955
10		B	143.60	151.48	10	0.788
11		C	170.43	179.41	10	0.898
12		D	170.95	180.51	10	0.956
13	10	A	165.82	175.48	10	0.966
14		B	164.43	172.90	10	0.847
15		C	171.25	180.44	10	0.919
16		D	173.73	182.13	10	0.84
17	25	A	179.38	187.29	10	0.791
18		B	167.82	175.97	10	0.815
19		C	153.96	162.91	10	0.895
20		D	170.56	179.02	10	0.846
21	50	A	163.67	172.34	10	0.867
22		B	164.35	173.92	10	0.957
23		C	172.46	181.42	10	0.896
24		D	174.57	184.77	10	~~0~~ 1.02
25	100	A	155.14	164.69	10	0.955
26		B	151.66	161.18	10	0.952
27		C	164.04	173.75	10	0,971
28		D	172.46	182.34	10	0.988
QA 1			160.89	160.90		
QA 2			162.69	162.70		
QA 3			169.42	169.414		
Balance ID			#1	#1		

Appendix G

Test Data and Summary of Statistics for the Reference Toxicant Evaluation of the *Menidia beryllina*

Chronic Larval Fish Survival and Growth Test — **Pacific EcoRisk**

Batch ID:	07-1664-1438	**Test Type:**	Growth-Survival (7d)	**Analyst:**	Rivian Villanueva
Start Date:	26 Feb-09 13:50	**Protocol:**	EPA/821/R/02/014 (2002)	**Diluent:**	Laboratory Water
Ending Date:	05 Mar-09 09:35	**Species:**	Menidia beryllina	**Brine:**	Crystal Sea
Duration:	6d 20h	**Source:**	Aquatic Biosystems, CO	**Age:**	11

Sample ID:	15-4866-4699	**Code:**	KCl	**Client:**	Reference Toxicant
Sample Date:	26 Feb-09 13:50	**Material:**	Potassium chloride	**Project:**	14501
Receive Date:	26 Feb-09 13:50	**Source:**	Reference Toxicant		
Sample Age:	N/A (25.7 °C)	**Station:**	In House		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
17-6451-2614	7d Survival Rate	1	1.25	1.12	10.3%		Steel Many-One Rank Test
09-1484-7067	Mean Dry Biomass-mg	1	1.25	1.12	19.2%		Dunnett's Multiple Comparison Test

Point Estimate Summary

Analysis ID	Endpoint	Level	g/L	95% LCL	95% UCL	TU	Method
04-4565-4745	7d Survival Rate	EC5	0.91	0.801	0.981		Linear Regression (MLE)
		EC10	0.966	0.869	1.03		
		EC15	1.01	0.917	1.07		
		EC20	1.04	0.956	1.09		
		EC25	1.07	0.991	1.12		
		EC40	1.14	1.08	1.19		
		EC50	1.19	1.14	1.25		
13-7333-3884	Mean Dry Biomass-mg	IC5	0.68	N/A	1.23		Linear Interpolation (ICPIN)
		IC10	0.915	0.27	1.14		
		IC15	1.02	0.535	1.14		
		IC20	1.06	0.785	1.19		
		IC25	1.09	0.973	1.24		
		IC40	1.2	1.08	1.35		
		IC50	1.27	1.13	1.37		

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	1	1	1	1	1	0	0	0.0%	0.0%
0.5		4	1	1	1	1	1	0	0	0.0%	0.0%
1		4	0.85	0.813	0.887	0.8	1	0.0183	0.1	11.8%	15.0%
1.25		4	0.4	0.347	0.453	0.3	0.6	0.0258	0.141	35.4%	60.0%
1.5		4	0.075	0.0563	0.0937	0	0.1	0.00913	0.05	66.7%	92.5%
2		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	1.03	0.989	1.08	0.898	1.13	0.0213	0.116	11.3%	0.0%
0.5		4	1.03	0.995	1.06	0.952	1.11	0.0152	0.0831	8.1%	0.65%
1		4	0.912	0.873	0.952	0.821	1.06	0.0194	0.106	11.7%	11.7%
1.25		4	0.543	0.477	0.61	0.407	0.791	0.0324	0.177	32.6%	47.4%
1.5		4	0.125	0.0932	0.157	0	0.187	0.0157	0.086	68.6%	87.9%
2		4	0	0	0	0	0	0	0		100.0%

Chronic Larval Fish Survival and Growth Test — Pacific EcoRisk

7d Survival Rate Detail

Conc-g/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Water Contr	1	1	1	1
0.5		1	1	1	1
1		0.8	1	0.8	0.8
1.25		0.3	0.6	0.4	0.3
1.5		0	0.1	0.1	0.1
2		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	1.13	0.973	0.898	1.13
0.5		1.09	0.952	1.11	0.957
1		0.863	1.06	0.901	0.821
1.25		0.424	0.791	0.552	0.407
1.5		0	0.175	0.139	0.187
2		0	0	0	0

7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Reference Toxicant
Test Material: Potassium Chloride
Test ID#: 32193 Project #: 14501
Test Date: 2/26/09 PAsource # 4.4.6

Organism Log#: 4409 Age: PA 10d 11d
Organism Supplier: ABS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 708

Treatment (g KCl/L)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.7	7.97		7.4		25.1	[illegible]	10	10	10	Date 2/26/09
0.5	25.7	7.99		7.3		25.8	10	10	10	10	Sample ID —
1	25.7	7.98		7.7		24.8	10	10	10	10	Test Solution Prep PA
1.25	25.7	8.01		7.6		26.3	10	10	10	10	New WQ MJM
1.5	25.7	8.00		8.0		27.2	10	10	10	10	Initiation Time 1380
2	25.7	8.01		7.6		26.5	10	10	10	10	Initiation Signoff JL
Meter ID	24A	pH11		DO14		EC04					
Control	25.5	8.03	7.75	6.7	6.6	25.1	10	10	10	10	Date 2/27/09
0.5	26.6	8.05	7.81	6.9	5.7	25.7	10	10	10	10	Sample ID —
1	25.5	8.05	7.79	7.0	5.7	26.1	8	10	10	10	Test Solution Prep KO
1.25	26.5	8.06	7.80	7.2	5.6	26.4	9	10	10	10	New WQ [illegible]
1.5	26.5	8.06	7.80	7.1	5.8	26.5	5	6	6	6	Renewal Time 1030
2	26.5	8.06	7.76	7.1	5.6	27.1	0	0	0	0	Renewal Signoff JL
											Old WQ MJM
Meter ID	24A	pH [illegible]		DO10	DO10	EC05					
Control	25.5	8.45	8.03	7.7	6.4	25.2	10	10	10	10	Date 2/28/09
0.5	25.5	8.46	8.03	7.7	6.1	25.8	10	10	10	10	Sample ID —
1	25.5	8.46	8.04	7.8	6.1	26.2	8	10	9	10	Test Solution Prep JL
1.25	25.5	8.46	8.03	7.9	5.0	26.3	6	7	7	9	New WQ JPL
1.5	25.5	8.46	8.07	7.9	6.1	26.6	2	3	4	5	Renewal Time 1000
2	—	—	—	—	—	—	[illegible]	[illegible]	[illegible]	[illegible]	Renewal Signoff KO
											Old WQ JPL
Meter ID	24A	pH03	pH12	DO10	DO10	EC05					
Control	25.6	7.90	7.67	8.4	7.4	24.3	10	10	10	10	Date 3/1/09
0.5	25.6	7.91	7.61	8.5	6.6	25.0	10	10	10	10	Sample ID —
1	25.6	7.90	7.67	8.4	6.4	25.6	8	10	9	10	Test Solution Prep PA
1.25	25.6	7.90	7.68	8.5	6.5	25.7	5	6	7	9	New WQ SL
1.5	25.6	7.89	7.66	8.6	6.0	25.8	2	1	2	3	Renewal Time 930
2	—	—	—	—	—	—	—	—	—	—	Renewal Signoff PA
											Old WQ SL
Meter ID	24A	pH 11	pH12	DO10	DO12	EC01					

7 Day Chronic Inland Silverside (*M. beryllina*) Toxicity Test Data

Client: Reference Toxicant
Test Material: Potassium Chloride
Test ID#: 32193 Project #: 14501
Test Date: 2/26/09 RANDOMIZE # 4.6.6

Organism Log#: 4409 Age: 10d 11d
Organism Supplier: ABS
Control/Diluent: DI + Crystal Sea @ 25 ppt
Control Water Batch: 708

Treatment (g KCl/L)	Temp (°C)	pH new	pH old	D.O. (mg/L) new	D.O. (mg/L) old	Salinity (ppt)	# Live Organisms A	B	C	D	SIGN-OFF
Control	25.5	7.77	7.48	9.2	5.7	24.4	10	10	10	10	Date 3/2/09
0.5	25.5	7.80	7.48	9.3	5.6	25.0	10	10	10	10	Sample ID -
1	25.5	7.80	7.49	9.5	5.6	25.6	8	10	9	10	Test Solution Prep RV
1.25	25.5	7.81	7.53	9.6	5.9	25.9	5	6	5	6	New WQ MJM
1.5	25.5	7.81	7.55	9.5	5.9	26.1	0	1	1	2	Renewal Time 1015
2	–	–	–	–	–	–	–	–	–	–	Renewal Signoff SH
											Old WQ MJM
Meter ID	24A	PH12	PH03	DO14	DO10	EC05					
Control	25.5	7.98	7.56	7.8	6.5	24.8	10	10	10	10	Date 3/3/09
0.5	25.5	7.96	7.65	7.9	6.3	25.2	10	10	10	10	Sample ID —
1	25.5	7.96	7.67	7.9	6.3	25.8	8	10	9	10	Test Solution Prep
1.25	25.5	7.97	7.73	8.0	6.5	26.0	4	6	5	6	New WQ DGV
1.5	25.5	7.97	7.83	8.0	6.8	26.3	—	1	1	2	Renewal Time 0945
2	–	–	–	–	–	–	–	–	–	–	Renewal Signoff SLR
											Old WQ MEC
Meter ID	24A	PH11	PH03	DO10	DO12	EC04					
Control	25.1	7.97	7.49	9.0	5.6	24.6	10	10	10	10	Date 3/4/09
0.5	25.1	8.02	7.54	7.7	5.6	25.2	10	10	10	10	Sample ID —
1	25.1	8.02	7.58	7.8	5.6	25.8	8	10	9	9	Test Solution Prep
1.25	25.1	8.04	7.63	7.7	5.7	26.1	4	6	5	6	New WQ MJM
1.5	25.1	8.03	7.71	7.9	5.8	26.3	—	1	1	2	Renewal Time 1545
2	–	–	–	–	–	–	–	–	–	–	Renewal Signoff AA
											Old WQ MJM
Meter ID	24A	PH11	PH11	DO12	DO12	EC01					
Control	25.5		7.72		6.6	25.2	10	10	10	10	Date 3/5/09
0.5	25.5		7.73		6.2	25.8	10	10	10	10	Termination Time 0935
1	25.5		7.76		6.6	26.4	8	10	8	8	Termination Signoff RV
1.25	25.5		7.71		6.4	26.7	3	6	4	3	Old WQ 4K
1.5	25.5		7.76		6.1	26.6	—	1	1	1	
2	–		–		–	–	–	–	–	–	
Meter ID	24A		PH11		DO12	EC04					