

Technical Report 427-272

## **SITE-SPECIFIC SULFIDE CRITERION FOR PRODUCED-WATER DISCHARGES AT FIVE CALIFORNIA OCS PLATFORMS**



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## 1.0 SUMMARY OF FINDINGS

This investigation establishes 12 µg/L as the undissociated-sulfide concentration that is protective of 95% of the marine genera residing in the water column at the boundary of the 100-m produced-water mixing zone surrounding Platforms Harvest, Hermosa, Hidalgo, Irene, and Gina. It departs from the national water-quality criterion of 2 µg/L initially promulgated in the General Permit for use in produced-water reasonable-potential analyses for California OCS platforms. During the data collection phase of the permit, the operators were allowed to submit relevant studies to USEPA Region IX for reconsideration of the criteria for H<sub>2</sub>S discharges to the Southern California OCS. This report recommends that the reasonable-potential analysis of produced-water discharges from the five subject platforms be evaluated based on a H<sub>2</sub>S criterion of 12 µg/L.

The current national water-quality criterion (WQC)<sup>a</sup> for undissociated sulfide (H<sub>2</sub>S) was developed using an extremely limited number of dated bioassay studies, conducted primarily on freshwater organisms, or on organisms exposed to H<sub>2</sub>S in a complex chemical mixture. Because H<sub>2</sub>S toxicity is closely related to the physicochemical properties of water, particularly pH and salinity, the freshwater data can greatly overestimate toxicity. Since this initial WQC designation in 1976, a significant amount of research on H<sub>2</sub>S has been produced using marine species, including new tests that were specifically conducted as part of this investigation.

The site-specific WQC development consisted of the following tasks.

- Evaluation of the suitability of available H<sub>2</sub>S toxicity data
- Identification of data gaps based on USEPA (1985) guidelines for WQC development
- Development and implementation of appropriate toxicity tests to fill the data gaps
- Integration and combination of the acceptable literature endpoints with new project-specific bioassay results to create a comprehensive database
- Calculation of a site-specific WQC applicable to the five California OCS platforms.

The resulting database consisted of a large number of modern bioassay tests, including tests conducted specifically for this study. Analysis of this comprehensive database demonstrated that the current national WQC of 2 µg/L was well below the effects concentrations reported in all acceptable studies, and did not accurately reflect site-specific conditions within produced-water discharges along the California OCS. Instead, a probabilistic risk assessment (PRA) of the database determined 12 µg/L to be a site-specific WQC for H<sub>2</sub>S exposure to marine organisms that is protective of multiple species at the 95% protection level specified in the USEPA (1985) criteria-development guidelines.

The literature review (Weston 2005) evaluated the results from 70 acute toxicity tests representing 31 marine species, and 27 chronic tests representing 19 marine species. For various reasons, all of chronic test values and approximately half of the acute test values were subject to

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<sup>a</sup> A list of acronyms and definitions is provided in Appendix II.

qualifications, but not necessarily elimination from the database. The qualifying factors included the following.

- Irregularities with test organisms (e.g., testing with single-celled organisms, non-water column species, or highly tolerant species)
- Variations in exposures (e.g., pulsed or short dosing periods, varied sulfide concentrations)
- Testing of freshwater or benthic species
- The presence of confounding factors (e.g., low dissolved oxygen, complex mixtures)
- Endpoints that consisted of subjective behavioral or subcellular responses
- Insufficient replication

Additional bioassays were conducted as part of this study to augment the literature database, particularly with respect to WQC development guidelines (USEPA 1985). Those guidelines require a minimum level of phylogenetic diversity within a variety of test types, including the following.

- 8 Acute Tests, using species within different specified families,
- 3 Chronic Tests,
- 2 Macroalgae Tests, and
- 1 Bioaccumulation Assessment

The guidelines also suggest inclusion of various standard species and tests in the criteria development. Based on the guideline requirements and a review of the available literature, a series of additional bioassay tests were conducted as part of this study. Together, the new bioassay results and the acceptable literature endpoints provided a more robust dataset for establishing the site-specific H<sub>2</sub>S WQC. The bioassays conducted as part of this investigation included a series of acute toxicity tests with three fish species, a mysid, an amphipod, a polychaete worm, and two species of larval molluscs. In order to allow for the calculation of an acute-to-chronic ratio (ACR), chronic tests were conducted with two of the fish species and the mysid that were included in the acute species list. Tests were conducted with continuous-flow and static-renewal exposures, using a dose-delivery system specially designed to offset the short half-life of sulfide in seawater.

A number of unusual characteristics associated with hydrogen-sulfide toxicity were discovered during the bioassay testing conducted as part of this investigation. Some of these characteristics caused the bioassay-testing phase of the study to be more time consuming than originally anticipated. For example, identifying and overcoming the source of rapid sulfide degradation within test chambers required a labor-intensive effort that extended the duration of the testing well beyond the original schedule. Similarly, investigating the unusual nature and timing of the toxic response to H<sub>2</sub>S led to further delays in reporting of the results. In the end, each of the unanticipated challenges encountered was addressed, and the bioassays provided the following noteworthy findings with regard to H<sub>2</sub>S toxicity in marine systems.

- Mysids and larval mollusks were the most sensitive test organisms. All three fish species and the amphipod were slightly less sensitive, while the polychaete worm was the least sensitive test species.
- All of the marine organisms tested in the recent bioassays were more sensitive than their counterparts that were reported in the literature.
- Results from toxicity tests that measured both acute and chronic endpoints on the same species, and conducted during the same testing period, established that the acute-to-chronic ratio (ACR) is approximately 1:1. This response indicates that H<sub>2</sub>S has a lethal effect that is rapid, and does not increase with increasing exposure durations.
- Dissolved sulfides and H<sub>2</sub>S in treatment systems are not persistent and the half-life in static systems is one hour or less depending upon the test concentration. Consequently, in static systems, the measured toxic threshold concentrations are generally underestimated.
- The dose-response of larval mollusks exposed to a single H<sub>2</sub>S spike at test initiation was essentially the same as static-renewal exposures over the entire test period. This indicates a very rapid mechanism of action for H<sub>2</sub>S toxicity. It also suggests that the results for spike tests that were reported in the literature (e.g., Knezovich et al. 1996) provide valid data for criterion development.
- Mollusk larvae that were exposed to single spikes at different developmental periods showed little or no sensitivity to H<sub>2</sub>S, indicating that larval sensitivity was only associated with the initial phase of development when rapid cell division dominates. Subsequent developmental phases involving cell differentiation and organ development were substantially less sensitive. This indicates that a criterion based on the initial larval endpoint may be over protective of fully developed zooplankton.
- The larval test results are consistent with a toxic mechanism that interferes with energy production for aerobic metabolism. The rapid mode of action is consistent with a toxic mechanism involving interference with the cytochrome *c* oxidase energy system. This type of interference results in the consumption of available ATP, and the simultaneous production of lactic acid. Therefore, the higher the rate of energy use, the faster the organism succumbs to the toxicant. The most sensitive tests (larval exposures) showed arrested development as a response to an initial high dose of H<sub>2</sub>S. Later dosing did not have the same type of response, and the toxicant was significantly less damaging.
- All of these test results are similar to the responses characterized by cyanide exposure, which results in a similarly rapid impact to test organisms. The cyanide WQC was developed based upon 304(a) aquatic life criteria as a criterion continuous concentration (CCC) developed with guidance provided by USEPA (1985).

These observations led to several conclusions about the protective nature of site-specific WQC criteria developed in this study. First, because H<sub>2</sub>S concentrations naturally undergo a rapid decline in aerated systems, measured H<sub>2</sub>S concentrations reported in the literature and in the bioassays conducted as part of this investigation, are likely to be lower than the initial peak concentrations that caused a toxic response. Consequently, the database contains conservative

estimates of the true exposure concentrations. This means that criteria developed from the database are likely to over-predict true effects in the environment.

Second, the more sensitive organisms that were tested as part of this study (mussels and abalone larvae) had a response to spiked H<sub>2</sub>S concentrations that was similar to a more constant concentration exposure. This means that literature data originally excluded because they only reported pulsed H<sub>2</sub>S exposures, could now be legitimately included in the database used for criterion development.

Third, an ACR of 1:1 indicates that both acute and chronic responses demonstrate the same rapid mode of toxic action, and that longer duration exposure periods do not result in greater effects. This means that data representing shorter-than-normal testing periods, but at least 24-h in duration, can be included in the database for criterion development.

Fourth, the effects of H<sub>2</sub>S are immediate and responses are based on the maximum exposure concentration. However, the doses that are typically reported in the bioassays represent average concentrations measured throughout the testing period. Because these values under-predict the actual (peak) threshold concentrations, it is appropriate to include all of the endpoints, both acute and chronic, in a single dataset to be used to develop the site-specific WQC.

The resulting comprehensive dataset included 102 toxicity values for marine organisms exposed to H<sub>2</sub>S. This dataset is substantially larger than datasets used to establish most other WQC. A PRA was conducted on the dataset to establish the H<sub>2</sub>S concentration that is analogous to the CCC because it is protective of 95% of the species evaluated, as suggested in the guidance provided by USEPA (2004). The PRA uses a Monte Carlo technique to select the most representative distribution for the data set being evaluated, and then provides a range of risk estimates at the 5% to 95% levels. Each of the distributions is further evaluated for *goodness of fit* parameters. The resulting risk estimates for 5% to 95% are then modified to illustrate the possible uncertainties in the risk estimates.

## 2.0 BACKGROUND

This study specifies a site-specific criterion for undissociated sulfide<sup>a</sup> that is protective of marine organisms residing in the water column at the boundary of the 100-m produced-water mixing zone surrounding Platforms Harvest, Hermosa, Hidalgo, Irene, and Gina. The satellite image on the cover of this report shows the locations of these five platforms.<sup>b</sup> Four of the platforms are operated by Plains Exploration and Production Company/Arguello Inc (PXP), and are located north of Point Conception near Point Arguello. Dos Cuadras Offshore Resources, LLC (DCOR) operates Platform Gina, which is located in the eastern Santa Barbara Channel.

Produced water is extracted along with oil and gas as a by-product of petroleum production. It is either re-injected back into the formation or disposed of, in some cases by discharge into the marine environment. Depending on the formation, dissolved sulfides may be present in produced water discharged from production platforms on the Southern California OCS. Undissociated sulfide is the dissolved fraction of hydrogen sulfide (H<sub>2</sub>S) in aqueous solutions, and has the potential to cause adverse ecological impacts. Like the toxicity of ammonia, sulfide toxicity is highly dependent on receiving-water properties. Namely, the dissociation of sulfide into H<sub>2</sub>S is a function of receiving-seawater temperature, pH, and salinity. Sulfide dissociation decreases with increasing pH, and in ocean waters offshore California, the toxic, undissociated form of sulfide comprises approximately 3% to 8% of the total sulfide concentration shortly after produced water begins mixing with seawater.

### 2.1 SCOPE

The determination of the site-specific criterion encompassed the following tasks.

- A comprehensive assessment of existing bioassay data on sulfide exposure
- The design, acquisition, testing, and calibration of dosing apparatus, measurement devices, and safety equipment for bioassay test chambers
- Chronic and acute bioassay testing of selected test species
- Exploratory retesting to investigate the timing and mechanism of the observed toxicity
- Determination of a site-specific CCC based on a comprehensive database including literature data and the result of the project-specific tests conducted for this investigation

A description of the overall approach and a list of candidate species was provided to USEPA Region-IX staff on 2 December 2005 for informal comment (MRS 2005). A revised list of candidate test species was provided to USEPA in February 2006 (MRS 2006). Revisions were necessary because some of the proposed test species on the original list were not available in a timely manner.

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<sup>a</sup> This report refers to undissociated sulfide and hydrogen sulfide (H<sub>2</sub>S) interchangeably as the toxic fraction of total dissolved sulfide within an aqueous solution.

<sup>b</sup> The image was adapted from <http://www.id.ucsb.edu/lovelab/viewvideos.html>

The bioassay facility began testing of organisms in December 2005 after completing an extensive design, acquisition, testing, and calibration of dosing apparatus, measurement devices, and safety equipment for the test chambers. The bioassay tables were fitted with these test and treatment apparatus, as well as the personnel protection sensors. The testing facility, which is located in Port Gamble, Washington, was large enough to conduct tests on multiple species concurrently.

Rather than using hydrogen-sulfide gas to dose the test chambers, as originally proposed, sodium-sulfide salts were spiked into de-oxygenated distilled water. The dissolved sulfide concentration in this stock solution remained stable over several days and was combined with seawater using peristaltic pumps capable of precisely controlling the injection rate. The resulting system was capable of delivering a relatively constant dose of hydrogen sulfide within the range of expected biological effects, without unduly modifying the pH, salinity, or dissolved oxygen within the test chambers. An ultraviolet-visible spectrophotometer was used to monitor dissolved sulfide concentrations in the test chambers. Additional test modifications were added during the period of testing to accommodate specific requirements of test organisms and to increase the consistency of toxicant dosing.

## 2.2 REGULATORY SETTING

### 2.2.1 General Permit

There are 19 produced-water discharges<sup>a</sup> located on the California OCS that are presently covered by the General National Pollutant Discharge Elimination System (NPDES) permit No. CAG280000<sup>b</sup> issued by Region IX of the USEPA. The General Permit currently specifies an undissociated sulfide criterion of 2 µg/L to be used in produced-water reasonable potential determinations.<sup>c</sup> Previous permits have not included monitoring for undissociated sulfide, so the occurrence and quantity of sulfide within the produced-water discharges along the California OCS was previously unknown. During the recently completed reasonable-potential sampling phase of the General Permit, all the platforms analyzed produced-water samples for total sulfide concentrations to establish the reasonable potential for the treated discharges to cause or contribute to a WQC exceedance of the undissociated-sulfide criterion at the boundary of the 100-m mixing zone.

Historically, most NPDES permits for discharges into the open-ocean waters offshore California did not include requirements for effluent monitoring of sulfide, largely because of its rapid

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<sup>a</sup> Table 7 on Page 22 of the General Permit at <http://www.epa.gov/Region9/water/npdes/ocsgeneralpermit904.pdf>

<sup>b</sup> Hereinafter referred to as the “General Permit” or “Permit.” The General Permit No. CAG280000 authorizes discharges to federal waters offshore Southern California from oil and gas exploration, development, and production facilities. It is available at: <http://www.epa.gov/Region9/water/npdes/ocsgeneralpermit904.pdf>

<sup>c</sup> Because undissociated sulfide cannot be directly measured, the General Permit recommends using EPA Method 376.1 (or equivalent method published in Standard Methods) to analyze for total sulfide. The undissociated-sulfide fraction is calculated based on the pH, temperature and salinity of both the end-of-pipe sample and ambient ocean conditions at the platform. In the absence of other information, an ambient ocean pH of 8.1 and salinity of 30 g/kg may be used.

oxidization by natural biological systems. For example, the California Ocean Plan (COP) does not prescribe a quantitative limit on sulfide, but states that: “*The dissolved sulfide concentration of waters in and near sediments shall not be significantly increased above that present under natural conditions*” as a result of the discharge (SWRCB 2001). The State criterion was directed at potentially anoxic conditions within the sediments of semi-enclosed bays and estuaries, where oxygen renewal is limited.

### 2.2.2 National Sulfide Criteria

The USEPA Red Book (1976) established a chronic marine criterion<sup>a</sup> (CCC) of 2 µg/L for undissociated sulfide based on bioassays conducted largely on freshwater species. The criterion was based on responses from a total of seven freshwater species and one marine species, a salmonid fish. These studies represented the available data at the time the 1976 WQC were promulgated. The Red Book derivation further stated that an acute limit for sulfide is inappropriate largely because aquatic organisms exhibit a strong avoidance reaction to sulfide. The CCC was reiterated in the USEPA Gold Book (1986) without significant further reassessment. Thus, based on a review of limited studies, undissociated-sulfide concentrations in excess of 2 µg/L were also considered to constitute a long-term hazard to marine organisms.

The use of data based on freshwater tests is problematic due to the effects that physicochemical properties have on H<sub>2</sub>S toxicity, particularly pH and salinity. H<sub>2</sub>S toxicity increases dramatically with lower pH. This effect is most apparent between the range of freshwater pH (~7) and marine water pH (~8), with a far greater proportion of the more toxic form sulfide in freshwater.

The use of the single marine test that was available in the development of a marine criterion is problematic for several reasons. Holland et al. (1960) found that a total dissolved sulfide concentration of 1,000 µg/L caused in 100% mortality in smolted Pacific Salmon after 72 hours (Holland et al. 1960).<sup>b</sup> This single result was used to support the use of results from tests on freshwater aquatic organisms in the development of the marine criterion. After application of pH, temperature, and salinity adjustments to the total-sulfide concentration determined by Holland et al. (1960), an H<sub>2</sub>S concentration that would not be considered hazardous to most marine fish was established at 2 µg/L. However, the 46-year-old study was based on exposure to a complex mixture of sulfite waste liquor containing numerous contaminants and abnormal pH values. USEPA guidance (1985, 2005) specifies that tests with complex mixtures of contaminants should not be used as primary data, and that they may only be used as ancillary information in the development of criteria.

In the 30 years since the national sulfide CCC was established, a significant amount of additional research has been conducted on sulfide toxicity to a diverse range of marine organisms. This research indicates that a re-evaluation of the marine criterion is warranted. The current H<sub>2</sub>S

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<sup>a</sup> The CCC is the USEPA saltwater Criterion Continuous Concentration that establishes the highest (dissolved) concentration of a pollutant in saltwater, to which an aquatic community can be exposed indefinitely (chronic limit) without resulting in an unacceptable effect.

<sup>b</sup> See the critique of historical studies provided in Section 3.1.3.

criterion of 2 µg/L is well below the effects concentrations reported in all studies examined in this review.

### 2.2.3 Site-Specific Sulfide Criteria

The General Permit relies on the CCC to establish an undissociated-sulfide concentration of 2 µg/L as the limiting criterion applicable to produced-water discharges, after dilution and at the edge of the 100 meter mixing zone. However, as described above, the Permit allows a permittee to submit a request for a recalculated criterion based on site-specific studies and analyses that consider ambient factors and the nature of the discharge.<sup>a</sup> PXP made such a request based on a screening recalculation that determined 3.2 µg/L to be a conservative representation of the USEPA Saltwater Criterion Maximum Concentration<sup>b</sup> (CMC) criterion (Tetra Tech 1999).

The Tetra Tech criterion was based on a cursory review of literature data from acute tests performed on marine species, including sensitive benthic invertebrates. Because the review was cursory in nature and lacked applicable data, the concentration was considered conservative. Moreover, as described above, the USEPA never promulgated an acute national criterion for sulfide because of the accepted avoidance response. They recommended developing a site-specific chronic criterion for produced water using the USEPA (1985) guidelines for criteria development. They further recommended that the proposed criterion should be submitted to the USEPA at the same time, or prior to, the submission of the reasonable-potential assessment. This report is a response to that request.

A site-specific water-quality criterion (WQC) is advisable because it is more applicable to the unique open-ocean conditions (i.e., seawater of comparable salinity, stable pH, and high dissolved oxygen concentrations) where the produced water is discharged. USEPA (1994) provides for a modification of National Level Criteria by states in order to accommodate the development of site-specific water-quality values using a water effects ratio (WER). The process presented in WER methodology indicates that these values can be developed for either CCC or CMC criteria. Historically, USEPA used available freshwater and marine data to develop an H<sub>2</sub>S CCC for National WQC (1976, 1986, 2002a, and 2006). Based on this regulatory precedent and the nature of hydrogen sulfide toxicity discussed below, a wide range of acceptable and available data can be used to develop a revised CCC value that is conservatively protective of the open-ocean environment where produced water is discharged.

Hydrogen-sulfide concentrations in solution are strongly influenced by water-quality parameters, particularly pH, temperature, and salinity. Because of these influences, it is difficult to apply responses from freshwater test organisms to the prediction of responses in marine organisms. For this reason alone, a separate evaluation for marine species is strongly indicated. Moreover, it has

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<sup>a</sup> Footnote 2 on Table 4 of the General Permit states that “A permittee may submit a request for a recalculated criterion based on site-specific studies and analyses that consider ambient factors and the nature of the discharge.”

<sup>b</sup> The USEPA Saltwater Criterion Maximum Concentration (CMC) is an estimate of the highest concentration of a pollutant in saltwater, to which an aquatic community can be exposed briefly (acute limit) without resulting in an unacceptable effect.



also become evident that hydrogen sulfide toxicity is often compounded by the presence of low dissolved oxygen concentrations. A number of studies used in the development of the original national criteria included this confounding factor. Finally, the process for developing WQC has become more rigorous since the criterion was first established, suggesting that the H<sub>2</sub>S WQC should be re-evaluated using modern toxicity data generated for marine species.

#### **2.2.4 Criterion Development Requirements**

Development of WQC by USEPA requires that the selected species for saltwater tests include acceptable acute tests with at least one species of saltwater animal from at least eight different families (USEPA 1985). Neither the Tetra Tech, nor the 1976 and 1986 USEPA national criteria for hydrogen sulfide meet this required level of diversity. However, since these original criteria were established, additional toxicity data have become available. With the addition of the bioassays conducted as part of this study, a large number of studies on a broad suite of organisms can be included in the criterion development.

At a minimum, the 1985 criterion-development guidelines require the inclusion of the following groups of marine organisms.

- 1) Two families in phylum chordata
- 2) A family in a phylum other than arthropoda or chordata
- 3) The mysidae or penaeidae family
- 4) Three other families not in the phylum chordata (may include mysidae or penaeidae, whichever was not used before)
- 5) A representative organism from at least one other family

The guidelines also require acceptable performance in three chronic tests with species used during the acute testing in order to develop an ACR. An acceptable test with a saltwater alga or vascular plant is also required, and if plants are found to be the more sensitive species, then a second plant species needs to be examined. Finally, a bioaccumulation assessment is generally required for testing contaminant-related impacts on bioaccumulable toxicants. Bagarinao and Vetter (1989) demonstrated that sulfide is not an accumulative contaminant in the tissues of marine organisms, so its effects are not compounded over time from repeated exposures.

Even though the USEPA guidelines require testing on a number of different species from several different major taxonomic groups, selection of the appropriate test organisms is crucial for evaluating produced-water discharges. One of the important results that arose out of the fish-exposure study by MRS (2005) is that the produced-water plumes discharged by the five platforms under consideration do not impinge on the seafloor (Figure 1). Thus, this study was not directed at specifically evaluating impacts to marine benthic infauna. Rather, the selected test species represented water-column organisms, or organisms that would be potentially exposed on structures in the water column.

## 2.3 UNDISSOCIATED SULFIDE PROPERTIES

A direct analytical test is not available to determine the amount of undissociated  $\text{H}_2\text{S}$  dissolved within aqueous solutions. Instead, chemistry laboratories must use an alternative test for the analysis of total sulfide by first testing for sulfide (as  $\text{S}^{2-}$ ) using the titrimetric method, and then calculating the amount of undissociated hydrogen sulfide present based on the physicochemical properties of the sample.

### 2.3.1 Sulfur in Seawater

Hydrogen sulfide (undissociated  $\text{H}_2\text{S}$ ) is a soluble, highly toxic compound that occurs naturally in anaerobic marine environments where biological and/or chemical oxygen demand exceeds the available dissolved oxygen. It can also be introduced directly into the marine environment through the discharge of domestic wastes, industrial wastes (e.g., tanneries, paper mills, chemical plants) and produced waters from the oil and gas industry (USEPA 1986, Verschueren 1983). It is a highly soluble compound ( $<4,000$  mg/L at  $20^\circ\text{C}$ ) and is biologically and chemically reactive.

There are three chemical states of sulfur in seawater (APHA 1998). Total sulfide includes dissolved sulfide as well as acid-soluble sulfides present in suspended matter. Dissolved sulfide consists largely of undissociated hydrogen sulfide ( $\text{H}_2\text{S}$ ) and bisulfide ions ( $\text{HS}^-$ ) in equilibrium with hydrogen ions ( $\text{H}_2\text{S} \rightleftharpoons \text{H}^+ + \text{HS}^-$ ). Only the undissociated ( $\text{H}_2\text{S}$ ) portions of dissolved sulfides are considered toxic to marine organisms. The degree of dissociation is directly dependent on the physicochemical properties of the seawater, namely, pH, temperature, and salinity.

For the ranges in seawater properties near the five platforms, the undissociated  $\text{H}_2\text{S}$  fraction is generally expected to be 3% to 8% of the sulfide concentration present in the produced-water plumes after dilution, and when they have taken on the properties of ambient seawater. Variations in ambient pH exert the greatest influence on the dissociation of sulfide. Variations in temperature are much less influential, while salinity variations have a negligible influence on dissociation. In comparison to the other seawater properties, variations in pH can have a much greater influence on  $\text{H}_2\text{S}$  dissociation. However, pH measurements in seawater are complex and the reported values can be in error if the instruments are not properly calibrated (Sigel et al. 1991). In addition, the highly buffered carbonate system in the ocean keeps pH relatively stable. Consequently, typical seawater pH in the upper ocean ranges between 8.1 and 8.3 due to equilibration with the atmosphere and stabilization by the carbonate system. As a result, the undissociated  $\text{H}_2\text{S}$  fraction is expected to be comparable to the dissociation achieved in the test chambers used in the bioassays conducted in this study, which was around 10%.

### 2.3.2 Sulfur in Living Systems

Reduced sulfur compounds in the form of proteins, coenzymes and major cellular metabolites control structure, binding site characteristics, transport system function, and protect systems from oxygen toxicity and numerous other contaminants. Oxidized forms of sulfur provide anionic binding sites, structural functions and maintenance of charge separation (Mitchell and Nickson 1993). One of the better understood 'detoxification mechanisms' is the induction of

metallothionein in response to elevated metals concentrations in the blood stream. This protein contains either the amino acid, cysteine, or cysteic acid, which are composed of ~15% sulfur. Another protective mechanism is mucous, which protects and lubricates internal and external surfaces of organisms. The mucin molecule contained in mucous is rich in cysteines, which are likely to be involved in establishing disulfide linkages within and among mucin monomers. Sulfur is, therefore, an essential and irreplaceable part of all living systems acting in energy storage and transfer that are also fundamental to cellular and organism communication, defense processes within the body and maintenance of the biological structure of the organism (Mitchell 1996).

Although sulfur is important for certain cellular functions, certain forms of sulfur can also be toxic. Hydrogen sulfide is known to compromise mitochondrial respiration through inhibition of cytochrome *c* oxidase in a manner similar to hydrogen cyanide (Smith et al. 1977, Holland and Kozlowski 1986). The suggested mechanism for this toxicity involves the binding of the heme iron of the enzyme resulting in nearly complete inhibition of aerobic metabolism followed by depleted levels of ATP and accumulation of lactic acid (Beauchamp et al. 1984). This process would debilitate cellular energy stores. It has also been reported that hydrogen sulfide exposure can result in altering the permeability of the cell membrane, potentially accelerating the impacts to the mitochondrial respiration (Thompson et al. 2003). Because the primary mechanism is inhibition of aerobic mitochondrial respiration and occurs rapidly, the potential for longer term, chronic impacts is minimal. This implies that effects associated with H<sub>2</sub>S toxicity will likely be evident with short-term exposures.

### 2.3.3 Sulfide Degradation

Dissolved sulfide concentrations rapidly degrade when introduced into oxygenated seawater. Dissolved sulfide is removed from test waters by microbial uptake, oxidation, speciation, coprecipitation, and volatilization. A principal sulfide-removal mechanism is chemical oxidation and the formation of nontoxic thiosulfates. Millero (1986), Millero and Hershey (1989), and Nriagu (1978) have demonstrated that the half-life of H<sub>2</sub>S in oxygenated seawater is between 24 minutes and 3 hours.

In addition to this chemical degradation, laboratory tests conducted as part of this study demonstrated that sulfides are also rapidly oxidized into sulfates and elemental sulfur in well-aerated water by biological systems. For the concentration ranges used to demonstrate effects in various organisms assess in this study, the half-life of total sulfides was approximately 1 hour, while the half-life of H<sub>2</sub>S itself was generally around 20 minutes. At the lowest H<sub>2</sub>S concentrations, the half-life was even shorter.

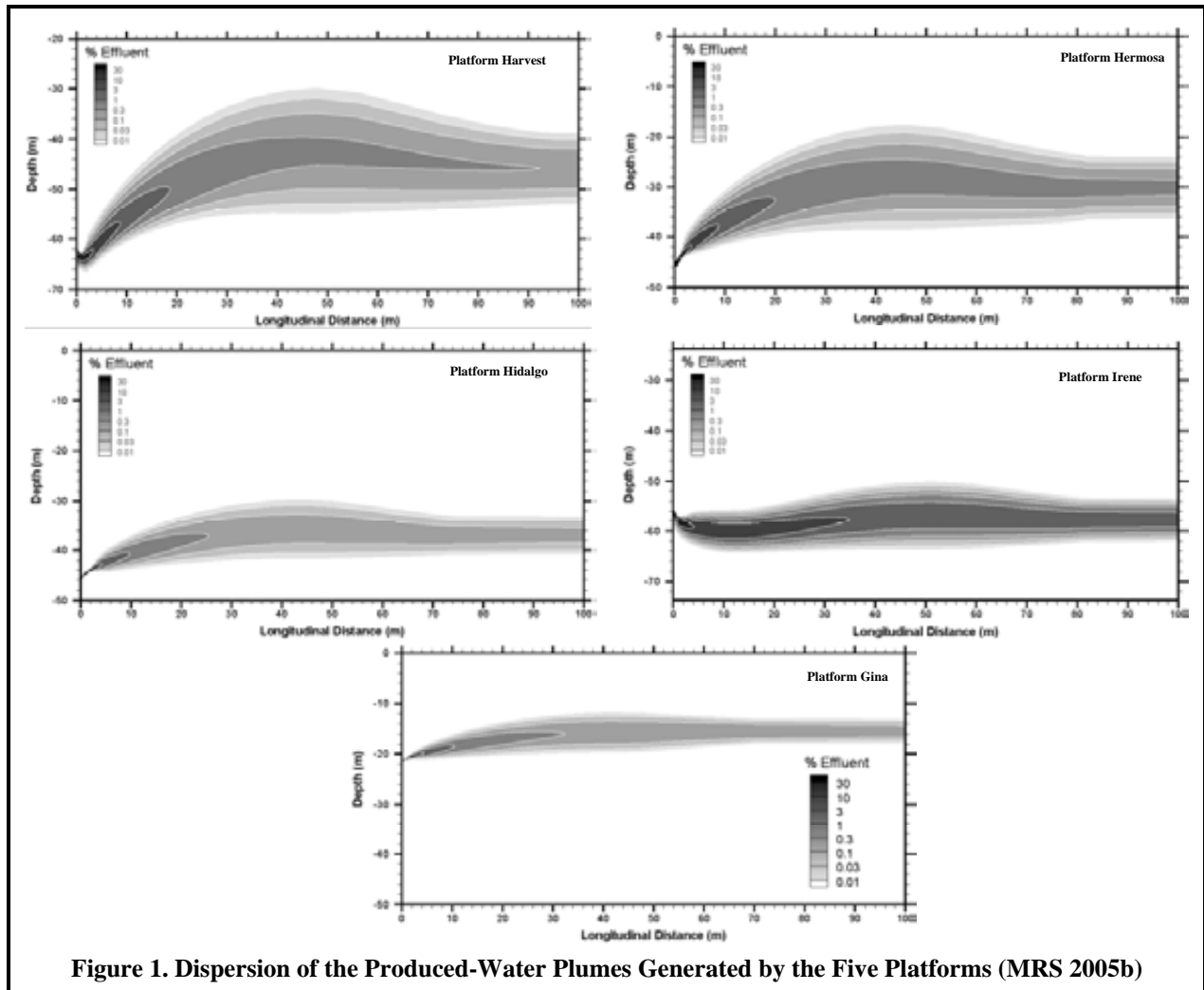


Figure 1. Dispersion of the Produced-Water Plumes Generated by the Five Platforms (MRS 2005b)

### Within Produced-Water Discharges

Rapid sulfide degradation complicates the interpretation of a chronic sulfide limit for open-ocean conditions. The 96-hour duration of a typical acute toxicity test is long relative to the expected longevity of sulfide exposure in the open ocean. Chronic assays test continuous exposures over longer durations. Hence, the general applicability of chronic test results for open-ocean conditions is questionable. However, the USEPA (2005) discounted the importance of sulfide degradation for produced water discharges because they generate a continuous supply undissociated sulfide at the edge of the 100-m mixing zone. Moreover, the nominal transit time across the mixing zone, which is around 15 minutes, limits the amount of degradation possible from the time of discharge.

### Within Bioassay Test Chambers

Rapid sulfide degradation in seawater also makes bioassay tests challenging to conduct. In the bioassays performed as part of this study, significant effort was expended in designing continuous-flow dosing apparatus to ensure a continuous sustained dose was maintained in the test chambers. In contrast to other contaminants, a consistent hydrogen-sulfide dose is difficult to maintain because of the natural decay in the concentration of dissolved sulfide within test

chambers. Because half of the dissolved sulfide is removed by oxidation alone in as little as 24 minutes, much of the dissolved sulfide could be depleted during the typical 96-hour bioassay unless concentrations are regularly replenished. However, a constant dose is difficult to maintain, particularly during static or periodic-renewal tests. Even in continuous-flow tests, repeated timely measurements of the sulfide concentrations are required to maintain the exposure levels throughout the test. Furthermore, the total sulfide concentration must be in conjunction with the pH, temperature, and salinity within the test chambers. Only with these ancillary physicochemical properties measured simultaneously can the appropriate dissociation factor be precisely determined.

As revealed in the detailed experiments conducted on sulfide concentrations in association with this study, decay is especially rapid at low sulfide concentrations, when half-lives of less than 20 minutes were observed. The resulting uncertainty in the overall continuous dose during individual tests was one of the primary reasons that most of the historically available bioassays were initially rejected for use in the development of a site-specific criterion (Weston 2005). This was also part of the rationale which led to the additional continuous-flow toxicity testing conducted as part of this study. Before the additional testing was initiated, only one of the tests reported in the literature was thought to be suitable for establishing a site-specific criterion for hydrogen sulfide in produced water.

### **Effect on the Validity of Historical Bioassay Results**

While the degradation rate appears to be quite rapid in static and static-renewal exposure systems, data collected during this study indicated that H<sub>2</sub>S toxicity is also rapid. Moreover, exposure to slightly lower concentrations of undissociated sulfide, even for extended periods, does not elicit an acute or even a chronic response. Consequently, the peak initial concentration may be the controlling factor in determining the toxicity of undissociated sulfide to marine organisms. Perhaps more importantly, these tests showed that the resulting ACR for undissociated sulfide was close to 1:1.

This highly unusual mode of action has profound implications for produced-water discharges. First, the debate over whether an acute (CMC) or chronic (CCC) criterion is more applicable, may not apply to H<sub>2</sub>S. Second, many of the historical bioassays reported in the literature, which were initially thought to be flawed because they reported nominal (possibly degraded) exposure concentrations, could be considered valid conservative measures of toxic response to undissociated sulfide. Accordingly, the site-specific criterion identified in this report could be developed using a combination of the results from recent bioassays, and the results from a wider range of appropriate bioassays reported in the literature. This resulted in a large number of literature values could now be included in the criterion determination.

### 3.0 CRITERION DEVELOPMENT

#### 3.1 REVIEW OF EXISTING LITERATURE

In November 2005, Weston (2005) reviewed available data on the response of marine organisms to H<sub>2</sub>S exposure. The review assessed literature sources that pertain to marine H<sub>2</sub>S exposure and identified additional test species that were appropriate for the development of a site-specific WQC applicable to produced-water discharges.

##### 3.1.1 Information Sources

Documents containing information on the toxic effects of hydrogen sulfide were identified from electronic and research library sources such as the ECOTOX database.<sup>a</sup> Approximately 70 acute toxicity test results were reviewed representing 31 separate marine species, including one diatom. Chronic toxicity tests (27) were reviewed which represented 19 marine species. Six species had both acute and chronic test results, including Chinook salmon (*Oncorhynchus tshawytscha*), Atlantic salmon (*Salmo salar*), white sea urchin (*Lytechinus pictus*), bay mussel (*Mytilus edulis*), Pacific oyster (*Crassostrea gigas*), and the common shrimp (*Crangon crangon*).

One study addressed the bioconcentration of elemental sulfur in fish tissues in the form of thiosulfate (Bagarinao and Vetter 1989). Although that study demonstrated evidence of bioaccumulation in five fish species, the hydrogen sulfide exposure concentrations (680 µg/L) far exceeded the levels evaluated in this investigation for toxic response. It is currently unclear whether significant bioaccumulation occurs at lower exposure levels.

##### 3.1.2 Suitability of Studies Reported in the Literature

At first site, the available studies represent sufficient phylogenetic diversity for computation of a revised chronic criterion for hydrogen sulfide. However, a more careful review of each document revealed that some tests were not appropriate for the development of the criterion. The next section presents summaries of some of the more important studies that have been conducted on the toxic effects of hydrogen sulfide. They are presented in alphabetical order by author.

Historical studies that are potentially pertinent to the development of a site-specific H<sub>2</sub>S criterion are summarized in Appendix III, Table 27 through Table 29. The tables evaluate information on acute, chronic, and bioaccumulative responses of marine organisms to H<sub>2</sub>S. The tables include comments regarding the level of applicability of each dataset. Specific data were excluded for the following reasons.

- Results were only reported for concentrations that did not elicit effects (NOEC's)
- Endpoints were behavioral, metabolic, or biomarker-related
- Test species were inappropriate

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<sup>a</sup> <http://mountain.epa.gov/ecotox/> The ECOTOX database was formerly known as AQUIRE.

- Toxicant exposure was through an ‘indirect’ pathways, such as a sediment exposure

The tables also provide an estimate of the H<sub>2</sub>S concentration in µg/L for the reported endpoint. In many cases, this required conversion of molar concentrations.<sup>a</sup> When only the total sulfide concentration was reported, the undissociated fraction was determined analytically from pH, if that was also reported. Otherwise, the H<sub>2</sub>S concentration was assumed to be 10% of the total sulfide concentration in order to reflect nominal seawater conditions within the test chambers.

Results reported in Table 29 for the sulfur bioaccumulation study also required conversion. The original exposure concentration was reported as 200 µM total sulfide. After conversion to 6.8 mg/kg by multiplying by the molecular weight of hydrogen sulfide (34 g), a 90% dissociation factor yielded an estimated H<sub>2</sub>S exposure concentration of 680 µg/L. Similarly, the tissue burdens that were originally reported as µM thiosulfate were converted to represent the accumulation of elemental sulfur in tissues (mg-sulfur/kg-tissue).

### 3.1.3 Critique of Selected Marine Studies

The following critique of selected toxicity studies was conducted on marine organisms that have potential applicability to the development of the site-specific sulfide criterion. The studies in the literature reported undissociated sulfide concentrations based on averages rather than peak initial doses. As such, they provide conservative estimates of the actual toxicity of undissociated sulfides to marine organisms.

#### **Abel (1976) *Effect of some pollutants on the filtration rate of Mytilus***

This short communication tested the effects of a group of toxicants, including sulfides, on survival and respiration rates of adult mussels. Respiration tests were conducted over short intervals (approximately 10 min) and were evaluated by measuring dye transport across the gills. It is unclear how long mussels were exposed to each of the toxicants prior to conducting the respiration experiments. The survival endpoint was conducted as a static, 96 h test, with no water exchanges. The author acknowledged that the concentration of sulfides probably changed over the test period.

The usefulness of this paper is limited because it is unclear what duration of exposure was associated with the respiration rate response. In addition, adult mussels are known to have a low sensitivity to H<sub>2</sub>S.

#### **Bagarinao and Vetter (1989) *Sulfide tolerance and detoxification in shallow-water marine fishes***

This paper provides information on a well-designed study that addressed the toxicity of hydrogen sulfide to various species of juvenile fish. The dosing mechanism used peristaltic pumps to create a continuous-flow system with well-aerated water (100 mL/min) and sulfide-dosed water being introduced at 1 to 2 mL/min from various stock solutions. The test subjects were held in small 4.5-L aquaria. The flow system was therefore designed to replace the test volume of water every 45 minutes. This rapid, continuous flow system reduced the potential for

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<sup>a</sup> 1 µg/L (ppb) = µM/L × 34.08 = µM/mL × 34.08 × 1000 = mM/L × 34.08 × 1000 = mg/L (ppm) × 1000

loss of the test concentrations of hydrogen sulfide and maintaining water quality for the relatively high density of fish that were retained in the test solutions. ASTM guidance (E729, 2000) indicates that under continuous flow conditions, stocking biomass in the test containers should not exceed 1 g/L of water flowing through the tank over a 24 h period, and should never exceed a biomass of 10 g/L at any one time. Under the first requirement, the total mass of all fish allowed in the aquaria would be 144 g representing the 144 L of water flowing through the tank per 24 hours. This is equivalent to the probable stocking biomasses provided for the test. However, the maximum biomass of fish at any time is only supposed to be 10 g/L and this is equaled or exceeded in all cases. This might indicate additional stress on the test organisms and the concentration/effects relationships may be more sensitive than if tested at lower biomass concentrations. The dose of hydrogen sulfide was determined after measuring the total dissolved sulfides and accounting for salinity, pH, and temperature relationships. Under the test conditions of 20°C, pH of 8.3, and salinity of 34‰, hydrogen sulfide represents 3.4% of the total dissolved sulfides. This is close to the concentration reported by the authors.

These data were used for the development of the site-specific WQC with the qualification that the results be validated, and the possibility of bias caused by overstocking be investigated.

**Breteler, Buhl, and Maki (1991) *The effect of dissolved H<sub>2</sub>S and CO<sub>2</sub> on short-term photosynthesis of *Skeletonema Costatum*, a marine diatom***

The purpose of this study was to assess the toxicity of CO<sub>2</sub> and H<sub>2</sub>S to the planktonic diatom *Skeletonema costatum*. The gasses were evaluated in both the total dissolved and undissociated molecular forms. Under controlled laboratory conditions, the diatoms were subjected to 4 h exposures to various concentrations of the gases. Measurements of photosynthesis inhibition during the exposure period were used to evaluate toxicity. Cells were obtained during an exponential growth phase from a culture of *Skeletonema costatum* that was grown in artificial, sterile seawater. Concentrated stock solution of either gas was added to flasks inoculated with a dense suspension of *S. costatum*. Experiments were conducted at 20°C and 30‰ salinity. The target pH range for the H<sub>2</sub>S treatments was  $7.9 \pm 0.2$ , and measurements were conducted at 0 and 4 h. Gas concentrations were also measured at the beginning and end of the tests. H<sub>2</sub>S was analyzed using a UV/VIS spectrophotometer (suspended algal cells were filtered out to remove interference with the absorbance readings). The fraction of molecular, aqueous H<sub>2</sub>S was then calculated to account for the conditions of the test system (pH, temperature, and salinity). EC<sub>50</sub> values were calculated using biological response expressed as a percent reduction in photosynthesis compared to controls.

Results indicated that H<sub>2</sub>S concentrations dropped by 30% to 50% during the test due to chemical oxidation. “Algal photosynthesis dropped abruptly at total dissolved sulfide concentrations in excess of 24 μM and stopped completely at  $\geq 74$  μM total sulfide. The estimated EC<sub>50</sub> value was 48 μM total HS<sup>-</sup>, or 104 μg/L molecular H<sub>2</sub>S.” This data is directly applicable to the development of the site-specific WQC.

**Caldwell (1975) *Hydrogen sulfide effects on selected larval and adult marine invertebrates***

This study addressed the influence of hydrogen sulfide on six species of marine invertebrates including larval and juvenile crab and oysters. The dosing mechanism added sodium sulfide solutions to test containers. The concentrations of free dissolved sulfides were then measured



and expressed in the document as hydrogen sulfide. In tests with amphipods, isopods, juvenile oysters and clams, and crab post-larvae, dissolved oxygen decreased as  $H_2S$  concentrations increased. In the larval tests with crab zoeae and *Crassostrea gigas*, hydrogen sulfide occurred in the presence of normal dissolved oxygen concentrations, pH, and temperature; however, exposure periods were limited to short pulsed exposures.

The species that were selected for study were a mixture of benthic species and water column species. As indicated by the author and Theede et al. (1969), species that live in the sediments are exposed to elevated hydrogen sulfide concentrations more often than other marine biota, and are generally more tolerant of them. These data were deemed acceptable for criteria development.

**Chapman and Fletcher (2002) *Differential effects of sediment on survival and growth of *Fucus serratus* embryos***

The goal of this study was to evaluate the effects of sediment and sediment chemistry on the survival and growth of brown alga embryos, *Fucus serratus*. *Fucus* is a common marine alga found in the rocky intertidal zones, but can also be found at intertidal elevations on man-made structures, such as pilings and docks. Chapman and Fletcher placed fertilized eggs onto glass slides and allowed the embryos to develop under sediment with a variety of exposure variables: light intensity, sediment grain size, low dissolved oxygen (DO), and presence of sulfides (concentration 0.5 mM of sulfides produced by the addition of  $Na_2S$ ). Sulfide exposures were made in static test chambers under low dissolved oxygen conditions, and no measurements of sulfide or water exchanges were made. The calculations of  $H_2S$  were based on 0.5 mM of sulfur multiplied by the atomic weight of sulfur (32) multiplied by the estimated fraction of sulfides as  $H_2S$  (3.5%). This yields an exposure level for undissociated sulfide of 560  $\mu g/L$ . Endpoints were survivorship of the *Fucus* zygotes and growth as measured by the length of embryo body.

The presence of sulfide resulted in significantly increased mortality and decreased growth when compared to the controls and the low DO treatments (regardless of whether sediment was present or absent). Survival in the hydrogen sulfide treatments ranged from 0% to 20%. Growth in the surviving zygotes, in the absence of sediment, was reduced 50% to 65% relative to both the controls and the low DO treatments. Direct correlation between sulfides and effects was difficult to interpret, since sulfides occurred in the presence of low DO and sulfide levels were not measured at all during the five-day exposure period. However, the treatments exposed only to low DO were not significantly affected compared to the controls, providing a compelling argument for sulfide-related effects. Because *Fucus* grows intertidally, it is unlikely to be exposed to produced water. Nevertheless, this study indicates that brown algae embryos may be sensitive to sulfide exposure. This study indicated that kelp germination and growth tests with the brown alga, *Macrocystis* sp. could be appropriate tests for evaluating  $H_2S$  exposure from produced water, and was used in developing the site-specific  $H_2S$  WQC.

**Dillon, Moore, and Gibson (1993) *Development of a chronic sublethal bioassay for evaluating contaminated sediment with the marine polychaete worm *Nereis* (*Neanthes*) *arenaceodentata****

The authors exposed three-week-old polychaete worms (*Neanthes arenaceodentata*) to five concentrations of  $H_2S$  in low oxygen water (1.5 mg  $O_2/L$ ). Tests were conducted as static-

renewal exposures with once-daily exchanges. Sulfide concentrations decreased by approximately 50% overnight. Tests with low dissolved oxygen only indicated that the sulfide tests were conducted with oxygen levels that were at the effects threshold. The authors acknowledge that the low DO conditions may have altered or obscured the sulfide effects determination.

This paper was useful for establishing a lower threshold for sulfide tolerance in this particular benthic polychaete; however, it was qualified due to the potential for confounding effects from low DO.

**Gopakumar and Kuttyamma (1996) *Effect of hydrogen sulfide on two species of Penaeid Prawns *Penaeus indicus* and *Metapenaeus dobsoni****

Two penaeid shrimp species, *Penaeus indicus* and *Metapenaeus dobsoni*, were exposed to H<sub>2</sub>S under continuous-flow conditions at pHs of 6, 7, and 8. Sulfide salts (Na<sub>2</sub>S) were dissolved into de-oxygenated distilled water which was then stocked into seawater at varying concentrations. Different size ranges of shrimp were tested, allowing a comparison of the sensitivities of three age classes.

As with other references, toxicity was pH dependent and the most toxic form of sulfide was associated with the un-ionized form (H<sub>2</sub>S), which can move more easily across cell membranes. This study is particularly useful in that it isolated H<sub>2</sub>S, manipulated pH, and provided LC<sub>50</sub> data for several age groups. The authors also compared penaeid shrimp to other shrimp and non-shrimp species, noting that the older age classes appeared to be more sensitive to H<sub>2</sub>S toxicity. This is consistent with the responses observed by Vismann (1996) indicating that the adult *C. crangon* were more sensitive than the juvenile penaeid shrimp.

**Holland, Lasater, Neumann, and Eldrige (1960) *Toxic effect of organic and inorganic pollutants on young salmon and trout***

From 1953 to 1955 a series of studies were performed on the toxic effects of pulp mill wastes on juvenile salmonids. The fish ranged in age from 68 to 280 days and were primarily Chinook salmon (*Oncorhynchus tshawytscha*) that had been 'smolted' to marine conditions prior to testing. An additional test with Silver salmon (*O. kisutch*) in freshwater for a period of 30 days, and a short term lethality test using high concentrations of effluent on pink salmon (*O. gorbuscha*) were also included in this work. These latter tests were either not marine or were for very short exposures to high concentrations of pulp mill effluent. Therefore, they were not applicable to the determination of the site-specific sulfide criterion for produced-water exposure.

Toxic effects that were studied included mortality, weight, and length changes relative to various dilutions of waste effluent. The concentrations of H<sub>2</sub>S were calculated from measurements made in the original condensate, plus pH and salinity in the dilutions. Effects concentrations that are included in previous USEPA WQC (1976, 1986) are based on the calculated H<sub>2</sub>S in these complex mixtures from the synthesized Kraft waste effluent.

These data were qualified by the following considerations.

- The exposure was from a mixture of toxicants from waste effluent, therefore the toxicity of the other contaminants on the fish cannot be ruled out; USEPA water quality criteria guidance disallows responses to complex mixtures
- There is an unknown stress associated with the length of time between smolting and testing

This document summarized one additional test that demonstrated greater levels of effects from lower concentrations of H<sub>2</sub>S for an unknown reason; however, these data were qualified by the investigators. The effects-based concentrations are probably overestimates of H<sub>2</sub>S toxicity; these values exceeded 70 µg/L for acute and chronic mortalities and/or sublethal effects on growth. Use of these data in the development of the new water quality criterion were subject to qualification and only used for comparison to tests that were specifically applicable to hydrogen sulfide toxicity.

Additional data from a separate test with Chinook salmon on 280-day old smolts produced 72 h toxicity data at a pH of 7.7, with an estimated H<sub>2</sub>S effects ranging from 60 µg/L to 106.8 µg/L. This experiment involved direct exposure of organisms to sodium sulfide rather than pulp-mill waste effluent, and therefore, those data were included in the development of the site-specific WQC.

**Holmer and Bondgaard (2001) *Photosynthetic and growth response of eelgrass to low oxygen and high sulfide concentrations during hypoxic events***

This study evaluated the effects of both low oxygen and hydrogen sulfide on the shallow subtidal marine plant, *Zostera marina* (eelgrass). Two exposure concentrations were used in combination with low oxygen and no oxygen waters: 10 µM to 100 µM in low oxygen and 100 µM to 1000 µM with no oxygen. Eelgrass was incubated for three weeks, with some measurements being made at six days. The authors evaluated photosynthetic activity, survival, shoot growth, chlorophyll-a concentration in the shoots, and sugar, starch and carbohydrate concentrations in roots and rhizomes.

This study provides good evidence that eelgrass survival, growth, and its energy transfer capabilities are impacted by the presence of sulfides. However, the following qualifications were noted.

- This study confounded the cause and effect relationship by combining sulfide exposures with low (or no) dissolved oxygen concentrations and the impacts of H<sub>2</sub>S exposure alone were not clear.
- Although this study suggests that eelgrass may be an appropriate test species for evaluating sulfide effects in the nearshore environment, it is unlikely that nearshore areas will be exposed to H<sub>2</sub>S associated with produced-water discharges on the OCS.

**Ivanov, Usenko, and Parkhomenko (1976) *Effect of hydrogen sulfide on the survival rate of eggs and embryonal mitosis of the Black Sea turbot***

Artificially fertilized eggs of the Black Sea turbot, *Rhombus maeoticus*, were exposed to seawater containing various concentrations of hydrogen sulfide for 24 hours. H<sub>2</sub>S levels were determined using an iodine and hydrochloric acid titration method just prior to placing the eggs

in one-liter bowls. There was no exchange of water during the test. After 24 hours, the number of dead eggs was counted in the experimental and control bowls. Results showed that hydrogen sulfide concentrations equal to or greater than 2.4 to 3.1 mg/L are lethal to developing Black Sea turbot eggs. The method of analyses described in the paper appears to measure total dissolved sulfides and not H<sub>2</sub>S. We have modified the effects-based concentration by assuming only 10% of the total dissolved sulfides are in the form of H<sub>2</sub>S. At the end of the test, the authors also fixed the specimens for cytological analysis and counted the number of chromosomal aberrations. Results indicated that sublethal concentrations of H<sub>2</sub>S in seawater did not have a cytogenic effect.

This study had limited applicability to the evaluation of the effects of H<sub>2</sub>S from produced water. It is possible that the high ambient concentrations of H<sub>2</sub>S at depth in the Black Sea have increased resistance in native species. Therefore, data from *Rhombus maeoticus* may not be representative of effects expected from species off the coast of California.

**Kiemer, Black, Lussot, Bullock, and Ezzi (1995) *The effects of chronic and acute exposure to hydrogen sulphide on Atlantic salmon (Salmo salar)***

In this study, Atlantic salmon smolts were exposed to hydrogen sulfide in marine water. Both acute and chronic tests were conducted, each with one concentration of H<sub>2</sub>S in two replicate tanks. One replicate tank was used for a control. H<sub>2</sub>S exposures were conducted as “pulses” (ten-minute pulses every six hours, with H<sub>2</sub>S concentration dropping during a three-hour period) at sublethal concentrations (0 µg/L to 265 µg/L for the chronic tests and 670 µg/L and 950 µg/L for the acute tests).

The authors observed no mortality at these test concentrations, and slightly increased growth during the chronic exposures. Gill lamellae in the acute exposures were thickened and fused at 670 µg/L H<sub>2</sub>S. Some gill damage was observed in the chronic exposures; however, this was reversible. Liver necrosis was observed in the 265 µg/L treatments, with >80% abnormal livers after 18 weeks of exposure.

Possible qualifications of these results from this study were that the number of replicates was limited (this study used only two replicates of one concentration for each test) and that the H<sub>2</sub>S concentrations were not held constant throughout the test.

Results from this study were used to estimate an NOEC for mortality and a LOEC for gill and liver damage. Of particular interest was the comparison of sensitivity in Atlantic salmon in freshwater (96 h LC<sub>50</sub> = 2 µg/L) versus saltwater (NOEC = 265 µg/L). Additionally, results from the pulsed exposures appear to be representative of the ‘all or none’ acute toxicity caused by H<sub>2</sub>S.

**Knezovich, Steichen, Jelinski, and Anderson (1996) *Sulfide tolerance of four marine species used to evaluate sediment and pore-water toxicity***

The authors of this study addressed the potential effects of sulfides on sediment toxicity evaluations, and therefore, were targeting species used in standardized sediment tests. Test organisms included two species of marine amphipod (*Eohaustorius estuarius* and *Rhepoxynius abronius*) and two larval forms commonly used in toxicity testing, the bay mussel (*Mytilus* sp.)

and purple sea urchin (*Strongylocentrotus purpuratus*). The test systems included sealed static and continuous flow test chambers. In addition to 48 h continuous flow testing, the *M. edulis* embryos were exposed to sodium sulfide in short duration exposures of 2 to 10 h, after which they were exposed to clean seawater for the duration of the test (48 h). Test results were expressed as total sulfides (measured using colorimetric detection) and were presented with a calculated percentage H<sub>2</sub>S (9% to 10% of total).

The water-only toxicity tests, which exposed the test organisms to sulfides throughout the exposure period were deemed appropriate for evaluating the water-quality criteria. The larvae are generally used to represent zooplankton. The amphipod tests, while performed on benthic organisms, are appropriate surrogates for marine amphipods and isopods that live in marine algae and kelp. The short-duration exposure tests demonstrated sensitivity to H<sub>2</sub>S over time.

**Llanso (1991) *Tolerance of low dissolved oxygen and hydrogen sulfide by the polychaete *Streblospio benedicti* (Webster)***

This study evaluated the combined effects of low dissolved oxygen and elevated sulfide concentrations on the survival of the polychaete, *Streblospio benedicti*. Measurements were made with a specific ion electrode that provided the concentrations of S<sup>-2</sup> and calculations were made for the concentration of H<sub>2</sub>S. The authors discovered that there was no increased mortality to polychaetes over a 55 h period under anoxic conditions even with H<sub>2</sub>S concentrations as high as 66 µM, which is equivalent to 2,244 µg/L. This is essentially a NOEC for survival over 55 h exposure, but the combined effect of anoxia and increased H<sub>2</sub>S may have been masked by the high mortality of the test organisms under anoxia-only conditions (100%).

**Losso, Novelli, Picone, Ghirardini, Ghetti, Rudello, and Ugo (2004) *Sulfide as a Confounding Factor in Toxicity Tests with the Sea Urchin *Paracentrotus lividus*: Comparisons with Chemical Analysis Data***

This study evaluated the toxicity of sulfides on the sea urchin, *Paracentrotus lividus*, using the sperm cell test and larval development test. Tests were conducted under aerobic conditions in static test containers. Sulfide concentrations were monitored during the exposure period and the calculated half-life in the test chambers was estimated at 50 minutes when test organisms were present. Based on the calculated half-life, the sperm cell test was less affected by the decreasing sulfide concentrations than the 72 h embryo test, which likely had little to no sulfides present for a substantial portion of the test.

Results were expressed as total sulfides and were determined using voltammetric measurements. Using a conversion of 7.89% for H<sub>2</sub>S, the median effective concentrations for the sperm cell test and embryo test were 94.7 µg/L and 34 µg/L, respectively. Based on the calculated half-life, the sperm cell test was less affected by the decreasing sulfide concentrations than the 72-h embryo test, which likely had little to no sulfides for a substantial portion of the test. The authors point out that impurities in the artificial water may have resulted in an increased half-life for total sulfide. EC<sub>50s</sub> appear to have been calculated based on measured sulfide concentrations at test initiation.

**Main and Nelson (1988) *Tolerance of the Sabellariid polychaete *Phragmatopoma lapidonsa* Kinberg to burial, turbidity, and hydrogen sulfide***

Authors exposed the tropical coral tube worm, *Phragmatopoma lapidonsa*, to a series of laboratory manipulations, including sediment burial, turbidity, and hydrogen sulfide. Hydrogen sulfide experiments were conducted with two concentrations of sulfide (4.3 mg/L and 0.048 mg/L) under aerobic and anaerobic conditions in sealed static test chambers. We applied a correction factor of 10% to estimate the H<sub>2</sub>S concentration from the reported total sulfide concentrations. Survival was determined after 24- and 48-h water-only exposures. No significant differences were noted in any of the 24 h treatments. In the 48 h tests, significant decreases in survival were only noted in the 4.3 mg/L treatment under anaerobic conditions, with a 50% decrease in survival, relative to the controls. No significant decrease in survival was noted at this concentration under aerobic conditions.

This study tested a species that may be representative of encrusting worms on structures. The authors noted that hydrogen sulfide concentrations were not controlled and likely decreased throughout the exposure period.

**Marcus, Lutz, and Chanton (1997) *Impact of anoxia and sulfide on the viability of eggs of three planktonic copepods***

The eggs of three species of calanoid copepods were exposed to anoxia, and anoxia combined with the presence of sulfide. The viability of the eggs over exposure periods of up to 32 days was determined in the presence of anoxia alone, and also in the presence of anoxia and 283 µM to 352 µM sulfide concentrations. According to the authors, there were no statistically significant differences in the time of hatching of eggs under anoxia, or anoxic conditions combined with these high sulfide concentrations. However, examination of the data presented in the paper led to a different conclusion by this review. It appears that there is a consistent decrease in time to egg mortality under sulfide addition, and extrapolations from the sulfide concentrations to H<sub>2</sub>S concentrations for these effects average approximately 765 µg/L using a conversion rate of 7.5% for a pH level of 8. This concentration approximates an LOEC for hatching success.

**Miron and Kristensen (1993) *Behavioral response of three Nereid polychaetes to injection of sulfide inside burrows***

This study presents data on behavioral responses of three species of polychaete worms to H<sub>2</sub>S injected into burrow holes. Adult polychaetes (*Nereis virens*, *N. diversicolor*, and *N. succinea*) were placed individually into exposure tubes. After the worms had established a burrow tube from one end of the V-tube to the other, baseline ventilation rates were established. Varying concentrations of sulfides were injected directly into the burrow holes and behavioral responses were recorded. Endpoints used for this test were ventilation rates, duration of ventilation, and duration of rest period. Neither the concentrations of H<sub>2</sub>S nor pH values were determined by the authors, so a 10% correction factor was applied to the reported total dissolved-sulfide concentrations.

There was little change in ventilation rate for any of the species evaluated. During injection, the duration of ventilation increased for *N. virens* and *N. succinea*. Rest periods also increased; however, this may be in response to the increase of ventilation periods. This data shows a potential stress response by polychaetes exposed to sulfides, indicating that polychaetes may be

sensitive to H<sub>2</sub>S. Marine worms are part of the fouling community on offshore structures, and may be a valuable representative species. However, USEPA disallows behavioral endpoints as justification for criteria development. Therefore, these data were disqualified from the development of a site-specific H<sub>2</sub>S criterion, because the significance of the ventilation endpoint is difficult to interpret.

**Theede, Ponat, Hiroki, and Schlieper (1969) *Studies on the resistance of marine bottom invertebrates to oxygen-deficiency and hydrogen sulphide***

Although this is a seminal paper in bringing to light issues of sulfide toxicity and tolerance in animals naturally exposed to sulfides, the authors only tested one concentration of hydrogen sulfide under low oxygen conditions. Because this study was conducted in low oxygen environments and used only one exposure concentration, it was not applicable to this evaluation.

**Thompson, Bay, Greenstein, and Laughlin (1991) *Sublethal effects of hydrogen sulfide in sediment on the urchin *Lytechinus pictus****

This paper presents the results of 49-d sediment toxicity tests exposing adult White sea urchins (*L. pictus*) to sand infused with various concentrations of H<sub>2</sub>S. The authors also conducted a 96 h water-only toxicity test with the adult urchins. An H<sub>2</sub>S solution was diffused up through bedded sediment (clean sand) in a continuous flow test chamber. Urchins were exposed to sulfides in the sediment, as well as to lower overlying water sulfides. Total sulfides were measured in the porewater and overlying water using the methylene blue colorimetric method; and H<sub>2</sub>S concentrations were calculated based on pH, temperature, and salinity. For the sediment test, porewater H<sub>2</sub>S concentrations were 165.8, 91.8, 32.9, and 1.0 µM/L, and overlying water concentrations were 0.273, 0.094, 0.026, and 0.021 µM/L. For the water-only test, H<sub>2</sub>S concentrations were 1.46, 0.63, 0.31, and <0.042 µM/L. Test endpoints for the sediment tests included mortality, avoidance, growth, and gonad growth. The endpoint for the water-only test was mortality.

Adult sea urchins are a species that would be potentially exposed to H<sub>2</sub>S from produced water and would be an appropriate species for establishing WQC. However, the sediment exposures conducted for this literature study are not applicable because sediment is not a likely exposure scenario for produced-water discharges, and it is difficult to translate the dose-response for direct contact to sulfide-spiked sediment with that of an in-water exposure. Although the reported dose response was not used to establish the site-specific H<sub>2</sub>S criterion, potential sublethal responses in adult sea urchins are indicated. The water-only response is an appropriate measure and can be used in an evaluation of H<sub>2</sub>S WQC. However, the species used in this study is a sediment dweller, which is less likely to be exposed to produced water. Adult sea urchins of the genus *Strongylocentrotus* would be a more appropriate candidate species because they form part of the fouling community on offshore structures. Moreover, they are generally considered to be similar in sensitivity to *Lytechinus*. The development of a site-specific criterion would suggest *Strongylocentrotus* as a test species rather than *Lytechinus*.

**Vismann (1996) *Sulfide species and total sulfide toxicity in the shrimp *Crangon crangon*. J. Exp. Mar. Biol. and Ecol.* 204: 141-154.**

Vismann exposed the common shrimp, *Crangon crangon*, to sulfides at varying pH, thus controlling the proportion of undissociated sulfide species. In addition, the author exposed

shrimp at varying sulfide concentrations at a fixed pH of 6.9. Exposures were staged, with 30 minutes of acclimation to test chambers, 30 minutes of pH regulation, 60 minutes of oxygen regulation, 40 minutes of sulfide regulation, and then a 60 minute recovery period. After the recovery period, mortality was determined. Experiments varied the sulfide exposure period, resulting in calculated  $LT_{50}$ 's.

The *C. crangon* shrimp were sensitive to sulfides, with a 1 h  $LC_{50}$  of 20  $\mu$ M total sulfide (37  $\mu$ g/L  $H_2S$ ) at a pH of 8. The link to pH is confirmed with no toxicity at pH 9.8, where there is no sulfide present as  $H_2S$ . Toxicity was directly correlated to pH, and consequently to the  $H_2S$  concentration, indicating that toxicity was due to  $H_2S$ . Behavioral responses were swimming, panic, and paralysis. Threshold concentrations were calculated for each behavior. This experiment indicates that shrimp are sensitive to sulfides. Crangonid and penaeid shrimp are likely to be exposed to produced water and both species lend themselves to laboratory testing. However, data representing behavioral endpoints were rejected for inclusion in WQC development.

### 3.1.4 Selection of Species for Additional Bioassay Testing

Based on the extensive review of pertinent literature in consideration of criteria-development guidelines, a list of species was identified for bioassay testing as part of this investigation. Candidate species for acute toxicity tests were selected based on the 1985 EPA guidelines for developing WQC, and perceived gaps in the existing data in the literature on sulfide toxicity. For marine WQC, at least one species of saltwater animal in at least eight different families must be included. The guidelines specify that test results for the following familial diversity are needed for the development of an acute criterion.

- 1) Two families in phylum chordata
- 2) A family in a phylum other than arthropoda or chordata
- 3) The mysidae or penaeidae family
- 4) Three other families not in the phylum chordata (may include mysidae or penaeidae, whichever was not used before)
- 5) A representative organism from at least one other family

The WQC development also requires the inclusion of chronic endpoints. This guidance suggests that some of the species evaluated for the acute endpoints be evaluated for chronic response in order to develop an acute-to-chronic ratio (ACR). Chronic tests are conducted under continuous flow conditions for a 7-d period.

The results from acute and chronic bioassay tests serve to improve the development of a site-specific criterion for  $H_2S$  exposure from produced-water discharges. In that regard, it is noteworthy that based on other EPA guidance and the nature of  $H_2S$  toxicity, some of the acute tests may also be considered indicative of a chronic endpoint. A list of selected species, life stages, and test endpoints are compared with test requirements in Table 1.



Table 1. Species Recommended for Additional Bioassay Testing

Saltwater Criterion Derivation Requirement		Phylum	Common Name	Species	Life Stage/Size	Test/ Duration
ACUTE						
Results of acceptable acute tests with at least one species of saltwater animal in at least eight different families such that all of the following taxa are included						
Two families in the phylum Chordata	Family 1	Chordata	Topsmelt	<i>Atherinops affinis</i>	7 d to 15 d	96-h acute
	Family 2	Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	Larvae or Juvenile	96-h acute
A family in a phylum other than Arthropoda or Chordata		Annelida	Nereidae	<i>Neanthes arenaceodentata</i>	Juvenile	96-h acute
Either the Mysidae or Penaeidae family <sup>a</sup>		Arthropoda	Penaeid Shrimp	<i>Penaeus</i> sp.	Juvenile	96-h acute
Three families not in the phylum Chordata (may include Mysidae or Penaeidae, whichever was not used above)	Family 1	Arthropoda	Mysid Shrimp	<i>Americamysis bahia</i>	1 d to 5 d	96-h acute
	Family 2	Arthropoda	Amphipod	<i>Ampelisca abdita</i>	Pre-mature Adult	96-h acute
	Family 3	Mollusca	Mussel	<i>Mytilus</i> sp.	Larval development	48-h acute
			Red Abalone	<i>Haliotis rufescens</i>	Larvae	48-h Acute
Any other family		Chordata	Bony Fish	<i>Menidia beryllina</i>	Juvenile	96-h acute
CHRONIC						
Acute-chronic ratios with species of aquatic animals in at least three different families provided that of the three species, at least one of the following taxa is included						
Fish		Chordata	Topsmelt	<i>Atherinops affinis</i>	9 d to 15 d	7-d Survival and Growth
Invertebrate		Arthropoda	Mysid Shrimp	<i>Americamysis bahia</i>	7 d	7-d Survival and Growth
Fish		Chordata	Inland Silverside	<i>Menidia beryllina</i>	9 d to 15 d	7-d Survival and Growth
PLANTS						
Results of at least one acceptable test with a saltwater alga or vascular plant. If plants are among the aquatic organisms most sensitive to the material, results of a test with a plant in another phylum should also be available.						
Plant 1		Bacillariophyta	Diatom	<i>Skeletonema costatum</i>	NA	96-h
Plant 2		Phaeophyta		<i>Fucus serratus</i>	Sporophyte	48-h Germination and growth (germ-tube length)
BIOACCUMULATION						
At least one acceptable bioconcentration factor determined with an appropriate saltwater species, if a maximum permissible tissue concentration is available						
Fish		Chordata	California Killifish	<i>Fundulus parvipinnis</i>	Adult	28-d

<sup>a</sup> An acceptable value exists in literature

### 3.2 RESULTS OF BIOASSAY TESTING

#### 3.2.1 Test Species

The following species were tested under acute conditions: Topsmelt (*Atherinops affinis*), Sheepshead Minnow (*Cyprinodon variegatus*), Polychaete worms (*Neanthes arenaceodentata*), Amphipod (*Ampelisca abdita*), Mysid (*Americamysis bahia*), Bay mussel (*Mytilus* sp.), Inland silverside (*Menidia beryllina*), and Red Abalone (*Haliotis rufescens*). The details of the test regime and the sources of the test organisms are listed in Table 2.

**Table 2. Species Tested for Acute Response**

Test Species	Phylum and EPA Category	Test Regime	Suppliers
Topsmelt <i>Atherinops affinis</i>	Chordata (1)	96-h Flow-through	Aquatic Biosystems Fort Collins, CO
Sheepshead Minnow <i>Cyprinodon variegatus</i>	Chordata (1)	96-h Flow-through	Aquatic Biosystems Fort Collins, CO
Polychaete Worm <i>Neanthes arenaceodentata</i>	Annelida (2)	96-h Flow-through	Don Reish Santa Barbara, CA
Amphipod <i>Ampelisca abdita</i>	Arthropoda (4)	96-h Flow-through	Aquatic Research Organisms, NH
Mysid <i>Americamysis bahia</i>	Arthropoda (4)	96-h Flow-through	Aquatic Biosystems Fort Collins, CO
Bay mussel <i>Mytilus</i> sp.	Mollusca (4)	48-h Static-renewal/ 48-h Spike Test	Carlsbad Aquafarms Carlsbad, CA
Inland silverside <i>Menidia beryllina</i>	Chordata (5)	96-h Flow-through	Aquatic Biosystems Fort Collins, CO
Red Abalone <i>Haliotis rufescens</i>	Mollusca (5)	48-h Spike Test	U.S. Abalone Santa Cruz, CA

Two fish species, *M. beryllina* and *C. variegatus*, and a mysid, *A. bahia*, were selected for chronic exposure testing to meet the requirement for ACR data. Chronic tests were conducted under continuous-flow conditions for a 7-d period (Table 3). Bioaccumulation data for a marine species was adequately represented in (Bagarinao and Vetter (1989); no further bioaccumulation tests were performed.

**Table 3. Species Tested for Chronic Response**

Test Species	Phylum and EPA Category	Test Regime	Suppliers
Inland silverside <i>Menidia beryllina</i>	Chordata (5)	7-d Flow-through	Aquatic Biosystems Fort Collins, CO
Mysid <i>Americamysis bahia</i>	Arthropoda (4)	7-d Flow-through	Aquatic Biosystems Fort Collins, CO
Sheepshead Minnow <i>Cyprinodon variegatus</i>	Chordata (1)	7-d Flow-through	Aquatic Biosystems Fort Collins, CO

### 3.2.2 Exposure Systems

Sulfides rapidly dissipate naturally from marine systems. Consequently, a series of experiments were conducted prior to toxicity testing to determine the half-life of total sulfides at varying concentrations. These studies indicated that the half-life of total sulfides at  $\text{H}_2\text{S}$  concentrations of 30  $\mu\text{g/L}$  and higher was approximately one hour. In contrast, the half-life was much shorter, around 20 minutes, when  $\text{H}_2\text{S}$  concentrations were lower, between 2  $\mu\text{g/L}$  and 20  $\mu\text{g/L}$ . A continuous-flow test system was designed in order to maintain consistent exposure concentrations throughout the 96-hour acute and 7-day chronic tests.

The primary elements of the experimental array are presented schematically in Figure 2. Photographs of the actual test components are shown in Figure 3. The dosing system for the continuous-flow tests consisted of three water bath tables, approximately 12 ft in length and 4 ft wide, which were capable of holding water to a depth of 6 in. A Lexan® hood that was constructed to be vapor-proof covered each table. The custom hoods were sealed onto the tops of the tables by a foam gasket that prevented the escape of any volatilized sulfides that might have been released from the test system. Air was removed from the system via ventilation ducts fitted with exhaust fans that pushed the air through an activated-carbon air treatment system prior to being exhausted outdoors. One-way valves allowed clean air to enter the hoods. Insulated doors allowed access to test containers, pumps, and stock solutions. Seawater at the target test temperature was circulated around the outside of the test chambers to maintain constant test temperature. Waste from flow-through test containers flowed into the water bath, which was designed to overflow into a waste tank. The contents of the waste tank were then pumped to a treatment system to remove sulfides.

One stock solution was prepared for each nominal test concentration. To prepare each concentration of stock solution, the required amount of deionized (DI) water was measured using a 2000 ml graduated cylinder and poured into a large volume glass carboy. The DI water was then bubbled with nitrogen gas and dissolved oxygen content was measured until a level of 0.5 mg/L or less was achieved. Sodium sulfide nonahydrate-hydrated crystals were measured using a Denver TB-215D microbalance and added to the de-oxygenated DI water. The solution was bubbled for approximately 10 minutes to insure adequate mixing of the stock. The stock carboys were covered with parafilm to prevent reentry of any oxygen into the stock solutions.

Test concentrations were generated by combining sulfide stock solutions with aerated seawater. A dosing system was created using two 24-channel peristaltic pumps, one for seawater and one for the stock solution. Each pump delivered either seawater or stock solution from the carboys to test chambers via Tygon® flexible tubing (Figure 3). The pumps were designed to produce a constant flow rate for each channel. In this way, a constant volume of seawater and stock solution was delivered to each test chamber. The test concentrations thus varied according to concentration of the stock solutions rather than delivery flow rate. Each peristaltic pump was calibrated before test initiation.

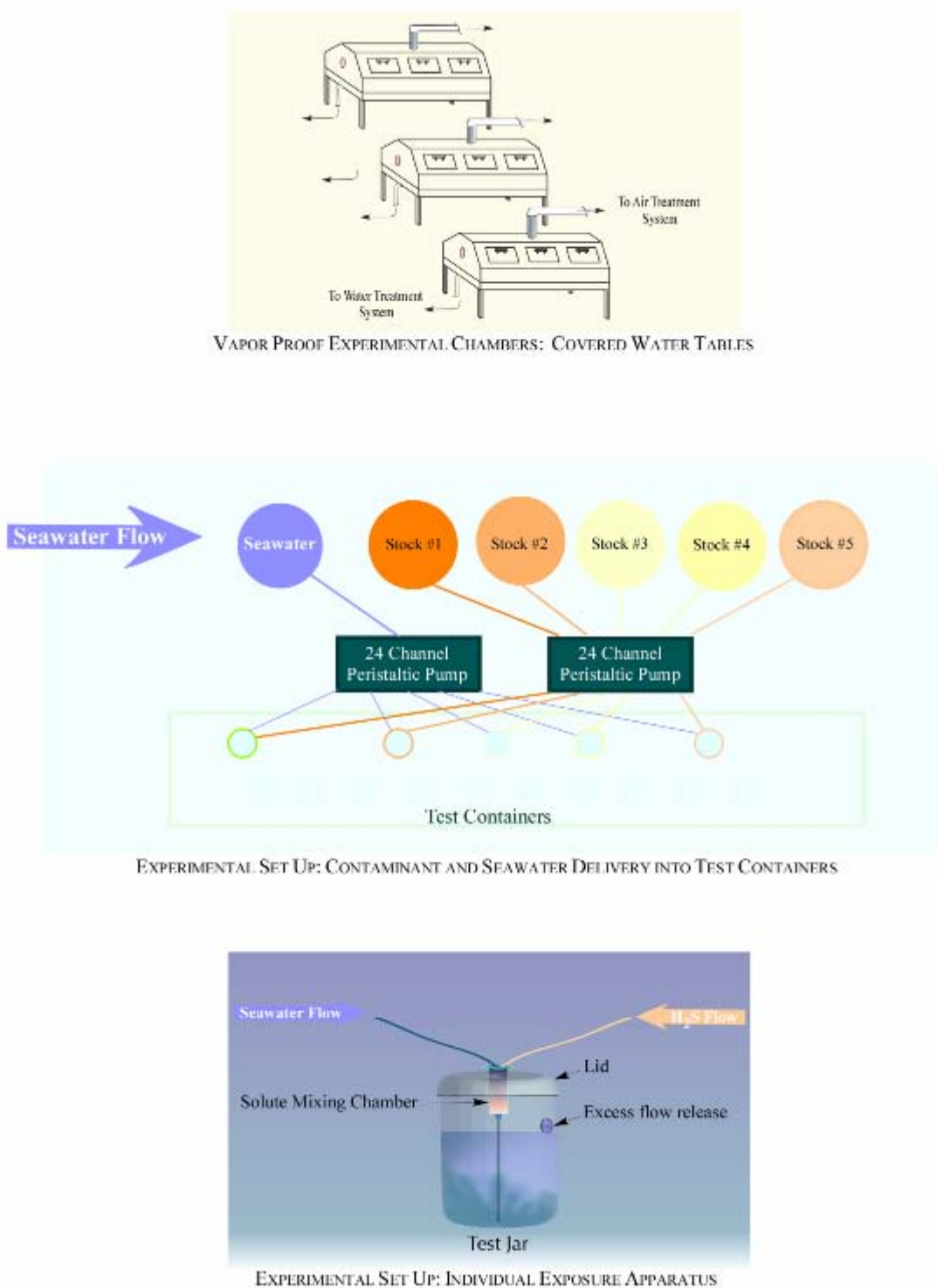
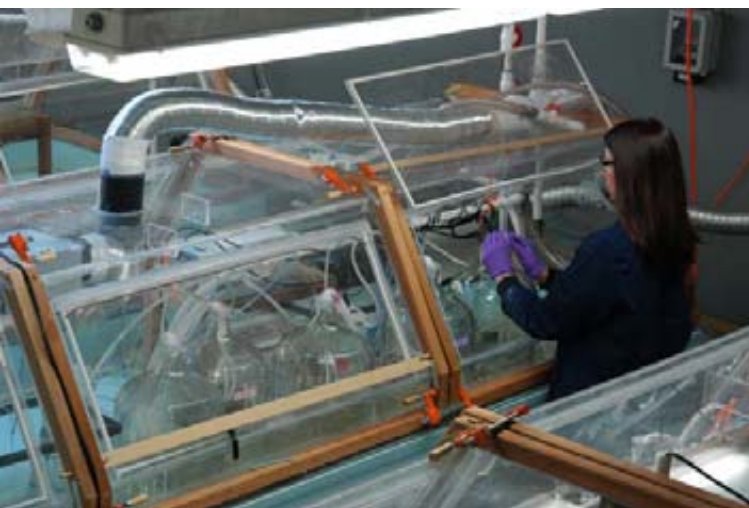


Figure 2. Schematic Representation of Experimental Array



**Figure 3. Photographs of Experimental Array**

For each pump, the channels were numbered 1 through 24, and that number corresponded to the position number for test chambers on the table. For example, the number one position on both the saltwater and the stock solution pumps, when combined, would supply the total flow to the test chamber in position one. Randomization was achieved in two ways. First, the stock concentrations were randomly allocated among the channel numbers. Because position numbers of test chambers on the table corresponded to channel numbers, the randomization of concentrations among channels also resulted in a randomization of the treatment chambers on the table.

Early experiments using a premixed combination of seawater and stock solution in the carboys indicated substantial sulfide loss in the dosing system tubing after the seawater and sulfide stock solution were combined. Based on these observations, the seawater and stock solutions were mixed immediately prior to placement in the test chambers. To allow for adequate mixing prior to delivery to the test chambers, tubing for the stock solution and the saltwater were directed into a plastic syringe prior to entering the test chamber (Figure 3). Mixing in the test chamber was facilitated by delivering the test solution to the bottom of the test chamber, which would then displace water out of the screened exit port at the top of the test chamber. To accommodate for the short half-life, the total flow to each test chamber was sufficient enough to exchange the test volume in 30 minutes.

### 3.2.3 Sulfide Monitoring

Total sulfides were measured using Method 8131 (Hach) with a HACH Ultra Violet Visual DR/4000V Spectrophotometer. This method is modeled after the Methylene Blue Method (ASTM Method 4500-S-D, and USEPA 376.2). The limits of detection for the instrument and method are 5 to 800  $\mu\text{g/L}$  sulfides with a 95% precision around a range. The method measures the concentrations of  $\text{H}_2\text{S}$  and acid-soluble sulfides that react with N,N-dimethyl-p-phenylene diamine sulfate to form methylene blue where the intensity of blue coloration measured at 665 nm is proportional to the total sulfide concentration.

In order to measure total sulfides, a sub-sample of test solution was withdrawn in a 25-mL glass pipette and placed in a clean, solvent-rinsed glass cuvette. Because the instrument measures total sulfides by ultra-violet light, the glass sides of the cuvette were cleaned in order to provide accurate and precise measurements. When measuring total sulfides, the upper detection limit for the HACH DR/4000V Spectrophotometer is 880  $\mu\text{g/L}$ . On occasion, total sulfide concentrations were greater than 880  $\mu\text{g/L}$ . When this occurred, the stock solution was diluted with DI water and the result was multiplied by the ratio of dilution. For high-concentration treatments, a 10-mL or 5-mL aliquot was collected and brought to a total volume of 25 mL using DI. One mL of sulfuric acid was added to each cuvette to decrease the pH. One mL of N,N-dimethyl-p-phenylene diamine sulfate was added to form methylene blue. The contents were then mixed thoroughly by inverting the cuvette 4 to 5 times. After 5 minutes, total sulfides were measured.

Sulfide concentrations were monitored throughout each of the tests. For the flow-through test, total sulfides were measured in each chamber at test initiation, in one replicate twice daily for test days 1 through 3, and in all chambers at test termination. Following measurement of total



sulfides, undissociated  $\text{H}_2\text{S}$  was calculated based on the temperature, salinity, and pH measurements taken at the time of sulfide measurement.

### 3.2.4 Acute and Chronic Bioassay Methods

The acute toxicity of  $\text{H}_2\text{S}$  was evaluated using both 48-h and 96-h exposures. In general, the toxicity testing program followed EPA guidelines for both acute and chronic testing, as described in the manuals: “Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to West Coast Marine and Estuarine Organisms”; “Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms”; and “Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Marine and Estuarine Organisms” (USEPA 1995, 2002bc). Chronic toxicity tests were conducted as 7-day, continuous-flow exposures and were tested using the continuous-flow exposure system. Methods generally followed EPA guidelines, as described in “Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Marine and Estuarine Organisms” (EPA 2002c).

#### Acute 96-hour exposures

96-h acute toxicity tests were conducted as continuous flow-through tests. Four replicate test chambers were prepared for each test treatment. Tests were conducted in 500-mL or 1-L acid-washed, solvent-rinsed glass containers fitted with a screened port at the 300-mL or 800-mL level. Test containers were labeled with predetermined position numbers. The containers were placed in randomly assigned positions in a temperature-controlled water bath where they were allowed to equilibrate for at least one hour. Immediately prior to test initiation, total sulfides and water quality parameters were measured in each test chamber. Water quality observations included DO, temperature, pH, and salinity. Water quality parameters were monitored in all replicates at initiation and termination, and in one replicate on test days 1 through 3. Target test parameters were as follows.

- DO:  $\geq 4.0$  mg/L
- pH:  $8.00 \pm 0.5$  units
- Temperature:  $20^\circ\text{C} \pm 2^\circ\text{C}$
- Salinity:  $28\text{‰} \pm 2\text{‰}$

Total sulfides were measured once or twice daily<sup>a</sup> following the methods presented in Section 3.2.3.

The tests were initiated by randomly allocating 10 test organisms into each test chamber, ensuring that each of the test organisms appeared healthy. Daily observations included number alive, number dead or missing, and general observations regarding the test system and test organisms' health. Mysids were fed <24-h brine shrimp, *Artemia nauplii*, twice daily and each of the fish species were fed <24-h *A. nauplii*, once, at 48-h after test initiation. At test termination, water quality and total sulfides were measured in each test chamber. The number of surviving

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<sup>a</sup> Measurements were generally recorded in the morning and afternoon of each day.

and dead test organisms were also determined at that time. A water-only, 4-day reference-toxicant test with cadmium chloride or copper sulfate was conducted concurrently with each batch of test organisms.

### **Acute 48-hour Larval Bioassay**

The H<sub>2</sub>S sensitivity of *Mytilus galloprovincialis* larvae was evaluated using 48-h, static-renewal tests. While these were considered static-renewal tests, it is important to note that no test solution was removed from the test chamber. Rather, concentrated stock was added to the test chambers at 1.5-h intervals. The amount of stock solution added to each test chamber was increased incrementally as the test proceeded in order to maintain the target H<sub>2</sub>S concentration.

Static-renewal tests were conducted in 600-mL acid-cleaned, solvent-rinsed glass beakers. Four replicate chambers were prepared for each test concentration. Each chamber was then placed in a predetermined, randomly-assigned position in a temperature-controlled room at 16°C. To prepare the test exposures, a concentrated sulfide stock solution was prepared in de-oxygenated deionized water, which was then spiked into 200 mL of seawater provided by the Port Gamble Laboratory's continuous-flow seawater system. Each chamber was then placed in a predetermined randomly-assigned position in a temperature-controlled room at 16°C.

To collect gametes for each test, mussels were placed in clean seawater and acclimated at 12°C for approximately 20 minutes. The water bath temperature was then increased over the period of 15 minutes to 20°C. Mussels were held at 20°C and monitored for spawning individuals. Spawning females were removed from the water bath and placed in individual containers with seawater. Spawning males were removed and placed in a separate water bath with other males. Gametes from at least two males and one female were used to initiate the test. Once sufficient eggs and sperm had been collected, the eggs were screened through 60-µm mesh to remove any detritus or feces, and a homogenized sperm solution was added to the egg solutions. Egg-sperm solutions were periodically homogenized with a perforated plunger during the fertilization process. Approximately one hour after fertilization, embryo solutions were checked for fertilization rate. Only those embryo stocks with >90% fertilization were used to initiate the tests. Embryo solutions were rinsed free of excess sperm on 20-µm mesh and then combined to create one embryo stock solution. Density of the embryo stock solution was determined by counting the number of embryos in a subsample of stock solution. This was used to determine the volume of embryo stock solution to deliver approximately 6,000 embryos to each test chamber.

The test was initiated by randomly allocating an aliquot of the embryo stock solution into each test chamber within two hours of egg fertilization. Embryos were held in suspension during initiation using a perforated plunger. The target stocking density was 20 to 40 embryos/mL.



Temperature, salinity, pH, and total sulfides were measured immediately following each renewal (every 1½ hours). Dissolved oxygen was measured daily. Target test parameters were as follows.

- DO: 60% saturation ( $\geq 4.3$  mg/L)
- pH:  $8.00 \pm 1.00$  units
- Temperature:  $16^{\circ}\text{C} \pm 2^{\circ}\text{C}$
- Salinity:  $30\text{‰} \pm 2\text{‰}$

The test was terminated approximately 48 hours after initiation, when 90% of the control larvae had achieved the prodissoconch I stage. To terminate the larval tests, the contents of each chamber were homogenized using a perforated plunger, a 10 mL subsample was transferred to a scintillation vial, and then preserved in 5% buffered formalin. The numbers of normal and abnormal larvae were enumerated using an inverted microscope. Normal larvae included all D-shaped prodissoconch I stage larvae. Abnormal larvae included abnormally shaped prodissoconch I larvae and all early stage larvae. A 48-h water-only reference-toxicant test with copper sulfate was conducted concurrently with each round of sediment tests.

#### Acute 48-Hour Spike Test

The H<sub>2</sub>S sensitivity of two larval species was evaluated at different life stages by using a spike test. Spike tests were conducted in 20-mL certified clean scintillation vials. Three replicate chambers were prepared for each test concentration at each time interval (0, 6, 12, 24, and 36 hours). Each chamber was then placed in a predetermined, randomly-assigned position in a temperature-controlled room at 16°C and inoculated with the embryo stock solution that was created following the methods presented in Section 4.4.1. To prepare the test exposures, a concentrated sulfide stock solution was prepared in deoxygenated deionized water which, at each time interval, was then spiked into 150 mL of seawater, provided by the Port Gamble Laboratory's continuous-flow seawater system, at doses specific to desired test concentrations. A 10 mL spike of each test concentration was added to the appropriate scintillation vial, according to the time period. The remaining solutions were then diluted by half with saltwater, to mimic the actual test chambers, and water quality and total sulfide were analyzed. The test was terminated approximately 48 hours after initiation.

#### Chronic 7-day Bioassays

The 7-d chronic toxicity tests were conducted as continuous-flow tests. Tests were conducted following a study design similar to that of the 96-hour acute toxicity tests. Water-quality observations included dissolved oxygen (DO), temperature, pH, and salinity and were monitored in all replicates at initiation, termination, and in one replicate on test days 1 through 3. Target test parameters were as follows.

- DO:  $\geq 4.0$  mg/L
- pH:  $8.00 \pm 0.5$  units
- Temperature:  $20^{\circ}\text{C} \pm 2^{\circ}\text{C}$
- Salinity:  $28\text{‰} \pm 2\text{‰}$

Total sulfides were measured once or twice daily following the methods presented in Section 3.2.3.

The tests were initiated by randomly allocating 10 test organisms into each test chamber, ensuring that each of the test organisms appeared healthy. Daily observations included number alive, number dead or missing, and general observations regarding the test system and test organisms' health. All chronic tests were fed <24-h brine shrimp, *Artemia nauplii*, twice daily. At test termination, water quality and total sulfides were measured in each test chamber. The numbers of surviving and dead test organisms were then determined. All surviving test organisms were then placed on tared weigh boats and dried to a stable weight at 60°C (generally 24 hours). Weight of the test organisms was then determined using a Denver TB-215D microbalance. A water-only, 7-d reference-toxicant test with cadmium chloride or copper sulfate was conducted concurrently with each batch of test organisms.

### Data Entry and QA/QC

All water quality and endpoint data were entered into excel spreadsheets. Water-quality parameters were summarized by calculating the mean, minimum, and maximum values for each test treatment. Endpoint data was calculated for each replicate, and then mean values and standard deviations were calculated for each test treatment.

All hand-entered data was reviewed for data entry errors, which were corrected prior to summary calculations. A minimum of 10% of all calculations and data sorting were reviewed for errors. Any apparent outliers were recounted.

### 3.2.5 Results of the Acute Toxicity Tests

#### 96-h Tests with *Menidia beryllina*

Five definitive continuous-flow hydrogen sulfide tests were conducted with the inland silverside (*M. beryllina*). The results of these tests are summarized in Table 4, and a summary of the related water-quality observations is presented in Table 5. The first test was performed on 13 January 2006 and the last test was conducted on 14 March 2006. The tests spanned a range of nominal exposure concentrations that were defined by initial range-testing bioassays. Nominal exposure concentrations in the definitive tests ranged from 0.2 µg/L H<sub>2</sub>S to 133 µg/L H<sub>2</sub>S. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.A.

The three of the acute tests with *M. beryllina* had mean control survival that exceeded 90%, validating these tests. Mean percentage survival in the controls for Test 2 was 85%. This was driven primarily by elevated mortality in two replicates. A clear dose-response was observed in this test and it was considered a valid test. Mean percentage survival in the controls for the last test was 83.3%. However, survival in the 4 µg/L treatment was 96.7%, indicating that the test conditions were suitable for acceptable *M. beryllina* survival. Control survival in the reference-toxicant tests was >90%, further indicating that the test organisms were healthy and suitable for testing. The LC<sub>50</sub> for the cadmium reference-toxicant test for the four tests ranged from 96.7 to 254 mg Cd/L, which was within the control chart limits (40.0 to 291 mg Cd/L), indicating that the test organisms used in this study were similar in sensitivity to those populations previously tested at Weston's Port Gamble Laboratory.

Table 4. Results of Acute Tests on *Menidia beryllina*

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
Test #1 Initiated on 13 January 2006						
Control	1	0.5	0.2	100	92.5	9.6
	2	0.1		100		
	3	0.1		90		
	4	0.1		80		
2.5	1	3.4	4.4	100	95.0	10.0
	2	2.4		100		
	3	6.3		100		
	4	5.3		80		
5	1	14.7	18.8	80	92.5	9.6
	2	15.4		100		
	3	22.7		100		
	4	22.5		90		
10	1	25.1	21.2	100	100.0	0.0
	2	24.7		100		
	3	21.1		100		
	4	13.6		100		
25	1	76.0	63.4	0	<u>0.0</u> <sup>a</sup>	0.0
	2	59.3		0		
	3	41.0		0		
	4	77.2		0		
50	1	133	120.8	0	<u>0.0</u>	0.0
	2	101		0		
	3	129		0		
	4	120		0		
Test #2 Initiated on 19 January 2006						
Control	1	0.2	0.2	100	85.0	12.9
	2	0.2		80		
	3	0.1		70		
	4	0.1		90		
2.5	1	0.9	0.6	70	85.0	12.9
	2	0.7		90		
	3	0.7		80		
	4	0.2		100		
5	1	2.3	2.2	90	90.0	0.0
	2	1.6		90		
	3	1.9		90		
	4	3.0		90		
10	1	9.6	7.8	90	87.5	9.6
	2	7.8		80		
	3	6.3		80		
	4	7.6		100		

<sup>a</sup> Bolded underlined values are statistically significantly different from control

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
15	1	19.1	16.8	60	60.0	14.1
	2	15.0		70		
	3	16.0		40		
	4	16.9		70		
20	1	29.4	25.5	10	<u>27.5</u>	15.0
	2	24.8		40		
	3	23.7		40		
	4	24.0		20		
Test #3 Initiated on 25 January 2006						
Control	1	0.1	0.1	90	96.7	5.8
	2	0.3		100		
	3	0.0		ND <sup>a</sup>		
	4	0.1		100		
2.5	1	0.9	1.0	90	95.0	5.8
	2	1.4		100		
	3	1.0		100		
	4	0.9		90		
5	1	5.9	4.9	100	97.5	5.0
	2	6.7		90		
	3	4.5		100		
	4	2.4		100		
10	1	16.1	15.4	100	95.0	5.8
	2	17.9		90		
	3	13.3		90		
	4	14.4		100		
15	1	43.1	39.6	90	90.0	8.2
	2	41.7		80		
	3	45.7		100		
	4	27.9		90		
20	1	48.7	53.6	80	<u>72.5</u>	22.2
	2	51.9		60		
	3	54.9		100		
	4	59.1		50		
Test #4 Initiated on 13 March 2006						
Control	1	0.0	0.1	90	95	5.8
	2	0.1		100		
	3	0.1		100		
	4	0.2		90		
4	1	1.5	2.1	100	100	0.0
	2	2.1		100		
	3	2.6		100		
	4	2.0		100		
8	1	8.7	7.7	90	95	5.8

<sup>a</sup> ND = No Data

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
	2	7.5		100		
	3	8.4		90		
	4	6.2		100		
16	1	14.3	12.0	100	97.5	5.0
	2	13.7		100		
	3	10.8		90		
	4	9.1		100		
24	1	25.2	21.4	80	85	5.8
	2	23.6		90		
	3	18.9		90		
	4	17.8		80		
32	1	36.0	30.2	70	80	14.1
	2	33.4		100		
	3	27.1		80		
	4	24.3		70		
48	1	53.4	49.8	20	<u>27.5</u>	15.0
	2	60.3		10		
	3	44.2		40		
	4	41.3		40		
Test #5 Initiated on 14 March 2006						
Control	1	0.0	0.2	80	83.3	5.8
	2	0.0		90		
	3	0.5		80		
4	1	2.2	2.1	100	96.7	5.8
	2	2.1		90		
	3	2.0		100		
8	1	10.7	8.8	100	90.0	10.0
	2	6.2		80		
	3	9.6		90		
16	1	17.2	12.8	90	83.3	11.5
	2	10.4		90		
	3	10.7		70		
24	1	21.2	17.3	80	80.0	20.0
	2	16.2		60		
	3	14.6		100		
32	1	30.1	21.9	40	<u>43.3</u>	15.3
	2	15.6		60		
	3	20.1		30		
48	1	34.3	26.7	30	<u>40.0</u>	10.0
	2	22.8		50		
	3	23.0		40		

Table 5. Summary of Water-Quality Observations during the *Menidia beryllina* Bioassays

Treatment	Dissolved Oxygen (mg/L)			Temperature (°C)			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1 Initiated on 13 January 2006</b>												
<b>Control</b>	6.3	5.2	7.2	20.6	20.0	21.0	27.5	25.0	29.0	7.7	7.4	7.9
<b>2.5</b>	5.6	3.4	6.9	20.8	20.2	21.0	24.8	24.0	26.0	7.7	7.5	8.0
<b>5</b>	5.6	4.1	6.8	20.8	20.3	21.0	24.6	23.0	26.0	7.8	7.6	8.1
<b>10</b>	5.7	4.7	6.9	20.8	20.4	21.0	24.4	23.0	26.0	7.9	7.8	8.1
<b>15</b>	5.5	3.4	6.6	20.8	20.4	21.0	24.7	23.0	27.0	8.0	7.7	8.3
<b>20</b>	5.3	1.9	6.6	20.7	20.1	20.9	23.8	22.0	26.0	8.4	7.9	8.8
<b>Test #2 Initiated on 19 January 2006</b>												
<b>Control</b>	6.2	4.6	7.7	19.6	19.4	19.6	30.0	30.0	30.0	7.6	7.3	7.9
<b>2.5</b>	6.0	4.6	7.5	19.6	19.4	19.8	26.9	26.0	28.0	7.7	7.5	7.9
<b>5</b>	5.7	2.7	7.6	19.7	19.6	19.8	27.1	26.0	28.0	7.7	7.5	8.0
<b>10</b>	5.6	3.7	7.5	19.7	19.6	19.8	27.0	26.0	28.0	7.8	7.6	8.0
<b>15</b>	5.3	3.1	7.3	19.7	19.5	19.8	26.7	26.0	27.0	7.8	7.6	8.0
<b>20</b>	5.2	2.3	7.3	19.7	19.6	19.8	26.6	26.0	28.0	7.9	7.6	8.1
<b>Test #3 Initiated on 25 January 2006</b>												
<b>Control</b>	6.6	6.1	6.9	19.2	18.1	19.7	29.4	28.0	30.0	7.6	7.5	7.7
<b>2.5</b>	6.1	5.4	6.5	19.3	18.2	19.8	27.0	26.0	28.0	7.7	7.3	7.8
<b>5</b>	6.5	5.9	7.1	19.2	18.4	19.7	27.1	26.0	28.0	7.7	7.7	7.8
<b>10</b>	5.9	5.5	6.4	19.3	18.4	19.7	27.6	27.0	30.0	7.8	7.7	7.9
<b>15</b>	5.7	5.2	6.8	19.3	18.5	19.7	27.2	27.0	28.0	7.8	7.6	7.9
<b>20</b>	5.3	4.4	6.6	19.2	18.0	19.7	27.1	27.0	28.0	7.9	7.7	8.0
<b>Test #4 Initiated on 13 March 2006</b>												
<b>Control</b>	7.6	6.1	8.4	19.3	18.8	19.7	31.1	31.0	32.0	7.7	7.7	7.9
<b>4</b>	7.7	6.6	8.1	19.4	19.0	19.8	30.0	30.0	30.0	7.8	7.7	7.9
<b>8</b>	7.7	7.1	8.1	19.6	19.3	19.9	30.0	30.0	30.0	7.8	7.7	7.9
<b>16</b>	7.7	7.0	8.1	19.7	19.3	20.0	30.0	30.0	30.0	7.8	7.8	8.0
<b>24</b>	7.7	7.1	8.1	19.7	19.4	19.9	30.0	30.0	30.0	7.9	7.7	8.0
<b>32</b>	7.6	7.0	8.0	19.7	19.4	19.9	30.0	30.0	30.0	7.9	7.8	7.9
<b>48</b>	7.6	7.1	8.0	19.6	19.4	19.8	30.0	30.0	30.0	7.9	7.8	7.9
<b>Test #5 Initiated on 14 March 2006</b>												
<b>Control</b>	7.6	7.2	8.1	19.4	19.0	19.7	30.4	30.0	31.0	7.8	7.7	7.9
<b>4</b>	7.5	7.2	8.0	19.6	19.4	19.8	30.0	30.0	30.0	7.8	7.7	8.0
<b>8</b>	7.5	7.2	8.0	19.7	19.4	19.9	30.0	30.0	30.0	7.8	7.7	8.0
<b>16</b>	7.4	7.0	7.9	19.6	18.9	20.0	30.0	30.0	30.0	7.8	7.8	8.0
<b>24</b>	7.5	7.2	7.9	19.7	19.3	20.0	30.0	30.0	30.0	7.8	7.7	8.0
<b>32</b>	7.4	7.0	7.9	19.6	18.5	20.0	30.0	30.0	30.0	7.9	7.8	8.0
<b>48</b>	7.4	6.9	7.9	19.8	19.3	20.0	30.0	30.0	30.0	7.8	7.7	8.0

As documented in Table 5, temperature and pH were within acceptable ranges throughout the 96-h tests. Dissolved oxygen was equal to or greater than the target limit of 4.4 mg/L in tests 2 through 5; however, minimum DO values recorded for the first test were below the limit for nominal concentrations 2.5, 5, 15, and 20 µg/L H<sub>2</sub>S. Deviations from the target for these concentrations were 1.0, 0.3, 1.0, and 2.5 mg/L, respectively. Aeration was provided to each of the test chambers during this test. Based on the acceptable survival in some of the treatments with low DO, this deviation did not appear to have affected interpretations of dose-response. Salinity values were maintained within acceptable ranges for all tests, with the exception of low values recorded during Test 1 and a slightly elevated value for the Test 4 control treatment. Salinity remained within the tolerance range for this species (EPA 2002c) throughout the test and it is not likely that the reported deviations affected the test response.

For each of the *M. beryllina* tests, the H<sub>2</sub>S concentration in the control seawater was <1 µg/L. During Test 1, the average measured sulfide concentrations in the test treatments were 4.4, 18.8, 21.2, 63.4, and 120.8 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *M. beryllina* exposed to concentrations of 4.4, 18.8, and 21.2 µg/L; whereas complete mortality was observed in the 63.4 and 120.8 µg/L exposures, with mean survival of 0%. The calculated LOEC was 63.4 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 34.6 µg/L H<sub>2</sub>S.

In Test 2, the average measured sulfide concentrations in the test treatments were <1.0, 2.2, 7.8, 16.8, and 25.5 µg/L H<sub>2</sub>S. There was minimal mortality in the control and first three treatments, with mean percentage survival ranging from 85 to 90%. Mortality was observed for the 16.8 and 25.5 µg/L treatments, with mean percentages survival of 60% and 27.5%, respectively. The calculated LOEC was 25.5 µg/L and the LC<sub>50</sub> was 20.8 µg/L H<sub>2</sub>S.

The average measured sulfide concentrations during Test 3 were 1.0, 4.9, 15.4, 39.6, and 53.6 µg/L H<sub>2</sub>S. All treatments except the 53.6 µg/L exposure had mean survival ranging from 90% to 97.5%. The highest exposure concentration, 53.6 µg/L, had a mean percentage survival of 72.5%. The calculated LOEC for Test 3 was 53.7 µg/L and the LC<sub>50</sub> was 65.0 µg/L H<sub>2</sub>S.

During Test 4, the average measured sulfide concentrations in the test treatments were 2.1, 7.7, 12.0, 21.4, 30.2, and 49.8 µg/L H<sub>2</sub>S. No significant mortality was observed in the *M. beryllina* exposed to concentrations up to 12.0 µg/L. Slight mortality was observed in the 21.4 and 30.2 µg/L exposures, with mean survivals of 85% and 80%, respectively. Statistically significant mortality was observed in the highest exposure concentration (49.8 µg/L H<sub>2</sub>S), with a mean percentage survival of 27.5%. The calculated LOEC was 49.8 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 40.4 µg/L H<sub>2</sub>S.

In the fifth test, the average measured sulfide concentrations in the test treatments were 2.1, 8.8, 12.8, 17.3, 21.9, and 26.7 µg/L H<sub>2</sub>S. Slight mortality was observed for the control, 12.8, and 17.3 µg/L treatments, with mean percentages survival of 83.3%, 83.3%, and 80%, respectively. Mortality was higher for the 21.9 µg/L and 26.0 µg/L exposures, with mean percentage survival of 43.3% and 40%, respectively. The calculated LOEC was 21.9 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 24.1 µg/L H<sub>2</sub>S.

### 96-h Tests with *Atherinops affinis*

Two continuous-flow hydrogen sulfide tests were conducted with the topsmelt, *A. affinis*, on 20 and 21 March 2006, with nominal concentrations of 10, 20, 30, 40, and 50 µg/L H<sub>2</sub>S. A summary of *A. affinis* survival is presented in Table 6, and a summary of water-quality observations is presented in Table 7. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.B.

Both tests were validated with >90% survival in the controls. The LC<sub>50</sub> for the copper sulfate reference-toxicant test was 167.48 µgCu/L, which is within the control chart limits (120.32 to 190.0 µgCu/L), indicating that the test organisms used in this study were similar in sensitivity to those populations previously tested at the Port Gamble Laboratory. Temperature, dissolved oxygen, and pH were within acceptable ranges throughout the 96-h tests. Salinity was generally within the target range of 28‰ ± 2‰ for both tests. Deviations from this range included elevated salinity values in the last treatment of Test 1 and the first treatment of Test 2. In both cases, the observed salinity was within the tolerance range for this fish (ranging from 5‰ to 34‰; EPA 1995), so these deviations were not believed to have affected test subject survival.

The observed H<sub>2</sub>S concentration in the control seawater for Tests 1 and 2 was <1 µg/L. During Test 1, the average measured sulfide concentrations in the test treatments were 11.4, 22.6, 35.9, 47.7, and 68.5 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *A. affinis* control treatment; whereas some mortality was observed in all other exposures, with mean survival ranging from 80% in the 22.6 µg/L exposure to 7.5% in the 68.5 µg/L exposure. The calculated LOEC was 11.4 µg/L H<sub>2</sub>S, and the EC<sub>50</sub> was 41.6 µg/L H<sub>2</sub>S.

In Test 2, the average measured sulfide concentrations in the test treatments were 9.4, 21.3, 34.5, 44.0, and 69.5 µg/L H<sub>2</sub>S. There was no significant mortality observed in exposures up to 21.3 µg/L. Mean percent survival was 46.7% in the 34.5 and 44.0 µg/L exposures and 10% in the 69.5 µg/L exposure. The calculated LOEC was 34.5 µg/L H<sub>2</sub>S, and the EC<sub>50</sub> was 37.8 µg/L H<sub>2</sub>S.

**Table 6. Results of Acute Tests on *Atherinops affinis***

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
<b>Test #1 Initiated on 20 March 2006</b>						
Control	1	0.4	0.4	90	90.0	0.0
	2	0.2		90		
	3	0.4		90		
	4	0.6		90		
10	1	11.4	11.4	70	<b>70.0</b>	8.2
	2	10.0		70		
	3	12.6		80		
	4	11.6		60		
20	1	23.3	22.6	80	<b>80.0</b>	0.0
	2	19.5		80		
	3	24.5		80		
	4	22.9		80		
30	1	35.8	35.9	40		18.3



Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
	2	31.2		70	<u>50.0</u>	
	3	40.1		60		
	4	36.3		30		
40	1	49.3	47.7	30	<u>30.0</u>	8.2
	2	47.2		20		
	3	51.7		30		
	4	42.8		40		
50	1	68.5	68.5	10	<u>7.5</u>	9.6
	2	66.8		20		
	3	72.9		0		
	4	66.0		0		
Test #2 Initiated on 21 March 2006						
Control	1	0.2	0.1	100	100.0	0.0
	2	0.0		100		
	3	0.1		100		
	4	0.0		100		
10	1	12.7	9.4	100	100.0	0.0
	2	7.9		100		
	3	10.0		100		
	4	7.2		ND		
20	1	28.9	21.3	90	90.0	0.0
	2	16.4		90		
	3	23.1		90		
	4	16.8		ND		
30	1	39.6	34.5	50	<u>46.7</u>	5.8
	2	29.6		40		
	3	38.0		50		
	4	30.9		ND		
40	1	45.1	44.0	40	<u>46.7</u>	11.5
	2	42.5		60		
	3	49.8		40		
	4	38.5		ND		
50	1	71.9	69.5	30	<u>10.0</u>	17.3
	2	64.0		0		
	3	72.2		0		
	4	70.0		ND		

Table 7. Summary of Water-Quality Observations during the *Atherinops affinis* Bioassays

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1 Initiated on 20 March 2006</b>												
<b>Control</b>	7.4	7.1	7.6	20.6	19.5	21.0	29.6	29.0	30.0	7.8	7.8	7.9
<b>10</b>	7.3	7.0	7.6	20.6	19.6	21.0	29.6	29.0	30.0	7.9	7.8	8.0
<b>20</b>	7.3	6.9	7.5	20.8	19.8	21.0	29.6	29.0	30.0	7.9	7.8	8.0
<b>30</b>	7.2	6.9	7.6	20.7	19.9	21.0	29.6	29.0	30.0	7.9	7.8	8.0

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>40</b>	7.2	6.8	7.5	20.7	19.9	21.1	29.6	29.0	30.0	8.0	7.8	8.1
<b>50</b>	7.4	7.1	7.6	20.4	19.7	21.0	30.6	30.0	31.0	7.8	7.7	7.8
<b>Test #2 Initiated on 21 March 2006</b>												
<b>Control</b>	7.0	6.6	7.3	19.2	18.5	20.1	31.1	31.0	32.0	7.7	7.6	7.9
<b>10</b>	6.9	6.5	7.3	19.5	18.9	20.6	30.0	30.0	30.0	7.8	7.7	7.9
<b>20</b>	6.9	6.6	7.3	19.6	19.0	20.9	30.0	30.0	30.0	7.9	7.8	8.0
<b>30</b>	6.9	6.7	7.3	19.6	18.8	20.8	30.0	30.0	30.0	7.9	7.8	8.1
<b>40</b>	6.7	6.3	7.3	19.4	18.7	20.7	30.0	30.0	30.0	7.9	7.8	8.1
<b>50</b>	6.7	6.3	7.3	19.5	18.8	20.7	30.0	30.0	30.0	7.9	7.9	8.1

### 96-h Tests with *Cyprinodon variegatus*

Three continuous-flow hydrogen sulfide tests were conducted with the sheepshead minnow, *C. variegatus*. The first test occurred on 10 February 2006, with nominal concentrations of 2.5, 5, 10, 15, and 20 µg/L H<sub>2</sub>S. Tests 2 and 3 were performed on 15 February 2006 with nominal concentrations of 15, 25, 50, 75, and 100 µg/L H<sub>2</sub>S. A summary of *C. variegatus* survival is presented in Table 8. and a summary of water-quality observations is presented in Table 9. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.C.

Test 1 was validated with a mean control survival of 97.5%. Mean percentage survival in the controls for Tests 2 and 3 was 82.5% and 85%. This slight deviation from target survival did not appear to affect the determination of a dose-response. It may also indicate that the observed responses represent a conservative estimate of toxicity. There were an insufficient number of test organisms available from the supplier to allow for a concurrent reference toxicant test.

Temperature and pH were within acceptable ranges throughout the 96-h tests. Salinity was slightly below the target range; however, it remained within the tolerance range for *C. variegatus* (5% to 32‰) throughout the test and did not affect test results. Dissolved oxygen was above the target limit of 4.4 mg/L for all treatments, with the exception of the 50, 75, and 100 µg/L treatments in Test 2 (Days 3 and 4). Trickle-flow aeration was provided for each of the test chambers on Day 3. Because some test chambers with decreased dissolved oxygen had survival rates of 80% to 90%, it is unlikely that the deviations affected test results.

During Test 1, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 2.6, 13.6, 16.6, 25.4, and 40.1 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *C. variegatus* exposed to any of the treatments during Test 1. The calculated LOEC and estimated LC<sub>50</sub> were >40.1 µg/L H<sub>2</sub>S.

In Test 2, the control seawater mean H<sub>2</sub>S concentration was <1 µg/L. Mean measured exposure concentrations were 35.4, 44.9, 69.2, 118, and 114 µg/L H<sub>2</sub>S. Mean percentage survival for these treatments was ranged from 82.5% in the 44.9 µg/L treatment to 60% in the 118 µg/L treatment. The calculated LOEC and estimated LC<sub>50</sub> was >118 µg/L H<sub>2</sub>S.

Table 8. Results of Acute Tests on *Cyprinodon variegatus*

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
Test #1 Initiated on 10 February 2006						
Control	1	0.3	0.2	95	97.5	2.9
	2	0.2		95		
	3	0.1		100		
	4	0.1		100		
2.5	1	2.1	2.6	95	98.8	2.5
	2	2.9		100		
	3	4.1		100		
	4	1.4		100		
5	1	10.3	13.6	100	100.0	0.0
	2	8.6		100		
	3	29.8		100		
	4	5.7		100		
10	1	23.3	16.6	100	100.0	0.0
	2	16.9		100		
	3	12.9		100		
	4	13.1		100		
15	1	27.2	25.4	95	95.0	4.1
	2	26.2		100		
	3	26.4		95		
	4	21.9		90		
20	1	49.4	40.1	100	97.5	2.9
	2	46.7		95		
	3	28.2		100		
	4	36.1		95		
Test #2 Initiated on 15 February 2006						
Control	1	0.6	0.5	70	82.5	15.0
	2	0.8		90		
	3	0.3		100		
	4	0.2		70		
15	1	30.9	35.4	70	77.5	9.6
	2	38.1		80		
	3	39.4		90		
	4	33.4		70		
25	1	37.1	44.9	80	82.5	5.0
	2	55.5		80		
	3	53.5		80		
	4	33.3		90		
50	1	67.9	69.2	70	80.0	8.2
	2	85.6		80		
	3	71.7		80		
	4	51.6		90		
75	1	107.7	117.6	50	67.5	17.1
	2	146.1		70		
	3	112.1		90		

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
	4	104.7		60		
100	1	115.0	113.9	30	60.0	24.5
	2	144.5		80		
	3	111.0		50		
	4	85.0		80		
Test #3 Initiated on 15 February 2006						
Control	1	0.4	0.5	100	85.0	12.9
	2	1.1		70		
	3	0.2		80		
	4	0.3		90		
15	1	25.3	25.7	80	85.0	5.8
	2	28.5		80		
	3	21.5		90		
	4	27.6		90		
25	1	37.5	38.9	90	77.5	22.2
	2	53.8		70		
	3	38.2		100		
	4	26.0		50		
50	1	58.2	60.3	80	85.0	5.8
	2	72.1		90		
	3	60.7		90		
	4	50.2		80		
75	1	97.6	103.7	60	<b><u>60.0</u></b>	0.0
	2	134.5		60		
	3	105.3		60		
	4	77.3		60		
100	1	115.6	102.0	20	<b><u>57.5</u></b>	26.3
	2	108.4		60		
	3	106.9		80		
	4	76.9		70		

Table 9. Summary of Water-Quality Observations during the *Cyprinodon variegatus* Bioassays

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1 Initiated on 10 February 2006</b>												
Control	7.7	7.1	8.2	19.3	18.7	19.9	30.0	29.0	31.0	7.9	7.6	8.1
2.5	7.6	7.1	7.9	19.5	19.0	19.8	27.5	26.0	30.0	8.0	7.5	8.1
5	7.5	6.2	7.9	19.5	18.6	19.9	27.4	26.0	28.0	8.0	7.7	8.2
10	7.4	6.5	7.9	19.6	19.0	19.9	27.5	26.0	30.0	8.0	7.7	8.2
15	7.3	6.2	7.9	19.5	18.8	19.8	27.5	26.0	30.0	8.1	7.7	8.3
20	7.2	6.0	7.8	19.5	19.2	19.8	27.5	27.0	30.0	8.1	7.8	8.3
<b>Test #2 Initiated on 15 February 2006</b>												
Control	7.4	6.7	8.2	19.2	18.5	20.3	29.7	29.0	31.0	7.9	7.7	8.1
15	7.0	5.0	7.7	19.3	18.5	20.4	27.4	25.0	29.0	8.0	7.8	8.2
25	6.8	5.9	7.5	19.4	18.5	20.4	27.2	26.0	29.0	8.1	7.9	8.3

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>50</b>	6.4	0.2	8.2	19.3	18.0	20.4	27.3	26.0	29.0	8.2	8.1	8.4
<b>75</b>	6.4	3.2	7.3	19.2	18.0	20.4	27.1	25.0	29.0	8.4	8.2	8.6
<b>100</b>	6.0	0.2	7.9	19.2	18.0	20.4	27.5	26.0	29.0	8.5	8.3	8.7
<b>Test #3 Initiated on 15 February 2006</b>												
<b>Control</b>	7.4	6.8	8.6	19.2	18.5	20.0	29.7	29.0	31.0	7.9	7.7	8.1
<b>15</b>	6.9	5.5	8.2	19.3	18.5	20.4	27.4	25.0	29.0	8.0	7.8	8.2
<b>25</b>	6.6	4.8	7.7	19.4	18.5	20.5	27.2	26.0	29.0	8.1	7.8	8.3
<b>50</b>	6.2	3.2	7.5	19.3	18.0	20.4	27.3	26.0	29.0	8.2	8.0	8.4
<b>75</b>	5.7	2.3	6.9	19.3	18.0	20.4	27.1	25.0	29.0	8.3	8.1	8.6
<b>100</b>	5.8	1.3	7.0	19.2	17.9	20.3	27.5	26.0	29.0	8.5	8.3	8.7

Test 3 had a mean observed H<sub>2</sub>S concentration in the control seawater of <1 µg/L. The average measured sulfide concentrations in the test treatments were 25.7, 38.9, 60.3, 104, and 102 µg/L H<sub>2</sub>S. Corresponding survival percentages for these treatments were 85%, 77.5%, 85%, 60%, and 57.5%. The computed LOEC was 60.3 µg/L and the LC<sub>50</sub> was 83.9 µg/L H<sub>2</sub>S.

#### 96-h Tests with *Americamysis bahia*

Two continuous-flow hydrogen sulfide tests were conducted with the mysid, *A. bahia*, on 25 January and 5 February 2006. The first test was performed with nominal concentrations of 2.5, 5, 10, 15, and 20 µg/L H<sub>2</sub>S, while the second test had nominal concentrations of 2, 4, 6, 8, and 12 µg/L H<sub>2</sub>S. A summary of *A. bahia* survival is presented in Table 10 and a summary of water-quality observations is presented in Table 11. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.D.

Both tests were validated with >90% mean survival in the controls. The LC<sub>50</sub>'s for the copper sulfate reference-toxicant tests were 245 µgCu/L and 356 µgCu/L, which are within the control chart limits (181 to 392 µgCu/L), indicating that the test organisms used in this study were similar in sensitivity to those populations previously tested at the Port Gamble Laboratory. Temperature and pH were within acceptable ranges throughout the 96-h tests. Dissolved oxygen was slightly below the target limit of 4.4 mg/L in Test 2, with deviations ranging from 0.4 to 2.2 mg/L. Trickle-flow aeration was provided to these test treatments, increasing the DO to acceptable levels. Salinity was within the target range during the first test, but minimum measured values for the second test were below target in four replicates. Salinity remained within the tolerance range for *A. mysis* (20‰ to 32‰). These deviations did not significantly impact the mean salinity values, and were not likely to affect the test results.

During Test 1, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 3.6, 14.6, 33.0, 59.0, and 78.3 µg/L H<sub>2</sub>S. Significant mortality was observed in the *A. bahia* exposed to 14.6 µg/L exposure, resulting in a mean percentage survival of 2.5%. Complete mortality, or 0% survival, occurred in the remaining three exposure concentrations. The calculated LOEC was 14.6 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 7.2 µg/L H<sub>2</sub>S.

Table 10. Results of Acute Tests on *Americamysis bahia*

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
Test #1 Initiated on 25 January 2006						
Control	1	0.0	0.2	80	90.0	11.5
	2	0.8		100		
	3	0.1		100		
	4	0.1		80		
2.5	1	3.0	3.6	80	87.5	9.6
	2	3.7		90		
	3	4.2		80		
	4	3.5		100		
5	1	13.7	14.6	0	<u>2.5</u>	5.0
	2	17.8		0		
	3	14.4		0		
	4	12.5		10		
10	1	29.6	33.0	0	<u>0.0</u>	0.0
	2	31.2		0		
	3	41.0		0		
	4	30.4		0		
15	1	57.3	59.0	0	<u>0.0</u>	0.0
	2	61.3		0		
	3	59.5		0		
	4	57.9		0		
20	1	84.5	78.3	0	<u>0.0</u>	0.0
	2	82.2		0		
	3	84.3		0		
	4	62.2		0		
Test #2 Initiated on 5 February 2006						
Control	1	0.3	0.2	100	97.5	5.0
	2	0.2		100		
	3	0.3		100		
	4	0.2		90		
2	1	1.2	2.1	90	85.0	10.0
	2	1.6		90		
	3	1.9		90		
	4	3.6		70		
4	1	4.0	5.5	100	90.0	8.2
	2	3.0		90		
	3	8.7		90		
	4	6.2		80		
6	1	10.6	9.9	60	<u>35.0</u>	23.8
	2	11.2		20		
	3	8.3		50		
	4	9.5		10		
8	1	14.1	14.5	0	<u>20.0</u>	24.5
	2	14.3		0		
	3	17.0		50		

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
	4	12.6		30		
12	1	34.5	29.1	0	<b>40.0</b>	40.8
	2	15.7		10		
	3	29.8		70		
	4	36.3		80		

Table 11. Summary of Water-Quality Observations during the *Americamysis bahia* Bioassays

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1 Initiated on 25 January 2006</b>												
<b>Control</b>	6.8	6.5	7.1	19.1	18.3	19.7	29.4	28.0	30.0	7.6	7.3	7.7
<b>2.5</b>	6.5	5.3	6.8	19.3	18.2	19.7	27.0	26.0	28.0	7.7	7.6	7.8
<b>5</b>	6.8	6.4	7.1	19.2	18.4	19.7	27.1	26.0	28.0	7.8	7.7	7.8
<b>10</b>	6.6	5.8	7.0	19.2	18.3	19.6	27.6	27.0	30.0	7.8	7.7	7.9
<b>15</b>	6.5	6.2	7.0	19.2	18.5	19.7	27.2	27.0	28.0	7.9	7.8	7.9
<b>20</b>	6.3	5.6	6.9	19.2	18.0	19.7	27.1	27.0	28.0	7.9	7.8	8.0
<b>Test #2 Initiated on 5 February 2006</b>												
<b>Control</b>	6.9	3.2	8.6	19.1	18.5	19.7	29.9	29.0	31.0	7.8	7.6	8.0
<b>2</b>	6.8	4.0	8.8	19.4	18.9	19.9	26.6	23.0	28.0	7.8	7.5	8.1
<b>4</b>	6.8	4.6	8.2	19.5	18.9	19.9	26.8	25.0	27.0	7.9	7.6	8.1
<b>6</b>	7.2	6.3	8.2	19.5	18.9	20.0	26.8	24.0	28.0	8.0	7.7	8.1
<b>8</b>	7.2	6.1	8.2	19.4	19.0	20.0	26.8	25.0	28.0	8.0	7.7	8.1
<b>12</b>	6.9	2.2	8.4	19.5	19.1	20.0	27.4	26.5	29.0	7.9	7.4	8.1

In Test 2, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 2.1, 5.5, 9.9, 14.5, and 29.1 µg/L H<sub>2</sub>S. There was slight mortality observed in the 2.1 µg/L exposure, with a mean survival of 85%. No significant mortality was observed for the 5.5 µg/L treatment. The mean percentage survival was 35%, 20%, and 40% for the 9.9, 14.5, and 29.1 µg/L exposures, respectively. The calculated LOEC was 9.9 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 11.1 µg/L H<sub>2</sub>S.

#### 96-h Tests with *Neanthes arenaceodentata*

Two continuous-flow hydrogen sulfide tests were conducted with the polychaete *N. arenaceodentata* on 17 February 2006, with nominal concentrations of 2.5, 5, 10, 25, and 50 µg/L H<sub>2</sub>S. A summary of *N. arenaceodentata* survival is presented in Table 12 and a summary of water-quality observations is presented in Table 13. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.E.

Table 12. Results of Acute Tests on *Neanthes arenaceodentata*

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
Test #1 Initiated on 17 February 2006						
Control	1	0.3	0.5	100	100.0	0.0
	2	0.7		100		
	3	0.6		100		
	4	0.5		100		
2.5	1	3.3	7.7	100	100.0	0.0
	2	7.9		100		
	3	6.7		100		
	4	13.0		100		
5	1	25.2	15.2	100	95.0	10.0
	2	4.8		80		
	3	10.3		100		
	4	20.3		100		
10	1	50.3	38.0	100	100.0	0.0
	2	33.0		100		
	3	37.3		100		
	4	31.5		100		
25	1	120	99.6	100	97.5	5.0
	2	83.2		90		
	3	97.0		100		
	4	98.3		100		
50	1	71.9	123.4	100	92.5	9.6
	2	147		80		
	3	128		90		
	4	147		100		
Test #2 Initiated on 17 February 2006						
Control	1	0.6	0.6	100	100.0	0.0
	2	0.6		100		
	3	0.7		100		
	4	0.5		100		
2.5	1	2.0	4.3	100	95.0	5.8
	2	5.0		90		
	3	4.3		90		
	4	5.7		100		
5	1	13.3	8.0	100	100.0	0.0
	2	3.9		100		
	3	6.2		100		
	4	8.8		100		
10	1	31.7	23.9	100	97.5	5.0
	2	27.4		90		
	3	20.0		100		
	4	16.4		100		
25	1	117.1	81.3	90	95.0	5.8
	2	67.8		90		
	3	64.4		100		



Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
	4	75.8		100		
50	1	59.5	86.6	100	97.5	5.0
	2	87.7		90		
	3	83.1		100		
	4	116		100		

Table 13. Summary of Water-Quality Observations during the *Neanthes arenaceodentata* Bioassays

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1 Initiated on 17 February 2006</b>												
<b>Control</b>	7.8	7.0	9.4	19.1	18.7	19.5	30.1	30.0	31.0	7.8	7.7	7.9
<b>2.5</b>	7.8	7.0	9.4	19.5	19.1	19.7	27.4	26.0	30.0	7.9	7.8	8.0
<b>5</b>	7.7	6.4	9.7	19.6	19.3	19.8	28.2	26.0	30.0	8.0	7.8	9.8
<b>10</b>	7.5	6.4	8.7	19.5	19.2	19.8	27.6	26.0	29.0	7.9	7.8	8.0
<b>25</b>	7.2	5.2	9.4	19.5	18.9	19.8	27.8	26.0	30.0	8.0	7.8	8.2
<b>50</b>	7.3	5.6	9.1	19.5	19.0	19.9	28.0	26.0	30.0	8.4	7.8	11.5
<b>Test #2 Initiated on 17 February 2006</b>												
<b>Control</b>	7.8	7.0	9.1	19.2	18.5	19.5	30.1	30.0	31.0	7.8	7.7	7.9
<b>2.5</b>	7.6	6.7	9.2	19.4	19.1	19.6	27.4	26.0	30.0	7.8	7.7	7.9
<b>5</b>	7.7	6.6	9.6	19.5	19.2	19.7	28.2	26.0	30.0	7.8	7.7	8.0
<b>10</b>	7.1	6.1	8.1	19.4	19.2	19.6	27.6	26.0	29.0	7.8	7.7	8.0
<b>25</b>	6.6	4.1	9.1	19.4	19.1	19.7	27.8	26.0	30.0	7.9	7.6	8.0
<b>50</b>	6.4	4.1	8.0	19.5	19.2	19.9	28.0	26.0	30.0	8.1	7.9	8.3

Both tests were validated with mean control survival of 100%. A reference-toxicant test was not conducted with the February 17 tests, due to insufficient availability of worms. Temperature was within the acceptable range throughout the 96-h tests. The minimum measured dissolved oxygen value was slightly below the target limit of 4.4 mg/L in two treatments of the second test. This deviation of 0.3 mg/L was not believed to have affected survival. Salinity was slightly elevated in the control for both tests, but survival was not affected.

During both tests, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the first test treatments were 7.7, 15.2, 38.0, 99.6, and 123.4 µg/L H<sub>2</sub>S. For Test 2, average measured sulfide concentrations were 4.3, 8.0, 23.9, 81.3, and 86.6 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *N. arenaceodentata* in any of the H<sub>2</sub>S exposures during either test. Mean percentage survival was >90% for all treatments. In Test 1, the calculated LOEC and LC<sub>50</sub> were >123.5 µg/L H<sub>2</sub>S. Test 2 had a calculated LOEC and LC<sub>50</sub> of >86.6 µg/L H<sub>2</sub>S.

### 96-h Tests with *Ampelisca abdita*

Two continuous-flow hydrogen sulfide tests were conducted with the amphipod *A. abdita* on 23 February 2006, with nominal concentrations of 2.5, 5, 10, 15, and 20 µg/L H<sub>2</sub>S. A summary of *A. abdita* survival is presented in Table 14, and a summary of water-quality observations is presented in Table 15. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.F.

Test 1 was validated with a mean control survival of 95%. Mean percentage survival in the controls for Test 2 was 77.5%. This was driven primarily by high mortality in one replicate. Survival was 92.5% in the 2.5 µg/L treatment, indicating that the test conditions were acceptable for *A. abdita* survival. The LC<sub>50</sub> for the cadmium reference-toxicant test was 0.64 mg Cd/L, which is within the control chart limits (0.00 to 0.86 mg Cd/L), indicating that the test organisms used in this study were similar in sensitivity to those populations previously tested at the Port Gamble Laboratory. Temperature, salinity, and pH were within acceptable ranges throughout the 96-h tests. Dissolved oxygen was slightly below the target limit of 4.4 mg/L in the 20 µg/L treatment. Trickle-flow aeration was provided to these treatments. These small deviations were not expected to have affected the test results.

During Test 1, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 1.4, 9.4, 22.2, 45.3, and 66.4 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *A. abdita* exposed to concentrations of 1.4 and 9.4 µg/L; whereas significant mortality was observed in the 22.2, 45.3, and 66.4 µg/L exposures, with mean survival of 65%, 47.5%, and 20%, respectively. The calculated LOEC was 22.2 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 40.2 µg/L H<sub>2</sub>S.

In Test 2, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were <1.0, 6.7, 17.7, 38.2, and 55.7 µg/L H<sub>2</sub>S. There was no significant mortality observed in any of the H<sub>2</sub>S exposures, with mean survival ranging from 80.0 to 92.5 in the test treatments. Test 2 had a calculated LOEC and LC<sub>50</sub> of >55.7 µg/L H<sub>2</sub>S.

**Table 14. Results of Acute Tests on *Ampelisca abdita***

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
<b>Test #1 Initiated on 23 February 2006</b>						
Control	1	0.3	0.2	100	95.0	10.0
	2	0.2		100		
	3	0.2		100		
	4	0.2		80		
2.5	1	1.9	1.4	90	85.0	10.0
	2	1.2		70		
	3	1.2		90		
	4	1.1		90		
5	1	11.3	9.4	100	87.5	9.6
	2	9.3		80		

Treatment	Replicate	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percentage Survival	Mean Survival	Standard Deviation
	3	8.6		80		
	4	8.5		90		
10	1	27.2	22.2	70	65.0	17.3
	2	20.5		40		
	3	20.4		80		
	4	20.7		70		
15	1	45.9	45.3	60	47.5	15.0
	2	45.1		60		
	3	46.5		40		
	4	43.8		30		
20	1	76.2	66.4	40	20.0	23.1
	2	69.9		40		
	3	66.0		0		
	4	53.6		0		
Test #2 Initiated on 23 February 2006						
Control	1	0.2	0.3	100	77.5	20.6
	2	0.5		80		
	3	0.1		80		
	4	0.3		50		
2.5	1	1.2	0.8	100	92.5	9.6
	2	0.5		90		
	3	0.8		100		
	4	0.9		80		
5	1	8.6	6.7	100	87.5	25.0
	2	6.9		100		
	3	5.7		100		
	4	5.4		50		
10	1	19.3	17.7	100	90.0	14.1
	2	19.0		70		
	3	15.8		100		
	4	16.7		90		
15	1	34.6	38.2	100	82.5	20.6
	2	47.5		60		
	3	37.0		100		
	4	33.6		70		
20	1	65.5	55.7	60	80.0	16.3
	2	62.6		80		
	3	52.7		100		
	4	42.0		80		

**Table 15. Summary of Water-Quality Observations during the *Ampelisca abdita* Bioassays**

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1 Initiated on 23 February 2006</b>												
<b>Control</b>	7.6	6.7	8.1	18.9	17.8	19.6	30.1	29.0	31.0	7.8	7.8	7.9
<b>2.5</b>	7.6	6.9	8.0	19.0	17.7	19.7	27.7	27.0	28.0	7.9	7.8	8.0
<b>5</b>	7.5	6.8	8.0	19.0	17.8	19.6	27.6	27.0	28.0	7.9	7.8	8.0
<b>10</b>	7.4	6.2	8.0	19.0	17.6	19.8	27.4	27.0	28.0	7.9	7.8	8.1
<b>15</b>	7.1	4.1	7.9	19.0	17.6	19.7	27.6	27.0	28.0	8.0	8.0	8.1
<b>20</b>	7.0	5.6	8.0	19.0	17.5	19.7	27.5	27.0	28.0	8.1	8.0	8.2
<b>Test #2 Initiated on 23 February 2006</b>												
<b>Control</b>	7.5	6.7	8.0	19.0	17.8	19.7	30.1	29.0	31.0	7.8	7.7	7.9
<b>2.5</b>	7.4	6.8	7.9	19.1	17.8	19.7	27.7	27.0	28.0	7.9	7.8	7.9
<b>5</b>	7.3	6.5	7.8	19.1	17.9	19.6	27.6	27.0	28.0	7.9	7.8	8.0
<b>10</b>	7.0	5.6	7.8	19.1	17.6	19.8	27.5	27.0	28.0	7.9	7.8	8.0
<b>15</b>	6.7	5.3	7.8	19.0	17.7	19.7	27.6	27.0	28.0	8.0	7.9	8.0
<b>20</b>	6.1	3.2	7.9	19.0	17.6	19.7	27.5	27.0	28.0	8.0	8.0	8.1

**48-h Tests with *Mytilus* spp.**

Two static renewal hydrogen sulfide tests were conducted with larvae of the mussel, *Mytilus* spp. on 22 February 2006, with nominal concentrations of 2, 4, 6, 8, and 15 µg/L H<sub>2</sub>S. A summary of *Mytilus* spp. normal survival is presented in Table 16, and a summary of water-quality observations is presented in Table 17. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.G.

Both tests were validated with a mean normal survival of >99.6% in the controls. The LC<sub>50</sub> for the copper reference-toxicant test was 15.6 µgCu/L, which is within the control chart limits (1.68 to 20.7 µgCu/L), indicating that the test organisms used in this study were similar in sensitivity to those populations previously tested at the Port Gamble Laboratory. Dissolved oxygen, temperature, and salinity were within acceptable ranges throughout the test, and pH ranged from 7.58 to 8.00.

During Test 1, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 2.1, 3.5, 5.5, 7.5, and 13.3 µg/L H<sub>2</sub>S. Mean normal survival was >90% in the 2.1 and 3.5 µg/L exposures and was 88.5% in the 5.5 µg/L exposures. Mean normal survival in the 7.5 and 13.3 µg/L exposures was significantly reduced, with 5.1% and 0.0% mean percent normal, respectively. The calculated LOEC based on the mean measured concentration was 3.5 µg/L H<sub>2</sub>S and the EC<sub>50</sub> was 6.3 µg/L H<sub>2</sub>S.

In Test 2, the control seawater mean H<sub>2</sub>S concentration was <1 µg/L. Mean measured exposure concentrations were 2.1, 2.8, 5.2, 7.7, and 14.6 µg/L H<sub>2</sub>S. Mean percentage normal survival was >90% for both the 2.1 and 2.8 µg/L treatments and was 84.7% in the 5.2 µg/L exposure. Mean normal survival was 3.7% and 0.0% in the 7.7 and 14.6 µg/L treatments, respectively. The calculated LOEC was 7.7 µg/L H<sub>2</sub>S and the EC<sub>50</sub> was 6.2 µg/L H<sub>2</sub>S.

Table 16. Results of 48-h Bivalve Larval Tests on *M. galloprovincialis*

Treatment	Rep	Mean H <sub>2</sub> S (µg/L)	Normal	Abnormal	Total	Percentage Normal	Mean Percentage Normal	SD
Test #1								
Control	1	0.0	125	0	125	100.0	100.0	0.0
	2	0.0	158	0	158	100.0		
	3	0.0	125	0	125	100.0		
	4	0.0	129	0	129	100.0		
2	1	2.1	94	1	95	98.9	99.7	0.5
	2	1.9	88	0	88	100.0		
	3	2.0	119	0	119	100.0		
	4	2.9	87	0	87	100.0		
4	1	3.3	100	4	104	96.2	<u>97.4</u>	1.0
	2	3.0	102	3	105	97.1		
	3	4.8	118	2	120	98.3		
	4	3.0	103	2	105	98.1		
6	1	5.4	77	8	85	90.6	<u>88.5</u>	7.6
	2	6.7	69	20	89	77.5		
	3	4.8	98	10	108	90.7		
	4	5.2	98	5	103	95.1		
8	1	8.8	8	86	94	8.5	<u>5.1</u>	2.8
	2	6.9	6	92	98	6.1		
	3	6.6	2	85	87	2.3		
	4	7.7	3	86	89	3.4		
15	1	15.0	0	80	80	0.0	<u>0.0</u>	0.0
	2	13.3	0	106	106	0.0		
	3	10.4	0	103	103	0.0		
	4	14.6	0	105	105	0.0		
Test #2								
Control	1	0.0	127	0	127	100.0	99.6	0.5
	2	0.0	117	1	118	99.2		
	3	0.0	108	0	108	100.0		
	4	0.0	110	1	111	99.1		
2	1	2.3	93	1	94	98.9	99.0	1.4
	2	2.0	65	2	67	97.0		
	3	2.3	92	0	92	100.0		
	4	1.6	26	0	26	100.0		
4	1	2.4	93	2	95	97.9	99.1	1.1
	2	2.7	68	1	69	98.6		
	3	3.2	60	0	60	100.0		
	4	2.9	51	0	51	100.0		
6	1	5.3	89	21	110	80.9	84.7	3.0
	2	4.9	107	20	127	84.3		
	3	5.3	72	12	84	85.7		
	4	5.3	73	10	83	88.0		
8	1	8.2	5	85	90	5.6	<u>3.7</u>	1.8
	2	7.9	5	94	99	5.1		
	3	6.1	2	78	80	2.5		
	4	8.5	2	106	108	1.9		

Treatment	Rep	Mean H <sub>2</sub> S (µg/L)	Normal	Abnormal	Total	Percentage Normal	Mean Percentage Normal	SD
15	1	16.4	0	95	95	0.0	<u>0.0</u>	0.0
	2	14.3	0	86	86	0.0		
	3	13.0	0	102	102	0.0		
	4	14.8	0	108	108	0.0		

Table 17. Summary of Water-Quality Observations during the 48-h *M. galloprovincialis* Bioassays

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1</b>												
<b>Control</b>	8.5	—	—	15.2	15.2	15.2	32.0	31.0	32.0	7.72	7.72	7.72
<b>2</b>	8.5	—	—	14.8	14.8	14.8	32.0	31.0	32.0	7.77	7.95	7.55
<b>4</b>	8.5	—	—	14.7	14.8	14.7	32.0	31.0	32.0	7.79	7.96	7.58
<b>6</b>	8.5	—	—	14.7	14.9	14.7	32.0	31.0	32.0	7.81	7.96	7.60
<b>8</b>	8.5	—	—	14.8	14.9	14.8	32.0	31.0	32.0	7.82	7.98	7.65
<b>15</b>	8.5	—	—	14.7	14.9	14.7	32.0	31.0	32.0	7.84	8.00	7.68
<b>Test #2</b>												
<b>Control</b>	8.5	—	—	14.9	14.9	14.7	32.0	31.0	32.0	7.70	7.70	7.68
<b>2</b>	8.5	—	—	14.8	15.0	14.7	32.0	31.0	32.0	7.79	7.95	7.60
<b>4</b>	8.5	—	—	14.7	15.0	14.6	32.0	31.0	32.0	7.79	7.96	7.58
<b>6</b>	8.5	—	—	14.6	14.9	14.5	32.0	31.0	32.0	7.81	7.96	7.63
<b>8</b>	8.5	—	—	14.6	14.9	14.5	32.0	31.0	32.0	7.82	7.97	7.66
<b>15</b>	8.5	—	—	14.6	14.9	14.5	32.0	31.0	32.0	7.85	8.00	7.68

**Spike Tests with *Mytilus* spp.**

Spike tests were conducted with larval *Mytilus* spp. on 22 March 2006, with nominal concentrations of 4, 6, 8, 10, 12, 14, 16, and 20 µg/L. Spiked concentrations of H<sub>2</sub>S were added once to each treatment at time intervals at 0, 6, 12, 24, and 36 hours after test initiation. A summary of *Mytilus* spp. normal survival for each concentration, and at each time period, is presented in Table 18. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.G.

Spikes for each time event were performed as individual events, and as such, exposure concentrations differed slightly at each time interval. The actual exposure concentrations for each time event are presented in Table Table 18. The results of each spike event were validated by >70% normal development in the controls. Water-quality parameters remained within acceptable limits throughout the 48-h exposures. The NOEC's increased with increasing time. At the 0-h spike event, the NOEC/LOEC threshold was between 4.1 and 6.8 µg/L. This effects threshold increased in concentration at the 6-h spike to between 9.1 and 12.8 µg/L. No threshold was observed for the concentration ranges at the 12-h, 24-h, and 36-h spike events. There was a calculable LOEC at the 36-h spike event; however, this was largely due to slightly higher percent normal development in the controls.

### Spike Tests with *Haliotis rufescens*

Spike tests were conducted with larval abalone, *H. rufescens*, on April 4 2006, with nominal concentrations of 4, 6, 8, 10, 12, 14, 16, and 20 µg/L. Spikes were added once to each treatment at time intervals at 0, 6, 12, 24, and 36 hours after test initiation. A summary of abalone normal survival for each concentration, and at each time period, is presented in Table 19. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.H.

As in the spike tests with *Mytilus* sp., the spike concentrations for each time event were performed as individual events. The actual exposure concentrations for each time event are presented in Table 19. Control normal development was below the target limit of >70% normal development; however, mean normal survival in the lowest H<sub>2</sub>S exposure (4 µg/L nominal) was >70%, indicating that the test conditions and dilution water were acceptable for normal abalone development. Water-quality parameters remained within acceptable limits throughout the 48-h exposures. As in the spike test with *Mytilus* sp., the NOEC's increased with increasing time. At the 0-h spike event, the NOEC/LOEC threshold was between 13.7 and 16.0 µg/L. No threshold was observed that the concentration ranges at the 6-h, 12-h, 24-h, and 36-h spike events. However, mean normal development in the 36-h spike event ranged between 47% and 61%. The cause of this decreased normal development rate is unclear; however, it does not appear to be associated with H<sub>2</sub>S, because there was a lack of dose-response with increasing H<sub>2</sub>S concentration.

### 3.2.6 Summary of Acute Toxicity Tests

A total of 29 acute toxicity tests were conducted with eight marine species. Of the three fish species, *A. affinis* and *M. beryllina* were similarly sensitive to H<sub>2</sub>S, with a mean EC<sub>50</sub> concentration for the two species of 40.1 µg/L, and a range of 20.8 to 65.8 µg/L. The sheepshead minnow, *C. variegatus*, was considerably less sensitive, with no calculable EC<sub>50</sub>'s, and mean percent survival 98%, 60%, and 58% in the highest tested concentrations (40.1 µg/L, 114 µg/L, and 102 µg/L H<sub>2</sub>S, respectively).

The sensitivity of *A. affinis* and *M. beryllina* in the continuous-flow exposures was greater than nearly all of the fish species observed in the literature. Tests with a variety of nearshore and open-water fish species (California killifish, Striped mullet, California halibut, Kelp bass, Bay blenny, Long-jaw mudsucker) had effects concentrations ranging from 34 µg/L to >1,802 µg/L H<sub>2</sub>S (see Table 27). While exposures were generally less than 24 hours for many species, it appears that the dose-response to hydrogen sulfide can occur over relatively short exposure periods. 96-hour LC<sub>50</sub> endpoints were calculated for the California killifish (*Fundulus parvipinnis*) and the Long-jawed mudsucker (*Gillichthys mirabilis*), with LC<sub>50</sub> values of 833 µg/L and 625 µg/L H<sub>2</sub>S, respectively.

Table 18. Results of Spike Test with Larval *Mytilus* sp.

Treatment	0 Hour Spike					6 Hour Spike					12 Hour Spike					24 Hour Spike					36 Hour Spike				
	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean
0	<1.0	79	84	78	80	<1.0	82	79	77	79	<1.0	73	76	72	74	<1.0	76	74	75	75	<1.0	83	85	81	83
4	1.7	79	78	76	78	6.5	77	81	82	80	0.6	62	58	64	61	2.7	79	78	81	79	3.1	78	79	73	77
6	1.9	77	80	81	79	7.8	65	73	82	73	3.1	62	71	70	68	3.2	77	82	83	81	4.8	78	83	76	79
8	4.1	81	78	75	78	9.1	63	68	76	69	3.8	59	63	72	65	4.2	77	83	69	76	7.7	78	75	81	78
10	6.8	45	28	47	<u>40</u>	12.8	49	46	52	<u>49</u>	8.9	58	68	75	67	8.3	73	72	81	75	10.8	69	77	72	73
12	7.7	27	27	26	<u>27</u>	12.1	61	46	51	<u>53</u>	10.7	70	62	62	65	11.2	73	77	73	74	12.1	75	79	79	78
14	10.3	0	0	1	<u>0</u>	14.1	49	40	34	<u>41</u>	12.8	74	69	72	72	12.7	77	79	70	75	17.1	80	81	77	79
16	16.9	0	0	0	<u>0</u>	17.7	48	42	46	<u>45</u>	17.8	64	78	66	69	15.7	70	75	81	75	22.7	79	74	83	79
20	18.9	0	0	0	<u>0</u>	18.4	41	49	38	<u>43</u>	23.6	71	76	70	72	14.1	64	69	78	70	35.0	69	78	78	<u>75</u>



Table 19. Results of Spike Test with Larval *Haliotis rufescens*

Treatment	0 Hour Spike					6 Hour Spike					12 Hour Spike					24 Hour Spike					36 Hour Spike				
	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean	H <sub>2</sub> S (µg/L)	Rep 1	Rep 2	Rep 3	Mean
0	<1.0				80	<1.0				79				74	<1.0				75	75	<1.0				83
4	5.11	90	90	92	91	2.57	85	86	89	87	82	88	89	86	5.68	79	78	77	78	79	3.07	58	56	55	56
6	6.38	90	92	88	90	6.61	85	87	88	87	88	85	80	84	7.85	84	74	81	80	81	4.74	40	49	53	47
8	7.70	89	88	84	87	7.92	88	83	83	85	88	86	85	86	20.0	80	76	86	81	76	6.74	55	47	56	53
10	13.7	87	83	76	<u>82</u>	10.9	89	89	86	88	89	83	88	87	15.2	75	77	77	76	75	10.1	62	53	68	61
12	16.0	76	75	75	<u>75</u>	9.15	88	82	81	83	87	81	88	85	17.2	78	78	79	78	74	10.2	53	58	49	53
14	20.6	65	72	64	<u>67</u>	11.5	86	82	79	82	86	88	88	87	20.2	77	70	77	75	75	12.8	55	54	48	52
16	20.5	59	62	55	<u>59</u>	12.6	85	87	81	85	86	87	83	85	21.7	74	73	77	75	75	13.7	58	53	58	56
20	29.3	31	39	43	<u>38</u>	14.8	82	82	88	84	84	93	83	86	26.6	75	69	72	72	70	18.3	62	50	40	51

Tests with Chinook salmon (*Oncorhynchus tshawytscha*) were sensitive at concentrations ranging from 11 µg/L to 300 µg/L H<sub>2</sub>S; however, these tests were conducted in a complex mixture of pulp mill effluent. The synergistic effects of other effluent constituents are unknown and as such make this comparison difficult to interpret and do not meet EPA criteria for inclusion in WQC development. Using only 96-h exposure values, the fish 96-h LC<sub>50</sub> for four species was 384 µg/L H<sub>2</sub>S. This mean response increases to 550 µg/L H<sub>2</sub>S (34 µg/L to 1,428 µg/L) for LC<sub>50</sub> values for exposure periods ranging from 2 to 192 hours.

Three invertebrate species were tested in 96-h exposures: *A. bahia*, *A. abdita*, and *N. arenaceodentata*. The mysid, *A. bahia*, was the most sensitive of the invertebrate species tested, with a mean estimated LC<sub>50</sub> of 9.0 µg/L and a range of 6.5 to 11.1 µg/L. The amphipod, *A. abdita*, was somewhat less sensitive, with LC<sub>50</sub>'s of 40.2 µg/L and >55.7 µg/L. The polychaete worm, *N. arenaceodentata* was among the least sensitive test organisms with LC<sub>50</sub>'s exceeding the test concentrations (>123 µg/L H<sub>2</sub>S).

*Peneus*, *Crangon*, and *Metapenaeus* shrimp were less sensitive to H<sub>2</sub>S than the mysids, with mean 96-h LC<sub>50</sub>'s of 312 µg/L, 71.5 µg/L, and 359 µg/L H<sub>2</sub>S for each respective genera at pHs of 8 to 8.3. The mean LC<sub>50</sub> concentration for the three genera of shrimp was 248 µg/L H<sub>2</sub>S.

The amphipod, *A. abdita*, was more sensitive than the amphipod species previously tested with H<sub>2</sub>S. The amphipods *Anisogammarus confervicola*, *Rhepoxynius abronius*, and *Eohaustorius estuarius* had mean LC<sub>50</sub> concentrations of 200 µg/L, 153 µg/L, and 262 µg/L H<sub>2</sub>S, respectively. These tests were conducted as 48-h tests and exposures were pulsed throughout the test. The tests with *A. abdita* were conducted for 96-h exposure periods with sustained H<sub>2</sub>S concentrations. The apparent increased sensitivity of *A. abdita* may be due to the differences in exposure rather than species or genera differences in sensitivity. The LC<sub>50</sub> for amphipods was 133 µg/L H<sub>2</sub>S.

Annelids were generally less sensitive to H<sub>2</sub>S than other invertebrate species. Previous tests with *Neanthes arenaceodentata* indicate a threshold between 517 µg/L and 2,035 µg/L, with an estimated LC<sub>50</sub> of 780 µg/L. This is consistent with the responses observed in this study. Other studies with polychaetes had behavioral endpoints and were therefore not used in criterion development.

The larval tests with *Mytilus* sp. were the most sensitive endpoint evaluated during this investigation. The 48-h EC<sub>50</sub> for normal development was 6.25 µg/L. Based on the results of the spike test, this level of H<sub>2</sub>S sensitivity occurred only during the very early phase of this test, when the embryonic development is dominated by cell division. During embryo development, cell differentiation starts after a period of very rapid cell division, for perhaps the first 6 to 12 hours of development. Once the embryo passes the morula stage (a ball of cells), development is dominated by both cell division and cell differentiation, with the formation of the mouth, gut, and anus. Shell development follows after approximately 24 hours of development. Based on the spike tests, the EC<sub>50</sub> was 6.9 µg/L when larvae were exposed at 0 hours. The EC<sub>50</sub> increased dramatically at 6 hours, with an EC<sub>50</sub> of 17.2 µg/L. At ≥12 hours, no significant decreases in normal development were observed at the highest test concentrations (15.7 to 35 µg/L H<sub>2</sub>S).

Based on the result of the spike test, the mussel larvae appear to be acutely sensitive to H<sub>2</sub>S only during the short, initial burst of development involving very high cell division.

Larval tests with abalone, *H. rufescens*, showed a similar pattern to that of the *Mytilus* spike test, however, the abalone larvae were less sensitive than the mussel with an estimated EC<sub>50</sub> of 26.1 at the 0-h time interval. At the 6-h time interval, there was a calculable LOEC of 14.8 µg/L; however, there was no calculable EC<sub>50</sub>.

The sensitivity of *Mytilus* sp. and *H. rufescens* was similar to that of other larval tests conducted with sea urchin, oyster, and mussel larvae. Median effective values for sea urchins ranged from 19 µg/L (*Strongylocentrotus purpuratus*) to 94.7 µg/L (*Paracentrotus lividus*). Bivalve larval EC<sub>50</sub>'s ranged from 10 µg/L (*Mytilus edulis*) to 320 µg/L (EC<sub>65</sub> for *Crassostrea gigas*). The mean EC<sub>50</sub> for all larval species combined, including the results of this study, was 74.3 µg/L H<sub>2</sub>S.

Based on the spike test, it appears that the larval tests with *Mytilus* and *Haliotis* may have somewhat limited applicability in WQC development. Typically the larval test is included as an indication of potential toxicity to small, highly sensitive water-column organisms, such as zooplankton. If cell division is the primarily mechanism affected by H<sub>2</sub>S, it would not be representative of acute toxicity to fully formed zooplankton. Rather, the later spike endpoints, during differentiation, may be more appropriate as acute endpoints. While the larval endpoint is included in the WQC development, it appears that it is a very conservative estimate of H<sub>2</sub>S toxicity to water-column organisms.

### 3.2.7 Results of the Chronic Toxicity Tests

#### 7-day Tests with *Menidia beryllina*

Two continuous-flow, chronic, hydrogen sulfide tests were conducted with the fish, *M. beryllina* on 13 and 14 March 2006, with nominal concentrations of 4, 8, 16, 24, 32, and 48 µg/L H<sub>2</sub>S. A summary of *M. beryllina* survival is presented in Table 20 and a summary of water-quality observations is presented in Table 21. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.I.

Both Tests 1 and 2 were validated, with greater than 80% mean survival and mean growth of greater than 0.5 mg/individual in the controls. The LC<sub>50</sub> for the cadmium reference-toxicant test was 166 µg Cu/L for combined survival and growth, which is within the control chart limits (156 to 168 µg Cu/L), indicating that the test organisms used in this study were similar in sensitivity to those populations previously tested at the Port Gamble Laboratory. Dissolved oxygen, temperature, salinity, and pH were within acceptable ranges throughout the 7-d tests.

Table 20. Results of Chronic Toxicity Tests on *Menidia beryllina*

Treatment	Rep	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percent Survival	Mean Percent Survival	SD	Individual Biomass (mg/ind.)	Mean Individual Biomass	SD	Combined Endpoint	Mean Combined Endpoint	SD
Test #1 Initiated on 13 March 2006												
Control	1	0.1	0.2	90	95.0	5.8	0.73	0.78	0.05	0.66	0.75	0.09
	2	0.2		100			0.80			0.80		
	3	0.2		100			0.85			0.85		
	4	0.3		90			0.75			0.68		
4	1	1.1	1.6	100	100.0	0.0	0.71	0.76	0.05	0.71	0.76	0.05
	2	1.7		100			0.83			0.83		
	3	2.1		100			0.74			0.74		
	4	1.6		100			0.75			0.75		
8	1	8.2	7.4	90	92.5	9.6	0.83	0.83	0.22	0.75	0.76	0.13
	2	7.6		100			0.79			0.79		
	3	8.0		80			1.12			0.90		
	4	5.8		100			0.59			0.59		
16	1	12.7	11.3	100	97.5	5.0	0.76	0.77	0.04	0.76	0.75	0.07
	2	12.8		100			0.81			0.81		
	3	10.6		90			0.72			0.64		
	4	9.1		100			0.79			0.79		
24	1	22.4	18.9	80	85.0	5.8	0.61	0.86	0.19	0.49	0.74	0.19
	2	20.7		90			0.85			0.77		
	3	16.2		90			1.06			0.95		
	4	16.4		80			0.92			0.74		
32	1	33.3	27.2	70	80.0	14.1	0.75	0.81	0.09	0.52	0.64	0.10
	2	30.0		100			0.76			0.76		
	3	24.5		80			0.79			0.63		
	4	20.8		70			0.93			0.65		
48	1	48.3	44.0	20	<u>17.5</u>	17.1	1.34	1.14	0.22	0.27	0.27	0.19
	2	50.0		0			NC <sup>a</sup>			NC		
	3	40.3		10			0.91			0.09		
	4	37.3		40			1.16			0.46		

<sup>a</sup> Not Calculable

Treatment	Rep	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percent Survival	Mean Percent Survival	SD	Individual Biomass (mg/ind.)	Mean Individual Biomass	SD	Combined Endpoint	Mean Combined Endpoint	SD
Test #2 Initiated on 14 March 2006												
Control	1	0.1	0.1	80	87.5	9.6	0.88	0.81	0.10	0.71	0.70	0.05
	2	0.0		90			0.86			0.77		
	3	0.5		80			0.85			0.68		
	4	0.0		100			0.66			0.66		
4	1	2.1	1.9	100	97.5	5.0	0.90	0.83	0.07	0.90	0.80	0.08
	2	2.1		90			0.84			0.75		
	3	2.0		100			0.84			0.84		
	4	1.5		100			0.73			0.73		
8	1	9.2	8.7	100	92.5	9.6	0.81	0.84	0.05	0.81	0.78	0.09
	2	6.2		80			0.88			0.70		
	3	9.6		90			0.79			0.71		
	4	9.7		100			0.89			0.89		
16	1	14.2	12.6	90	82.5	9.6	0.82	0.83	0.14	0.74	0.69	0.13
	2	10.4		90			0.77			0.69		
	3	10.7		70			0.72			0.51		
	4	14.9		80			1.03			0.82		
24	1	17.3	17.1	80	75.0	19.1	0.87	0.85	0.10	0.69	0.64	0.18
	2	16.2		60			0.96			0.58		
	3	14.6		100			0.86			0.86		
	4	20.3		60			0.71			0.43		
32	1	21.4	21.3	40	<u>45.0</u>	12.9	0.90	0.86	0.06	0.36	0.39	0.13
	2	15.6		60			0.93			0.56		
	3	20.1		30			0.83			0.25		
	4	28.0		50			0.79			0.40		
48	1	28.9	25.1	30	<u>35.0</u>	10.0	0.89	0.98	0.16	0.27	0.34	0.07
	2	22.8		50			0.86			0.43		
	3	23.0		30			0.95			0.28		
	4	25.8		30			1.20			0.36		

**Table 21. Summary of Water-Quality Observations during the 7-d Chronic Test with *Menidia beryllina* Bioassays**

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1</b>												
<b>Control</b>	7.4	6.1	8.4	19.4	18.8	19.7	30.9	30.0	32.0	7.7	7.6	7.9
<b>4</b>	7.6	6.6	8.1	19.5	19.0	19.8	30.1	30.0	32.0	7.8	7.7	7.9
<b>8</b>	7.5	7.1	8.1	19.7	19.3	19.9	30.0	30.0	30.0	7.8	7.7	7.9
<b>16</b>	7.5	7.0	8.1	19.7	19.3	20.0	30.0	30.0	30.0	7.8	7.7	8.0
<b>24</b>	7.4	7.0	8.1	19.7	19.4	19.9	30.0	30.0	30.0	7.8	7.7	8.0
<b>32</b>	7.4	7.0	8.0	19.7	19.4	20.0	30.0	30.0	30.0	7.9	7.8	7.9
<b>48</b>	7.3	6.8	8.0	19.6	19.4	19.8	30.0	30.0	30.0	7.9	7.8	7.9
<b>Test #2</b>												
<b>Control</b>	7.2	6.9	8.1	20.0	19.0	21.2	30.5	30.0	31.0	7.8	7.6	7.9
<b>4</b>	7.3	6.9	8.0	20.2	19.4	21.2	29.7	29.0	30.0	7.8	7.7	8.0
<b>8</b>	7.3	6.8	8.0	20.2	19.4	21.2	29.7	29.0	30.0	7.9	7.7	8.0
<b>16</b>	7.2	6.9	7.9	20.2	18.9	21.1	29.6	29.0	30.0	7.9	7.8	8.0
<b>24</b>	7.2	6.9	7.9	20.3	19.3	21.1	29.7	29.0	30.0	7.9	7.7	8.0
<b>32</b>	7.2	6.9	7.9	20.2	18.5	21.1	29.8	29.0	30.0	7.9	7.8	8.0
<b>48</b>	7.2	6.9	7.9	20.2	19.3	21.0	29.7	29.0	30.0	7.9	7.7	8.0

During Test 1, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 1.6, 7.4, 11.3, 18.9, 27.2, and 44.0 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *M. beryllina* exposed to concentrations up to 27.2 µg/L, with mean survival ranging from 80% to 100%. Statistically significant mortality was observed in the highest H<sub>2</sub>S exposure (44.0 µg/L), with a mean survival of 17.5%. The calculated LOEC was 44.0 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 33.9 µg/L H<sub>2</sub>S. There was no significant decrease in mean individual growth in any of the test concentrations, with an LOEC of >44.0 µg/L H<sub>2</sub>S.

In Test 2, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 1.9, 8.7, 12.6, 17.1, 21.3, and 25.1 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *M. beryllina* exposed to concentrations up to 17.1 µg/L; whereas significant mortality was observed in the 21.3 and 25.1 µg/L exposures, with mean survival of 45% and 35%, respectively. The calculated LOEC was 21.3 µg/L H<sub>2</sub>S, and the LC<sub>50</sub> was 23.3 µg/L H<sub>2</sub>S. There was no significant decrease in mean individual growth in any of the test concentrations, with an LOEC of >25.1 µg/L H<sub>2</sub>S.

#### **7-day Tests with *Cyprinodon variegatus***

Two continuous-flow hydrogen sulfide tests with the fish, *C. variegatus* were conducted concurrently on 15 February 2006, with nominal concentrations of 15, 25, 50, 75, and 100 µg/L H<sub>2</sub>S. A summary of *C. variegatus* survival is presented in Table 22, and a summary of water-quality observations is presented in Table 23. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.J.

Table 22. Results of Chronic Toxicity Tests on *Cyprinodon variegatus*

Treatment	Rep	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percent Survival	Mean Percent Survival	SD	Individual Biomass (mg/ind.)	Mean Individual Biomass	SD	Combined Endpoint	Mean Combined Endpoint	SD
Test #1 Initiated on 15 February 2006												
Control	1	0.4	0.4	60	72.5	18.9	0.53	0.64	0.12	0.32	0.46	0.12
	2	0.6		70			0.60			0.42		
	3	0.4		100			0.61			0.61		
	4	0.1		60			0.80			0.48		
15	1	30.8	32.4	70	72.5	17.1	0.51	0.60	0.08	0.35	0.44	0.14
	2	33.6		80			0.70			0.56		
	3	41.2		90			0.60			0.54		
	4	24.0		50			0.58			0.29		
25	1	49.7	50.4	80	82.5	5.0	0.72	0.67	0.03	0.57	0.55	0.03
	2	51.9		80			0.66			0.53		
	3	58.2		80			0.65			0.52		
	4	41.8		90			0.64			0.58		
50	1	77.0	75.1	70	70.0	8.2	0.51	0.63	0.13	0.36	0.44	0.13
	2	83.2		60			0.53			0.32		
	3	75.8		80			0.74			0.59		
	4	64.4		70			0.74			0.52		
75	1	119	123.1	50	60.0	14.1	0.59	0.46	0.08	0.29	0.27	0.05
	2	129		60			0.44			0.26		
	3	137		80			0.41			0.33		
	4	107.8		50			0.42			0.21		
100	1	122	131.1	0	<b>40.0</b>	29.4	NC	0.49	0.09	NC	0.26	0.04
	2	149		70			0.40			0.28		
	3	151		40			0.53			0.21		
	4	102		50			0.56			0.28		

Treatment	Rep	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percent Survival	Mean Percent Survival	SD	Individual Biomass (mg/ind.)	Mean Individual Biomass	SD	Combined Endpoint	Mean Combined Endpoint	SD
Test #2 Initiated on 15 February 2006												
Control	1	0.3	0.4	100	85.0	12.9	0.503	0.525	0.03	0.503	0.444	0.05
	2	0.8		70			0.569			0.398		
	3	0.2		80			0.501			0.401		
	4	0.2		90			0.526			0.473		
15	1	35.8	30.3	80	85.0	5.8	0.522	0.522	0.03	0.418	0.443	0.03
	2	25.7		80			0.544			0.435		
	3	32.2		90			0.480			0.432		
	4	27.6		90			0.540			0.486		
25	1	34.9	35.4	90	77.5	22.2	0.542	0.543	0.15	0.488	0.416	0.13
	2	41.8		70			0.750			0.525		
	3	40.8		100			0.418			0.418		
	4	24.0		50			0.462			0.231		
50	1	61.7	57.8	80	85.0	5.8	0.440	<u>0.405</u>	0.03	0.352	0.344	0.03
	2	61.1		90			0.386			0.347		
	3	56.0		90			0.416			0.374		
	4	52.6		80			0.378			0.302		
75	1	102.7	94.1	60	<u>57.5</u>	5.0	0.432	<u>0.423</u>	0.02	0.259	0.243	0.02
	2	112.7		60			0.437			0.262		
	3	83.6		60			0.388			0.233		
	4	77.3		50			0.434			0.217		
100	1	94.7	85.7	20	<u>50.0</u>	21.6	0.415	<u>0.408</u>	0.05	0.083	0.204	0.10
	2	90.6		60			0.343			0.206		
	3	104.3		50			0.420			0.210		
	4	53.2		70			0.454			0.318		



**Table 23. Summary of Water-Quality Observations during the 7-d Chronic Test with *Cyprinodon variegatus* Bioassays**

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1</b>												
<b>Control</b>	7.1	6.4	8.2	19.1	18.0	20.3	29.8	29.0	31.0	7.9	7.7	8.1
<b>15</b>	6.8	5.0	7.7	19.2	18.5	20.4	27.4	25.0	29.0	8.0	7.7	8.2
<b>25</b>	6.5	5.5	7.5	19.3	18.5	20.4	27.2	26.0	29.0	8.1	7.8	8.3
<b>50</b>	6.1	0.2	8.2	19.2	18.0	20.4	27.2	26.0	30.0	8.2	8.0	8.4
<b>75</b>	5.8	2.9	7.3	19.2	18.0	20.4	27.1	25.0	29.0	8.4	8.2	8.6
<b>100</b>	5.8	0.2	7.9	19.2	18.0	20.4	27.6	26.0	29.0	8.5	8.2	8.7
<b>Test #2</b>												
<b>Control</b>	6.9	5.6	8.6	19.2	18.5	20.0	29.8	29.0	31.0	7.9	7.5	8.1
<b>15</b>	6.3	5.2	8.2	19.2	18.5	20.4	27.4	25.0	29.0	8.0	7.6	8.2
<b>25</b>	5.9	4.6	7.7	19.3	18.5	20.5	27.2	26.0	29.0	8.0	7.8	8.3
<b>50</b>	5.1	0.8	7.5	19.2	18.0	20.4	27.2	26.0	30.0	8.1	7.8	8.4
<b>75</b>	4.4	0.5	6.9	19.2	18.0	20.4	27.1	25.0	29.0	8.3	8.0	8.6
<b>100</b>	4.8	1.3	7.0	19.2	17.9	20.3	27.6	26.0	29.0	8.4	8.1	8.7

Mean percentage survival in the Test 1 controls was 72.5% and individual growth was 0.64 mg. Mean percentage in Test 2 was 85%, with mean individual growth of 0.53 mg. Although Test 1 controls did not meet the target of 80% survival, overall survival in the pooled controls (Test 1 and 2) combined was acceptable. Since the two tests were conducted concurrently, both tests were considered acceptable for evaluating H<sub>2</sub>S toxicity. There were insufficient organisms available for conducting a concurrent reference-toxicant test.

Temperature was within acceptable ranges throughout the 7-day tests. Salinity was below the acceptable, with salinities as low as 25‰; however, this salinity is still considered within the tolerance range for *C. variegatus* (USEPA 2000c) and was not likely to affect the results of this test. Dissolved oxygen was below 1 mg/L in the highest treatment concentrations and was likely due to the high total sulfide concentrations. However, the decreased dissolved oxygen levels did not appear to cause increased mortality, with 70% to 85% survival in the nominal 50 µg/L treatment. Aeration was provided to all test chambers with depressed dissolved oxygen concentrations. pH also appeared to be affected by the high sulfide concentrations, with pHs of 8.4 to 8.7 in the highest H<sub>2</sub>S treatments. As with DO, survival and growth did not appear to be decreased due to the increased pH. Furthermore, the EPA Gold Book guidance on pH lists the upper bound of the acceptable range for most marine fish at pH 9 (USEPA 1986). It should be noted that the elevated pH does affect the proportion of total sulfides that is in the unionized form (H<sub>2</sub>S), and that this was taken into account when determining the actual H<sub>2</sub>S concentrations.

During Test 1, the mean observed H<sub>2</sub>S concentration in the control seawater was <1 µg/L. The average measured sulfide concentrations in the test treatments were 32.4, 50.4, 75.1, 123, 131 µg/L H<sub>2</sub>S. There was no significant mortality observed in the *C. variegatus* exposed to concentrations up to 123 µg/L. Mean percentage survival in the 131 µg/L H<sub>2</sub>S exposure was 53.3% and was statistically significantly lower than the controls. The calculated LOEC was 131

$\mu\text{g/L H}_2\text{S}$ . There was no calculable  $\text{LC}_{50}$ . The LOEC for growth was  $123 \mu\text{g/L H}_2\text{S}$  and there was no calculable  $\text{EC}_{50}$  for the growth endpoint.

In Test 2, the mean observed  $\text{H}_2\text{S}$  concentration in the control seawater was  $<1 \mu\text{g/L}$ . The average measured sulfide concentrations in the test treatments were 30.3, 35.4, 57.8, 94.1, and  $85.7 \mu\text{g/L H}_2\text{S}$ . There was no statistically significant mortality observed in *C. variegatus* exposed to concentrations up to  $57.8 \mu\text{g/L}$ , with mean survival ranging from 77.5% to 85% in the test treatments. Mean percentage survival was approximately 50% in the  $85.7$  and  $94.1 \mu\text{g/L H}_2\text{S}$  exposures. Although the statistical LOEC was calculated at  $35.4 \mu\text{g/L}$ , the dose-response curve was not linear and the correct estimated LOEC was  $85.7 \mu\text{g/L H}_2\text{S}$ . There was no calculable  $\text{LC}_{50}$ . *C. variegatus* exposed to  $\text{H}_2\text{S}$  concentrations of 57.8 to  $94.1 \mu\text{g/L H}_2\text{S}$  showed some decrease in individual growth, ranging from 0.405 to 0.423 mg/ind. at test termination. The calculated LOEC for growth was  $57.8 \mu\text{g/L}$  and  $\text{EC}_{50}$  was estimated at  $91.4 \mu\text{g/L H}_2\text{S}$ .

### 7-day Tests with *Americamysis bahia*

One continuous-flow hydrogen sulfide test was conducted with the mysid, *A. bahia* on 1 February 2006, with nominal concentrations of 2.5, 5, 10, 15, and  $20 \mu\text{g/L H}_2\text{S}$ . A summary of *A. bahia* survival is presented in Table 24, and a summary of water-quality observations is presented Table 25. Termination and comprehensive water-quality data for each of the replicates are presented in Appendix IV.K.

The mysid chronic test was validated by  $>80\%$  survival and mean individual growth of  $>0.2$  mg/ind. The  $\text{LC}_{50}$  for the cadmium reference-toxicant test was  $177 \mu\text{g Cu/L}$ , which is within the control chart limits (146 to  $312 \mu\text{g Cu/L}$ ), indicating that the test organisms used in this study were similar in sensitivity to those populations previously tested at the Port Gamble Laboratory. Water-quality parameters were generally within range throughout the 7-day mysid tests. Temperature was slightly below the target range of 18 to  $20^\circ\text{C}$ ; however, this did not appear to adversely affect survival or growth. Salinity in the highest  $\text{H}_2\text{S}$  concentration was slightly below the target range of 26‰ to 30‰ during Test 1. However, this is still well within the tolerance range for this species (20‰ to 30‰; EPA 2002) and did not likely affect mysid survival.

During Test 1, the mean observed  $\text{H}_2\text{S}$  concentration in the control seawater was  $<1 \mu\text{g/L}$ . The average measured sulfide concentrations in the test treatments were 3.9, 11.8, 29.6, 64.4,  $71.2 \mu\text{g/L H}_2\text{S}$ . With the exception of  $3.9 \mu\text{g/L H}_2\text{S}$ , statistically significant mortality was observed in each of  $\text{H}_2\text{S}$  exposures. The calculated LOEC was  $12.0 \mu\text{g/L H}_2\text{S}$ , and the  $\text{LC}_{50}$  was  $8.4 \mu\text{g/L H}_2\text{S}$ . There was no statistically significant decrease in growth.

### 3.2.8 Summary of Chronic Toxicity Test Results

Six chronic tests were conducted with three marine species. As in the acute toxicity tests, *A. bahia* were the most sensitive species, with a 7-d  $\text{LC}_{50}$  of  $8.4 \mu\text{g/L}$ . However, it is important to note that no effects were observed in the growth endpoint, generally considered to be a more sensitive endpoint than survival. Furthermore, there was little change in survival from the acute toxicity tests, with a mean acute  $\text{LC}_{50}$  of  $9.0 \mu\text{g/L}$ . Additionally, the survival observed during the chronic tests changed little between the 96-h time interval and test termination at 168 h (7 days).

Table 24. Results of Chronic Toxicity Tests on *Americamysis bahia*

Treatment	Rep	Mean H <sub>2</sub> S (µg/L)	Treatment Mean H <sub>2</sub> S (µg/L)	Percent Survival	Mean Percent Survival	SD	Individual Biomass (mg/ind.)	Mean Individual Biomass	SD	Combined Endpoint	Mean Combined Endpoint	SD
Test #1 Initiated on 1 February 2006												
Control	1	0.1	0.1	80	87.5	9.6	0.26	0.27	0.04	0.21	0.23	0.02
	2	0.1		80			0.32			0.25		
	3	0.2		90			0.24			0.21		
	4	0.2		100			0.25			0.25		
2.5	1	4.8	3.9	90	92.5	9.6	0.26	0.23	0.03	0.24	0.21	0.04
	2	4.2		80			0.20			0.16		
	3	3.4		100			0.25			0.25		
	4	3.1		100			0.20			0.20		
5	1	12.4	11.8	20	<u>17.5</u>	12.6	0.19	0.30	0.18	0.04	0.07	0.03
	2	12.7		20			0.50			0.10		
	3	12.2		30			0.20			0.06		
	4	9.7		0			NC			NC		
10	1	36.6	29.6	0	<u>0.0</u>	0.0	NC	NC	NC	NC	NC	NC
	2	31.8		0			NC			NC		
	3	25.2		0			NC			NC		
	4	24.7		0			NC			NC		
15	1	60.5	64.4	0	<u>0.0</u>	0.0	NC	NC	NC	NC	NC	NC
	2	53.3		0			NC			NC		
	3	58.7		0			NC			NC		
	4	85.2		0			NC			NC		
20	1	75.4	71.2	0	<u>0.0</u>	0.0	NC	NC	NC	NC	NC	NC
	2	69.0		0			NC			NC		
	3	70.8		0			NC			NC		
	4	69.4		0			NC			NC		

**Table 25. Summary of Water-Quality Observations during the 7-d Chronic Test with *Americamysis bahia* Bioassays**

Treatment	DO mg/L			Temp °C			Salinity (‰)			pH		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
<b>Test #1</b>												
<b>Control</b>	7.8	7.2	8.5	19.5	18.6	19.9	28.8	28.0	30.0	7.8	7.5	7.9
<b>2.5</b>	7.6	7.1	8.2	19.7	19.1	20.3	26.9	26.0	28.0	7.9	7.7	8.0
<b>5</b>	7.7	7.3	8.2	19.8	19.3	20.2	26.8	26.0	29.0	7.9	7.7	8.1
<b>10</b>	7.6	7.1	8.2	19.8	19.4	20.3	26.9	26.0	28.0	7.9	7.7	8.1
<b>15</b>	7.3	5.2	8.1	19.9	19.5	20.1	26.9	26.0	29.0	8.0	7.8	8.2
<b>20</b>	7.3	6.1	8.1	19.8	19.4	20.1	25.8	24.0	29.0	8.0	7.9	8.3

There are several chronic data points in the literature for crustaceans. Significant decreases in copepod hatching success were observed at concentrations of 765 µg/L H<sub>2</sub>S. Tests with *C. crangon* and *Cancer antennarius* indicate some H<sub>2</sub>S sensitivity; however, these were analyzed for behavioral endpoints and therefore were not applicable for use in WQC development. Regardless, the survival endpoint for *A. bahia* observed during this study was more sensitive than any of the behavioral endpoints previously observed with shrimp and crab species.

Fish were sensitive to chronic exposures to H<sub>2</sub>S, with a mean 7-d LC<sub>50</sub> of 28.6 µg/L for *M. beryllina* and >112 µg/L for *C. variegatus*. As with the mysid, the growth endpoint was generally less sensitive than the survival endpoint. For the *M. beryllina* test there was no significant decrease in growth in the highest test concentration. One calculable EC<sub>50</sub> was observed in the chronic tests with *C. variegatus*. It had an EC<sub>50</sub> for the growth endpoint of 91.4 µg/L H<sub>2</sub>S.

Chronic endpoints observed in other fish species in the literature included liver damage and survival. In Atlantic salmon (*Salmo salar*), liver damage was observed in smolts at concentrations of 670 µg/L H<sub>2</sub>S. Ten percent mortality was observed in Chinook salmon exposed to H<sub>2</sub>S in a complex effluent mixture at concentrations of 95.5 µg/L and 143 µg/L H<sub>2</sub>S (Holland et al. 1960). However, both of these datasets are problematic for interpreting H<sub>2</sub>S toxicity. Smolting salmon may be highly variable in their sensitivity to contaminants due to physiological and anatomical changes that occur during the smolting process. While this data provides an indication of smolt sensitivity, it is not representative of fish that would likely be exposed to H<sub>2</sub>S in open marine waters. As noted in the previous discussion of acute toxicity, the Holland et al. (1960) study is inappropriate for use in WQC development because the H<sub>2</sub>S exposure in that study occurred in the presence of a complex mixture containing other potentially confounding contaminants.

### 3.3 SPECIFICATION OF CRITERIA

The use of bioassay data to establish WQC is based on the philosophy that the waters of interest should support aquatic biota naturally occurring in that water body. When toxicity data for species indigenous to a particular location are not available, the use of effects data for surrogate

or alternative species can be an acceptable approach if those species are likely to demonstrate similar responses to the toxicant of concern at that location. Often, however, surrogate species are selected for maximum sensitivity to chemical challenges. This study first reviewed the existing toxicity data for H<sub>2</sub>S, and then selected species that were both representative of the range of species found in waters of the California OCS and considered maximally sensitive to H<sub>2</sub>S exposure.

### 3.3.1 Approach

USEPA WQC are established to protect 95% of the species likely to encounter the chemical of concern (USEPA 2000). This strategy can be modified to protect a species that is commercially or ecologically important, but it is generally recognized that the criteria are not designed to be completely protective of all species.

However, confidence in the reliability of the calculation of the 95<sup>th</sup> percentile in an effects dataset is directly dependent on the number of observations in the dataset, among other things. In particular, attempting to specify the 95<sup>th</sup> percentile in very small data sets of less than 10 observations is of limited value. Guidance by USEPA (2005) on what constitutes an acceptable minimal number of data points suggests that when a large dataset is available, a probabilistic risk assessment (PRA) approach should be used to determine the concentration that protects 95% of the species because it also provides confidence limits around that point.

A fundamental assumption of the PRA is that the sensitivity to H<sub>2</sub>S is distributed across all species in the marine community in the same way that toxicant sensitivity is distributed across individual members of a species. Just as a species population is comprised of a very few sensitive individuals and a very few tolerant individuals, the majority of individuals center about the mid range. The marine community is also comprised of a small number of very sensitive and very tolerant species, with the majority of species falling near the mean. The width of the distribution curve (representing the distribution of the sensitivity of individual species within the marine community) determines the slope of the dose-response curve for species sensitivity to H<sub>2</sub>S. The slope of the dose-response curve can then be used to estimate a toxicity value that would be protective of 95% of the species in the data set. Because this investigation has shown the H<sub>2</sub>S acts in an acutely toxic manner rather than in a chronic fashion, it is appropriate to combine all the effects data when estimating a safe level for H<sub>2</sub>S. By using such a large and comprehensive dataset, with both acute and chronic endpoints combined, the estimate should be protective of all acceptable endpoints. For the purposes of this site-specific WQC determination, the toxicity value was based on the 5<sup>th</sup> percentile of species ranked from the most sensitive to least sensitive, to provide a criterion that would be protective of 95% of the marine community.

This study included both a comprehensive examination of available H<sub>2</sub>S toxicity data, and the generation of new data derived from numerous bioassays conducted at the Port Gamble Laboratory as part of this investigation. This approach resulted in a strong, documented database of 103 effects for dozens of aquatic species that should be representative of impacts to open-ocean marine organisms under several exposure scenarios. Based on this comprehensive dataset, the PRA identified a nominal H<sub>2</sub>S concentration of 12 µg/L that is protective of 95% of the species included in this study.

The bioassay endpoints used in the PRA were the effects concentrations, including the LOEC, LC<sub>50</sub> and EC<sub>50</sub>. These endpoints were selected because they represent a threshold above which adverse effects abruptly increase. NOEC values were not included in the evaluation even though they represent a sensitive endpoint.<sup>a</sup> Often, the NOEC simply reflects an inadequate concentration series that failed to span the effects range during a particular test. With an ideal concentration series, the LOEC would only be slightly higher than the NOEC. However, in many of the literature studies, there was a relatively large difference between the NOEC and LOEC, which is probably indicative of deficiencies in the design of these particular bioassay test series.

There are several aspects of PRA that serve to focus attention on the most critical range of effects data, namely, the most sensitive species.

- Bioassay response data can be represented by one of several distributions depending on the effects range of interest, and the PRA is capable of determining the optimal distribution.
- PRA provides a reasonable, representative estimate of the likelihood of effects on selected aquatic species because it accounts for the uncertainties in biological responses to contaminants, and addresses the individual variability in responses.
- The PRA provides an estimate of how representative the individual observed effects are within the more realistic range of effects for all of the species evaluated.
- Graphic presentations of the range of risks can be used to look for changes in the character of responses within and among various taxonomic groupings or response-type measurements, allowing a visual evaluation of different levels of sensitivity to the toxicant.
- Generally, differences in biological responses can be grouped and evaluated to estimate the most likely response. PRA can estimate the concentration(s) most likely to be protective at the 95% level for the test species of interest. By applying appropriate distributional assumptions, it is possible to determine the most likely 95<sup>th</sup> percentiles and the optimal protective level, while graphically illustrating the potential uncertainties associated with that determination.

The focus of this study was to determine the concentration of H<sub>2</sub>S that is protective of 95% of the species selected from the results of the historic data reviews and current bioassay tests. All of the relevant historic and current laboratory data (n=103) were considered in the development of the estimated protective water concentration of 12 µg/L H<sub>2</sub>S. These data, along with the PRA results are provided in Appendix V.

### 3.3.2 Methodology

The 95% protective level in these studies was developed using standard statistical approaches and probabilistic risk assessment (PRA) techniques. The PRA provides an estimate of the breakpoint in a data set at the confidence interval of interest (95%), as well as estimate of the uncertainty in the risk estimate. For this study, we used the commercial probabilistic risk software Crystal Ball® to estimate the range of possible outcomes of the 95% protective H<sub>2</sub>S concentrations for the 103 test results used in this report. This software uses a Monte Carlo

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<sup>a</sup> Because the NOEC is the highest concentration where there is no statistically significant increase in adverse effects relative to the control populations

randomization of each of the entries in a dataset and estimates the “most likely” set of risk outcomes for a given dataset. The Monte Carlo technique randomly generates values for uncertain variables over and over (10,000 times in this study) to simulate a model calculating the results of a series of equations with a range of values for each data point. The outcomes generated by this technique more closely reflect the range of exposures and effects (toxicity) likely to occur in the environment than more rudimentary analysis techniques.

The current national criterion for H<sub>2</sub>S was determined by a standard statistical method that estimates species sensitivity using extrapolations of a paired data set. In order to estimate the protective level, the approach directly exams only the lowest values in an overall ranking of species-specific toxicity values. Each species with a documented toxicity value is given a percentile sensitivity ranking using the formula: Percent =  $(100 \cdot n / N + 1)$ , where n is the rank of the species in terms of sensitivity to the chemical and N = the total number of species evaluated in the data set (Figure 4). A log-linear regression is then performed on the lowest toxicity concentrations, or on the four concentrations that lie closest to the 95<sup>th</sup> percentile ranking. This results in a direct approximation using the relative sensitivities of only a few of the species in the entire dataset. It often lacks accuracy due to an inexact fit, particularly when one of the four values is an outlier. Errors in the fit can be particularly problematic when there are less than 20 observations and the protective level is extrapolated from the lowest four values, rather than being interpolated from intermediate values.

PRA improves on the estimation of a protective level by employing Monte Carlo estimates of likely responses for each of the data points in a data set. The result is a much more representative and accurate reflection of the relationships between species and toxicity, after factoring in the uncertainties associated with each observation. The PRA is particularly useful for determining the 5% and 95% estimates of a data set because these values are dependent on a relatively precise match of the distributional curves to the data. Therefore, PRA provides a much more accurate representation of the distribution of species and their sensitivity to the chemical of concern.

The PRA software selects the most representative distribution for the data set being evaluated, and then provides a range of risk estimates at the 5% to 95% levels. Each distribution is further evaluated for *goodness of fit* parameters, and the resulting risk estimates for 5% to 95% are then modified to illustrate the possible uncertainties in the risk estimates due to skewness and overall lack of fit between the data and the theoretical distribution. The resulting graphs provide a visualization of the protective value and the impact of the fit on the results.

The PRA software compared results of all 103 H<sub>2</sub>S toxicity values in the comprehensive data set using a variety of computer-generated distributional assumptions that included the normal, lognormal, triangular, Weibull, and beta distributions. The PRA then calculated the most representative set of endpoints by a precise fit of the distribution to the dataset.

The dataset included all of the appropriate literature data and the experimental test results produced as part of the bioassays conducted as part of this study. Test species that appeared to be comparatively insensitive to H<sub>2</sub>S exposure were excluded from the PRA analyses because H<sub>2</sub>S toxicity does not appear to be a primary concern for those species. Since the purpose of the PRA

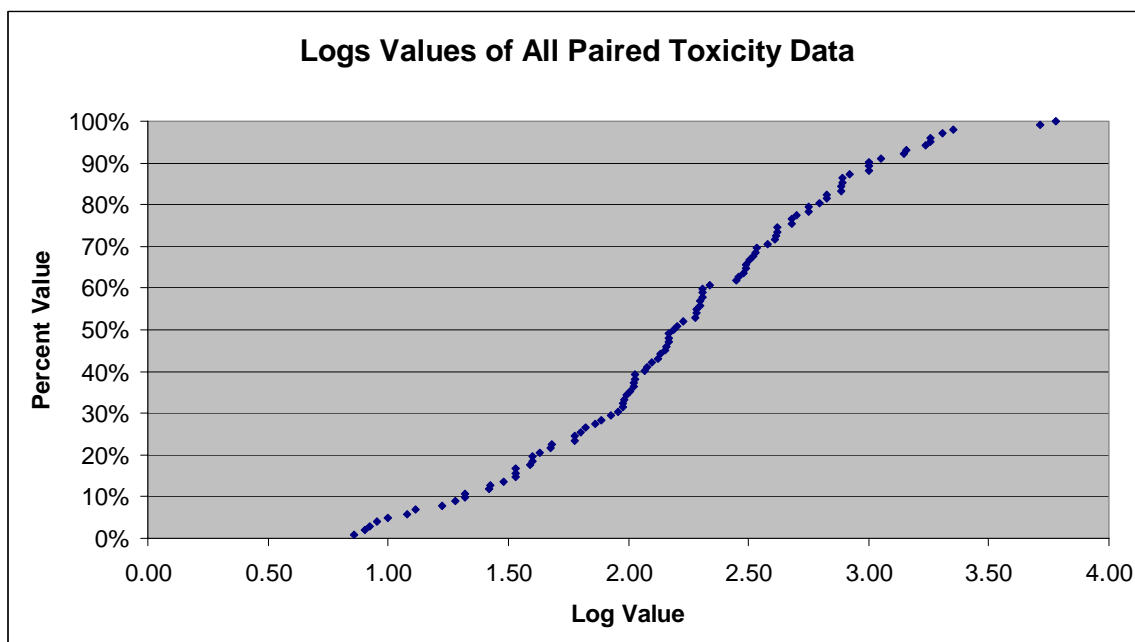


Figure 4. Simple Ranking of Log-Concentration for the H<sub>2</sub>S Dataset

approach was to develop a conservative level of protection for 95% of the species likely to be impacted, including wholly insensitive species in the PRA would skew the outcome inappropriately upward. All results were then compared and plotted to illustrate the break point (5%) that defined the predicted protective level for 95% of the species in the data set.

### 3.3.3 Results

The estimated 95% protective level included the results of the bioassays conducted at the Port Gamble Laboratory and the data from the literature. These data were entered into Excel<sup>®</sup> spreadsheets and collated according to species and endpoints. The results of the information in the spreadsheets were then incorporated into the Crystal Ball<sup>®</sup> software to determine the most representative distributions. Each of the distributions generated in Crystal Ball<sup>®</sup> was given a ranking (best-fit) and prioritized to recommend the range of likely outcomes of the 95% protective estimates.

Because the data used in the PRA represent a large range of reported acute and chronic endpoints for a wide variety of species, the resulting risk estimates should be adequately protective of the marine aquatic species likely to be exposed to H<sub>2</sub>S at an offshore site. Analysis of the entire dataset resulted in arithmetic and geometric mean effects concentrations of 454 µg/L and 162 µg/L, respectively. The 95% protective level was determined to be 12 µg/L. Figure 5 shows a screenshot of the output of the Crystal Ball<sup>®</sup> software showing a PRA of the H<sub>2</sub>S database. The predicted log-concentrations at the 95% breakpoint generated by the Monte Carlo randomization ranges from 1.01188 to 1.14055. The mean of these values (the estimated central 95% breakpoint) is 1.076215. Taking the antilog of this value results in a 95% protective concentration of 12 µg/L.



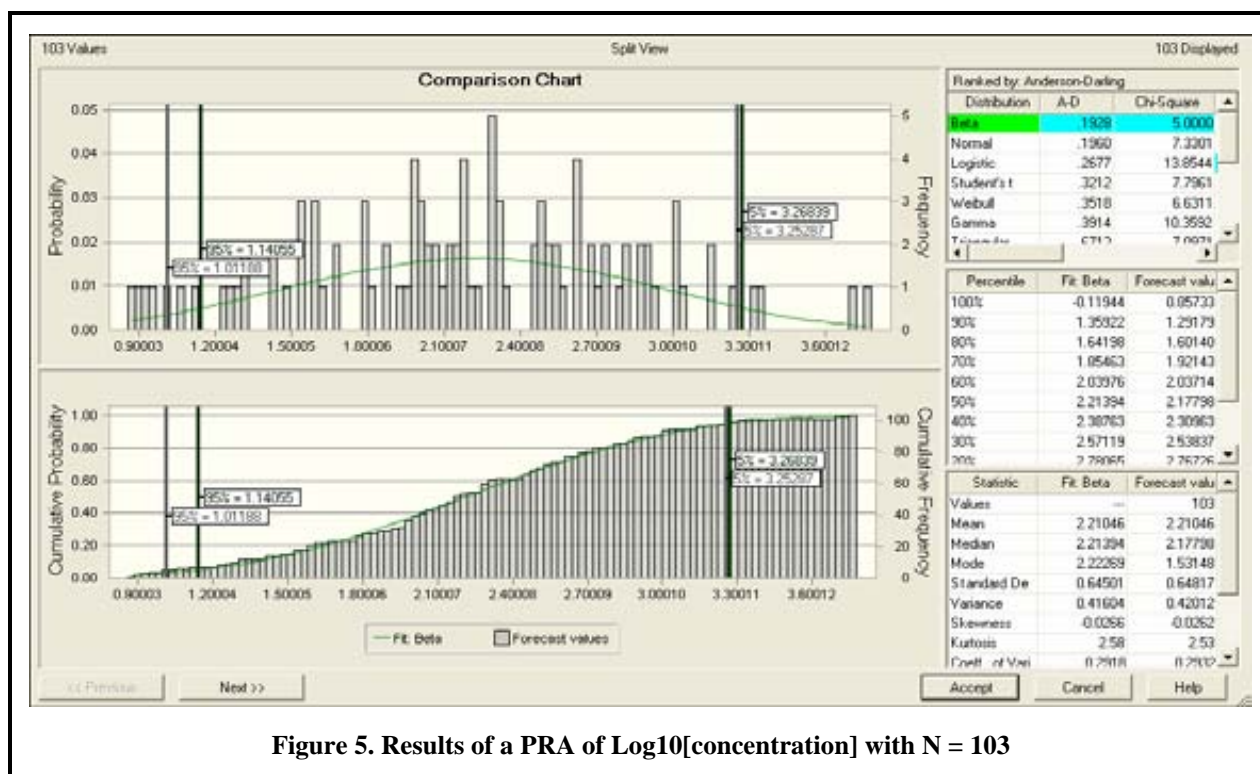


Figure 5. Results of a PRA of Log10[concentration] with N = 103

The bioassays conducted at the Port Gamble Laboratory as part of this study found that the LOEC and EC<sub>50</sub> endpoints for H<sub>2</sub>S exposure for six of the eight reported species are relatively high (Table 26). Within the entire dataset, the two most conservative mean species values for the LOEC and EC<sub>50</sub> were 11.1 µg/L for the mysid shrimp (*Americamysis bahia*) and 7.6 µg/L for the larval bay mussel (*Mytilus galloprovincialis*) spike tests. These are the lowest mean species endpoints generated by the PRA in this study, and are the only data that fall below the 95% protective concentration determined from the 103 values included in the effects database.

Table 26. Protective Estimates of H<sub>2</sub>S Toxicity for Individual Species based on the data generated in the Port Gamble Laboratory Bioassays

Test Species	Group(s)	Endpoint	Concentration (µg/L)	
			Mean Effect	Geometric Mean
<i>Americamysis bahia</i>	Acute	LOEC-EC <sub>50</sub>	11.5	11.1
<i>Ampelisca abdita</i>	Acute	LOEC-EC <sub>50</sub>	43.5	43.2
<i>Atherinops affinis</i>	Acute	LOEC-EC <sub>50</sub>	31.4	30.2
<i>Cyprinodon variegatus</i>	Acute	LOEC-EC <sub>50</sub>	105.5	101.9
<i>Menidia beryllina</i>	Acute	LOEC-EC <sub>50</sub>	34.2	33.2
<i>Neanthes arenaceodentata</i>	Acute	LOEC-EC <sub>50</sub>	105.1	105.1
<i>Mytilus galloprovincialis</i>	Acute	LOEC-EC <sub>50</sub>	7.6	7.6
<i>Haliotis rufescens</i>	Acute	LOEC-EC <sub>50</sub>	17.25	17.25

The analysis used to generate the 95% protective level of H<sub>2</sub>S is appropriate and conservative for several reasons. The dataset used to establish the WQC was large, which leads to a high degree of confidence in the result. Additionally, the site-specific criterion is based on a more sound

scientific approach applied to a more comprehensive and representative dataset than those used to establish the national criterion.

The results of this evaluation are conservative because the comprehensive data set includes larval development endpoints that are the most susceptible to the mechanism of H<sub>2</sub>S toxicity. The inclusion of bioassay results reported from the literature adds another level of conservatism to the WQC. Several of the literature studies potentially over-predict the effects from H<sub>2</sub>S because they include exposure of test organisms to media that contain other potentially toxic compounds in addition to H<sub>2</sub>S, thereby confounding how much of the reported toxicity was due to H<sub>2</sub>S and how much was due to the other influences within the exposure media. In particular, the salmonid bioassay data that was used to support the original WQC was determined using a complex mixture of sulfite waste liquor used as test media. The database is also conservative because it includes very sensitive species of juvenile/larval crustaceans that are more susceptible to H<sub>2</sub>S exposure than other species of similar phylogenetic standing.

A more important form of conservatism arises because the reported effects concentrations that were included in the database represent the average exposures rather than the highest concentrations that are the likely cause of the response. Thus, the effects-based concentrations reported in the literature often underestimate of the actual exposure concentration because of the short half-life of the H<sub>2</sub>S contaminant. Additionally, the exposure periods in some cases greatly exceed the current standard exposure periods of 24 to 96 hours or 7 to 10 days.

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**5.0 APPENDIX I: LIST OF CONTRIBUTORS**

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## 6.0 APPENDIX II: LIST OF ACRONYMS AND DEFINITIONS

<b>ACR</b>	The acute-to-chronic ratio (ACR) expresses the relationship between the H <sub>2</sub> S concentration causing acute toxicity to a species (expressed as an acute toxicity endpoint such as an LC <sub>50</sub> ) and the H <sub>2</sub> S concentration causing chronic toxicity to the same species (expressed as a chronic toxicity endpoint such as an NOEC or its equivalent, i.e., $ACR = ATE/CTE$ or $LC_{50}/NOEC$ ). An ACR is commonly used to extrapolate to a “chronic toxicity” concentration using exposure considerations and available acute toxicity data when chronic toxicity data for the species, chemical, or effluent of concern are unavailable. The ACR should normally be greater than one, since the ratio compares an acute effect concentration with a chronic effect concentration. However, in the case of H <sub>2</sub> S, the ACR was found to be close to unity.
<b>Acute test</b>	A comparative study in which organisms, that are subjected to different treatments, are observed for a short period usually not constituting a substantial portion of their life span. Acute tests often utilize mortality as the only measured effect; chronic tests usually include additional measures of effect such as growth or reproduction
<b>AML</b>	The calculated average monthly limit (AML) of wasteload allocation assigned by a State of EPA for a particular facility.
<b>APHA</b>	American Public Health Association
<b>ATP</b>	An adenosine-derived nucleotide, C <sub>10</sub> H <sub>16</sub> N <sub>5</sub> O <sub>13</sub> P <sub>3</sub> , that supplies large amounts of energy to cells for various biochemical processes, including muscle contraction and sugar metabolism, through its hydrolysis to ADP.
<b>BAF</b>	Bioaccumulation Factor
<b>Bioaccumulation</b>	The net accumulation of a substance by an organism from all environmental sources.
<b>Bioaccumulation Factor (BAF)</b>	The quotient obtained by dividing the concentration of a substance in an organism (or specified tissue) by its concentration in a specified exposure medium, for example, air, food, sediment, soil, water, when several media are possible sources.
<b>Bioassay</b>	An experiment that uses living whole organisms, tissues or cells to measure the presence, the concentration or the relative potency of one or more chemicals.
<b>Bioconcentration</b>	The net accumulation of a substance by an aquatic organism as a result of uptake directly from aqueous solution.

<b>CCC</b>	WQC for aquatic life contain two expressions of allowable magnitude a CMC (Criteria Maximum Concentration) to protect against acute (short-term) effects and a CCC (Criterion Continuous Concentration) to protect against chronic (long-term) effects. EPA derives acute criteria from 48- to 96-hour tests of lethality or immobilization. EPA derives chronic criteria from longer-term (often greater than 28-day) tests that measure survival, growth, reproduction, or in some cases, bioconcentration.
<b>Chronic test</b>	A comparative study in which organisms, that are subjected to different treatments, are observed for a long period or a substantial portion of their life span. Acute tests often utilize mortality as the only measured effect; chronic tests usually include additional measures of effect such as growth or reproduction. Note Cost-effective short-term chronic tests have been designed which involve exposure of sensitive life stages (e.g. larval fish, juvenile invertebrates) for a relatively brief period (e.g. seven days).
<b>CMC</b>	See CCC
<b>Confidence limits (or intervals)</b>	These are probability limits, based on the data set and statistical model employed, that the "true value" lies within the limits specified. Typically limits are expressed at the 95% or 99% probability levels.
<b>Control chart</b>	A cumulative summary chart of results from QA tests with reference materials (e.g. reference toxicants). The results of a given QA test are compared to the control chart mean value and acceptance limits (typically 95% confidence limits, i.e. mean + 2 standard deviations) or warning limits (typically 99% confidence limits, i.e. mean + 3 standard deviations).
<b>Control, negative</b>	Measures taken to insure that a test, its components, or the environment do not cause undesired effects, or produce incorrect test results. In toxicity tests a negative control typically consists of clean water or sediment that is essentially free of contaminants or the test material. The response of organisms in the negative control is used to determine test acceptability and as a baseline for gauging adverse effects among animals exposed to treatments.
<b>Control, positive</b>	Measures taken to insure that a test and/or its components are working properly and producing correct or expected results from positive test subjects. In toxicity tests positive controls typically consist of reference toxicants.
<b>COP</b>	California Ocean Plan (SWRCB, 2001)

<b>CV</b>	A coefficient of variation is used to compare the relative amounts of variation in populations having different means. It is defined as the standard deviation expressed as a percentage of the mean.
<b>CWA</b>	Clean Water Act
<b>DCOR</b>	Dos Cuadras Offshore Resources, LLC
<b>DO</b>	Dissolved Oxygen
<b>Effect Concentration (EC)</b>	A point estimate of the toxicant concentration that would cause an observable adverse effect (e.g., death, immobilization, or serious incapacitation) in a given percent of the test organisms, calculated from a continuous model (e.g., Probit Model). EC <sub>25</sub> is a point estimate of the toxicant concentration that would cause an observable adverse effect in 25 percent of the organisms. The lower the EC <sub>25</sub> , the more toxic the chemical or effluent sample. Other EC values, e.g. the EC <sub>50</sub> or EC <sub>10</sub> may also be calculated to determine concentrations causing effect in a greater or lesser proportion of the population.
<b>EPA</b>	Environmental Protection Agency
<b>IC</b>	Inhibition Concentration (IC) A point estimate of the toxicant concentration that would cause a given percent reduction in a non-lethal biological measurement (e.g., reproduction or growth), calculated from a continuous model (e.g., Interpolation Method). IC <sub>25</sub> is a point estimate of the toxicant concentration that would cause a 25-percent reduction in a non-lethal biological measurement. The lower the IC <sub>25</sub> , the more toxic the chemical or effluent sample. Other IC values, e.g. the IC <sub>50</sub> or IC <sub>10</sub> may also be calculated to determine concentrations causing a more or less severe effect.
<b>Indigenous Species</b>	A species that is likely, due to historical presence, to occur at a specified site for some portion of its life span.
<b>LC<sub>50</sub> (lethal concentration, 50 percent)</b>	The toxicant or effluent concentration that would cause death in 50% of the test organisms. The concentration is calculated from the data set using statistical or graphical models. The lower the LC <sub>50</sub> , the more toxic the chemical or effluent sample. Other LC values, e.g. the LC <sub>90</sub> or LC <sub>5</sub> may also be calculated to determine concentrations causing more or less mortality to the population. Note The LC value must always be associated with the duration of exposure. Thus a 48 h LC <sub>50</sub> , 96 h LC <sub>50</sub> , etc. is calculated.
<b>Life-cycle Test</b>	A comparative study in which organisms, that are subjected to different treatments, are observed at least from a life stage in one generation to the same life stage in the next generation.

<b>LOEC (Lowest-observed-effect-concentration)</b>	The lowest concentration of an effluent or toxicant that results in adverse effects on the test organisms (i.e., where the values for the observed endpoints are statistically different from the control).
<b>Maximum Daily Limit (MDL)</b>	The calculated maximum WLA assigned by a State or EPA for a particular facility.
<b>Minimum Significant Difference (MSD)</b>	The minimum difference which can exist between a test treatment and the controls in a particular test and be statistically significant; a measure of test sensitivity. The lower the MSD the more sensitive the test. Related to test power, the MSD is decreased by increasing the number of replicates and/or decreasing the amount of between-replicate variability in the controls and treatments.
<b>Mixing Zone</b>	Area where an effluent discharge undergoes initial dilution and is extended to cover the secondary mixing in the ambient water body. A mixing zone is an allocated impact zone where WQC can be exceeded as long as acutely toxic conditions are prevented.
<b>MRS</b>	Marine Research Specialists
<b>NOAEC (No-observed-acute-effect-concentration)</b>	The NOEC for acute effects (typically survival); see NOEC.
<b>NOEC (No-observed-effect-concentration)</b>	The highest concentration of an effluent or toxicant that causes no observable adverse effects on the test organisms (i.e., the highest concentration of toxicant at which the values for the observed responses are not statistically different from the control).
<b>NPDES</b>	National Pollutant Discharge Elimination System, created under the Clean Water Act. The permitting system under which point source discharges are regulated to eliminate or minimize the discharge of toxicants into surface waters. States frequently oversee their own programs which must comply with (i.e. be equally or more stringent) the national permit program.
<b>OCS</b>	Pacific Outer Continental Shelf
<b>Power (1-<math>\beta</math>)</b>	The probability correctly rejecting the null hypothesis depending on the significance criterion, the variability of the sample results, and the size of the impact. The probability of correctly detecting an actual toxic effect.
<b>ppb</b>	Parts per billion = $\mu\text{g/L}$ in solution , or $\mu\text{g/Kg}$ dry weight
<b>ppm</b>	Parts per million = $\text{mg/L}$ in solution , or $\text{mg/Kg} = \mu\text{g/g}$ dry weight

<b>PRA</b>	Probabilistic Risk Assessment (PRA) uses Monte Carlo techniques to estimate the range of possible outcomes of a study and a “most likely” response to the exposure concentration. The use of PRA provides an estimate of risk that incorporates both the range of exposure outcomes and an evaluation of the uncertainties in the risk parameters.
<b>Precision</b>	A measure of the reproducibility within a data set. Precision can be measured both within a laboratory and between laboratories using the same test method and toxicant.
<b>p-value</b>	Low <i>p</i> -values (<0.05) indicate a high degree of confidence that the computed quantity is statistically significant.
<b>PXP</b>	Plains Exploration and Production Company/Arguello Inc
<b>Quality Assurance</b>	An integrated system of activities involving planning, quality control, quality assessment, reporting and quality improvement to ensure that a product or service meets defined standards of quality with a stated level of confidence.
<b>Quality Control</b>	The overall system of technical activities whose purpose is to measure and control the quality of a product or service so that it meets the needs of users.
<b>Reference Toxicant</b>	The toxicant used in performing toxicity tests to indicate the sensitivity of a test organism and to demonstrate the laboratory's ability to perform the test correctly and obtain consistent results.
<b>Replicate</b>	Each of several experimental units that are tested simultaneously using the same experimental conditions.
<b>Resident Species</b>	A species that is regularly present at a specified site for some portion of its life span.
<b>RP</b>	Reasonable Potential
<b>RWC</b>	Receiving Water Concentration is the measured or projected exposure concentration of a toxicant in the receiving water after mixing. The RWC is calculated at the edge of a mixing zone. It must be less than the most-limiting WQ standard (CMC or CCC).
<b>Surrogate Species</b>	A species that is tested to estimate responses of other species, for which direct testing is impractical.
<b>SWRCB</b>	California State Water Resources Control Board
<b>TAC (Test Acceptability Criteria)</b>	Specific criteria for determining whether toxicity test results are acceptable (EPA, 2001). For example, a TAC for minimum control survival is typically specified in toxicity test methods.

<b>Toxicity</b>	The property of a chemical, or combination of chemicals, to adversely affect organisms, tissues or cells.
<b>Toxicity Test</b>	A procedure to determine the toxicity of a chemical or an effluent using living organisms. A toxicity test measures the degree of effect of a specific chemical or effluent on exposed test organisms.
<b>TU</b>	Toxic units. Expresses the relative toxicity of an effluent in such a manner that the larger the toxic unit value the more toxic the effluent. $TU_a = 100/LC_{50}$ . $TU_c = 100/NOEC$ . Examples $LC_{50} = 25\%$ , $TU_a = 4.0$ and $NOEC = 12.5\%$ , $TU_c = 8.0$ . Permits with whole effluent toxicity limits (WET limits) often specify a TU value that may not be exceeded.
<b>TU<sub>a</sub></b>	Acute Toxicity Units (See TU)
<b>TU<sub>c</sub></b>	Chronic Toxicity Units (See TU)
<b>Type I Error (<math>\alpha</math>)</b>	The rejection of the null hypothesis ( $H_0$ ) when it is, in fact, true (i.e., determining that the effluent is toxic when the effluent is not toxic).
<b>Type II Error (<math>\beta</math>)</b>	The acceptance of the null hypothesis ( $H_0$ ) when it is not true (i.e., determining that the effluent is not toxic when the effluent is toxic).
<b>USEPA</b>	U.S. Environmental Protection Agency
<b>Water Effects Ratio (WER)</b>	A correction factor used to adjust a state or regional water quality standard to account for differences in the toxicity of a specific pollutant between laboratory water and site water.
<b>Whole Effluent Toxicity (WET)</b>	The total toxic effect of an effluent measured directly with a toxicity test.
<b>WQC</b>	Water Quality Criterion
<b>Weston</b>	Weston Solutions, Incorporated
<b>WSPA</b>	Western States Petroleum Association

## 7.0 APPENDIX III: SUMMARY OF DATA REPORTED IN THE LITERATURE

**Table 27. Summary of Literature Data on Acute Responses of Marine Organisms to Hydrogen Sulfide**

Phylum/ Family	Common Name	Species	Life Stage	Effect	Concentration (µg/L)	Effect Level	Qualification	Reference
Bacillariophyta	Diatom	<i>Skeletonema costatum</i>	Population	Photosynthesis	105	4h EC <sub>50</sub>	I <sup>a</sup> : Single cell organism	Breteler et al. 1991
Bacillariophyta	Diatom	<i>Skeletonema costatum</i>	Population	Photosynthesis	41	4h NOEC	R: Single cell organism	Breteler et al. 1991
Arthropoda	Isopoda	<i>Gnorimosphaeroma oregonensis</i>	Adult	Survival	5,200	96h LC <sub>50</sub>	I: Low dissolved oxygen	Caldwell 1975
Arthropoda	Amphipoda	<i>Anisogammarus confervicola</i>	Adult	Survival	200	96h LC <sub>50</sub>	I: Low dissolved oxygen	Caldwell 1975
Arthropoda	Amphipoda	<i>Corophium salmonis</i>	Adult	Survival	<1,000	96h LC <sub>50</sub>	I: Low dissolved oxygen	Caldwell, 1975
Arthropoda	Amphipoda	<i>Rhepoxynius abronius</i>	Adult	Survival	160	48h LC <sub>50</sub>	I: Pulsed/Shortened exposure period	Knezovich et al. 1996
Arthropoda	Amphipoda	<i>Rhepoxynius abronius</i>	Adult	Survival	≈147 <sup>b</sup>	48h LOEC	I: Pulsed/Shortened exposure period	Knezovich et al. 1996
Arthropoda	Amphipoda	<i>Eohaustorius estuarius</i>	Adult	Survival	≈332	48h LC <sub>50</sub>	I: Pulsed/Shortened exposure period	Knezovich et al. 1996
Arthropoda	Amphipoda	<i>Eohaustorius estuarius</i>	Adult	Survival	≈192	48h LOEC	I: Pulsed/Shortened exposure period	Knezovich et al. 1996
Arthropoda	Common Shrimp	<i>Crangon crangon</i>	Adult	Survival	71.5	1h LC <sub>50</sub>	R: Exposure period is too short	Vismann 1996
Arthropoda	Peneaid Shrimp	<i>Penaeus indicus</i>	Post Larvae (20-25 mm)	Survival	117 (pH 6-6.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Penaeus indicus</i>	Post Larvae (20-25 mm)	Survival	189 (pH 7-7.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Penaeus indicus</i>	Post Larvae (20-25 mm)	Survival	342 (pH 8-8.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Penaeus indicus</i>	Juv. (35-40 mm)	Survival	63 (pH 6-6.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Penaeus indicus</i>	Juv. (35-40 mm)	Survival	119 (pH 7-7.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996

<sup>a</sup> Studies qualified with an “I” are considered suitable for inclusion in the site-specific criterion development. Those qualified with an “R” were rejected, and were not included in the criterion development for the reasons indicated.

<sup>b</sup> “≈” indicates that data from paper was used to estimate or calculate H<sub>2</sub>S concentration based on fraction of total dissolved sulfides being either 10% where pH data were missing or use of a pH dependant percentage where pH data was available.



Phylum/ Family	Common Name	Species	Life Stage	Effect	Concentration (µg/L)	Effect Level	Qualification	Reference
Arthropoda	Peneaid Shrimp	<i>Penaeus indicus</i>	Juv. (35-40 mm)	Survival	281 (pH 8-8.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Penaeus indicus</i>	Juv. (85-90 mm)	Survival	144 (pH 8-8.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Metapenaeus dobsoni</i>	Post Larvae (20-25 mm)	Survival	125 (pH 6-6.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Metapenaeus dobsoni</i>	Post Larvae (20-25 mm)	Survival	219 (pH 7-7.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Metapenaeus dobsoni</i>	Post Larvae (20-25 mm)	Survival	378 (pH 8-8.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Metapenaeus dobsoni</i>	Juv. (35-40 mm)	Survival	77 (pH 6-6.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Metapenaeus dobsoni</i>	Juv. (35-40 mm)	Survival	147 (pH 7-7.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Peneaid Shrimp	<i>Metapenaeus dobsoni</i>	Juv. (35-40 mm)	Survival	340 (pH 8-8.3)	96h LC <sub>50</sub>	I: Meets WQC requirements	Gopakumar and Kuttyamma 1996
Arthropoda	Dungeness Crab	<i>Cancer magister</i>	Post Larval	Survival	1,000	96h LC <sub>50</sub>	I: Low dissolved oxygen	Caldwell 1975
Arthropoda	Dungeness Crab	<i>Cancer magister</i>	Zoea	Survival	500	96h LC <sub>50</sub>	I: Meets WQC requirements	Caldwell 1975
Mollusca	Tellinidae Clam	<i>Macoma balthica</i>	Adult	Survival	6,000	96h LC <sub>50</sub>	I: Low dissolved oxygen.	Caldwell 1975
Mollusca	Pacific Oyster	<i>Crassostrea gigas</i>	Larvae	Survival	1,400	96h LC <sub>50</sub>	I: Pulsed/Shortened exposure period	Caldwell 1975
Mollusca	Bay Mussel	<i>Mytilus edulis</i>	Adult	Survival	>≈2,100	96h LC <sub>50</sub>	R: Variable sulfide concentration	Abel, 1976
Mollusca	Bay Mussel	<i>Mytilus edulis</i>	Adult	Filtration rate	≈80	96h EC <sub>50</sub>	R: Variable sulfide concentration, Behavioral endpoint	Abel, 1976
Echinodermata	White Sea Urchin	<i>Lytechinus pictus</i>	Adult	Survival	43	96h LC <sub>50</sub>	I: Non-water-column species	Thompson et al. 1991
Echinodermata	White Sea Urchin	<i>Lytechinus pictus</i>	Adult	Survival	21	96h LOEC	I: Non-water-column species	Thompson et al. 1991
Annelida	Nereidae	<i>Neanthes arenaceodentata</i>	2-3 weeks old	Survival	2,035	96h LOEC	I: Low dissolved oxygen	Dillon et al. 1993
Annelida	Nereidae	<i>Neanthes arenaceodentata</i>	2-3 weeks old	Survival	780	Estimated LC <sub>50</sub>	I: Low dissolved oxygen	Dillon et al. 1993

Phylum/ Family	Common Name	Species	Life Stage	Effect	Concentration (µg/L)	Effect Level	Qualification	Reference
Annelida	Spionidae	<i>Streblospio benedicti</i>	Adult	Survival	2,244	24h EC <sub>50</sub>	I: Low dissolved oxygen	Llanos 1991
Annelida	Capitellidae	<i>Capitella capitata</i>	Larvae	Settlement time	1,724	3h LOEC	I: Meets WQC requirements	Dubilier, 1988 <sup>a</sup>
Annelida	Sabellariid	<i>Phragmatopoma lapidosa</i>	Adult	Survival	4300	NOEC	R: No effect concentration	Main and Nelson 1988
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	250d	Mortality	≈80	7d NOEC	R: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	250d	Mortality	≈200	7d LC <sub>30</sub>	I: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	250d	Mortality	≈300	7d LC <sub>100</sub>	I: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	260d	Mortality	≈9	16d NOEC	R: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	260d	Mortality	≈11	16d LC <sub>10</sub>	R: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	260d	Mortality	≈15	16d LC <sub>35</sub>	R: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	260d	Mortality	≈23	16d LC <sub>60</sub>	R: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	260d	Mortality	≈45	16d LC <sub>100</sub>	R: Complex mixture, no direct measurement of H <sub>2</sub> S	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	280d	Mortality	≈107	72h LC <sub>90</sub>	I: Meets WQC requirements	Holland, et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	280d	Mortality	≈60	72h LC <sub>30</sub>	I: Meets WQC requirements	Holland, et al. 1960
Chordata	Long-Jawed Mudsucker	<i>Gillichthys mirabilis</i>	4-15g	Mortality	>1,802	>20h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Long-Jawed Mudsucker	<i>Gillichthys mirabilis</i>	4-15g	Mortality	625	96h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Long-Jawed Mudsucker	<i>Gillichthys mirabilis</i>	4-15g	Mortality	417	192h LC <sub>70</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	California Killifish	<i>Fundulus parvipinnis</i>	3-8g	Mortality	>1,802	>20h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	California Killifish	<i>Fundulus parvipinnis</i>	3-8g	Mortality	833	96h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989

<sup>a</sup> As referenced in Sims and Moore 1995

Phylum/ Family	Common Name	Species	Life Stage	Effect	Concentration (µg/L)	Effect Level	Qualification	Reference
Chordata	California Killifish	<i>Fundulus parvipinnis</i>	3-8g	Mortality	417	192h NOEC	R: High test organism density	Bagarinao and Vetter 1989
Chordata	Striped Mullet	<i>Mugil cephalus</i>	8-21g	Mortality	1,428	12h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	California Halibut	<i>Paralichthys californicus</i>	80-114g	Mortality	1,122	10h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Bay Blenny	<i>Hypsoblennius gentilis</i>	8-19g	Mortality	782	10h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Kelp Bass	<i>Paralabrax clathratus</i>	10-40g	Mortality	476	8-9h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Sand Bass	<i>Paralabrax nebulifer</i>	8-32g	Mortality	476	7h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Black Surf Perch	<i>Embiotoca jacksoni</i>	20-30g	Mortality	170	6h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Giant Kelpfish	<i>Heterostichus rostratus</i>	5-21g	Mortality	136	4-5h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Speckled Sanddab	<i>Citharichthys stigmaeus</i>	11-40g	Mortality	102	2-3h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Northern Anchovy	<i>Engraulis mordax</i>	8-40g	Mortality	34	1-2h LC <sub>50</sub>	I: High test organism density	Bagarinao and Vetter 1989
Chordata	Atlantic salmon	<i>Salmo salar</i>	Smolt	Mortality	265	NOEC	R: Insufficient replicates, varying concentrations; no effect level	Kiemer et al. 1995
Chordata	Black Sea Turbot	<i>Rhombus maeoticus</i>	Eggs (large-cell morula stage)	Mortality	≈310	LOEC	I: Test species likely has high tolerance	Ivanov et al. 1973
Chordata	Black Sea Turbot	<i>Rhombus maeoticus</i>	Eggs (large-cell morula stage)	Mortality	≈220	NOEC	R: Test species likely has high tolerance; no effect level	Ivanov et al. 1973

Table 28. Summary of Literature Data on Chronic Responses of Marine Organisms to Hydrogen Sulfide

Phylum/Family	Common Name	Species	Life Stage	Effect	Concentration (µg/L)	Effect Level	Qualification	Reference
Mollusca	Bay Mussel	<i>Mytilus edulis</i>	Larvae	Combine mortality and abnormality	≈5	48h NOEC	R: Pulsed/Shortened exposure period no effect level	Knezovich et al. 1996
Mollusca	Bay Mussel	<i>Mytilus edulis</i>	Larvae	Combine mortality and abnormality	≈9	48h LOEC	I: Pulsed/Shortened exposure period	Knezovich et al. 1996
Mollusca	Bay Mussel	<i>Mytilus edulis</i>	Larvae	Combine mortality and abnormality	≈10	48h EC <sub>50</sub>	I: Pulsed/Shortened exposure period	Knezovich et al. 1996
Mollusca	Pacific Oyster	<i>Crassostrea gigas</i>	Larvae	Normal development	100	24h NOEC	R: Pulsed/Shortened exposure period – no effect level	Caldwell 1975
Mollusca	Pacific Oyster	<i>Crassostrea gigas</i>	Larvae	Normal development	320	24h EC <sub>65</sub>	I: Pulsed/Shortened exposure period	Caldwell 1975
Mollusca	Pacific Oyster	<i>Crassostrea gigas</i>	Larvae	Normal development	1,000	24h EC <sub>87</sub>	I: Pulsed/Shortened exposure period	Caldwell 1975
Annelida	Nereidae	<i>Nereis virens</i>	Adult	Duration of ventilation	1720	NOEC	R: Endpoint was behavioral – no effect and behavioral	Miron and Kristensen 1993
Annelida	Nereidae	<i>Nereis virens</i>	Adult	Duration of ventilation	3450	LOEC	R: Endpoint was behavioral	Miron and Kristensen 1993
Annelida	Nereidae	<i>Nereis diversicolor</i>	Adult	Duration of ventilation	3,400	NOEC	R: Endpoint was behavioral	Miron and Kristensen 1993
Annelida	Nereidae	<i>Nereis diversicolor</i>	Adult	Duration of ventilation	6,800	LOEC	R: Endpoint was behavioral	Miron and Kristensen 1993
Annelida	Nereidae	<i>Neanthes succinea</i>	Adult	Duration of ventilation	1720	NOEC	R: Endpoint was behavioral	Miron and Kristensen 1993
Annelida	Nereidae	<i>Neanthes succinea</i>	Adult	Duration of ventilation	3450	LOEC	R: Endpoint was behavioral	Miron and Kristensen 1993
Echinodermata	Sea Urchin	<i>Strongylocentrotus purpuratus</i>	Larvae	Combined survival and abnormality	≈10	48h NOEC	R: Only partial short-term duration test – no effect level	Knesovich et al. 1996
Echinodermata	Sea Urchin	<i>Strongylocentrotus purpuratus</i>	Larvae	Combined survival and abnormality	≈13	48h LOEC	I: Shortened exposure period	Knesovich et al. 1996
Echinodermata	Sea Urchin	<i>Strongylocentrotus purpuratus</i>	Larvae	Combined survival and abnormality	≈19	48h EC <sub>50</sub>	I: Shortened exposure period	Knesovich et al. 1996
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Behavior/avoidance	1,190	49d NOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Behavior/avoidance	3,094	49d LOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991

Phylum/Family	Common Name	Species	Life Stage	Effect	Concentration (µg/L)	Effect Level	Qualification	Reference
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Survival	1,190	49d NOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Survival	3,094	49d LOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Growth	<1,190	49d NOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Growth	1,190	49d LOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Male gonad growth	<1,190	49d NOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991
Echinodermata	Sea Urchin	<i>Lytechinus pictus</i>	Adult	Male gonad growth	1,190	49d LOEC	R: Indirect exposure to H <sub>2</sub> S	Thompson et al. 1991
Echinodermata	Sea Urchin	<i>Paracentrotus lividus</i>	Sperm	Fertilization	94.7	EC <sub>50</sub>	I: Meets WQC	Losso et al. 2004
Echinodermata	Sea Urchin	<i>Paracentrotus lividus</i>	Embryo	Development	34	EC <sub>50</sub>	I: Meets WQC	Losso et al. 2004
Chordata	Atlantic salmon	<i>Salmo salar</i>	Smolt	Liver Damage	670	10 week EC <sub>50</sub>	I: Varying concentrations	Kiemer et al. 1995
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	68d	Mortality	95.5	23d LC <sub>10</sub>	I: Additional stressors, no direct measurement of H <sub>2</sub> S	Holland et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	68d	Mortality	143	23d LC <sub>10</sub>	I: Additional stressors, no direct measurement of H <sub>2</sub> S	Holland et al. 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	68d	Mortality	287	23d LC <sub>70</sub>	I: Additional stressors, no direct measurement of H <sub>2</sub> S	Holland et al. 1960
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	Adult	Photosynthetic rate	408	6d EC <sub>50</sub>	I: Non-water-column species, no measurements	Holmer and Bondgaard 2001
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	Adult	Survival	408	21d LC <sub>50</sub>	I: Low DO; Benthic species, no measurements	Holmer and Bondgaard 2001
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	Adult	Leaf elongation	204	21d EC <sub>50</sub>	I: Low DO; Benthic species, no measurements	Holmer and Bondgaard 2001
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	Adult	Root sugars and starch	<204	21d NOEC	R: Low DO; Benthic species, no measurements – no effect level	Holmer and Bondgaard 2001
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	Adult	Root sugars and starch	<204	21d NOEC	R: Low DO; Benthic species, no measurements – no effect level	Holmer and Bondgaard 2001
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	Adult	Root sugars and starch	<204	21d NOEC	R: Low DO; Benthic species, no measurements – no effect level	Holmer and Bondgaard 2001
Phaeophyta	Brown Algae	<i>Fucus serratus</i>	Zygote	Growth	≈560	5d LOEC	I: Low dissolved oxygen, no measurements	Chapman and Fletcher 2002
Arthropoda	Common Shrimp	<i>Crangon crangon</i>	Adult	Behavior – swimming	306	LOEC	R: Endpoint was behavioral	Vismann, 1996

Phylum/Family	Common Name	Species	Life Stage	Effect	Concentration (µg/L)	Effect Level	Qualification	Reference
Arthropoda	Common Shrimp	<i>Crangon crangon</i>	Adult	Behavior – panic	414	LOEC	R: Endpoint was behavioral	Vismann, 1996
Arthropoda	Common Shrimp	<i>Crangon crangon</i>	Adult	Behavior - paralysis	551	LOEC	R: Endpoint was behavioral	Vismann, 1996
Arthropoda	Cancer Crab	<i>Cancer antennarius</i>	Adult	Heart rate	1,000	LOEC	R: Metabolic endpoint	Vetter et al., 1987 (as referenced in Bagarinao 1992)
Arthropoda	Copepods	<i>Acartia tonsa</i> <i>Centropages hammaties</i> <i>Labidocera aestiva</i>	Eggs	Hatching success	765	LOEC	I: Low dissolved oxygen	Marcus et al., 1997

Table 29. Bioaccumulation of Sulfur Concentrations in Fish Tissues after Exposure to a Constant Concentration of 680 µg/L H<sub>2</sub>S<sup>a</sup>

Common Name	Species	Duration of Exposure (time to death)	Tissue	Concentration of Sulfur in Tissue (mg/kg)	
				Unexposed	Sulfide-Killed
Longjaw Mudsucker	<i>Gillichthys mirabilis</i>	96h	Blood	0	3.2 ± 1.2 (5) <sup>b</sup>
			Spleen	0	4.0 ± 0.7 (2)
			Kidney	0	4.2 ± 3.2 (2)
			Liver	0	2.4 ± 0.3 (2)
California Killifish	<i>Fundulus parvipinnis</i>	96h	Blood	0	3.4 ± 1.0 (5)
			Spleen	0	8.0 ± 4.8 (3)
			Kidney	0.06 ± 0.06	2.6 ± 0.1 (3)
			Liver	0	3.7 ± 1.2 (3)
California Halibut	<i>Paralichthys californicus</i>	5-10h	Blood	0.4	9.7 ± 12.8 (2)
			Spleen	1.2	4.9 ± 5.4 (2)
			Kidney	1.5	8.2 ± 1.5 (2)
			Liver	—	—
Barred Sand Bass	<i>Paralabrax nebulifer</i>	3-7h	Blood	0	2.8
			Spleen	0	0.5
			Kidney	0	9
			Liver	0	3.8
Speckled Sanddab	<i>Citharichtys stigmaeus</i>	2-3h	Blood	0	1.6 ± 1.0 (4)
			Spleen	0.06 ± 0.06	0.7 ± 0.4 (4)
			Kidney	0.1 ± 0.1	0.4 ± 0.2 (5)
			Liver	0.03 ± 0.03	0

<sup>a</sup> Bagarinao et al. 1989<sup>b</sup> (n) = number of fish analyzed

Table 29. Bioaccumulation of Sulfur Concentrations in Fish Tissues after Exposure to a Constant Concentration of 680 µg/L H<sub>2</sub>S<sup>a</sup>

Common Name	Species	Duration of Exposure (time to death)	Tissue	Concentration of Sulfur in Tissue (mg/kg)	
				Unexposed	Sulfide-Killed
Longjaw Mudsucker	<i>Gillichthys mirabilis</i>	96h	Blood	0	3.2 ± 1.2 (5) <sup>b</sup>
			Spleen	0	4.0 ± 0.7 (2)
			Kidney	0	4.2 ± 3.2 (2)
			Liver	0	2.4 ± 0.3 (2)
California Killifish	<i>Fundulus parvipinnis</i>	96h	Blood	0	3.4 ± 1.0 (5)
			Spleen	0	8.0 ± 4.8 (3)
			Kidney	0.06 ± 0.06	2.6 ± 0.1 (3)
			Liver	0	3.7 ± 1.2 (3)
California Halibut	<i>Paralichthys californicus</i>	5-10h	Blood	0.4	9.7 ± 12.8 (2)
			Spleen	1.2	4.9 ± 5.4 (2)
			Kidney	1.5	8.2 ± 1.5 (2)
			Liver	—	—
Barred Sand Bass	<i>Paralabrax nebulifer</i>	3-7h	Blood	0	2.8
			Spleen	0	0.5
			Kidney	0	9
			Liver	0	3.8
Speckled Sanddab	<i>Citharichtys stigmaeus</i>	2-3h	Blood	0	1.6 ± 1.0 (4)
			Spleen	0.06 ± 0.06	0.7 ± 0.4 (4)
			Kidney	0.1 ± 0.1	0.4 ± 0.2 (5)
			Liver	0.03 ± 0.03	0

<sup>a</sup> Bagarinao et al. 1989<sup>b</sup> (n) = number of fish analyzed



## 8.0 APPENDIX IV: BIOASSAY TEST DATA AND ANALYSES

**Table A1. Acute *Menidia beryllina* (13 Jan 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	0.3	0.8				1.1		0.0	<b>0.5</b>	100	100	100	<b>100</b>
	2	0.2		0.2				0.2	0.0	<b>0.1</b>	110	100	100	<b>100</b>
	3	0.2			0.0				0.0	<b>0.1</b>	100	100	90	<b>90</b>
	4	0.2				0.2			0.0	<b>0.1</b>	100	80	80	<b>80</b>
<b>2.5</b>	1	4.8	2.1				0.3*		0.0*	<b>3.4</b>	100	100	100	<b>100</b>
	2	3.8		1.0				0.0*	0.0*	<b>2.4</b>	100	100	100	<b>100</b>
	3	6.3			0.9*				0.0*	<b>6.3</b>	100	100	100	<b>100</b>
	4	5.3				0.8*			0.0*	<b>5.3</b>	100	80	80	<b>80</b>
<b>5</b>	1	18.8	10.6				0.8*		0.0*	<b>14.7</b>	80	80	80	<b>80</b>
	2	22.8		7.9				0.0*	0.0*	<b>15.4</b>	100	100	100	<b>100</b>
	3	22.7			4.5*				0.1*	<b>22.7</b>	100	100	100	<b>100</b>
	4	22.5				0.5*			0.0*	<b>22.5</b>	100	100	90	<b>90</b>
<b>10</b>	1	18.1	32.1				0.2*		0.0*	<b>25.1</b>	100	100	100	<b>100</b>
	2	14.8		34.7				0.0*	0.2*	<b>24.7</b>	100	100	100	<b>100</b>
	3	17.3			25.0				0.2*	<b>21.1</b>	100	100	100	<b>100</b>
	4	14.4				12.8			0.1*	<b>13.6</b>	100	100	100	<b>100</b>
<b>25</b>	1	76.0	ND							<b>76.0</b>	100	0	T	<b>T</b>
	2	62.3		56.4				53.4		<b>59.3</b>	90	20	10	<b>0</b>
	3	63.7			18.2					<b>41.0</b>	90	0	T	<b>T</b>
	4	66.4				88.0				<b>77.2</b>	90	10	10	<b>0</b>
<b>50</b>	1	133	ND							<b>133</b>	90	40	0	<b>T</b>
	2	131		72.3						<b>101</b>	100	0	T	<b>T</b>
	3	129			23.8*					<b>129</b>	90	10	0	<b>T</b>
	4	112				128				<b>120</b>	90	40	0	<b>T</b>

ND = No data

T = Test terminated due to 0% survival

\* Sulfide concentrations decreased during test

# **Acute Fish Test-96 Hr Survival**

Start Date: 1/13/2006      Test ID: P051102.05      Sample ID:  
End Date: 1/17/2006      Lab ID: WESTON - Port Gamble      Sample Type:  
Sample Date:      Protocol: EPA/600/R      Test Species: Menidia beryllina  
Comments: Average

Conc-ug/L	1	2	3	4
Control	1.0000	1.0000	0.9000	0.8000
4.4	1.0000	1.0000	1.0000	0.8000
18.8	0.8000	0.1000	1.0000	0.9000
21.1	1.0000	1.0000	1.0000	1.0000
63.4	0.0000	0.0000	0.0000	0.0000
120.8	0.0000	0.0000	0.0000	0.0000

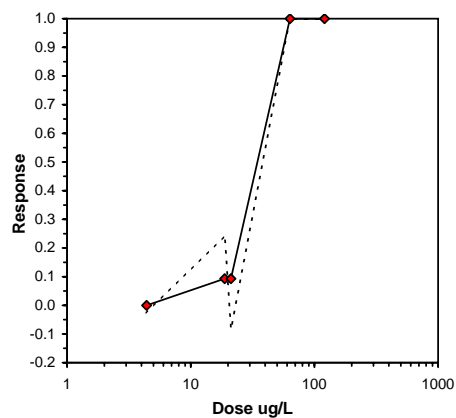
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%	N				
Control	0.9250	1.0000	1.2951	1.1071	1.4120	11.347	4			3	40
4.4	0.9500	1.0270	1.3358	1.1071	1.4120	11.411	4	19.50	10.00	2	40
18.8	0.7000	0.7568	1.0225	0.3218	1.4120	47.285	4	15.00	10.00	12	40
21.1	1.0000	1.0811	1.4120	1.4120	1.4120	0.000	4	22.00	10.00	0	40
*63.4	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	10.00	40	40
*120.8	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	10.00	40	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.742237	0.884	-1.964618	8.1661828
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	Chv	TU
Steel's Many-One Rank Test	21.1	63.4	36.575128	

Treatments vs Control

Trimmed Spearman-Kärber			
Trim Level	EC50	95% CL	
0.0%	32.120	29.319	35.189
5.0%	33.859	31.087	36.879
10.0%	34.562	32.498	36.756
20.0%	34.562	32.498	36.756
Auto-0.0%	32.120	29.319	35.189





## 4-Day Acute Toxicity Test

**Table A2. Acute *Menidia beryllina* (13 Jan 2006)**

Weston Test ID: P051102.05	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10			
	2		11	10	10	10			
	3		10	10	9	9			
	4		10	8	8	8			
2.5	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	8	8	8			
5	1		8	8	8	8			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	8	9			
10	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
25	1		10	0	0	0			
	2		9	2	1	0			
	3		9	0	0	0			
	4		9	1	1	0			
50	1		9	4	0	0			
	2		5	0	0	0			
	3		9	1	0	0			
	4		9	4	0	0			
Date			1/14/06	1/15/06	1/16/06	1/17/06			
Time			1050	1014	1215	1530			
Initials			TS	TS	GZ	GZ			



## 4-Day Acute Toxicity Test

**Table A3. Acute *Menidia beryllina* (13 Jan 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen Sulfide	Date Test Started:	13-Jan-06
Client Sample ID:	N/A	Date Test Ended:	17-Jan-06
Weston Test ID:	P051102.05	Study Director:	Brian Hester
Species:	<i>Menidia beryllina</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 1/13/05  Time: 1945  Technician: BWG/BH	Control		1		7.1		20.9		26		7.9	3	0	3
			2		7.1		20.8		26		7.9	2	0	2
			3		7.2		20.5		25		7.8	2	0	2
			4		7.1		20.9		26		7.9	2	0	2
	2.5		1		6.9		20.8		24		8.0	70	0	70
			2		6.9		20.8		24		7.9	45	0	45
			3		6.9		20.9		24		7.9	74	0	74
			4		6.9		20.9		24		7.9	63	0	63
	5		1		6.8		20.9		23		8.1	339	0	339
			2		6.8		20.9		23		8.0	331	0	331
			3		6.6		20.8		24		8.0	332	0	332
			4		6.7		20.8		24		8.0	328	0	328
	10		1		6.9		20.8		23		8.1	326	0	326
			2		6.8		20.8		23		8.1	266	0	266
			3		6.7		20.7		23		8.1	311	0	311
			4		6.7		20.7		23		8.1	260	0	260
	25		1		6.4		20.8		23		8.3	426	1/5	2130
			2		6.6		20.8		23		8.3	349	1/5	1745
			3		6.6		20.8		23		8.3	357	1/5	1785
			4		6.0		20.8		23		8.3	372	1/5	1860
	50		1		6.0		20.8		23		8.6	292	1/25	7300
			2		6.1		20.9		22		8.6	285	1/25	7125
			3		5.5		20.1		22		8.8	443	1/25	11075
			4		5.6		20.8		22		8.6	244	1/25	6100
<b>Day 1</b> <b>AM</b> Date: 1/14/06 Time: 1034 Technician: TS	Control				6.8		20.6		28		7.8	8	0	8
	2.5				6.1		20.7		24		7.7	16	0	16
	5				6.6		20.7		24		7.8	101	0	101
	10				6.7		20.7		25		7.9	381	0	381
	25				6.3		20.8		25		7.8	249	1/25	6225
	50				6.6		20.9		24		7.9	752	1/25	18800
<b>Day 1</b> <b>PM</b> Date: 1/14/06 Time: 1627 Technician: TS	Control				6.9		20.3		27		7.8	2	0	2
	2.5				6.4		20.6		25		7.7	8	0	8
	5				6.2		20.6		25		7.9	94	0	94
	10				6.3		20.6		25		7.9	412	0	412
	25				5.0		20.8		25		8.3	64	1/25	1600
	50				4.6		20.8		25		8.6	161	1/25	4025



## 4-Day Acute Toxicity Test

**Table A3. Acute *Menidia beryllina* (13 Jan 2006)**

Weston Test ID:	P051102.05	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 1/15/06 Time: 0924 Technician: TS Feed Time: 0915	Control		3		6.0		20.2		27		7.6	0	0	0
	2.5				4.5		20.6		25		7.5	5	0	5
	5				4.3		20.6		25		7.7	35	0	35
	10				5.3		20.5		25		7.9	297	0	297
	25				4.6		20.6		25		7.8	7	1/25	175
	50				5.7		20.7		25		8.0	14	1/25	350
<b>Day 2</b> <b>PM</b> Date: 1/15/06 Time: 1552 Technician: JM	Control		4		6.4		20.0		28		7.8	2	0	2
	2.5				3.4		20.2		24		7.5	4	0	4
	5				4.1		20.3		25		7.6	3	0	3
	10				4.7		20.4		25		7.8	123	0	123
	25				3.4		20.4		26		7.9	42	1/25	1050
	50				1.9		20.4		25		8.6	285	1/25	7125
<b>Day 3</b> <b>AM</b> Date: 1/16/06 Time: 1009 Technician: GZ	Control		1		6.1		20.4		29		7.4	5	0	5
	2.5				6.3		20.6		26		7.6	2	0	2
	5				6.2		20.6		26		7.7	0	0	0
	10				5.6		20.6		26		7.8	2	0	2
	25				5.4		20.6		27		8.0	39	1/25	975
	50				5.7		20.7		26		8.1	37	1/25	925
<b>Day 3</b> <b>PM</b> Date: 1/16/06 Time: 1530 Technician: GZ	Control		2		5.9		20.4		27		7.5	1	0	1
	2.5				6.0		20.8		26		7.7	0	0	0
	5				5.6		20.8		25		7.7	0	0	0
	10				5.0		20.8		25		7.8	0	0	0
	25				5.3		20.8		26		8.0	789	0	789
	50				ND		ND		ND		ND	ND	ND	ND
<b>Day 4</b>  Date: 1/17/06  Time: 0910  Technician: GZ/JM	Control		1		5.2		20.3		29		7.5	0	0	0
			2		5.2		20.8		29		7.7	0	0	0
			3		5.2		20.9		29		7.7	0	0	0
			4		5.3		21.0		29		7.7	0	0	0
	2.5		1		4.9		21.0		26		7.6	0	0	0
			2		4.4		21.0		25		7.6	0	0	0
			3		4.3		21.0		25		7.6	0	0	0
			4		4.5		21.0		25		7.6	0	0	0
	5		1		4.5		20.9		25		7.7	0	0	0
			2		4.5		21.0		25		7.7	0	0	0
			3		4.8		20.8		25		7.8	1	0	1
			4		4.7		20.9		25		7.8	0	0	0
	10		1		4.9		20.9		24		7.9	0	0	0
			2		4.8		21.0		25		7.9	2	0	2
			3		4.9		21.0		25		7.9	2	0	2
			4		4.7		21.0		25		7.9	1	0	1
	25		1		ND		ND		ND		ND	ND	ND	ND
			2		5.3		20.9		25		7.7	1	0	1
			3		ND		ND		ND		ND	ND	ND	ND
			4		5.2		21.0		25		7.8	4	0	4
	50		1		ND		ND		ND		ND	ND	ND	ND
			2		ND		ND		ND		ND	ND	ND	ND
			3		ND		ND		ND		ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND

ND = No data; replicate previously terminated

**Table A4. Acute *Menidia beryllina* (19 Jan 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
Control	1	0.0	0.2				0.5		0.0	<b>0.2</b>	100	100	100	<b>100</b>
	2	0.0		0.1				0.4	0.2	<b>0.2</b>	100	90	80	<b>80</b>
	3	0.2			0.2				0.0	<b>0.1</b>	100	80	70	<b>70</b>
	4	0.0				0.2			0.2	<b>0.1</b>	100	90	90	<b>90</b>
2.5	1	1.3	0.5				1.5		0.3	<b>0.9</b>	90	70	70	<b>70</b>
	2	1.2		0.2				0.7	0.5	<b>0.7</b>	100	100	90	<b>90</b>
	3	0.3			1.6				0.2	<b>0.7</b>	100	90	80	<b>80</b>
	4	0.2				0.3			0.2	<b>0.2</b>	100	100	100	<b>100</b>
5	1	2.4	2.2				1.3*		0.3*	<b>2.3</b>	100	90	90	<b>90</b>
	2	1.7		1.6				2.9*	0.5*	<b>1.6</b>	90	90	90	<b>90</b>
	3	1.9			2.0				0.3*	<b>1.9</b>	100	90	90	<b>90</b>
	4	3.0				0.7*			0.0*	<b>3.0</b>	100	100	90	<b>90</b>
10	1	10.7	8.4				0.5*		0.6*	<b>9.6</b>	100	100	100	<b>90</b>
	2	9.5		6.1				0.7*	0.0*	<b>7.8</b>	90	90	80	<b>80</b>
	3	8.4			4.2				0.3*	<b>6.3</b>	100	90	80	<b>80</b>
	4	7.6				2.1*			0.1*	<b>7.6</b>	100	100	100	<b>100</b>
15	1	19.0	19.2				5.9*		0.0*	<b>19.1</b>	90	60	60	<b>60</b>
	2	15.6		14.5				2.6*	0.0*	<b>15.0</b>	80	70	70	<b>70</b>
	3	17.9			14.1				0.0*	<b>16.0</b>	60	40	40	<b>40</b>
	4	16.9				4.6*			0.7*	<b>16.9</b>	80	80	70	<b>70</b>
20	1	23.2	35.5				5.1*		0.6*	<b>29.4</b>	70	40	30	<b>10</b>
	2	22.6		27.1				1.6*	0.0*	<b>24.8</b>	60	50	40	<b>40</b>
	3	28.6			18.8				1.2*	<b>23.7</b>	80	60	60	<b>40</b>
	4	24.0				3.2*			0.0*	<b>24.0</b>	60	40	40	<b>20</b>

\* Sulfide concentrations decreased during test

Acute Fish Test-96 Hr Survival				
Start Date:	1/19/2006	Test ID:	P060103.02	Sample ID:
End Date:	1/23/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPA/600/R	Test Species:
Comments:	Average			
	Menidia beryllina			

Conc-ug/L	1	2	3	4
Control	1.0000	0.8000	0.7000	0.9000
0.6	0.7000	0.9000	0.8000	1.0000
2.2	0.9000	0.9000	0.9000	0.9000
7.8	0.9000	0.8000	0.8000	1.0000
16.8	0.6000	0.7000	0.4000	0.7000
25.5	0.1000	0.4000	0.4000	0.2000

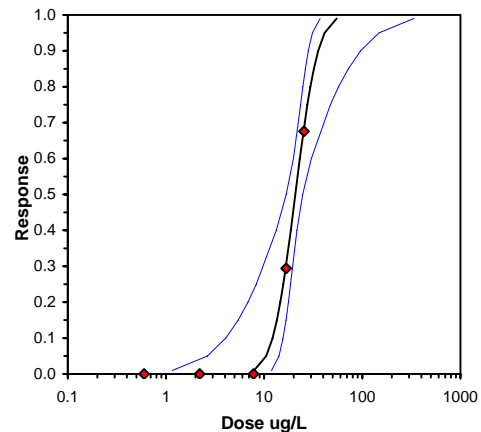
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%					
Control	0.8500	1.0000	1.1898	0.9912	1.4120	15.281	4			6	40
0.6	0.8500	1.0000	1.1898	0.9912	1.4120	15.281	4	18.00	10.00	6	40
2.2	0.9000	1.0588	1.2490	1.2490	1.2490	0.000	4	20.00	10.00	4	40
7.8	0.8750	1.0294	1.2188	1.1071	1.4120	11.906	4	19.00	10.00	5	40
16.8	0.6000	0.7059	0.8883	0.6847	0.9912	16.263	4	11.00	10.00	16	40
*25.5	0.2750	0.3235	0.5387	0.3218	0.6847	33.093	4	10.00	10.00	29	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.9505879	0.884	-0.013205	-0.904228
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	16.8	25.5	20.697826	

Treatments vs Control

Parameter	Value	SE	95% Fiducial Limits	Maximum Likelihood-Probit							
				Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter	
Slope	5.5158021	1.8439943	1.9015732 9.1300309	0.15	0.5208905	7.8147278	0.91	1.3180536	0.1812973	4	
Intercept	-2.270123	2.44674	-7.065734 2.5254875								
TSCR	0.1297067	0.027713	0.0753893 0.1840241								
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	7.8757708	1.1623027 11.878936								
EC05	3.355	10.467576	2.6365794 14.193305								
EC10	3.718	12.18181	4.0703025 15.643832								
EC15	3.964	13.494322	5.4451833 16.738156								
EC20	4.158	14.637604	6.8484353 17.697591								
EC25	4.326	15.695331	8.3175422 18.608328								
EC40	4.747	18.712099	13.260487 21.614278								
EC50	5.000	20.799536	16.797431 24.719311								
EC60	5.253	23.119839	19.863601 30.283099								
EC75	5.674	27.563656	23.549862 47.300718								
EC80	5.842	29.555432	24.828705 57.292649								
EC85	6.036	32.059462	26.300759 71.923386								
EC90	6.282	35.513662	28.178578 96.088074								
EC95	6.645	41.329594	31.091429 148.18127								
EC99	7.326	54.930594	37.180947 335.84697								





## 4-Day Acute Toxicity Test

**Table A5. Acute *Menidia beryllina* (19 Jan 2006)**

Weston Test ID: P060103.02	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10			
	2		10	9	8	8			
	3		10	8	7	7			
	4		10	9	9	9			
2.5	1		9	7	7	7			
	2		10	10	9	9			
	3		10	9	8	8			
	4		10	10	10	10			
5	1		10	9	9	9			
	2		9	9	9	9			
	3		10	9	9	9			
	4		10	10	9	9			
10	1		10	10	10	9			
	2		9	9	8	8			
	3		10	9	8	8			
	4		10	10	10	10			
15	1		9	6	6	6			
	2		8	7	7	7			
	3		6	4	4	4			
	4		8	8	7	7			
20	1		7	4	3	1			
	2		6	5	4	4			
	3		8	6	6	4			
	4		6	4	4	2			
Date			1/20/06	1/21/06	1/22/06	1/23/06			
Time			1112	1400	1400	1320			
Initials			TS	JM/GZ	GZ	JM/GZ			





## 4-Day Acute Toxicity Test

**Table A6. Acute *Menidia beryllina* (19 Jan 2006)**

Client	Marine Research Specialists
Project:	Hydrogen Sulfide
Client Sample ID:	N/A
Weston Test ID:	P060103.02
Species:	<i>Menidia beryllina</i>

Date Received:	N/A
Date Test Started:	19-Jan-06
Date Test Ended:	23-Jan-06
Study Director:	Brian Hester
# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 1/19/05  Time: 1326  Technician: JW	Control		1		7.7		19.6		30		7.8	0	0	0
			2		7.7		19.6		30		7.8	0	0	0
			3		7.7		19.6		30		7.9	2	0	2
			4		7.7		19.6		30		7.9	0	0	0
	2.5		1		7.4		19.7		26		7.9	16	0	16
			2		7.5		19.7		26		7.9	14	0	14
			3		7.5		19.7		26		7.9	3	0	3
			4		7.5		19.7		26		7.9	2	0	2
	5		1		7.6		19.6		26		8.0	36	0	36
			2		7.5		19.6		27		7.9	20	0	20
			3		7.4		19.6		26		8.0	28	0	28
			4		7.5		19.6		26		7.9	36	0	36
	10		1		7.5		19.6		26		8.0	158	0	158
			2		7.3		19.6		26		8.0	141	0	141
			3		7.5		19.6		26		8.0	124	0	124
			4		7.3		19.7		26		8.0	112	0	112
	15		1		7.2		19.7		26		8.0	56	1/5	280
			2		7.3		19.7		26		8.0	46	1/5	230
			3		7.1		19.7		26		8.0	53	1/5	265
			4		7.2		19.7		26		8.0	50	1/5	250
	20		1		7.1		19.8		26		8.1	85	1/5	425
			2		7.1		19.8		26		8.1	83	1/5	415
			3		7.1		19.7		26		8.1	105	1/5	525
			4		7.3		19.7		26		8.1	88	1/5	440
<b>Day 1</b> <b>AM</b> Date: 1/20/06 Time: 0822 Technician: TS/JM	Control		1		6.8		19.6		30		7.7	2	0	2
	2.5				6.3		19.7		28		7.8	5	0	5
	5				6.7		19.7		28		7.8	21	0	21
	10				6.6		19.7		27		7.9	101	0	101
	15				6.3		19.8		27		7.9	46	1/5	230
	20				6.3		19.8		26		8.0	105	1/5	525
<b>Day 1</b> <b>PM</b> Date: 1/20/06 Time: 1429 Technician: JM/GZ	Control		2		6.6		19.6		30		7.76	1	0	1
	2.5				6.5		19.7		27		7.79	2	0	2
	5				6.6		19.7		27		7.81	16	0	16
	10				6.5		19.7		27		7.77	56	0	56
	15				6.0		19.7		27		7.89	34	1/5	170
	20				6.3		19.6		27		7.98	77	1/5	385



## 4-Day Acute Toxicity Test

**Table A6. Acute *Menidia beryllina* (19 Jan 2006)**

Weston Test ID:	P060103.02	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 1/21/06 Time: 0911 Technician: JM/GZ Feed Time: 0900	Control		3		5.4		19.6		30		7.53	1	0	1
	2.5				5.3		19.8		27		7.64	11	0	11
	5				5.5		19.8		27		7.71	16	0	16
	10				4.6		19.7		27		7.71	34	0	34
	15				4.8		19.7		27		7.81	28	1/5	140
	20				3.5		19.7		27		7.82	38	1/5	190
<b>Day 2</b> <b>PM</b> Date: 1/21/06 Time: 1505 Technician: JM/GZ	Control		4		5.9		19.5		30		7.54	1	0	1
	2.5				5.0		19.7		27		7.58	2	0	2
	5				5.1		19.7		27		7.66	5	0	5
	10				5.0		19.6		28		7.71	17	0	17
	15				3.7		19.5		27		7.68	7	1/5	35
	20				3.5		19.7		27		7.69	5	1/5	25
<b>Day 3</b> <b>AM</b> Date: 1/22/06 Time: 0934 Technician: JM/GZ	Control		1		4.6		19.4		30		7.52	3	0	3
	2.5				5.4		19.5		27		7.63	10	0	10
	5				4.4		19.6		27		7.59	8	0	8
	10				4.1		19.7		27		7.60	3	0	3
	15				3.1		19.7		27		7.62	8	1/5	40
	20				2.3		19.8		27		7.63	7	1/5	35
<b>Day 3</b> <b>PM</b> Date: 1/22/06 Time: 1410 Technician: JM/GZ	Control		2		4.9		19.5		30		7.41	2	0	2
	2.5				4.6		19.6		28		7.50	4	0	4
	5				4.8		19.7		27		7.58	18	0	18
	10				5.0		19.7		28		7.72	6	0	6
	15				3.7		19.7		27		7.68	4	1/5	20
	20				7.0		19.8		27		7.77	3	1/5	15
<b>Day 4</b>  Date: 1/23/06  Time: 0915  Technician: GZ/JM	Control		1		5.4		19.5		30		7.28	0	0	0
			2		5.0		19.6		30		7.46	1	0	1
			3		5.5		19.6		30		7.53	0	0	0
			4		5.4		19.6		30		7.56	1	0	1
	2.5		1		5.2		19.4		27		7.61	2	0	2
			2		5.1		19.5		27		7.52	3	0	3
			3		5.2		19.6		27		7.59	1	0	1
			4		5.0		19.7		27		7.58	1	0	1
	5		1		4.8		19.6		28		7.54	2	0	2
			2		4.9		19.6		28		7.6	3	0	3
			3		2.7		19.8		27		7.65	2	0	2
			4		4.0		19.7		28		7.56	0	0	0
	10		1		4.0		19.8		28		7.6	4	0	4
			2		4.4		19.8		27		7.65	0	0	0
			3		3.7		19.7		28		7.62	2	0	2
			4		4.5		19.6		27		7.65	1	0	1
	15		1		4.8		19.8		27		7.7	0	1/5	0
			2		4.5		19.8		27		7.67	0	1/5	0
			3		4.7		19.8		27		7.69	0	1/5	0
			4		3.5		19.7		27		7.65	1	1/5	5
	20		1		5.2		19.7		27		7.75	1	1/5	5
			2		4.6		19.8		28		7.6	0	1/5	0
			3		2.9		19.7		26		7.72	2	1/5	10
			4		2.8		19.7		27		7.73	0	1/5	0

**Table A7. Acute *Menidia beryllina* (25 Jan 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
Control	1	0.0	0.2				0.1		0.0	<b>0.1</b>	100	100	100	<b>90</b>
	2	0.0		0.1				1.1	0.0	<b>0.3</b>	100	100	100	<b>100</b>
	3	0.0			0.0				0.1	<b>0.0</b>	ND	ND	ND	<b>ND</b>
	4	0.0				0.3			0.0	<b>0.1</b>	100	100	100	<b>100</b>
2.5	1	0.4	0.9				1.4		0.1*	<b>0.9</b>	100	100	100	<b>90</b>
	2	1.0		1.2				1.9	0.0*	<b>1.4</b>	100	100	100	<b>100</b>
	3	1.3			0.6				0.0*	<b>1.0</b>	100	100	100	<b>100</b>
	4	0.8				0.9			0.0*	<b>0.9</b>	100	90	90	<b>90</b>
5	1	4.1	7.8				0.4*		0.2*	<b>5.9</b>	100	100	100	<b>100</b>
	2	0.0*		6.7				2.5*	0.8*	<b>6.7</b>	100	100	100	<b>90</b>
	3	3.0			6.0				0.9*	<b>4.5</b>	100	100	100	<b>100</b>
	4	2.4				1.5*			0.6*	<b>2.4</b>	100	100	100	<b>100</b>
10	1	13.5	18.7				0.4*		6.3*	<b>16.1</b>	100	100	100	<b>100</b>
	2	15.2		20.5				3.8*	5.5*	<b>17.9</b>	100	90	90	<b>90</b>
	3	13.1			13.4				3.7*	<b>13.3</b>	100	90	90	<b>90</b>
	4	14.4				5.8*			1.3*	<b>14.4</b>	100	100	100	<b>100</b>
15	1	48.7	37.5				1.4*		11.6*	<b>43.1</b>	100	90	90	<b>90</b>
	2	47.6		35.9				1.6*	9.9*	<b>41.7</b>	100	90	90	<b>80</b>
	3	32.4			59.1				9.7*	<b>45.7</b>	100	100	100	<b>100</b>
	4	27.9				5.3*			4.6*	<b>27.9</b>	100	90	100	<b>90</b>
20	1	43.4	53.9				2.5*		15.6*	<b>48.7</b>	100	80	80	<b>80</b>
	2	47.4		56.3				3.3*	14.5*	<b>51.9</b>	100	80	60	<b>60</b>
	3	64.5			45.4				12.0*	<b>54.9</b>	100	100	100	<b>100</b>
	4	59.1				1.4*			13.3*	<b>59.1</b>	70	60	60	<b>50</b>

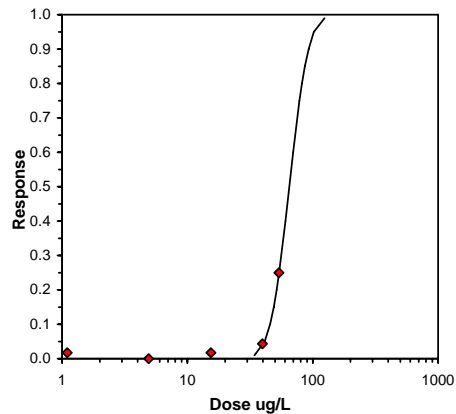
\* Sulfide concentrations decreased during test

Acute Fish Test-96 Hr Survival				
Start Date:	1/25/2006	Test ID:	P060103.04	Sample ID:
End Date:	1/29/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPA/600/R	Test Species:
Comments:	Average			
Conc-ug/L	1	2	3	4
Control	0.9000	1.0000	1.0000	
1.1	0.9000	1.0000	1.0000	0.9000
4.9	1.0000	0.9000	1.0000	1.0000
15.4	1.0000	0.9000	0.9000	1.0000
39.6	1.0000	0.8000	1.0000	0.9000
53.7	0.8000	0.6000	1.0000	0.5000

Transform: Arcsin Square Root											
Conc-ug/L	Mean	N-Mean	Mean	Min	Max	CV%	N	t-Stat	1-Tailed Critical	MSD	Number Resp
Control	0.9667	1.0000	1.3577	1.2490	1.4120	6.930	3				1
1.1	0.9500	0.9828	1.3305	1.2490	1.4120	7.072	4	0.236	2.567	0.2957	2
4.9	0.9750	1.0086	1.3713	1.2490	1.4120	5.942	4	-0.118	2.567	0.2957	1
15.4	0.9500	0.9828	1.3305	1.2490	1.4120	7.072	4	0.236	2.567	0.2957	2
39.6	0.9250	0.9569	1.2951	1.1071	1.4120	11.347	4	0.544	2.567	0.2957	3
*53.7	0.7250	0.7500	1.0477	0.7854	1.4120	26.497	4	2.691	2.567	0.2957	11
											40

Auxiliary Tests				Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)				0.9305108	0.881	0.4316294	1.4567967
Bartlett's Test indicates equal variances (p = 0.24)				6.7858467	15.086272		
Hypothesis Test (1-tail, 0.05)				NOEC	LOEC	ChV	TU
Bonferroni t Test				39.6	53.7	46.114206	
Treatments vs Control							

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	8.3336824	4.6322787	-0.745584	17.412949	0.0333333	0.4427081	7.8147278	0.93	1.8128875	0.119995	7
Intercept	-10.108028	7.9208895	-25.632972	5.4169155							
TSCR	0.0399999	0.0160001	0.0086398	0.0713601							
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	34.177273									
EC05	3.355	41.258487									
EC10	3.718	45.615004									
EC15	3.964	48.811333									
EC20	4.158	51.510664									
EC25	4.326	53.94511									
EC40	4.747	60.602018									
EC50	5.000	64.996125									
EC60	5.253	69.708839									
EC75	5.674	78.311013									
EC80	5.842	82.012072									
EC85	6.036	86.547449									
EC90	6.282	92.611985									
EC95	6.645	102.39096									
EC99	7.326	123.60543									





## 4-Day Acute Toxicity Test

**Table A8. Acute *Menidia beryllina* (25 Jan 2006)**

Weston Test ID: P060103.04	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	9			
	2		10	10	10	10			
	3		ND	ND	ND	ND			
	4		10	10	10	10			
2.5	1		10	10	10	9			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	9	9	9			
5	1		10	10	10	10			
	2		10	10	10	9			
	3		10	10	10	10			
	4		10	10	10	10			
10	1		10	10	10	10			
	2		10	9	9	9			
	3		10	9	9	9			
	4		10	10	10	10			
15	1		10	9	9	9			
	2		10	9	9	8			
	3		10	10	10	10			
	4		10	9	10	9			
20	1		10	8	8	8			
	2		10	8	6	6			
	3		10	10	10	10			
	4		7	6	6	5			
Date			1/26/06	1/27/06	1/28/06	1/29/06			
Time			1135	1353	1359	1420			
Initials			JM	JM/BH	BCG	JW			



## 4-Day Acute Toxicity Test

**Table A9. Acute *Menidia beryllina* (25 Jan 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen Sulfide	Date Test Started:	25-Jan-06
Client Sample ID:	N/A	Date Test Ended:	29-Jan-06
Weston Test ID:	P060103.04	Study Director:	Brian Hester
Species:	<i>Menidia beryllina</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 1/25/05  Time: 1435  Technician: JW/GZ	Control		1		6.7		19.3		29		7.59	0	0	0
			2		6.6		19.7		29		7.65	0	0	0
			3		6.1		19.5		29		7.62	0	0	0
			4		6.9		19.5		29		7.67	0	0	0
	2.5		1		5.9		19.5		26.5		7.67	3	0	3
			2		5.4		19.8		26.5		7.64	7	0	7
			3		6.2		19.6		27		7.30	5	0	5
			4		6.1		19.5		27		7.66	6	0	6
	5		1		6.4		19.5		26.5		7.73	34	0	34
			2		7.1		19.4		28		7.70	0	0	0
			3		6.0		19.6		27		7.70	24	0	24
			4		6.8		19.5		27		7.73	20	0	20
	10		1		5.9		19.3		27		7.69	105	0	105
			2		6.0		19.6		27		7.76	136	0	136
			3		6.3		19.6		27		7.80	127	0	127
			4		5.6		19.7		27		7.78	134	0	134
	15		1		5.7		19.5		27		7.73	82	1/5	410
			2		5.3		19.6		27		7.56	57	1/5	285
			3		6.0		19.7		27		7.83	67	1/5	335
			4		5.5		19.7		27		7.76	50	1/5	250
	20		1		5.7		19.4		27		7.90	104	1/5	520
			2		5.9		19.7		27		7.88	109	1/5	545
			3		6.1		19.6		27		7.67	96	1/5	480
			4		5.6		19.2		27		7.87	133	1/5	665
<b>Day 1</b> <b>AM</b> Date: 1/26/06 Time: 0856 Technician: JM	Control		1		6.6		19.5		29		7.60	1	0	1
	2.5				6.2		19.6		27		7.63	6	0	6
	5				6.7		19.6		28		7.73	66	0	66
	10				6.4		19.6		28		7.75	165	0	165
	15				6.5		19.6		28		7.83	78	1/5	390
	20				6.6		19.6		27		7.88	124	1/5	620
<b>Day 1</b> <b>PM</b> Date: 1/26/06 Time: 1720 Technician: JM/JW/BH	Control		2		6.4		18.8		29		7.63	1	0	1
	2.5				6.0		18.9		27		7.61	8	0	8
	5				6.7		18.7		27		7.77	61	0	61
	10				6.1		18.8		27		7.75	180	0	180
	15				6.8		18.7		27		7.91	88	1/5	440
	20				6.3		18.8		27		7.91	138	1/5	690



## 4-Day Acute Toxicity Test

**Table A9. Acute *Menidia beryllina* (25 Jan 2006)**

Weston Test ID:	P060103.04	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 1/27/06 Time: 0931 Technician: JM Feed Time:	Control		3		6.6		19.3		30		7.53	0	0	0
	2.5				6.5		19.3		28		7.64	4	0	4
	5				6.5		19.3		28		7.69	47	0	47
	10				5.8		19.3		27		7.73	113	0	113
	15				6.0		19.2		28		7.79	113	1/5	565
	20				5.9		19.3		27		7.83	94	1/5	470
<b>Day 2</b> <b>PM</b> Date: 1/27/06 Time: 1705 Technician: JM/JW	Control		4		6.5		18.7		28		7.61	2	0	2
	2.5				6.2		18.8		27		7.62	6	0	6
	5				6.9		18.4		27		7.71	12	0	12
	10				5.9		18.6		27		7.71	47	0	47
	15				5.2		18.6		27		7.69	41	0	41
	20				4.4		18.8		28		7.72	12	0	12
<b>Day 3</b> <b>AM</b> Date: 1/28/06 Time: 1015 Technician: JW	Control		1		6.2		18.7		30		7.64	1	0	1
	2.5				5.8		18.8		27		7.64	10	0	10
	5				5.9		18.9		27		7.69	3	0	3
	10				5.5		18.9		28		7.69	3	0	3
	15				6.0		18.9		27		7.68	11	0	11
	20				4.5		19.0		27		7.75	22	0	22
<b>Day 3</b> <b>PM</b> Date: 1/28/06 Time: 1715 Technician: JW	Control		2		6.6		18.1		30		7.58	7	0	7
	2.5				6.4		18.2		26		7.66	14	0	14
	5				6.6		18.4		26		7.75	22	0	22
	10				5.8		18.4		27		7.79	36	0	36
	15				5.6		18.5		27		7.85	17	0	17
	20				5.0		18.0		27		7.93	42	0	42
<b>Day 4</b>  Date: 1/29/06  Time: 0836  Technician: BG/JW	Control		1		6.7		19.5		30		7.70	0	0	0
			2		6.6		19.3		30		7.71	0	0	0
			3		6.7		19.7		30		7.71	1	0	1
			4		6.8		19.6		30		7.73	0	0	0
	2.5		1		6.2		19.7		27		7.79	1	0	1
			2		6.4		19.4		27		7.78	0	0	0
			3		6.4		19.6		28		7.76	0	0	0
			4		6.2		19.8		27		7.78	0	0	0
	5		1		6.4		19.7		27		7.82	2	0	2
			2		6.4		19.6		27		7.79	8	0	8
			3		6.5		19.3		27		7.82	9	0	9
			4		6.2		19.2		27		7.8	6	0	6
	10		1		5.8		19.5		30		7.85	69	0	69
			2		5.8		19.4		28		7.86	61	0	61
			3		5.5		19.5		28		7.85	40	0	40
			4		5.5		19.3		28		7.82	13	0	13
	15		1		5.3		19.7		27		7.89	136	0	136
			2		5.5		19.6		27		7.91	121	0	121
			3		5.4		19.2		28		7.89	115	0	115
			4		5.2		19.1		27		7.9	55	0	55
	20		1		4.8		19.7		27		7.93	199	0	199
			2		4.6		19.0		27		7.93	185	0	185
			3		4.9		19.4		28		7.95	161	0	161
			4		4.5		19.6		27		7.94	173	0	173

**Table A10. Acute *Menidia beryllina* (13 Mar 2006)**

Treatment	Rep	Day 0	Day 1	Day 2	Day 3	Day 4	Mean	Percentage Survival			
		PM	AM	AM	AM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	0.0	0.0				<b>0.0</b>	100	100	100	<b>90</b>
	2	0.1		0.0			<b>0.1</b>	100	100	100	<b>100</b>
	3	0.0			0.1		<b>0.1</b>	100	100	100	<b>100</b>
	4	0.3				0.0	<b>0.2</b>	100	100	90	<b>90</b>
<b>4</b>	1	2.3	0.7				<b>1.5</b>	100	100	100	<b>100</b>
	2	2.1		2.1			<b>2.1</b>	100	100	100	<b>100</b>
	3	4.2			1.1		<b>2.6</b>	100	100	100	<b>100</b>
	4	2.0				0.0*	<b>2.0</b>	100	100	100	<b>100</b>
<b>8</b>	1	8.7	8.8				<b>8.7</b>	90	90	90	<b>90</b>
	2	6.8		8.2			<b>7.5</b>	100	100	100	<b>100</b>
	3	9.7			7.2		<b>8.4</b>	100	90	90	<b>90</b>
	4	7.8				4.6	<b>6.2</b>	100	100	100	<b>100</b>
<b>16</b>	1	15.2	13.4				<b>14.3</b>	100	100	100	<b>100</b>
	2	13.8		13.6			<b>13.7</b>	100	100	100	<b>100</b>
	3	14.6			6.9		<b>10.8</b>	90	90	90	<b>90</b>
	4	12.5				5.6	<b>9.1</b>	100	100	100	<b>100</b>
<b>24</b>	1	25.5	24.9				<b>25.2</b>	90	90	80	<b>80</b>
	2	24.6		22.6			<b>23.6</b>	100	90	90	<b>90</b>
	3	25.7			12.1		<b>18.9</b>	100	90	90	<b>90</b>
	4	24.6				11.0	<b>17.8</b>	100	90	80	<b>80</b>
<b>32</b>	1	38.3	33.8				<b>36.0</b>	90	70	70	<b>70</b>
	2	36.1		30.7			<b>33.4</b>	100	100**	100	<b>100</b>
	3	35.0			19.3		<b>27.1</b>	100	80	80	<b>80</b>
	4	33.9				14.6	<b>24.3</b>	100	70	70	<b>70</b>
<b>48</b>	1	56.5	50.3				<b>53.4</b>	100	30	20	<b>20</b>
	2	59.0		61.6			<b>60.3</b>	80	10	10	<b>10</b>
	3	52.1			36.3		<b>44.2</b>	100	50	50	<b>40</b>
	4	53.7				28.8	<b>41.3</b>	80	40	40	<b>40</b>

\* Sulfide concentrations decreased during test

\*\* Survival miscounted



### Larval Fish Growth and Survival Test-96 Hr Survival

Start Date: 03/13/06	Test ID: Menidia T2	Sample ID:
End Date:	Lab ID: WESTON	Sample Type:
Sample Date:	Protocol: EPAW 95-EPA/600/R-95/136	Test Species: MB-Menidia beryllina
Comments: Max		

Conc-ug/L	1	2	3	4
Control	0.9000	1.0000	1.0000	0.9000
2.1	1.0000	1.0000	1.0000	1.0000
7.7	0.9000	1.0000	0.9000	1.0000
12	1.0000	1.0000	0.9000	1.0000
21.4	0.8000	0.9000	0.9000	0.8000
30.2	0.7000	1.0000	0.8000	0.7000
49.8	0.2000	0.1000	0.4000	0.4000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%	N				
Control	0.9500	1.0000	1.3305	1.2490	1.4120	7.072	4			2	40
2.1	1.0000	1.0526	1.4120	1.4120	1.4120	0.000	4	22.00	10.00	0	40
7.7	0.9500	1.0000	1.3305	1.2490	1.4120	7.072	4	18.00	10.00	2	40
12	0.9750	1.0263	1.3713	1.2490	1.4120	5.942	4	20.00	10.00	1	40
21.4	0.8500	0.8947	1.1781	1.1071	1.2490	6.954	4	12.00	10.00	6	40
30.2	0.8000	0.8421	1.1254	0.9912	1.4120	17.662	4	13.00	10.00	8	40
*49.8	0.2750	0.2895	0.5387	0.3218	0.6847	33.093	4	10.00	10.00	29	40

#### Auxiliary Tests

Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	Statistic: 0.965052	Critical: 0.896	Skew: 0.417004	Kurt: 0.657638
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Equality of variance cannot be confirmed

#### Hypothesis Test (1-tail, 0.05)

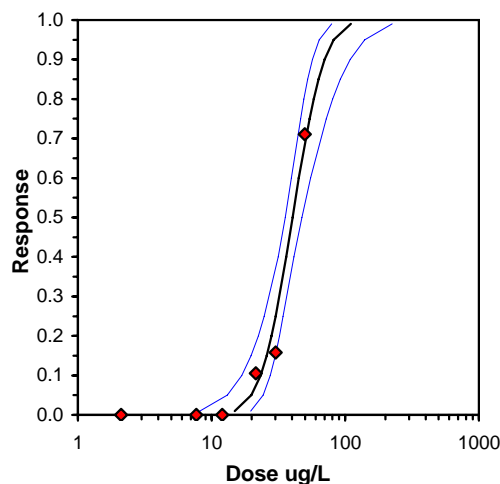
Steel's Many-One Rank Test	NOEC: 30.2	LOEC: 49.8	ChV: 38.78092	TU:
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Treatments vs Control

#### Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	5.355841	1.061035	3.276213	7.435469	0.05	4.187072	9.487729	0.38	1.606348	0.186712	4
Intercept	-3.60335	1.657669	-6.85238	-0.35431							
TSCR	0.032997	0.014434	0.004706	0.061287							

Point	Probits	ug/L	95% Fiducial Limits	
EC01	2.674	14.85905	8.189845	19.55392
EC05	3.355	19.91747	13.09725	24.37793
EC10	3.718	23.2845	16.75402	27.5302
EC15	3.964	25.87221	19.7167	29.98319
EC20	4.158	28.13243	22.36798	32.19201
EC25	4.326	30.22823	24.83729	34.33696
EC40	4.747	36.22806	31.56193	41.38927
EC50	5.000	40.3969	35.63928	47.37229
EC60	5.253	45.04546	39.62345	55.06848
EC75	5.674	53.98628	46.28762	72.20641
EC80	5.842	58.00813	49.0393	80.72119
EC85	6.036	63.07579	52.36972	92.06925
EC90	6.282	70.08566	56.78799	108.8233
EC95	6.645	81.9336	63.89134	139.7294
EC99	7.326	109.826	79.39449	224.1852





## 4-Day Acute Toxicity Test

**Table A11. Acute *Menidia beryllina* (13 Mar 2006)**

Weston Test ID: P060103.26	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	9			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	9	9			
4	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
8	1		9	9	9	9			
	2		10	10	10	10			
	3		10	9	9	9			
	4		10	10	10	10			
16	1		10	10	10	10			
	2		10	10	10	10			
	3		9	9	9	9			
	4		10	10	10	10			
24	1		9	9	8	8			
	2		10	9	9	9			
	3		10	9	9	9			
	4		10	9	8	8			
32	1		9	7	7	7			
	2		10	10**	10	10			
	3		10	8	8	8			
	4		10	7	7	7			
48	1		10	3	2	2			
	2		8	1	1	1			
	3		10	5	5	4			
	4		8	4	4	4			
Date			3/14/06	3/15/06	3/16/06	3/17/06			
Time			950	943	915	925			
Initials			GZ	JM	TS	GZ			

\*\* Survival miscounted



## 4-Day Acute Toxicity Test

**Table A12. Acute *Menidia beryllina* (13 Mar 2006)**

Client	Marine Research Specialists										Date Received:	N/A			
Project:	Hydrogen Sulfide										Date Test Started:	13-Mar-06			
Client Sample ID:	N/A										Date Test Ended:	20-Mar-06			
Weston Test ID:	P060103.26										Study Director:	Brian Hester			
Species:	<i>Menidia beryllina</i>										# Organisms/Chamber:	10			

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H2S
<b>Day 0</b>  Date: 3/13/06  Time: 1703  Technician: TS/GZ/JM	Control		1		8.4		19.0		31		7.70	0	0	0	0
			2		8.2		19.2		31		7.71	1	0	1	0.12
			3		8.1		19.5		31		7.77	0	0	0	0
			4		8.1		19.7		31		7.79	3	0	3	0.31
	4		1		8.1		19.2		30		7.83	24	0	24	2.35
			2		8.1		19.0		30		7.83	21	0	21	2.07
			3		8.1		19.1		30		7.75	36	0	36	4.17
			4		8.0		19.5		30		7.82	20	0	20	1.98
	8		1		8.1		19.4		30		7.84	91	0	91	8.66
			2		8.0		19.4		30		7.84	71	0	71	6.76
			3		8.0		19.3		30		7.81	95	0	95	9.66
			4		8.0		19.7		30		7.84	83	0	83	7.82
	16		1		8.1		19.5		30		7.85	82	1:2	164	15.24
			2		8.0		19.5		30		7.86	76	1:2	152	13.83
			3		8.1		19.3		30		7.79	69	1:2	138	14.63
			4		8.0		19.6		30		7.86	69	1:2	138	12.51
	24		1		8.1		19.5		30		7.87	143	1:2	286	25.48
			2		8.0		19.5		30		7.87	138	1:2	276	24.59
			3		8.0		19.4		30		7.86	141	1:2	282	25.74
			4		7.9		19.7		30		7.87	139	1:2	278	24.60
	32		1		8.0		19.5		30		7.90	229	1:2	458	38.29
			2		8.0		19.5		30		7.90	216	1:2	432	36.12
			3		8.0		19.4		30		7.88	200	1:2	400	35.01
			4		7.9		19.7		30		7.89	200	1:2	400	33.93
	48		1		8.0		19.4		30		7.93	359	1:2	718	56.51
			2		8.0		19.4		30		7.94	383	1:2	766	59.01
			3		8.0		19.5		30		7.92	325	1:2	650	52.08
			4		7.9		19.7		30		7.94	352	1:2	704	53.69
<b>Day 1</b> <b>AM</b> Date: 3/14/06 Time: 0902 Technician: TS/GZ	Control				8.0		19.2		31		7.76	0	0	0	0
	4				7.4		19.6		30		7.79	7	0	7	0.73
	8				7.1		19.8		30		7.82	90	0	90	8.82
	16				7.0		20.0		30		7.82	69	1:2	138	13.433
	24				7.1		19.9		30		7.85	136	1:2	272	24.943
	32				7.0		19.9		30		7.87	192	1:2	384	33.76
	48				7.1		19.7		30		7.92	316	1:2	632	50.30
<b>Day 1</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND



## 4-Day Acute Toxicity Test

**Table A12. Acute *Menidia beryllina* (13 Mar 2006)**

Weston Test ID:	P060103.26	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H2S
<b>Day 2</b> <b>AM</b> Date: 3/15/06 Time: 0832 Technician: GZ/JM	Control		2		6.9		18.9		32		7.68	0	0	0	0
	4				6.6		19.5		30		7.71	17	0	17	2.11
	8				7.4		19.8		30		7.74	71	0	71	8.21
	16				7.4		19.8		30		7.75	60	1:2	120	13.59
	24				7.5		19.9		30		7.74	98	1:2	196	22.592
	32				7.3		19.9		30		7.75	136	1:2	272	30.715
	48				7.4		19.7		30		7.81	307	1:2	614	61.628
<b>Day 2</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 3</b> <b>AM</b> Date: 3/16/06 Time: 1021 Technician: JM	Control		3		7.0		18.8		31		7.65	1	0	1	0.14
	4				7.6		19.3		30		7.77	10	0	10	1.10
	8				7.5		19.6		30		7.78	67	0	67	7.18
	16				7.3		19.8		30		7.79	66	0	66	6.88
	24				7.4		19.7		30		7.76	109	0	109	12.14
	32				7.4		19.7		30		7.80	188	0	188	19.27
	48				7.2		19.8		30		7.78	341	0	341	36.31
<b>Day 3</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 4</b> <b>AM</b> Date: 3/17/06 Time: 0845 Technician: GZ/TS	Control		4		6.1		19.7		31		7.88	0	0	0	0
	4				7.5		19.8		30		7.92	0	0	0	0
	8				7.4		19.9		30		7.94	60	0	60	4.55
	16				7.4		19.9		30		7.96	77	0	77	5.59
	24				7.4		19.8		30		7.98	157	0	157	10.95
	32				7.4		19.8		30		7.94	192	0	192	14.59
	48				7.4		19.8		30		7.94	379	0	379	28.81
<b>Day 4</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND

ND = No data, replicate previously terminated

**Table A13. Acute *Menidia beryllina* (14 Mar 2006)**

Treatment	Rep	Day 0	Day 1	Day 2	Day 3	Day 4	Mean	Percentage Survival			
		PM	AM	AM	AM	AM		Day 1	Day 2	Day 3	Day 4
Control	1		0.0				<b>0.0</b>	90	90	80	<b>80</b>
	2	0.0			0.0		<b>0.0</b>	90	90	90	<b>90</b>
	3			1.0		0.0	<b>0.5</b>	90	90	80	<b>80</b>
4	1		2.2				<b>2.2</b>	100	100	100	<b>100</b>
	2	2.1			0.0*		<b>2.1</b>	90	80	90	<b>90</b>
	3			2.0		0.5*	<b>2.0</b>	100**	100**	100	<b>100</b>
8	1		10.7				<b>10.7</b>	100	100	100	<b>100</b>
	2	8.2			4.1		<b>6.2</b>	90	90	80	<b>80</b>
	3			9.6		3.1*	<b>9.6</b>	90	90	90**	<b>90</b>
16	1		17.2				<b>17.2</b>	90	90	90	<b>90</b>
	2	14.7			6.1		<b>10.4</b>	100	90	90	<b>90</b>
	3			13.8		7.7	<b>10.7</b>	80	70	70	<b>70</b>
24	1		21.2				<b>21.2</b>	80	80	80**	<b>80</b>
	2	16.2			6.8*		<b>16.2</b>	100	90	60	<b>60</b>
	3			16.0		13.2	<b>14.6</b>	100	100	100	<b>100</b>
32	1		30.1				<b>30.1</b>	70	50	40	<b>40</b>
	2	19.3			11.9		<b>15.6</b>	90	80	80	<b>60</b>
	3			23.3		16.9	<b>20.1</b>	70	50	50	<b>30</b>
48	1		34.3				<b>34.3</b>	50	40	30**	<b>30</b>
	2	31.2			14.4		<b>22.8</b>	70	60	50	<b>50</b>
	3			25.3		20.7	<b>23.0</b>	70	70	50	<b>40</b>

\* Sulfide concentrations decreased during test

\*\* Survival miscounted

# **Larval Fish Growth and Survival Test-96 Hr Survival**

Start Date: 3/14/2006      Test ID: Men 3/14      Sample ID:  
 End Date:      Lab ID: WESTON      Sample Type:  
 Sample Date:      Protocol: EPAM 94-EPA/600/4-91/003      Test Species: MB-Menidia beryllina  
 Comments:

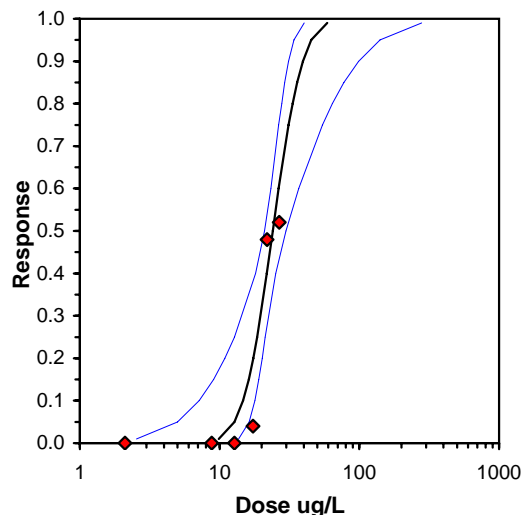
Conc-ug/L	1	2	3
Control	0.8000	0.9000	0.8000
2.1	1.0000	0.9000	1.0000
8.8	1.0000	0.8000	0.9000
12.8	0.9000	0.9000	0.7000
17.3	0.8000	0.6000	1.0000
21.9	0.4000	0.6000	0.3000
26.7	0.3000	0.5000	0.4000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%						
Control	0.8333	1.0000	1.1544	1.1071	1.2490	7.096	3				5	30
2.1	0.9667	1.1600	1.3577	1.2490	1.4120	6.930	3	-1.618	2.530	0.3177	1	30
8.8	0.9000	1.0800	1.2561	1.1071	1.4120	12.145	3	-0.809	2.530	0.3177	3	30
12.8	0.8333	1.0000	1.1631	0.9912	1.2490	12.802	3	-0.069	2.530	0.3177	5	30
17.3	0.8000	0.9600	1.1351	0.8861	1.4120	23.265	3	0.154	2.530	0.3177	6	30
*21.9	0.4333	0.5200	0.7168	0.5796	0.8861	21.724	3	3.485	2.530	0.3177	17	30
*26.7	0.4000	0.4800	0.6833	0.5796	0.7854	15.058	3	3.752	2.530	0.3177	18	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.988173	0.873	0.112005	-0.19767
Bartlett's Test indicates equal variances ( $p = 0.76$ )	3.396231	16.81189		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	17.3	21.9	19.46458		0.28519	0.340957	0.205828	0.023654	4.5E-04	6, 14
Treatments vs Control										

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	5.996611	1.888326	2.295492	9.697729	0.166667	4.033511	9.487729	0.4	1.381581	0.166761	6
Intercept	-3.2848	2.532672	-8.24884	1.679233							
TSCR	0.099844	0.032346	0.036445	0.163243							
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	9.854558	2.543874	13.74303							
EC05	3.355	12.80217	5.004137	16.2707							
EC10	3.718	14.71866	7.154234	17.86087							
EC15	3.964	16.17128	9.079081	19.07577							
EC20	4.158	17.42735	10.93677	20.16464							
EC25	4.326	18.58243	12.7782	21.23502							
EC40	4.747	21.84402	18.12	25.24969							
EC50	5.000	24.07581	21.02927	29.79077							
EC60	5.253	26.53563	23.31954	36.78558							
EC75	5.674	31.19317	26.53725	54.50465							
EC80	5.842	33.26063	27.77446	64.07469							
EC85	6.036	35.84408	29.23504	77.51472							
EC90	6.282	39.38162	31.12705	98.6752							
EC95	6.645	45.27705	34.08587	141.4171							
EC99	7.326	58.81997	40.27558	278.7353							





## 4-Day Acute Toxicity Test

**Table A14. Acute *Menidia beryllina* (14 Mar 2006)**

Weston Test ID: P060103.27	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		9	9	8	8			
	2		9	9	9	9			
	3		9	9	8	8			
	4		10	10	10	10			
4	1		10	10	10	10			
	2		9	8	9	9			
	3		10**	10**	10	10			
	4		10	10	10	10			
8	1		10	10	10	10			
	2		9	9	8	8			
	3		9	9	9**	9			
	4		10	10	10	10			
16	1		9	9	9	8			
	2		10	9	9	9			
	3		8	7	7	7			
	4		10	10	9	8			
24	1		8	8	8**	8			
	2		10	9	6	6			
	3		10	10	10	10			
	4		9	7	6	6			
32	1		7	5	4	4			
	2		9	8	8	6			
	3		7	5	5	3			
	4		8	6	6	5			
48	1		5	4	3**	3			
	2		7	6	5	5			
	3		7	7	5	4			
	4		9	7	5	3			
Date			3/15/06	3/16/06	3/17/06	3/18/06			
Time			930	1000	851	905			
Initials			DM	DM	TS	DM			

\*\* Survival miscounted



## 4-Day Acute Toxicity Test

**Table A15. Acute *Menidia beryllina* (14 Mar 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen Sulfide	Date Test Started:	14-Mar-06
Client Sample ID:	N/A	Date Test Ended:	21-Mar-06
Weston Test ID:	P060103.27	Study Director:	Brian Hester
Species:	<i>Menidia beryllina</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H2S
<b>Day 0</b>  Date: 3/14/06  Time: 1808  Technician: TS	Control		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		8.1		19.0		31		7.80	0	0	0	0.00
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	4		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		8.0		19.4		30		7.88	24	0	24	2.10
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	8		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		8.0		19.4		30		7.89	96	0	96	8.23
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	16		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.5		30		7.91	179	0	179	14.65
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	24		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.6		30		7.93	207	0	207	16.18
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	32		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.6		30		7.93	247	0	247	19.31
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	48		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.7		30		7.87	352	0	352	31.15
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 1</b> <b>AM</b> Date: 3/15/06 Time: 0815 Technician: DM	Control				7.8		19.4		30		7.79	0	0	0	0.00
	4				7.6		19.4		30		7.75	19	0	19	2.18
	8				7.8		19.4		30		7.77	97	0	97	10.68
	16				7.7		18.9		30		7.79	160	0	160	17.18
	24				7.7		19.3		30		7.84	222	0	222	21.20
	32				7.9		18.5		30		7.81	288	0	288	30.07
	48				7.5		19.3		30		7.81	337	0	337	34.27
<b>Day 1</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND





## 4-Day Acute Toxicity Test

**Table A15. Acute *Menidia beryllina* (14 Mar 2006)**

Weston Test ID:	P060103.27	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H2S
<b>Day 2</b> <b>AM</b> Date: 3/16/06 Time: 1100 Technician: DM/JM	Control		3		7.2		19.6		30		7.65	5	0	5	0.98
	4				7.2		19.8		30		7.70	23	0	23	2.01
	8				7.2		19.9		30		7.70	96	0	96	9.63
	16				7.2		19.9		30		7.75	101	0	101	13.78
	24				7.2		20.0		30		7.73	149	0	149	15.95
	32				7.1		20.0		30		7.75	176	0	176	23.30
	48				6.9		20.0		30		7.71	280	0	280	25.29
<b>Day 2</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 3</b> <b>AM</b> Date: 3/17/06 Time: 0900 Technician: GZ/TS	Control		2		7.4		19.3		31		7.92	0	0	0	0.00
	4				7.3		19.8		30		7.98	0	0	0	0.00
	8				7.2		19.7		30		7.97	58	0	58	4.15
	16				7.0		19.8		30		7.99	90	0	90	6.15
	24				7.3		19.8		30		7.99	99	0	99	6.76
	32				7.0		19.9		30		8.01	182	0	182	11.86
	48				7.2		19.9		30		8.00	216	0	216	14.39
<b>Day 3</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 4</b> <b>AM</b> Date: 3/18/06 Time: 0850 Technician: DM	Control		3		7.3		19.7		30		7.70	0	0	0	0.00
	4				7.6		19.5		30		7.70	4	0	4	0.51
	8				7.5		19.9		30		7.78	29	0	29	3.08
	16				7.2		20.0		30		7.79	74	0	74	7.67
	24				7.3		20.0		30		7.75	117	0	117	13.17
	32				7.2		20.0		30		7.77	156	0	156	16.85
	48				7.3		19.9		30		7.78	195	0	195	20.70
<b>Day 4</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND

ND = No data; replicate previously terminated

**Table B1. Acute *Atherinops affinis* (20 Mar 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
Control	1	0.5	0.2						0.5	<b>0.4</b>	100	100	100	<b>90</b>
	2	0.0					0.1		0.6	<b>0.2</b>	90	90	90	<b>90</b>
	3	0.4			0.1				0.7	<b>0.4</b>	100	100	100	<b>90</b>
	4	0.3							0.9	<b>0.6</b>	100	100	100	<b>90</b>
10	1	12.0	11.9						10.4	<b>11.4</b>	100	90	90	<b>70</b>
	2	16.4					5.2		8.3	<b>10.0</b>	100	100	80	<b>70</b>
	3	14.0			15.9				8.0	<b>12.6</b>	100	80	80	<b>80</b>
	4	14.1							9.1	<b>11.6</b>	100	100	90	<b>60</b>
20	1	24.5	26.8						18.7	<b>23.3</b>	90	90	90	<b>80</b>
	2	26.9					14.0		17.8	<b>19.5</b>	100	100	100	<b>80</b>
	3	25.6			27.8				20.1	<b>24.5</b>	100	90	90	<b>80</b>
	4	25.6							20.1	<b>22.9</b>	80	80**	80	<b>80</b>
30	1	38.0	39.8						29.6	<b>35.8</b>	70	60	60	<b>40</b>
	2	39.6					25.7		28.4	<b>31.2</b>	70	70	70	<b>70</b>
	3	43.9			43.3				33.2	<b>40.1</b>	100	80	70	<b>60</b>
	4	40.6							32.1	<b>36.3</b>	90	40	30	<b>30</b>
40	1	47.8	53.5						46.6	<b>49.3</b>	80	50	40	<b>30</b>
	2	52.8					43.7		44.9	<b>47.2</b>	70	50	20	<b>20</b>
	3	50.6			59.6				45.1	<b>51.7</b>	90	80	50	<b>30</b>
	4	45.0							40.5	<b>42.8</b>	80	60	50	<b>40</b>
50	1	70.2	71.3						63.9	<b>68.5</b>	30	10	10	<b>10</b>
	2	82.1					58.5		59.7	<b>66.8</b>	80	30	20	<b>20</b>
	3	68.4			83.3				67.0	<b>72.9</b>	40	0	T	<b>T</b>
	4	67.4							64.7	<b>66.0</b>	40	10	10	<b>0</b>

T = Test terminated due to 0% survival

\*\* Survival miscounted

Acute Fish Test-96 Hr Survival				
Start Date:	3/20/2006	Test ID:	P060103.28	Sample ID:
End Date:	3/24/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPAA 91-EPA/600/4-90/027F	Test Species:
Comments:	Average			
	Antherinops affinis			

Conc-ug/L	1	2	3	4
Control	0.9000	0.9000	0.9000	0.9000
11.4	0.7000	0.7000	0.8000	0.6000
22.6	0.8000	0.8000	0.8000	0.8000
35.9	0.4000	0.7000	0.6000	0.3000
47.8	0.3000	0.2000	0.3000	0.4000
68.6	0.1000	0.2000	0.0000	0.0000

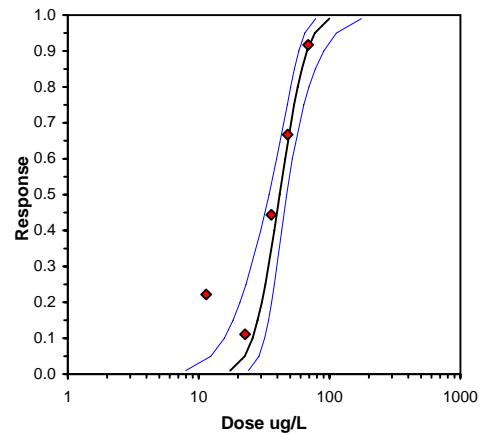
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%					
Control	0.9000	1.0000	1.2490	1.2490	1.2490	0.000	4			4	40
*11.4	0.7000	0.7778	0.9939	0.8861	1.1071	9.086	4	10.00	10.00	12	40
*22.6	0.8000	0.8889	1.1071	1.1071	1.1071	0.000	4	10.00	10.00	8	40
*35.9	0.5000	0.5556	0.7854	0.5796	0.9912	23.814	4	10.00	10.00	20	40
*47.8	0.3000	0.3333	0.5769	0.4636	0.6847	15.654	4	10.00	10.00	28	40
*68.6	0.0750	0.0833	0.2757	0.1588	0.4636	53.294	4	10.00	10.00	37	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.9122946	0.884	0.1807433	0.3226673
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	<11.4	11.4		

Treatments vs Control

Parameter	Value	SE	95% Fiducial Limits	Maximum Likelihood-Probit							
				Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter	
Slope	6.1465704	1.3454533	3.509482 8.7836589	0.1	3.4786801	7.8147278	0.32	1.6193737	0.1626924	7	
Intercept	-4.953594	2.2445724	-9.352956 -0.554232								
TSCR	0.1906934	0.0411726	0.1099951 0.2713918								
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	17.413845	7.9656418 24.040099								
EC05	3.355	22.478565	12.39106 28.894936								
EC10	3.718	25.755801	15.656958 31.923028								
EC15	3.964	28.232797	18.313543 34.181397								
EC20	4.158	30.370249	20.722643 36.124861								
EC25	4.326	32.332506	23.017877 37.917217								
EC40	4.747	37.85786	29.787001 43.13427								
EC50	5.000	41.626864	34.481942 47.020781								
EC60	5.253	45.771099	39.436343 51.882065								
EC75	5.674	53.592993	47.459024 63.466213								
EC80	5.842	57.055699	50.458089 69.595473								
EC85	6.036	61.375281	53.876817 77.946935								
EC90	6.282	67.277881	58.165853 90.423951								
EC95	6.645	77.08658	64.711165 113.46284								
EC99	7.326	99.506799	78.242017 175.45517								





## 4-Day Acute Toxicity Test

**Table B2. Acute *Atherinops affinis* (20 Mar 2006)**

Weston Test ID: P060103.28	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	9			
	2		9	9	9	9			
	3		10	10	10	9			
	4		10	10	10	9			
10	1		10	9	9	7			
	2		10	10	8	7			
	3		10	8	8	8			
	4		10	10	9	6			
20	1		9	9	9	8			
	2		10	10	10	8			
	3		10	9	9	8			
	4		8	8**	8	8			
30	1		7	6	6	4			
	2		7	7	7	7			
	3		10	8	7	6			
	4		9	4	3	3			
40	1		8	5	4	3			
	2		7	5	2	2			
	3		9	8	5	3			
	4		8	6	5	4			
50	1		3	1	1	1			
	2		8	3	2	2			
	3		2*	0	0	0			
	4		4	1	1	0			
Date			03/21/06	03/22/06	03/23/06	03/24/06			
Time			1110	910	935	1535			
Initials			JW	GZ	TS	JM/GZ			

\* Replicate understocked (only 5 fish added at start of test)

\*\* Survival miscounted



## 4-Day Acute Toxicity Test

**Table B3. Acute *Atherinops affinis* (20 Mar 2006)**

Client	Marine Research Specialists
Project:	Hydrogen Sulfide
Client Sample ID:	N/A
Weston Test ID:	P060103.28
Species:	<i>Atherinops affinis</i>

Date Received:	N/A
Date Test Started:	20-Mar-06
Date Test Ended:	24-Mar-06
Study Director:	Brian Hester
# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H2S
<b>Day 0</b>  Date: 3/20/06  Time: 1650  Technician: JM	Control		1		7.5		19.7		31		7.69	4	0	4	0.51
			2		7.5		20.4		31		7.83	0	0	0	0.00
			3		7.3		20.4		31		7.82	4	0	4	0.38
			4		7.4		20.3		31		7.84	3	0	3	0.28
	10		1		7.6		20.4		30		7.91	151	0	151	12.00
			2		7.6		19.5		30		7.84	173	0	173	16.42
			3		7.5		20.4		30		7.91	176	0	176	13.98
			4		7.5		20.5		30		7.91	178	0	178	14.09
	20		1		7.6		20.7		30		7.96	346	0	346	24.45
			2		7.6		19.6		30		7.95	359	0	359	26.89
			3		7.5		20.6		30		7.97	369	0	369	25.61
			4		7.5		20.8		30		7.97	372	0	372	25.65
	30		1		7.5		20.9		30		7.99	577	0	577	37.97
			2		7.5		19.8		30		7.98	568	0	568	39.63
			3		7.5		21.0		30		7.94	601	0	601	43.90
			4		7.4		20.9		30		7.97	591	0	591	40.61
	40		1		7.4		20.8		30		8.00	740	0	740	47.82
			2		7.6		19.9		30		8.00	793	0	793	52.81
			3		7.4		20.9		30		8.02	820	0	820	50.57
			4		7.4		20.7		30		8.03	741	0	741	45.02
	50		1		7.4		20.6		30		8.06	246	1:5	1230	70.24
			2		7.5		19.9		30		8.03	263	1:5	1315	82.07
			3		7.5		20.7		30		8.08	251	1:5	1255	68.38
			4		7.4		21.0		30		8.08	250	1:5	1250	67.42
<b>Day 1</b>  <b>AM</b> Date: 3/21/06 Time: 0900 Technician: BH	Control				7.1		20.0		31		7.81	2	0	2	0.20
	10				7.2		20.0		30		7.90	145	0	145	11.92
	20				7.3		20.2		30		7.93	350	0	350	26.82
	30				7.2		20.5		30		7.98	584	0	584	39.80
	40				7.2		20.3		30		8.02	850	0	850	53.49
	50				7.2		20.1		30		8.04	235	1:5	1175	71.28
<b>Day 1</b>  <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	10				ND		ND		ND		ND	ND	ND	ND	ND
	20				ND		ND		ND		ND	ND	ND	ND	ND
	30				ND		ND		ND		ND	ND	ND	ND	ND
	40				ND		ND		ND		ND	ND	ND	ND	ND
	50				ND		ND		ND		ND	ND	ND	ND	ND

ND = No data; replicate previously terminated



## 4-Day Acute Toxicity Test

**Table B3. Acute *Atherinops affinis* (20 Mar 2006)**

Weston Test ID:	P060103.28	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H <sub>2</sub> S
<b>Day 2</b> <b>AM</b> Date: 3/22/06 Time: 0841 Technician: TS/GZ Feed Time: 0820	Control		3		7.2		20.1		31		7.67	1	0	1	0.13
	10				7.2		20.5		30		7.79	156	0	156	15.91
	20				7.1		20.6		30		7.81	285	0	285	27.78
	30				7.0		20.8		30		7.85	486	0	486	43.27
	40				7.0		20.9		30		7.88	715	0	715	59.56
	50				7.1		20.8		30		7.92	217	1:5	1085	83.27
<b>Day 2</b> <b>PM</b> Date: Time: Technician:	Control		4		ND		ND		ND		ND	ND	ND	ND	ND
	10				ND		ND		ND		ND	ND	ND	ND	ND
	20				ND		ND		ND		ND	ND	ND	ND	ND
	30				ND		ND		ND		ND	ND	ND	ND	ND
	40				ND		ND		ND		ND	ND	ND	ND	ND
	50				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 3</b> <b>AM</b> Date: 3/23/06 Time: 0845 Technician: GZ	Control		1		7.2		20.5		31		7.69	1	0	1	0.12
	10				7.1		20.9		30		7.80	53	0	53	5.22
	20				7.0		20.9		30		7.81	145	0	145	14
	30				6.9		21.0		30		7.83	278	0	278	25.65
	40				6.9		21.0		30		7.86	505	0	505	43.74
	50				6.8		21.0		30		7.89	144	1:5	720	58.52
<b>Day 3</b> <b>PM</b> Date: Time: Technician:	Control		2		ND		ND		ND		ND	ND	ND	ND	ND
	10				ND		ND		ND		ND	ND	ND	ND	ND
	20				ND		ND		ND		ND	ND	ND	ND	ND
	30				ND		ND		ND		ND	ND	ND	ND	ND
	40				ND		ND		ND		ND	ND	ND	ND	ND
	50				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 4</b>  Date: 3/24/06  Time: 0850  Technician: GZ/JM	Control		1		7.6		20.2		30		7.67	4	0	4	0.53
			2		7.3		20.7		30		7.79	6	0	6	0.61
			3		7.4		20.9		30		7.74	6	0	6	0.67
			4		7.4		21.0		30		7.74	8	0	8	0.89
	10		1		7.3		21.0		29		7.76	97	0	97	10.4
			2		7.3		21.0		29		7.77	79	0	79	8.3
			3		7.3		21.0		29		7.77	76	0	76	7.98
			4		7.3		21.0		29		7.77	87	0	87	9.14
	20		1		7.2		21.0		29		7.79	186	0	186	18.75
			2		7.2		20.9		29		7.78	172	0	172	17.76
			3		7.2		20.8		29		7.79	198	0	198	20.08
			4		7.3		20.9		29		7.79	199	0	199	20.12
	30		1		7.2		20.9		29		7.81	305	0	305	29.58
			2		7.3		20.9		29		7.81	293	0	293	28.41
			3		7.4		21.0		29		7.80	336	0	336	33.16
			4		7.2		21.0		29		7.80	325	0	325	32.08
	40		1		7.2		20.9		29		7.83	501	0	501	46.59
			2		7.1		20.8		29		7.84	492	0	492	44.94
			3		7.1		21.0		29		7.82	476	0	476	45.06
			4		7.1		20.9		29		7.83	436	0	436	40.54
	50		1		7.0		20.8		29		7.84	140	1:5	700	63.94
			2		7.0		20.9		29		7.85	134	1:5	670	59.73
			3		6.9		21		29		7.86	154	1:5	770	67
			4		6.9		21.1		29		7.85	146	1:5	730	64.66

ND = No data; replicate previously terminated

**Table B4. Acute *Atherinops affinis* (21 Mar 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
Control	1	0.1	0.1						0.4	<b>0.2</b>	100	100	100	<b>100</b>
	2						0.0		0.0	<b>0.0</b>	100	100	100	<b>100</b>
	3				0.0				0.1	<b>0.1</b>	100	100	100	<b>100</b>
	4								0.0	<b>0.0</b>	100	100	100	<b>100</b>
10	1	15.1	17.6						5.5	<b>12.7</b>	100	100	100	<b>100</b>
	2						8.5		7.2	<b>7.9</b>	100	100	100	<b>100</b>
	3				13.7				6.2	<b>10.0</b>	100	100	100	<b>100</b>
	4								7.2	<b>7.2</b>	ND	ND	ND	<b>ND</b>
20	1	26.0	44.3						16.5	<b>28.9</b>	90	90	90	<b>90</b>
	2						17.3		15.5	<b>16.4</b>	90	90	90	<b>90</b>
	3				30.4				15.8	<b>23.1</b>	100	100	100	<b>90</b>
	4								16.8	<b>16.8</b>	ND	ND	ND	<b>ND</b>
30	1	37.1	50.6						31.2	<b>39.6</b>	70	60	50	<b>50</b>
	2						32.4		26.7	<b>29.6</b>	70	40	40	<b>40</b>
	3				49.4				26.7	<b>38.0</b>	70	50	50	<b>50</b>
	4								30.9	<b>30.9</b>	ND	ND	ND	<b>ND</b>
40	1	41.5	62.6						31.3	<b>45.1</b>	40	40	40	<b>40</b>
	2						47.3		37.7	<b>42.5</b>	70	70	70	<b>60</b>
	3				59.4				40.2	<b>49.8</b>	70	70	50	<b>40</b>
	4								38.5	<b>38.5</b>	ND	ND	ND	<b>ND</b>
50	1	57.4	88.2						70.1	<b>71.9</b>	50	40	40	<b>30</b>
	2						60.6		67.4	<b>64.0</b>	20	0	T	<b>T</b>
	3				81.4				63.0	<b>72.2</b>	0	T	T	<b>T</b>
	4								70.0	<b>70.0</b>	ND	ND	ND	<b>ND</b>

T = Test terminated due to 0% survival

Acute Fish Test-96 Hr Survival				
Start Date:	3/21/2006	Test ID:	P060103.29	Sample ID:
End Date:	3/25/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPAA 91-EPA/600/4-90/027F	Test Species:
Comments:	Average			
	Antherinops affinis			


Conc-ug/L	1	2	3
Control	1.0000	1.0000	1.0000
9.5	1.0000	1.0000	1.0000
21.3	0.9000	0.9000	0.9000
34.5	0.5000	0.4000	0.5000
44	0.4000	0.6000	0.4000
69.5	0.3000	0.0000	0.0000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%						
Control	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	3				0	30
9.5	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	3	0.000	2.500	0.2296	0	30
21.3	0.9000	0.9000	1.2490	1.2490	1.2490	0.000	3	1.774	2.500	0.2296	3	30
*34.5	0.4667	0.4667	0.7518	0.6847	0.7854	7.731	3	7.187	2.500	0.2296	16	30
*44	0.4667	0.4667	0.7518	0.6847	0.8861	15.463	3	7.187	2.500	0.2296	16	30
*69.5	0.1000	0.1000	0.2991	0.1588	0.5796	81.247	3	12.116	2.500	0.2296	27	30

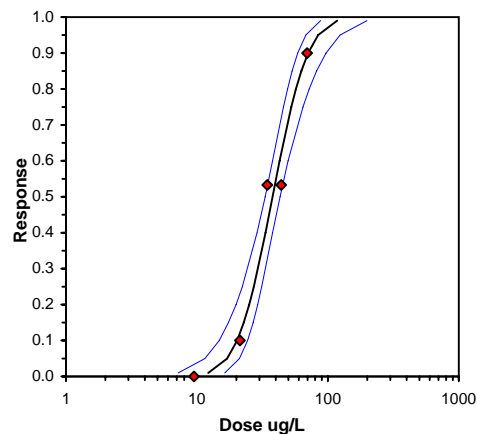
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.8144589	0.858	1.4227025	4.1176815
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	21.3	34.5	27.108117		0.1184307	0.1214674	0.6080664	0.0126558	1.6E-07	5, 12

Treatments vs Control

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	4.72763	0.7462803	3.2649206	6.1903395	0	2.6240579	7.8147278	0.45	1.5779789	0.2115225	3
Intercept	-2.4601	1.1849951	-4.782691	-0.13751							
TSCR						1.0					

Point	Probits	ug/L	95% Fiducial Limits	
EC01	2.674	12.187203	7.1795034	16.272082
EC05	3.355	16.984602	11.517307	21.135319
EC10	3.718	20.272249	14.774781	24.366976
EC15	3.964	22.842832	17.441121	26.87952
EC20	4.158	25.116383	19.860649	29.116628
EC25	4.326	27.246402	22.15773	31.246261
EC40	4.747	33.449551	28.799441	37.840919
EC50	5.000	37.84242	33.235233	43.079427
EC60	5.253	42.812197	37.843923	49.704469
EC75	5.674	52.559186	45.841547	64.588425
EC80	5.842	57.01652	49.197054	72.05493
EC85	6.036	62.691385	53.293905	82.047225
EC90	6.282	70.640834	58.791324	96.85037
EC95	6.645	84.314521	67.782918	124.23879
EC99	7.326	117.50432	88.043809	199.29716







## 4-Day Acute Toxicity Test

**Table B5. Acute Atherinops affinis (21 Mar 2006)**

Weston Test ID: P060103.29	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
10	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		0*	0*	0*	0*			
20	1		9	9	9	9			
	2		9	9	9	9			
	3		10	10	10	9			
	4		0*	0*	0*	0*			
30	1		7	6	5	5			
	2		7	4	4	4			
	3		7	5	5	5			
	4		0*	0*	0*	0*			
40	1		4	4	4	4			
	2		7	7	7	6			
	3		7	7	5	4			
	4		0	0	0	0			
50	1		5	4	4	3			
	2		2	0	0	0			
	3		0	0	0	0			
	4		0*	0*	0*	0*			
Date			03/22/06	03/23/06	03/24/06	03/25/06			
Time			840	935	1000	1540			
Initials			GZ	GZ	GZ	GZ			

\* appears no fish added



## 4-Day Acute Toxicity Test

**Table B6. Acute *Atherinops affinis* (21 Mar 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen Sulfide	Date Test Started:	21-Mar-06
Client Sample ID:	N/A	Date Test Ended:	25-Mar-06
Weston Test ID:	P060103.29	Study Director:	Brian Hester
Species:	<i>Atherinops affinis</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H2S
<b>Day 0</b>  Date: 3/21/06  Time: 1700  Technician: JW	Control		1		7.3		19.8		32		7.87	1	0	1	0.09
			2		ND		ND		ND		ND	ND	ND	ND	ND
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	10		1		7.3		19.8		30		7.94	198	0	198	15.05
			2		ND		ND		ND		ND	ND	ND	ND	ND
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	20		1		7.3		20.3		30		8.01	404	0	404	25.98
			2		ND		ND		ND		ND	ND	ND	ND	ND
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	30		1		7.3		20.4		30		8.05	632	0	632	37.14
			2		ND		ND		ND		ND	ND	ND	ND	ND
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	40		1		7.3		20.3		30		8.07	736	0	736	41.54
			2		ND		ND		ND		ND	ND	ND	ND	ND
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	50		1		7.3		20.2		30		8.12	226	1:5	1130	57.37
			2		ND		ND		ND		ND	ND	ND	ND	ND
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 1</b> <b>AM</b> Date: 3/22/06 Time: 0820 Technician: GZ/TS	Control				7.0		20.1		31		7.69	1	0	1	0.13
	10				7.0		20.6		30		7.79	173	0	173	17.58
	20				7.0		20.9		30		7.82	469	0	469	44.34
	30				7.2		20.8		30		7.87	593	0	593	50.62
	40				7.1		20.7		30		7.92	813	0	813	62.60
	50				7.0		20.7		30		7.93	234	1:5	1170	88.18
<b>Day 1</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	10				ND		ND		ND		ND	ND	ND	ND	ND
	20				ND		ND		ND		ND	ND	ND	ND	ND
	30				ND		ND		ND		ND	ND	ND	ND	ND
	40				ND		ND		ND		ND	ND	ND	ND	ND
	50				ND		ND		ND		ND	ND	ND	ND	ND

ND = No data; replicate previously terminated



## 4-Day Acute Toxicity Test

**Table B6. Acute *Atherinops affinis* (21 Mar 2006)**

Weston Test ID:	P060103.29	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H2S
<b>Day 2</b> <b>AM</b> Date: 3/23/06 Time: 0915 Technician: TS/GZ Feed Time: 0815	Control				6.6		19.1		31		7.56	0	0	0	0
	10				6.9		19.5		30		7.66	100	0	100	13.73
	20				7.0		19.5		30		7.75	266	0	266	30.43
	30				7.1		19.7		30		7.79	472	0	472	49.39
	40				6.8		19.6		30		7.83	615	0	615	59.4
	50				6.9		19.7		30		7.89	192	1:5	960	81.44
<b>Day 2</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	10				ND		ND		ND		ND	ND	ND	ND	ND
	20				ND		ND		ND		ND	ND	ND	ND	ND
	30				ND		ND		ND		ND	ND	ND	ND	ND
	40				ND		ND		ND		ND	ND	ND	ND	ND
	50				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 3</b> <b>AM</b> Date: 3/24/06 Time: 0832 Technician: GZ/JM	Control				6.6		19.5		31		7.65	0	0	0	0
	10				6.8		19.6		30		7.74	73	0	73	8.5
	20				6.9		19.5		30		7.79	164	0	164	17.27
	30				6.8		19.5		30		7.81	321	0	321	32.43
	40				6.8		19.1		30		7.79	443	0	443	47.27
	50				7.0		19.5		30		7.87	136	1:5	680	60.58
<b>Day 3</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	10				ND		ND		ND		ND	ND	ND	ND	ND
	20				ND		ND		ND		ND	ND	ND	ND	ND
	30				ND		ND		ND		ND	ND	ND	ND	ND
	40				ND		ND		ND		ND	ND	ND	ND	ND
	50				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 4</b>  Date: 3/25/06  Time: 1045  Technician: GZ	Control		1		7.1		18.5		31		7.67	3	0	3	0.42
			2		7.1		18.8		31		7.78	0	0	0	0
			3		7.1		18.5		31		7.78	1	0	1	0.11
			4		7.1		19.0		31		7.79	0	0	0	0
	10		1		6.8		19.2		30		7.82	55	0	55	5.5
			2		6.9		19.1		30		7.84	75	0	75	7.21
			3		6.5		19.1		30		7.78	57	0	57	6.21
			4		7.2		18.9		30		7.88	81	0	81	7.21
	20		1		6.6		19.2		30		7.88	187	0	187	16.47
			2		7.1		19.0		30		7.88	175	0	175	15.52
			3		6.7		19.0		30		7.82	157	0	157	15.79
			4		6.6		19.1		30		7.87	186	0	186	16.79
	30		1		6.7		19.2		30		7.88	354	0	354	31.19
			2		6.7		19.2		30		7.90	316	0	316	26.69
			3		6.7		19.1		30		7.90	315	0	315	26.69
			4		6.7		18.8		30		7.89	353	0	353	30.86
	40		1		6.4		18.7		30		7.93	388	0	388	31.26
			2		6.6		18.8		30		7.94	479	0	479	37.66
			3		6.3		18.7		30		7.92	489	0	489	40.25
			4		6.4		19.0		30		7.84	399	0	399	38.49
	50		1		6.3		19		30		7.90	165	1:5	825	70.14
			2		6.3		19.1		30		7.92	166	1:5	830	67.4
			3		6.4		19.1		30		7.94	162	1:5	810	63.04
			4		6.4		18.8		30		7.95	182	1:5	910	70.03

ND = No data; replicate previously terminated

**Table C1. Acute *Cyprinodon variegatus* Test 1 (10 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	0.0	0.3				0.5		0.0	<b>0.3</b>	ND	95	95	<b>95</b>
	2	0.0		0.2				0.5	0.0	<b>0.2</b>	ND	100	95	<b>95</b>
	3	0.0			0.1				0.0	<b>0.1</b>	ND	100	100	<b>100</b>
	4	0.0				0.2			0.0	<b>0.1</b>	ND	100	100	<b>100</b>
<b>2.5</b>	1	1.7	2.1				2.5		2.2	<b>2.1</b>	ND	95	95	<b>95</b>
	2	1.4		5.5				1.8	3.7	<b>2.9</b>	ND	100	100	<b>100</b>
	3	1.7			6.4				4.1	<b>4.1</b>	ND	100	100	<b>100</b>
	4	1.3				1.5			3.6	<b>1.4</b>	ND	100	100	<b>100</b>
<b>5</b>	1	6.5	7.8				16.7		12.4	<b>10.3</b>	ND	100	100	<b>100</b>
	2	6.9		9.9				8.8	10.4	<b>8.6</b>	ND	100	100	<b>100</b>
	3	6.8			52.7				12.2	<b>29.8</b>	ND	100	100	<b>100</b>
	4	6.5				4.9			10.8	<b>5.7</b>	ND	100	100	<b>100</b>
<b>10</b>	1	13.2	12.3				44.4		27.7	<b>23.3</b>	ND	100	100	<b>100</b>
	2	14.3		20.8				15.6	25.6	<b>16.9</b>	ND	100	100	<b>100</b>
	3	16.3			9.4				26.0	<b>12.9</b>	ND	100	100	<b>100</b>
	4	15.3				10.9			24.1	<b>13.1</b>	ND	100	100	<b>100</b>
<b>15</b>	1	23.0	22.3				36.1		51.7	<b>27.2</b>	ND	95	95	<b>95</b>
	2	22.1		146*				30.2	50.2	<b>26.2</b>	ND	100	100	<b>100</b>
	3	20.2			32.5				49.5	<b>26.4</b>	ND	100	100	<b>95</b>
	4	21.7				22.1			46.7	<b>21.9</b>	ND	95	90	<b>90</b>
<b>20</b>	1	44.3	35.5				68.6		52.5	<b>49.4</b>	ND	100	100	<b>100</b>
	2	42.0		56.4				41.7	62.0	<b>46.7</b>	ND	95	95	<b>95</b>
	3	43.8			12.6				61.2	<b>28.2</b>	ND	100	100	<b>100</b>
	4	41.7				30.6			68.3	<b>36.1</b>	ND	95	95	<b>95</b>

ND = No data

\* Sulfide concentrations decreased during test

Acute Fish Test-96 Hr Survival				
Start Date:	2/10/2006	Test ID:	P060103.12	Sample ID:
End Date:	2/14/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPAA 91-EPA/600/4-90/027F	Test Species:
Comments:	Average			
	CV-Cyprinodon variegatus			

Conc-ug/L	1	2	3	4
Control	0.9500	0.9500	1.0000	1.0000
1.625	0.9500	1.0000	1.0000	1.0000
13.6	1.0000	1.0000	1.0000	1.0000
16.55	1.0000	1.0000	1.0000	1.0000
25.4	0.9500	1.0000	0.9500	0.9000
40.1	1.0000	0.9500	1.0000	0.9500

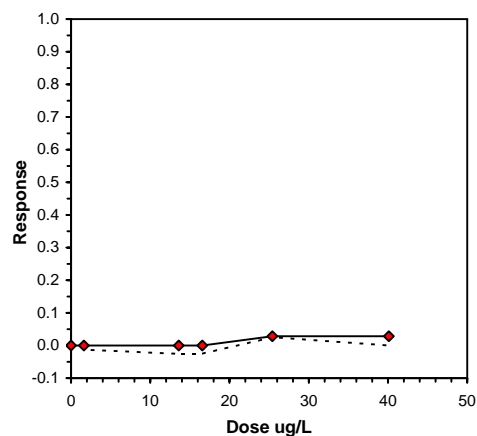
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	Rank Sum	1-Tailed Critical	Isotonic	
			Mean	Min	Max	CV%				Mean	N-Mean
Control	0.9750	1.0000	1.4020	1.3453	1.4588	4.673	4			0.9906	1.0000
1.625	0.9875	1.0128	1.4304	1.3453	1.4588	3.967	4	20.00	10.00	0.9906	1.0000
13.6	1.0000	1.0256	1.4588	1.4588	1.4588	0.000	4	22.00	10.00	0.9906	1.0000
16.55	1.0000	1.0256	1.4588	1.4588	1.4588	0.000	4	22.00	10.00	0.9906	1.0000
25.4	0.9500	0.9744	1.3496	1.2490	1.4588	6.354	4	15.00	10.00	0.9625	0.9716
40.1	0.9750	1.0000	1.4020	1.3453	1.4588	4.673	4	18.00	10.00	0.9625	0.9716

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.9380057	0.884	-0.100104	0.0149096
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	40.1	>40.1		

Treatments vs Control

Point	ug/L	SD	Linear Interpolation (200 Resamples)	
			95% CL(Exp)	Skew
IC05	>40.1			
IC10	>40.1			
IC15	>40.1			
IC20	>40.1			
IC25	>40.1			
IC40	>40.1			
IC50	>40.1			





## 4-Day Acute Toxicity Test

**Table C2. Acute *Cyprinodon variegatus*, Test 1 (10 Feb 2006)**

Weston Test ID: P060103.12	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		ND	9	9	9			
	2		ND	10	9	9			
	3		ND	10	10	10			
	4		ND	10	10	10			
2.5	1		ND	0	0	0			
	2		ND	7	9	9			
	3		ND	10	10	10			
	4		ND	10	10	10			
5	1		ND	8	8	8			
	2		ND	10	10	10			
	3		ND	1	1	1			
	4		ND	9	9	9			
10	1		ND	10	10	10			
	2		ND	8	8	8			
	3		ND	2	2	2			
	4		ND	1	1	1			
15	1		ND	0	1	1			
	2		ND	5	5	5			
	3		ND	0	0	0			
	4		ND	0	0	0			
20	1		ND	10	10	10			
	2		ND	2	2	2			
	3		ND	2	2	2			
	4		ND	7	7	7			
Date			02/11/06	02/12/06	02/13/06	02/14/06			
Time			1500	1600	1215	1110			
Initials			GZ	GZ/AM	GZ/AM	JW			

\* Organisms from Tests 1 and 1A were combined as one test for analyses  
 ND = No data



## 4-Day Acute Toxicity Test

**Table C2. Acute *Cyprinodon variegatus*, Test 1A (10 Feb 2006)**

Weston Test ID: P060103.11	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		ND	10	10	10			
	2		ND	10	10	10			
	3		ND	10	10	10			
	4		ND	10	10	10			
2.5	1		ND	19	19	19			
	2		ND	13	11	11			
	3		ND	10	10	10			
	4		ND	10	10	10			
5	1		ND	12	12	12			
	2		ND	10	10	10			
	3		ND	19	19	19			
	4		ND	11	11	11			
10	1		ND	10	10	10			
	2		ND	12	12	12			
	3		ND	18	18	18			
	4		ND	19	19	19			
15	1		ND	19	18	18			
	2		ND	15	15	15			
	3		ND	20	20	19			
	4		ND	19	18	18			
20	1		ND	10	10	10			
	2		ND	17	17	17			
	3		ND	18	18	18			
	4		ND	12	12	12			
Date			02/11/06	02/12/06	02/13/06	02/14/06			
Time			1500	1600	1215	1110			
Initials			GZ	GZ/AM	GZ/AM	JW			

\* Organisms from Tests 1 and 1A were combined as one test for analyses

ND = No data



## 4-Day Acute Toxicity Test

**Table C3. Acute *Cyprinodon variegatus*, Test 1 (10 Feb 2006)**

Client	Marine Research Specialists
Project:	Hydrogen Sulfide
Client Sample ID:	N/A
Weston Test ID:	P060103.12
Species:	<i>Cyprinodon variegatus</i>

Date Received:	N/A
Date Test Started:	10-Feb-06
Date Test Ended:	14-Feb-06
Study Director:	Brian Hester
# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/10/06  Time: 1053  Technician: CC/JW/GZ	Control		1		8.1		19.2		30		8.05	0	0	0
			2		8.1		19.2		30		8.07	0	0	0
			3		8.1		19.3		31		8.06	0	0	0
			4		8.1		19.4		30		8.07	0	0	0
	2.5		1		7.9		19.4		28		8.14	44	0	44
			2		7.9		19.3		28		8.13	35	0	35
			3		7.9		19.3		27		8.14	49	0	49
			4		7.9		19.5		28		8.13	33	0	33
	5		1		7.8		19.4		28		8.16	149	0	149
			2		7.9		19.3		28		8.17	187	0	187
			3		7.8		19.5		28		8.16	144	0	144
			4		7.9		19.4		28		8.15	146	0	146
	10		1		7.8		19.4		29		8.19	333	0	333
			2		7.8		19.3		27		8.21	369	0	369
			3		7.9		19.4		27		8.21	416	0	416
			4		7.8		19.4		28		8.22	394	0	394
	15		1		7.8		19.3		27		8.22	598	0	598
			2		7.8		19.3		28		8.26	633	0	633
			3		7.8		19.4		28		8.25	547	0	547
			4		7.8		19.5		28		8.22	532	0	532
	20		1		7.7		19.3		27		8.28	250	1/5	1250
			2		7.8		19.3		28		8.25	231	1/5	1155
			3		7.8		19.4		27		8.27	250	1/5	1250
			4		7.8		19.3		28		8.27	236	1/5	1180
<b>Day 1</b> <b>AM</b> Date: 2/11/06 Time: 0950 Technician: GZ/CC	Control		1		7.6		19.2		30		7.87	6	0	6
	2.5				7.8		19.0		26		7.98	33	0	33
	5				7.7		18.8		26		8.02	131	0	131
	10				7.7		19.0		26		8.01	38	1/5	190
	15				7.7		19.1		26		8.18	101	1/5	505
	20				7.6		19.2		27		8.21	167	1/5	835
<b>Day 1</b> <b>PM</b> Date: 2/11/06 Time: 1625 Technician: GZ	Control		2		8.0		18.8		31		7.98	5	0	5
	2.5				7.9		19.0		28		7.90	100	0	100
	5				7.8		19.1		28		8.14	242	0	242
	10				7.8		19.0		28		8.17	499	0	499
	15				7.8		19.2		28		8.22	772	1/5	3860
	20				7.7		19.2		28		8.22	299	1/5	1495





## 4-Day Acute Toxicity Test

**Table C3. Acute *Cyprinodon variegatus*, Test 1 (10 Feb 2006)**

Weston Test ID:	P060103.12	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 2/12/06 Time: 1100 Technician: CC/AM Feed Time: 1000	Control		3		7.7		19.2		30		7.93	4	0	4
	2.5				7.5		19.6		30		7.46	1	0	1
	5				7.1		19.7		28		8.02	776	0	776
	10				7.6		19.7		30		7.96	0	0	0
	15				7.7		19.7		30		7.98	0	1/5	0
	20				7.6		19.6		30		8.03	4	1/5	20
<b>Day 2</b> <b>PM</b> Date: 2/12/06 Time: 1530 Technician: CC	Control		4		7.6		19.5		29		8.02	3	0	3
	2.5				7.5		19.8		28		8.08	40	0	40
	5				7.6		19.9		28		8.11	27	1/5	135
	10				7.4		19.9		28		8.16	77	1/5	385
	15				7.5		19.8		28		8.22	130	1/5	650
	20				7.3		19.8		28		8.21	162	1/5	810
<b>Day 3</b> <b>AM</b> Date: 2/13/06 Time: 0937 Technician: AM/TS	Control		1		7.6		18.9		30		7.62	4	0	4
	2.5				7.6		19.6		27		7.72	31	0	31
	5				7.3		19.7		27		7.73	43	1/5	215
	10				7.2		19.8		27		7.72	85	1/5	425
	15				7.0		19.8		27		7.80	129	1/5	645
	20				7.1		19.7		27		7.84	194	1/5	970
<b>Day 3</b> <b>PM</b> Date: 2/13/06 Time: 1415 Technician: AM/GZ	Control		2		7.7		19.1		29		7.99	8	0	8
	2.5				7.7		19.5		28		8.11	35	0	35
	5				7.7		19.7		27		8.10	163	0	163
	10				7.8		19.7		27		8.13	308	0	308
	15				7.7		19.6		27		8.19	136	1/5	680
	20				7.6		19.6		27		8.21	196	1/5	980
<b>Day 4</b>  Date: 2/14/06  Time: 1030  Technician: GZ	Control		1		7.5		19.6		30		7.86	0	0	0
			2		7.5		19.7		30		7.84	0	0	0
			3		7.4		19.8		30		7.85	0	0	0
			4		7.4		19.8		30		7.87	0	0	0
	2.5		1		7.6		19.8		27		7.90	26	0	26
			2		7.3		19.8		27		7.89	43	0	43
			3		7.3		19.8		27		7.89	48	0	48
			4		7.4		19.8		27		7.90	43	0	43
	5		1		7.3		19.7		27		7.90	149	0	149
			2		7.4		19.7		27		7.89	122	0	122
			3		7.5		19.8		27		7.91	149	0	149
			4		7.5		19.8		27		7.90	129	0	129
	10		1		7.3		19.8		27		7.95	369	0	369
			2		7.4		19.8		27		7.92	320	0	320
			3		7.2		19.7		27		7.94	343	0	340
			4		7.2		19.8		27		7.97	336	0	336
	15		1		7.1		19.6		27		7.96	141	1/5	705
			2		7.2		19.6		27		8.00	149	1/5	745
			3		7.2		19.7		27		7.99	144	1/5	720
			4		7.1		19.7		27		7.97	130	1/5	650
	20		1		7.1		19.6		27		8.00	156	1/5	780
			2		7.0		19.7		27		8.00	184	1/5	920
			3		7.0		19.7		27		7.98	174	1/5	870
			4		7.1		19.8		27		7.92	171	1/5	855



## 4-Day Acute Toxicity Test

**Table C3. Acute *Cyprinodon variegatus*, Test 1A (10 Feb 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen sulfide	Date Test Started:	10-Feb-06
Client Sample ID:	N/A	Date Test Ended:	14-Feb-06
Weston Test ID:	P060103.11	Study Director:	Brian Hester
Species:	<i>Cyprinodon variegatus</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/10/06  Time: 1053  Technician: CC/JW/GZ	Control		1		8.2		19.0		30		8.04	0	0	0
			2		8.1		19.4		30		8.06	0	0	0
			3		8.1		19.2		31		8.04	0	0	0
			4		8.1		19.3		30		8.08	0	0	0
	2.5		1		7.9		19.3		28		8.13	24	0	24
			2		7.8		19.3		28		8.13	21	0	21
			3		7.9		19.3		27		8.13	19	0	19
			4		7.9		19.4		28		8.13	17	0	17
	5		1		7.8		19.4		28		8.15	123	0	123
			2		7.8		19.3		28		8.16	111	0	111
			3		7.9		19.4		28		8.17	149	0	149
			4		7.8		19.4		28		8.15	123	0	123
	10		1		7.8		19.4		29		8.20	273	0	273
			2		7.8		19.3		27		8.22	309	0	309
			3		7.8		19.3		27		8.21	352	0	352
			4		7.8		19.4		28		8.22	346	0	346
	15		1		7.9		19.2		27		8.25	544	0	544
			2		7.8		19.3		28		8.25	522	0	522
			3		7.8		19.4		28		8.23	476	0	476
			4		7.8		19.5		28		8.23	529	0	529
	20		1		7.7		19.3		27		8.27	231	1/5	1155
			2		7.7		19.3		28		8.26	207	1/5	1035
			3		7.7		19.3		27		8.28	225	1/5	1125
			4		7.7		19.3		28		8.26	209	1/5	1045
<b>Day 1</b> <b>AM</b> Date: 2/11/06 Time: 0950 Technician: GZ/CC	Control				7.1		18.7		30		8.04	1	0	1
	2.5				7.1		19.0		26		7.99	26	0	26
	5				6.2		18.6		26		8.02	109	0	109
	10				6.5		19.0		26		8.04	39	1/5	195
	15				7.2		18.8		26		8.21	101	1/5	505
	20				7.2		19.2		27		8.22	170	1/5	850
<b>Day 1</b> <b>PM</b> Date: 2/11/06 Time: 1625 Technician: GZ	Control				7.8		19.2		31		8.02	1	0	1
	2.5				7.9		19.0		28		8.09	50	0	50
	5				7.6		19.3		28		8.11	148	0	148
	10				7.6		19.3		28		8.16	390	0	390
	15				7.5		19.3		28		8.21	624	1/5	3120
	20				7.5		19.4		28		8.21	240	1/5	1200



## 4-Day Acute Toxicity Test

**Table C3. Acute *Cyprinodon variegatus*, Test 1A (10 Feb 2006)**

Weston Test ID:	P060103.11	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
Day 2	Control		3		7.5		18.8		30		7.99	3	0	3
AM	2.5				7.3		19.5		30		7.95	87	0	87
Date: 2/12/06	5				6.3		19.6		28		8.03	840	0	840
Time: 1100	10				7.5		19.6		30		7.96	130	0	130
Technician: CC/AM	15				7.4		19.6		30		7.94	86	1/5	430
Feed Time: 1000	20				7.3		19.6		30		8.08	45	1/5	225
Day 2	Control		4		7.5		19.1		29		7.98	3	0	3
PM	2.5				7.5		19.8		28		8.00	12	0	12
Date: 2/12/06	5				7.3		19.9		28		8.01	8	1/5	40
Time: 1530	10				6.8		19.9		28		8.08	13	1/5	65
Technician: CC	15				6.9		19.8		28		8.12	67	1/5	335
	20				6.4		19.8		28		8.01	82	1/5	410
Day 3	Control		1		7.5		19.0		30		7.66	3	0	3
AM	2.5				7.5		19.4		27		7.72	11	0	11
Date: 2/13/06	5				7.1		19.6		27		7.69	12	1/5	60
Time: 0937	10				6.8		19.7		27		7.70	59	1/5	295
Technician: AM/TS	15				6.2		19.8		27		7.74	10	1/5	50
	20				6.5		19.7		27		7.81	90	1/5	450
Day 3	Control		2		7.5		19.5		30		8.02	4	0	4
PM	2.5				7.5		19.6		27		8.07	18	0	18
Date: 2/13/06	5				7.5		19.8		27		8.09	43	0	43
Time: 1415	10				7.5		19.7		27		8.12	150	0	150
Technician: AM/GZ	15				7.0		19.6		27		8.12	50	1/5	250
	20				6.2		19.7		27		8.15	63	1/5	315
Day 4	Control		1		7.5		19.6		30		7.83	0	0	0
			2		7.4		19.8		30		7.85	0	0	0
			3		7.3		19.9		30		7.84	0	0	0
			4		7.4		19.8		30		7.87	0	0	0
	2.5		1		7.5		19.8		27		7.92	9	0	9
			2		7.2		19.8		27		7.89	6	0	6
			3		7.2		19.8		27		7.91	14	0	14
			4		7.4		19.8		27		7.88	24	0	24
	5		1		7.1		19.8		27		7.90	25	0	25
			2		7.4		19.7		27		7.89	43	0	43
			3		7.3		19.9		27		7.90	49	0	49
			4		7.2		19.8		27		7.89	26	0	26
	10		1		6.7		19.8		27		7.90	122	0	122
			2		7.0		19.9		27		7.93	91	0	91
			3		6.9		19.6		27		7.90	80	0	80
			4		6.8		19.8		27		7.95	16	0	16
	15		1		6.3		19.8		27		7.93	13	1/5	65
			2		6.4		19.7		27		7.97	17	1/5	85
			3		6.6		19.7		27		7.98	24	1/5	120
			4		6.4		19.7		27		7.93	42	1/5	210
20		1		6.3		19.7		27		7.98	17	1/5	85	
		2		6.2		19.7		27		7.97	16	1/5	80	
		3		6.0		19.7		27		7.94	21	1/5	105	
		4		6.2		19.8		27		7.92	22	1/5	110	

**Table C4. Acute *Cyprinodon variegatus*, Test 2 (15 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	0.2	1.4				0.1			<b>0.6</b>	100	100	80	<b>70</b>
	2	0.1		1.5				0.8		<b>0.8</b>	100	90	90	<b>90</b>
	3	0.2			0.6				0.0	<b>0.3</b>	100	100	100**	<b>100</b>
	4	0.2				0.2				<b>0.2</b>	100	100	70	<b>70</b>
<b>15</b>	1	26.0	47.2				19.6			<b>30.9</b>	100	90	70	<b>70</b>
	2	29.1		52.2				32.9		<b>38.1</b>	100	100	90	<b>80</b>
	3	34.0			54.4				29.7	<b>39.4</b>	100	100	100	<b>90</b>
	4	34.1				32.7				<b>33.4</b>	100	90	80	<b>70</b>
<b>25</b>	1	24.0	75.3				12.0			<b>37.1</b>	100	80	80	<b>80</b>
	2	28.3		67.7				70.6		<b>55.5</b>	100	100	80	<b>80</b>
	3	25.3			87.9				47.3	<b>53.5</b>	100	100	80	<b>80</b>
	4	29.0				37.7				<b>33.3</b>	100	90	90	<b>90</b>
<b>50</b>	1	31.5	132.4				39.9			<b>67.9</b>	70	70	70	<b>70</b>
	2	38.0		128.1				90.6		<b>85.6</b>	90	90	80	<b>80</b>
	3	42.9			122.3				49.8	<b>71.7</b>	80	80	80**	<b>80</b>
	4	40.9				62.3				<b>51.6</b>	100	100	100	<b>90</b>
<b>75</b>	1	84.5	205.1				33.4			<b>107.7</b>	100	80	50**	<b>50</b>
	2	74.7		216.3				147.2		<b>146.1</b>	100	100	80	<b>70</b>
	3	57.2			239.4				39.7	<b>112.1</b>	100	100	100	<b>90</b>
	4	97.9				111.5				<b>104.7</b>	100	100	90	<b>60</b>
<b>100</b>	1	80.3	214.0				50.7			<b>115.0</b>	100	90	90	<b>30</b>
	2	115.2		216.9				101.5		<b>144.5</b>	100	100	100	<b>80</b>
	3	52.5			231.1				49.4	<b>111.0</b>	100	100	80	<b>50</b>
	4	55.2				114.7				<b>85.0</b>	100	100	90	<b>80</b>

\*\* Survival miscounted

Acute Fish Test-96 Hr Survival				
Start Date:	2/15/2006	Test ID:	P060103.15	Sample ID:
End Date:	2/19/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPAA 91-EPA/600/4-90/027F	Test Species:
Comments:	Average			
	CV-Cyprinodon variegatus			

Conc-ug/L	1	2	3	4
Control	0.7000	0.9000	0.9000	0.7000
35.45	0.7000	0.8000	0.9000	0.7000
44.85	0.8000	0.8000	0.8000	0.9000
69.2	0.7000	0.8000	0.7000	0.9000
113.875	0.3000	0.8000	0.5000	0.8000
118.4	0.4000	0.7000	0.9000	0.6000

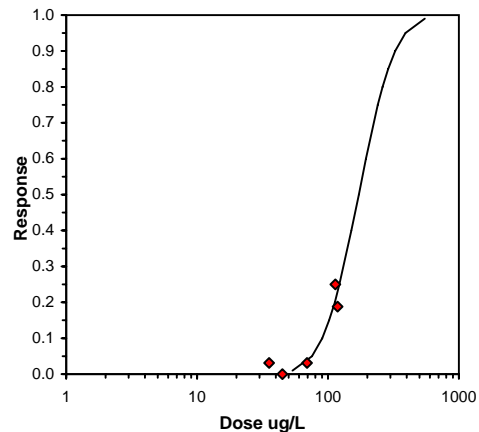
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%						
Control	0.8000	1.0000	1.1201	0.9912	1.2490	13.293	4				8	40
35.45	0.7750	0.9688	1.0846	0.9912	1.2490	11.294	4	0.290	2.410	0.2948	9	40
44.85	0.8250	1.0313	1.1426	1.1071	1.2490	6.209	4	-0.184	2.410	0.2948	7	40
69.2	0.7750	0.9688	1.0846	0.9912	1.2490	11.294	4	0.290	2.410	0.2948	9	40
113.875	0.6000	0.7500	0.8948	0.5796	1.1071	28.961	4	1.842	2.410	0.2948	16	40
118.4	0.6500	0.8125	0.9527	0.6847	1.2490	24.656	4	1.368	2.410	0.2948	14	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.9653639	0.884	-0.003288	-0.349468
Bartlett's Test indicates equal variances ( $p = 0.37$ )	5.4257598	15.086272		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	118.4	>118.4			0.2703985	0.3337181	0.0394846	0.0299259	0.3001582	5, 18

Treatments vs Control

Parameter	Value	SE	95% Fiducial Limits	Maximum Likelihood-Probit						
				Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	4.6047477	5.5427636	-6.259069 15.468564	0.2	0.7422063	7.8147278	0.86	2.2345338	0.2171672	13
Intercept	-5.289464	11.394598	-27.62288 17.043947							
TSCR	0.1992077	0.040882	0.119079 0.2793365							
Point	Probits	ug/L	95% Fiducial Limits							
EC01	2.674	53.620075								
EC05	3.355	75.392057								
EC10	3.718	90.411334								
EC15	3.964	102.20085								
EC20	4.158	112.65781								
EC25	4.326	122.47763								
EC40	4.747	151.18723								
EC50	5.000	171.60652								
EC60	5.253	194.78364								
EC75	5.674	240.44227								
EC80	5.842	261.40043								
EC85	6.036	288.14634								
EC90	6.282	325.72019								
EC95	6.645	390.60875								
EC99	7.326	549.21229								





## 4-Day Acute Toxicity Test

**Table C5. Acute *Cyprinodon variegatus*, Test 2 (15 Feb 2006)**

Weston Test ID: P060103.16	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	8	7			
	2		10	9	9	9			
	3		10	10	9	9			
	4		10	10	7	7			
15	1		10	9	7	7			
	2		10	10	9	8			
	3		10	10	10	9			
	4		10	9	8	7			
25	1		10	8	8	8			
	2		10	10	8	8			
	3		10	10	8	8			
	4		10	9	9	9			
50	1		7	7	7	7			
	2		9	9	8	8			
	3		8	8	7	7			
	4		10	10	10	9			
75	1		10	8	4	4			
	2		10	10	8	7			
	3		10	10	10	9			
	4		10	10	9	6			
100	1		10	9	9	3			
	2		10	10	10	8			
	3		10	10	8	5			
	4		10	10	9	8			
Date			2/16/06	2/17/06	2/18/06	2/19/06			
Time			1057	1710	1315	1110			
Initials			EB/JM	TS/JM/EB	TS/EB	TS/EB			



## 4-Day Acute Toxicity Test

**Table C6. Acute *Cyprinodon variegatus* , Test 2 (15 Feb 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen Sulfide	Date Test Started:	15-Feb-06
Client Sample ID:	NA	Date Test Ended:	19-Feb-06
Weston Test ID:	P060103.16	Study Director:	Brian Hester
Species:	<i>Cyprinodon variegatus</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/15/06  Time: 1030  Technician: GZ/TS	Control		1		8		19.4		29		8.04	3	0	3
			2		7.6		19.5		29		8.08	1	0	1
			3		7.5		19.4		29		8.03	3	0	3
			4		7.5		19.1		29		8.04	4	0	4
	15		1		7.6		18.9		26		8.24	130	1/5	650
			2		7.5		19.5		27		8.22	140	1/5	700
			3		7.3		19.5		25		8.19	151	1/5	755
			4		7.4		19.4		26		8.18	149	1/5	745
	25		1		7.4		19.4		26		8.24	120	1/5	600
			2		7.3		19.5		26		8.26	148	1/5	740
			3		7.3		19.4		26		8.28	138	1/5	690
			4		7.5		19.2		26		8.31	169	1/5	845
	50		1		7.3		19.4		26		8.42	235	1/5	1175
			2		7.3		19.4		26		8.39	265	1/5	1325
			3		7.4		19.3		26		8.38	293	1/5	1465
			4		7.4		19.3		26		8.40	292	1/5	1460
	75		1		7.2		19.4		25		8.55	168	1/25	4200
			2		7.3		19.2		26		8.54	146	1/25	3650
			3		7.3		19.3		26		8.56	117	1/25	2925
			4		7.3		19.4		26		8.51	179	1/25	4475
	100		1		7.3		19.3		26		8.66	206	1/25	5150
			2		7.3		19.3		26		8.65	289	1/25	7225
			3		7.3		19.1		26		8.62	123	1/25	3075
			4		7.3		19.3		26		8.67	145	1/25	3625
<b>Day 1</b> <b>AM</b> Date: 2/16/06 Time: 0950 Technician: JM/EB	Control				7.1		20.3		29		7.72	12	0	12
	15				6.9		20.4		27		7.95	126	1/5	630
	25				7.0		20.4		27		8.04	244	1/5	1220
	50				6.9		20.3		26		8.21	619	1/5	3095
	75				6.9		20.3		26		8.29	229	1/25	5725
	100				6.9		20.4		26		8.38	292	1/25	7300
<b>Day 1</b> <b>PM</b> Date: 2/16/06 Time: Technician:	Control				7.4		19.1		30		7.82	15	0	15
	15				7.4		19.3		28		7.93	134	1/5	670
	25				7.1		19.8		27		8.06	229	1/5	1145
	50				7.1		19.8		28		8.19	579	1/5	2895
	75				7.0		19.5		28		8.32	261	1/25	6525
	100				6.9		19.8		28		8.39	306	1/25	7650



## 4-Day Acute Toxicity Test

**Table C6. Acute *Cyprinodon variegatus* , Test 2 (15 Feb 2006)**

Weston Test ID:	P060103.16	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b>	Control		3	A	7.0		20.1		30		7.78	6	0	6
<b>AM</b>	15				6.8		20.4		28		7.91	134	1/5	670
Date: 2/17/06	25				6.4		20.4		28		7.92	221	1/5	1105
Time: 1100	50				6.6		20.4		29		8.10	456	1/5	2280
Technician: EB/JM	75				6.6		20.4		29		8.26	254	1/25	6350
	100				6.9		20.4		29		8.34	293	1/25	7325
<b>Day 2</b>	Control		4		7.1		18.7		30		7.78	2	0	2
<b>PM</b>	15				6.8		18.8		28		7.93	84	1/5	420
Date: 2/17/06	25				6.7		18.9		28		7.95	101	1/5	505
Time: 1623	50				6.5		18.9		28		8.15	258	1/5	1290
Technician: TS	75				6.5		18.8		28		8.28	123	1/25	3075
	100				6.3		18.9		29		8.38	159	1/25	3975
<b>Day 3</b>	Control		1		7.0		18.5		31		7.97	2	0	2
<b>AM</b>	15				6.3		18.5		29		8.06	334	0	334
Date: 2/18/06	25				6.3		18.7		28		8.04	39	1/5	195
Time: 1015	50				5.8		18.8		28		8.23	197	1/5	985
Technician: EB/TS	75				5.5		18.5		28		8.23	33	1/25	825
	100				2.0		18.5		29		8.50	92	1/25	2300
<b>Day 3</b>	Control		2		8.2		18.9		31		7.69	6	0	6
<b>PM</b>	15				7.7		18.5		29		7.81	66	1/5	330
Date: 2/18/06	25				6.0		18.5		28		7.88	163	1/5	815
Time: 1530	50				8.2		18.0		28		8.10	336	1/5	1680
Technician: TS/EB	75				6.1		18.0		28		8.20	136	1/25	3400
	100				7.9		18.0		29		8.42	154	1/25	3850
<b>Day 4</b>	Control		3		6.7		18.7		30		7.80	0	0	0
<b>AM</b>	15				5.0		18.6		28		7.89	70	1/5	350
Date: 2/19/06	25				5.9		18.7		29		8.01	145	1/5	725
Time: 0925	50				0.2		18.8		29		8.07	174	1/5	870
Technician: EB/TS	75				3.2		18.6		28		8.25	41	1/25	1025
	100				0.2		18.6		29		8.37	67	1/25	1675
<b>Day 4</b>	Control		4		6.6		18.0		31		7.70	0	0	0
<b>PM</b>	15				5.8		18.6		29		8.12	38	1/5	190
Date: 2/19/06	25				6.0		18.6		29		8.21	167	1/5	835
Time: 1530	50				6.6		18.5		30		8.28	241	1/5	1205
Technician: TS	75				5.3		18.6		29		8.45	56	1/25	1400
	100				6.9		18.6		29		8.56	134	1/25	3350



**Table C7. Acute *Cypridodon variegatus*, Test 3 (15 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	0.2	0.8				0.2			<b>0.4</b>	100	100	100	<b>100</b>
	2	0.2		2.3				0.7		<b>1.1</b>	100	90	70	<b>70</b>
	3	0.3			0.3				0.0	<b>0.2</b>	100	100	80	<b>80</b>
	4	0.3				0.4				<b>0.3</b>	100	100	100	<b>90</b>
<b>15</b>	1	24.3	47.3				4.3			<b>25.3</b>	80	90	90	<b>80</b>
	2	18.3		40.7				26.5		<b>28.5</b>	100	90	80	<b>80</b>
	3	32.7			31.0				1.0	<b>21.5</b>	100	90	90	<b>90</b>
	4	40.0				15.2				<b>27.6</b>	100	90	90	<b>90</b>
<b>25</b>	1	30.3	75.1				7.2			<b>37.5</b>	100	100	100	<b>90</b>
	2	34.5		79.1				47.9		<b>53.8</b>	100	90	70	<b>70</b>
	3	42.7			70.0				2.0	<b>38.2</b>	100	100	100	<b>100</b>
	4	32.2				19.9				<b>26.0</b>	100	70	60	<b>50</b>
<b>50</b>	1	36.0	131.3				7.3			<b>58.2</b>	90	90	80	<b>80</b>
	2	46.5		104.1				65.8		<b>72.1</b>	90	90	90	<b>90</b>
	3	44.1			96.5				41.6	<b>60.7</b>	90	90	90	<b>90</b>
	4	45.1				55.3				<b>50.2</b>	100	80	80	<b>80</b>
<b>75</b>	1	75.4	213.5				3.8			<b>97.6</b>	80	90	70	<b>60</b>
	2	62.3		213.3				128.1		<b>134.5</b>	100	90	80	<b>60</b>
	3	64.0			188.4				63.4	<b>105.3</b>	100	100	60	<b>60</b>
	4	66.7				87.9				<b>77.3</b>	90	90	70	<b>60</b>
<b>100</b>	1	86.2	241.4				19.3			<b>115.6</b>	90	90	30	<b>20</b>
	2	58.5		192.1				74.7		<b>108.4</b>	70	60	60	<b>60</b>
	3	73.9			207.5				39.4	<b>106.9</b>	100	100	80	<b>80</b>
	4	55.9				97.9				<b>76.9</b>	90	70**	70**	<b>70</b>

\*\* Survival miscounted

Acute Fish Test-96 Hr Survival				
Start Date:	2/15/2006 10:30	Test ID:	P060103.16	Sample ID:
End Date:	2/22/2006 11:10	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPAA 91-EPA/600/4-90/027F	Test Species:
Comments:	Average			
	CV-Cyprinodon variegatus			

Conc-ug/L	1	2	3	4
Control	1.0000	1.0000	1.0000	0.8000
25.7	0.9000	0.7000	0.9000	0.9000
38.9	1.0000	0.8000	0.8000	0.9000
60.3	0.7000	0.4000	0.8000	0.7000
102	0.6000	0.6000	0.4000	0.3000
103.7	0.4000	0.4000	0.0000	0.0000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%						
Control	0.9500	1.0000	1.3358	1.1071	1.4120	11.411	4				2	40
25.7	0.8500	0.8947	1.1846	0.9912	1.2490	10.885	4	1.145	2.410	0.3182	6	40
38.9	0.8750	0.9211	1.2188	1.1071	1.4120	11.906	4	0.886	2.410	0.3182	5	40
*60.3	0.6500	0.6842	0.9435	0.6847	1.1071	19.184	4	2.971	2.410	0.3182	14	40
*102	0.4750	0.5000	0.7591	0.5796	0.8861	20.120	4	4.367	2.410	0.3182	21	40
*103.7	0.2000	0.2105	0.4217	0.1588	0.6847	71.998	4	6.922	2.410	0.3182	32	40

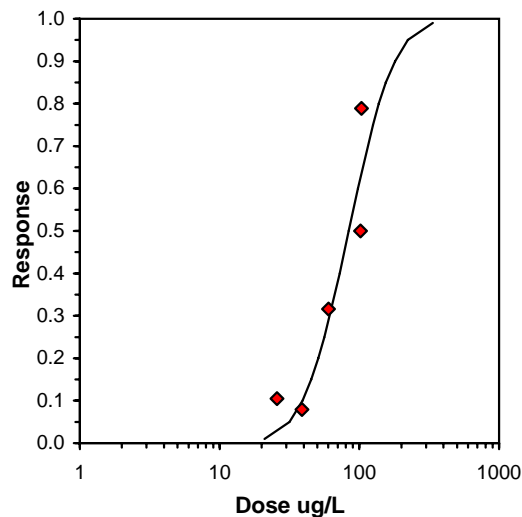
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.918433	0.884	-0.28886	-1.01471
Bartlett's Test indicates equal variances ( $p = 0.69$ )	3.03457	15.08627		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	38.9	60.3	48.43212		0.221858	0.234576	0.46976	0.03487	1.5E-05	5, 18
Treatments vs Control										

Parameter	Value	SE	95% Fiducial Limits	Maximum Likelihood-Probit							
				Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter	
Slope	3.87179	1.451667	-0.74806 8.491642	0.05	8.090409	7.814728	4.0E-02	1.923913	0.258278	11	
Intercept	-2.44899	2.76497	-11.2484 6.350383								
TSCR	0.074936	0.059659	-0.11493 0.264799								

Point	Probits	ug/L	95% Fiducial Limits
EC01	2.674	21.04104	
EC05	3.355	31.55604	
EC10	3.718	39.16657	
EC15	3.964	45.31316	
EC20	4.158	50.87918	
EC25	4.326	56.19615	
EC40	4.747	72.19027	
EC50	5.000	83.92914	
EC60	5.253	97.57687	
EC75	5.674	125.3484	
EC80	5.842	138.4476	
EC85	6.036	155.4538	
EC90	6.282	179.8498	
EC95	6.645	223.2251	
EC99	7.326	334.7792	

Significant heterogeneity detected ( $p = 4.00E-02$ )





## 4-Day Acute Toxicity Test

**Table C8. Acute *Cyprinodon variegatus* , Test 3 (15 Feb 2006)**

Weston Test ID: P060103.15	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10			
	2		10	9	7	7			
	3		10	10	8	8			
	4		10	10	10	9			
15	1		8	9	9	8			
	2		10	9	8	8			
	3		10	9	9	9			
	4		10	9	9	9			
25	1		10	10	10	9			
	2		10	9	7	7			
	3		10	10	10	10			
	4		10	7	6	5			
50	1		9	9	8	8			
	2		9	9	9	9			
	3		9	9	9	9			
	4		10	8	8	8			
75	1		8	9	7	6			
	2		10	9	8	6			
	3		10	10	6	6			
	4		9	9	7	6			
100	1		9	9	3	2			
	2		7	6	6	6			
	3		10	10	8	8			
	4		9	6	6	6			
Date			2/16/06	2/17/06	2/18/06	2/19/06			
Time			1057	1710	1315	1110			
Initials			EB/JM	TS/JM/EB	EB/TS	EB/TS			



## 4-Day Acute Toxicity Test

**Table C9. Acute *Cyprinodon variegatus*, Test 3 (15 Feb 2006)**

Client	Marine Research Specialists
Project:	Hydrogen Sulfide
Client Sample ID:	NA
Weston Test ID:	P060103.15
Species:	<i>Cyprinodon variegatus</i>

Date Received:	N/A
Date Test Started:	15-Feb-06
Date Test Ended:	19-Feb-06
Study Director:	Brian Hester
# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/15/06  Time: 1030  Technician: GZ,TS	Control		1		7.3		19.7		29		8.05	3	0	3
			2		7.4		19.6		29		8.08	4	0	4
			3		7.6		19.3		29		8.04	5	0	5
			4		7.4		19.4		29		8.08	3	0	5
	15		1		7.9		18.9		26		8.20	111	1:05	555
			2		7.5		19.5		27		8.22	88	1:05	440
			3		7.0		19.5		25		8.21	152	1:05	760
			4		7.2		19.4		26		8.19	179	1:05	895
	25		1		7.1		19.6		26		8.23	148	1:05	740
			2		7.1		19.5		26		8.26	180	1:05	900
			3		7.0		19.4		26		8.22	204	1:05	1,020
			4		7.3		19.2		26		8.32	192	1:05	960
	50		1		6.8		19.5		26		8.22	172	1:05	860
			2		7.0		19.2		26		8.41	339	1:05	1,695
			3		7.2		19.4		26		8.36	288	1:05	1,440
			4		7.4		19.4		26		8.40	322	1:05	1,610
	75		1		6.9		19.3		25		8.55	150	1:25	3,750
			2		6.6		19.4		26		8.42	93	1:25	2,325
			3		6.8		19.4		26		8.51	117	1:25	2,925
			4		6.9		19.4		26		8.51	122	1:25	3,050
	100		1		7.0		19.4		26		8.62	202	1:25	5,050
			2		7.0		19.3		26		8.72	172	1:25	4,300
			3		7.0		19.4		26		8.69	203	1:25	5,075
			4		6.9		19.2		26		8.70	157	1:25	3,925
<b>Day 1</b> <b>AM</b> Date: 2/16/06 Time: 0940 Technician: EB/JM	Control		1		7.1		20.0		29		7.77	7	0	7
	15				6.8		20.4		27		7.93	121	1:05	605
	25				6.8		20.5		27		8.01	228	1:05	1,140
	50				6.8		20.4		26		8.19	587	1:05	2,935
	75				6.7		20.3		26		8.26	223	1:25	5,575
	100				6.7		20.3		26		8.36	315	1:25	7,875
<b>Day 1</b> <b>PM</b> Date: Time: Technician:	Control		2		7.2		19.6		30		7.74	20	0	20
	15				7.1		19.8		28		7.97	114	1:05	570
	25				6.9		19.9		27		8.04	256	1:05	1,280
	50				6.7		20.1		28		8.22	503	1:05	2,515
	75				6.1		19.9		28		8.22	206	1:25	5,150
	100				6.4		19.8		28		8.39	271	1:25	6,775



## 4-Day Acute Toxicity Test

**Table C9. Acute *Cyprinodon variegatus*, Test 3 (15 Feb 2006)**

Weston Test ID:	P060103.13	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b>	Control		3		7.3		19.2		30		7.76	3	0	3
<b>AM</b>	15				6.4		20.3		28		7.87	70	1:05	350
Date: 2/17/06	25				6.3		20.4		28		7.92	176	1:05	880
Time: 1100	50				6.2		20.4		29		8.07	337	1:05	1,685
Technician: EB/JM	75				6.4		20.4		29		8.23	187	1:25	4,675
	100				6.6		20.3		29		8.34	263	1:25	6,575
<b>Day 2</b>	Control		4		7.1		18.8		30		7.81	4	0	4
<b>PM</b>	15				6.3		18.8		28		7.88	35	1:05	175
Date: 2/17/06	25				6.2		18.9		28		7.91	49	1:05	245
Time: 1623	50				5.5		18.8		28		8.13	219	1:05	1,095
Technician: TS	75				5.5		18.8		28		8.28	97	1:25	2,425
	100				6.2		19.0		29		8.40	142	1:25	3,550
<b>Day 3</b>	Control		1		7.1		18.5		31		7.83	2	0	2
<b>AM</b>	15				6.1		18.5		29		8.03	69	0	69
Date: 2/18/06	25				5.7		18.7		28		8.01	22	1:05	110
Time: 1015	50				4.4		18.7		28		8.13	29	1:05	145
Technician: TS/EB	75				2.3		18.5		28		8.36	5	1:25	125
	100				1.9		18.5		29		8.50	35	1:25	875
<b>Day 3</b>	Control		2		8.6		18.8		31		7.66	5	0	5
<b>PM</b>	15				8.2		18.5		29		7.80	52	1:05	260
Date: 2/18/06	25				7.7		18.5		28		7.86	106	1:05	530
Time: 1530	50				7.5		18.0		28		8.05	219	1:05	1,095
Technician: TS/EB	75				4.6		18.0		28		8.07	89	1:25	2,225
	100				6.3		17.9		29		8.36	99	1:25	2,475
<b>Day 4</b>	Control		3		6.8		18.5		30		7.75	0	0	0
<b>AM</b>	15				5.5		18.6		28		7.83	2	1:05	10
Date: 2/19/06	25				4.8		18.6		29		7.80	4	1:05	20
Time: 0925	50				3.2		18.8		29		8.03	133	1:05	665
Technician: EB/TS	75				3.9		18.5		28		8.24	64	1:25	1,600
	100				1.3		18.6		29		8.34	50	1:25	1,250
<b>Day 4</b>	Control		4		6.8		18.5		31		7.97	0	0	0
<b>PM</b>	15				5.5		18.6		29		8.05	5	1:05	25
Date: 2/19/06	25				5.1		18.6		29		8.11	7	1:05	35
Time: 1530	50				5.6		18.5		30		8.27	209	1:05	1,045
Technician: TS	75				4.3		18.5		29		8.38	3	1:25	75
	100				5.2		18.6		29		8.57	76	1:25	1,900

**Table D1. Acute *Americamysis bahia* (25 Jan 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
Control	1	0.0	0.0				0.1		0.0	0.0	90	80	80	80
	2	0.0		0.5				2.6	0.0	0.8	100	100	100	100
	3	0.0			0.3				0.0	0.1	100	100	100	100
	4	0.0				0.2			0.0	0.1	100	90	90	80
2.5	1	0.1*	3.8				1.5		3.8	3.0	100	80	80	80
	2	2.2		4.4				5.3	3.0	3.7	100	90	90	90
	3	4.5			5.7				2.2	4.2	100	90	90	80
	4	4.7				3.9			1.9	3.5	100	100	100	100
5	1	12.2	15.1							13.7	70	40	0	T
	2	0.0*		20.6				15.0		17.8	80	60	20	0
	3	9.9			19.0					14.4	100	80	0	T
	4	11.7				13.4			7.0*	12.5	100	60	40	10
10	1	34.0	25.1							29.6	90	10	0	T
	2	36.4		25.9						31.2	70	0	T	T
	3	36.2			45.8					41.0	100	20	0	T
	4	30.8				30.0				30.4	90	0	T	T
15	1	57.3								57.3	30	0	T	T
	2	61.3								61.3	0	T	T	T
	3	59.5								59.5	70	0	T	T
	4	57.9								57.9	50	0	T	T
20	1	84.5								84.5	0	T	T	T
	2	82.2								82.2	0	T	T	T
	3	84.3								84.3	0	T	T	T
	4	62.2								62.2	0	T	T	T

T = Test terminated due to 0% survival

\* Sulfide concentrations decreased during test

Acute Fish Test-96 Hr Survival				
Start Date:	1/25/2006	Test ID:	P060103.03	Sample ID:
End Date:	1/29/2006	Lab ID:	PGL- Port Gamble Laboratory	Sample Type:
Sample Date:		Protocol:	EPAM 94-EPA/600/4-91/003	Test Species:
Comments:	MY-Mysidopsis bahia			

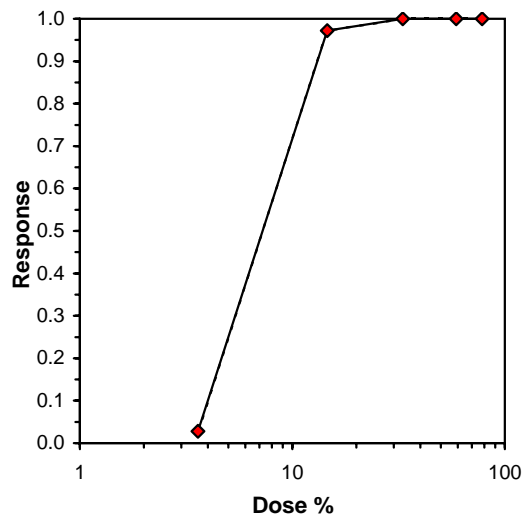
Conc-%	1	2	3	4
Control	0.8000	1.0000	1.0000	0.8000
3.6	0.8000	0.9000	0.8000	1.0000
14.6	0.0000	0.0000	0.0000	0.1000
33.1	0.0000	0.0000	0.0000	0.0000
59	0.0000	0.0000	0.0000	0.0000
78.3	0.0000	0.0000	0.0000	0.0000

Conc-%	Mean	N-Mean	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%	N				
Control	0.9000	1.0000	1.2596	1.1071	1.4120	13.974	4			4	40
3.6	0.8750	0.9722	1.2188	1.1071	1.4120	11.906	4	17.00	10.00	5	40
*14.6	0.0250	0.0278	0.1995	0.1588	0.3218	40.840	4	10.00	10.00	39	40
*33.1	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	10.00	40	40
*59	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	10.00	40	40
*78.3	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	10.00	40	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.870282	0.884	0.430144	0.512542
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	3.6	14.6	7.249828	27.77778
Treatments vs Control				

Trimmed Spearman-Kärber			
Trim Level	EC50	95% CL	
0.0%			
5.0%	7.2498	6.8655	7.6557
10.0%	7.2498	6.8655	7.6557
20.0%	7.2498	6.8655	7.6557
Auto-2.8%	7.2498	6.8655	7.6557





## 4-Day Acute Toxicity Test

**Table D2. Acute *Americamysis bahia* (25 Jan 2006)**

Weston Test ID: P060103.03	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		9	8	8	8			
	2		10	10	10	10			
	3		10*	10	10	10			
	4		10	9	9	8			
2.5	1		10	8	8	8			
	2		10	9	9	9			
	3		10	9	9	8			
	4		10	10	10	10			
5	1		7	4	0	0			
	2		8	6	2	0			
	3		10	8	0	0			
	4		10	6	4	1			
10	1		9	1	0	0			
	2		7	0	0	0			
	3		10	2	0	0			
	4		9	0	0	0			
15	1		3	0	0	0			
	2		0	0	0	0			
	3		7	0	0	0			
	4		5	0	0	0			
20	1		0	0	0	0			
	2		0	0	0	0			
	3		0	0	0	0			
	4		0	0	0	0			
Date			1/26/2006	1/27/2006	1/28/2006	1/29/2006			
Time			1315	1353	1359	1420			
Initials			JM	JM/BH	BCG	JW			

\* Replicate was initially stoked with 20 but 10 were removed





## 4-Day Acute Toxicity Test

**Table D3. Acute *Americamysis bahia* (25 Jan 2006)**

Client	Marine Research Specialists
Project:	Hydrogen Sulfide
Client Sample ID:	N/A
Weston Test ID:	P060103.03
Species:	<i>Americamysis bahia</i>

Date Received:	N/A
Date Test Started:	25-Jan-06
Date Test Ended:	29-Jan-06
Study Director:	Brian Hester
# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 1/25/06  Time: 1435  Technician: JW/GZ	Control		1		7.0		19.2		29		7.54	0	0	0
			2		6.7		19.7		29		7.63	0	0	0
			3		7.1		19.3		29		7.66	0	0	0
			4		6.8		19.5		29		7.68	0	0	0
	2.5		1		6.6		19.5		26.5		7.71	1	0	1
			2		5.3		19.7		26.5		7.70	17	0	17
			3		6.7		19.6		27		7.72	37	0	37
			4		6.8		19.5		27		7.76	42	0	42
	5		1		6.8		19.4		26.5		7.76	109	0	109
			2		7.1		19.4		28		7.71	0	0	0
			3		7.1		19.6		27		7.78	92	0	92
			4		7.1		19.6		27		7.79	111	0	111
	10		1		6.7		19.3		27		7.79	324	0	324
			2		6.7		19.6		27		7.81	361	0	361
			3		6.8		19.6		27		7.83	374	0	374
			4		6.8		19.6		27		7.83	319	0	319
	15		1		6.8		19.6		27		7.87	129	1/5	645
			2		6.3		19.6		27		7.87	138	1/5	690
			3		6.8		19.7		27		7.86	131	1/5	655
			4		6.6		19.7		27		7.89	136	1/5	680
	20		1		6.6		19.5		27		7.93	216	1/5	1080
			2		6.8		19.5		27		7.93	210	1/5	1050
			3		6.7		19.7		27		7.92	211	1/5	1055
			4		6.8		19.4		27		7.95	166	1/5	830
<b>Day 1 AM</b> Date: 1/26/06 Time: 0856 Technician: JM	Control				6.8		19.2		29		7.50	0	0	0
	2.5				6.4		19.6		27		7.63	26	0	26
	5				6.9		19.5		28		7.70	120	0	120
	10				6.5		19.5		28		7.75	221	0	221
	15				7.0		19.3		28		7.82	101	1/5	505
	20				6.7		19.6		27		7.86	166	1/5	830
<b>Day 1 PM</b> Date: 1/26/06 Time: 1720 Technician: JM/JW/BH	Control				6.9		18.7		29		7.57	3	0	3
	2.5				6.7		18.9		27		7.69	34	0	34
	5				7.0		18.7		27		7.73	173	0	173
	10				6.9		18.8		27		7.80	252	0	252
	15				6.9		18.8		27		7.89	222	1/5	1110
	20				6.9		18.8		27		7.91	254	1/5	1270



## 4-Day Acute Toxicity Test

**Table D3. Acute *Americamysis bahia* (25 Jan 2006)**

Weston Test ID:	P060103.03	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 1/27/06 Time: 0931 Technician: JM Feed Time: 0920	Control		3		6.7		19.3		30		7.32	1	0	1
	2.5				6.7		19.3		28		7.61	38	0	38
	5				6.8		19.3		28		7.69	148	0	148
	10				6.5		19.3		27		7.75	401	0	401
	15				6.7		19.1		28		7.79	120	1/5	600
	20				6.6		19.2		27		7.86	171	1/5	855
<b>Day 2</b> <b>PM</b> Date: 1/27/06 Time: 1705 Technician: JM/JW	Control		4		6.6		18.4		28		7.57	1	0	1
	2.5				6.6		18.7		27		7.63	27	0	27
	5				6.5		18.4		27		7.76	120	0	120
	10				7.0		18.4		27		7.76	268	0	268
	15				6.2		18.5		27		7.80	115	1/5	575
	20				5.6		18.6		28		7.87	158	1/5	790
<b>Day 3</b> <b>AM</b> Date: 1/28/06 Time: 1015 Technician: JW	Control		1		6.5		18.6		30		7.64	1	0	1
	2.5				6.0		18.8		27		7.66	11	0	11
	5				6.4		18.8		27		7.72	55	0	55
	10				5.8		18.9		28		7.72	140	0	140
	15				6.3		18.9		27		7.77	330	0	330
	20				5.6		19.0		27		7.84	135	1/5	675
<b>Day 3</b> <b>PM</b> Date: 1/28/06 Time: 1359 Technician: BG	Control		2		6.8		18.3		30		7.49	14	0	14
	2.5				6.8		18.2		26		7.70	42	0	42
	5				6.8		18.4		26		7.77	136	0	136
	10				6.6		18.3		27		7.81	336	0	336
	15				6.7		18.5		27		7.92	714	0	714
	20				6.6		18.0		27		7.94	215	1/5	1075
<b>Day 4</b>  Date: 1/29/06  Time: 0836  Technician: BG/JW	Control		1		6.9		19.4		30		7.74	0	0	0
			2		6.8		19.3		30		7.70	0	0	0
			3		6.7		19.7		30		7.72	0	0	0
			4		7.1		19.4		30		7.73	0	0	0
	2.5		1		6.5		19.6		27		7.80	37	0	37
			2		6.6		19.4		27		7.78	28	0	28
			3		6.7		19.5		28		7.78	21	0	21
			4		6.4		19.7		27		7.77	17	0	17
	5		1		6.7		19.7		27		7.83	126	0	126
			2		6.8		19.6		27		7.83	130	0	130
			3		6.8		19.4		27		7.81	114	0	114
			4		6.7		19.2		27		7.80	68	0	68
	10		1		6.5		19.4		30		7.82	336	0	336
			2		6.5		19.3		28		7.85	339	0	339
			3		6.4		19.5		28		7.87	315	0	315
			4		6.4		19.3		28		7.90	287	0	287
	15		1		6.2		19.6		27		7.89	563	0	563
			2		6.4		19.6		27		7.90	455	0	455
			3		6.3		19.2		28		7.94	647	0	647
			4		6.2		19.1		27		7.92	515	0	515
	20		1		5.9		19.6		27		7.95	211	1/5	1055
			2		6.1		19.4		27		7.95	134	1/5	670
			3		6.0		19.3		28		7.96	194	1/5	970
			4		5.8		19.5		27		7.97	143	1/5	715

**Table D4. Acute *Americamysis bahia* (5 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	0.2	0.2				0.0		0.6	<b>0.3</b>	100	100	100	<b>100</b>
	2	0.3		0.0				0.0	0.4	<b>0.2</b>	100	100	100	<b>100</b>
	3	0.5			0.1				0.3	<b>0.3</b>	100	100	100	<b>100</b>
	4	0.2				0.1			0.5	<b>0.2</b>	100	90	90	<b>90</b>
<b>2</b>	1	2.2	0.9				0.4		1.3	<b>1.2</b>	100	100	100	<b>90</b>
	2	1.5		0.2				1.0	3.6	<b>1.6</b>	100	90	90	<b>90</b>
	3	2.5			2.3				0.9	<b>1.9</b>	100	90	90	<b>90</b>
	4	1.9				1.0			7.8	<b>3.6</b>	100	100	100	<b>70</b>
<b>4</b>	1	7.4	4.6				1.2		2.9	<b>4.0</b>	100	100	100	<b>100</b>
	2	6.9		2.5				2.5	0.1	<b>3.0</b>	100	100	90	<b>90</b>
	3	8.0			4.0				14.2	<b>8.7</b>	100	100	100	<b>90</b>
	4	7.4				2.9			8.2	<b>6.2</b>	100**	100	80	<b>80</b>
<b>6</b>	1	13.6	12.5				6.2		10.3	<b>10.6</b>	100	90	90	<b>60</b>
	2	13.4		7.3				5.4	18.4	<b>11.2</b>	90	60	30	<b>20</b>
	3	13.2			8.6				3.2	<b>8.3</b>	100	90	70	<b>50</b>
	4	16.0				5.3			7.2	<b>9.5</b>	90	60	10	<b>10</b>
<b>8</b>	1	19.0	22.0				5.1		10.4	<b>14.1</b>	60	40	0	<b>T</b>
	2	11.2		6.5				8.8	30.8	<b>14.3</b>	100	100	100	<b>0</b>
	3	17.5			13.9				19.5	<b>17.0</b>	100	100	100	<b>50</b>
	4	9.9				10.8			17.2	<b>12.6</b>	100	100	70	<b>30</b>
<b>12</b>	1	31.2	22.1				16.3		68.3	<b>34.5</b>	80	50	40	<b>0</b>
	2	25.0		8.1				10.1	19.8	<b>15.7</b>	100	100	50	<b>10</b>
	3	20.4			18.9				50.2	<b>29.8</b>	100	100	100	<b>70</b>
	4	0.2*				0.1*			36.3	<b>36.3</b>	90	90	80	<b>80</b>

T = Test terminated due to 0% survival

\* Sulfide concentrations decreased during test

\*\* Survival miscounted

Mysid Survival, Growth and Fecundity Test-96 Hr Survival				
Start Date:	2/5/2006	Test ID:	P060103.09	Sample ID:
End Date:	2/9/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPAM 94-EPA/600/4-91/003	Test Species:
Comments:	Average			
	MY-Mysidopsis bahia			

Conc-ug/L	1	2	3	4
Control	1.0000	1.0000	1.0000	0.9000
2.1	0.9000	0.9000	0.9000	0.7000
5.5	1.0000	0.9000	0.9000	0.8000
9.9	0.6000	0.2000	0.5000	0.1000
14.5	0.0000	0.0000	0.5000	0.3000
29.1	0.0000	0.1000	0.7000	0.8000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%	N					
Control	0.9750	1.0000	1.3713	1.2490	1.4120	5.942	4				1	40
2.1	0.8500	0.8718	1.1846	0.9912	1.2490	10.885	4	0.983	2.410	0.4575	6	40
5.5	0.9000	0.9231	1.2543	1.1071	1.4120	9.935	4	0.616	2.410	0.4575	4	40
*9.9	0.3500	0.3590	0.6142	0.3218	0.8861	43.220	4	3.988	2.410	0.4575	26	40
*14.5	0.2000	0.2051	0.4206	0.1588	0.7854	74.606	4	5.007	2.410	0.4575	32	40
*29.1	0.4000	0.4103	0.6447	0.1588	1.1071	73.537	4	3.827	2.410	0.4575	24	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.9794255	0.884	-0.006949	-0.393614
Bartlett's Test indicates equal variances ( $p = 0.06$ )	10.517562	15.086272		

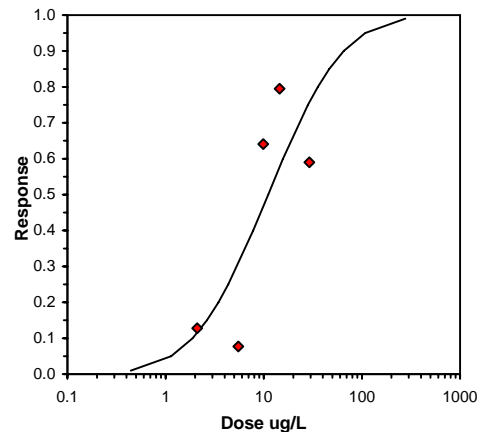
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	5.5	9.9	7.3790243		0.3337887	0.3474373	0.6431189	0.0720876	2.1E-04	5, 18

Treatments vs Control

				Maximum Likelihood-Probit						
Parameter	Value	SE	95% Fiducial Limits	Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	1.6633573	0.8904387	-1.170416 4.4971306	0.025	27.136082	7.8147278	5.5E-06	1.0444803	0.6011938	3
Intercept	3.262656	0.9422673	0.2639411 6.2613709							
TSCR	0.0231924	0.0711727	-0.203311 0.2496957		1.0					

Point	Probits	ug/L	95% Fiducial Limits
EC01	2.674	0.4424809	
EC05	3.355	1.136592	
EC10	3.718	1.8794014	
EC15	3.964	2.6386579	
EC20	4.158	3.4554365	
EC25	4.326	4.3549313	
EC40	4.747	7.8013479	
EC50	5.000	11.078484	
EC60	5.253	15.732258	
EC75	5.674	28.18249	
EC80	5.842	35.518759	
EC85	6.036	46.513355	
EC90	6.282	65.30419	
EC95	6.645	107.98315	
EC99	7.326	277.37439	

Significant heterogeneity detected ( $p = 5.51E-06$ )





## 4-Day Acute Toxicity Test

**Table D5. Acute *Americamysis bahia* (5 Feb 2006)**

Weston Test ID: P060103.09	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	9	9	9			
2	1		10	10	10	9			
	2		10	9	9	9			
	3		10	9	9	9			
	4		10	10	10	7			
4	1		10	10	10	10			
	2		10	10	9	9			
	3		10	10	10	9			
	4		10**	10	8	8			
6	1		10	9	9	6			
	2		9	6	3	2			
	3		10	9	7	5			
	4		9	6	1	1			
8	1		6	4	0	0			
	2		10	10	10	0			
	3		10	10	10	5			
	4		10	10	7	3			
12	1		8	5	4	0			
	2		10	10	5	1			
	3		10	10	10	7			
	4		9	9	8	8			
Date			2/6/06	2/7/06	2/8/06	2/9/06			
Time			1142	1206	1015	1510			
Initials			TS	TS	GZ/AM	TS/CC/GZ			

\*\* Survival miscounted



## 4-Day Acute Toxicity Test

**Table D6. Acute *Americamysis bahia* (5 Feb 2006)**

Client	Marine Research Specialists
Project:	Hydrogen Sulfide
Client Sample ID:	N/A
Weston Test ID:	P060103.09
Species:	<i>Americamysis bahia</i>

Date Received:	N/A
Date Test Started:	5-Feb-06
Date Test Ended:	9-Feb-06
Study Director:	Brian Hester
# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/5/06  Time: 1144  Technician: BH/JM	Control		1		8.6		18.6		30		7.76	2	0	2
			2		8.5		18.8		30		7.87	3	0	3
			3		8.5		18.9		30		7.89	6	0	6
			4		8.4		19.0		30		7.89	2	0	2
	2		1		8.2		18.9		27		7.94	29	0	29
			2		8.1		19.1		27		8.04	25	0	25
			3		8.2		19.2		28		7.95	34	0	34
			4		8.8		19.0		27		8.05	31	0	31
	4		1		8.1		19.2		27		8.08	130	0	130
			2		8.1		19.0		27		8.08	122	0	122
			3		8.1		19.1		27		8.05	132	0	132
			4		8.2		18.9		27		8.03	117	0	117
	6		1		8.1		19.1		27		8.03	215	0	215
			2		8.1		19.0		28		8.03	214	0	214
			3		8.2		19.2		27		8.05	219	0	219
			4		8.1		19.2		27		8.07	276	0	276
	8		1		8.1		19.1		27		8.08	336	0	336
			2		8.2		19.2		28		8.03	179	0	179
			3		8.1		19.1		27		8.07	302	0	302
			4		8.1		19.0		27		8.05	163	0	163
	12		1		8.0		19.2		27		8.11	589	0	589
			2		8.1		19.2		28		8.11	474	0	474
			3		8.1		19.1		27		8.09	386	0	368
			4		8.4		19.1		28		7.99	3	0	3
<b>Day 1 AM</b> Date: 2/6/06 Time: 1026 Technician: TS/GZ/JM	Control				8.1		18.9		29		7.77	2	0	2
	2				7.1		19.8		27		7.78	8	0	8
	4				7.6		19.7		27		7.84	49	0	49
	6				7.6		19.9		27		7.85	135	0	135
	8				7.6		19.9		27		7.89	257	0	257
	12				7.9		19.9		27		7.90	264	0	264
<b>Day 1 PM</b> Date: 2/6/06 Time: 1625 Technician: TS/GZ	Control				7.6		19.3		30		7.80	0	0	0
	2				7.5		19.7		27		7.80	2	0	2
	4				6.3		19.8		27		7.70	20	0	20
	6				7.3		19.9		26		7.80	71	0	71
	8				6.7		19.9		27		7.80	63	0	63
	12				7.2		19.9		27		8.00	120	0	120



## 4-Day Acute Toxicity Test

**Table D6. Acute *Americamysis bahia* (5 Feb 2006)**

Weston Test ID:	P060103.09	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 2/7/06 Time: 1015 Technician: JW/GZ Feed Time:	Control				6.6		19.5		29		7.73	1	0	1
	2				6.6		19.2		27		7.84	24	0	24
	4				6.9		19.8		27		7.88	46	0	46
	6				6.9		19.8		27		7.88	99	0	99
	8				6.9		19.8		27		7.94	182	0	182
	12				7.0		19.8		27		7.67	141	0	141
			3											
<b>Day 2</b> <b>PM</b> Date: 2/7/06 Time: 1408 Technician: TS/JM	Control				6.6		19.7		30		8.00	1	0	1
	2				6.8		19.9		27		8.00	15	0	15
	4				6.9		19.9		27		8.00	43	0	43
	6				6.7		20.0		27		8.10	97	0	97
	8				6.7		20.0		27		8.10	199	0	199
	12				6.9		20.0		29		8.10	1	0	1
			4											
<b>Day 3</b> <b>AM</b> Date: 2/8/06 Time: 0850 Technician: JM/AM	Control				6.7		19.3		31		7.99	0	0	0
	2				7.0		19.6		28		7.98	6	0	6
	4				6.2		19.9		27		8.07	21	0	21
	6				7.0		19.7		27		8.04	101	0	101
	8				7.0		19.7		27		8.06	86	0	86
	12				7.0		19.9		28		8.13	323	0	323
			1											
<b>Day 3</b> <b>PM</b> Date: 2/8/06 Time: 1434 Technician: JM/TS/AM	Control				6.9		19.3		30		7.91	0	0	0
	2				7.1		19.6		26		7.97	14	0	14
	4				7.0		19.8		27		8.01	38	0	38
	6				6.9		19.9		27		7.95	72	0	72
	8				7.0		19.9		27		8.01	134	0	134
	12				6.6		20.0		27		8.00	150	0	150
			2											
<b>Day 4</b>  Date: 2/9/06  Time: 0900  Technician: CC	Control		1		3.2		19.1		30.0		7.6	4	0	4
			2		6.3		18.5		31.0		7.7	3	0	3
			3		6.5		19.2		29.0		7.8	3	0	3
			4		4.7		19.2		30.0		7.6	3	0	3
	2		1		5.4		19.5		27.0		7.5	7	0	7
			2		6.0		19.4		24.0		7.9	43	0	43
			3		4.5		19.5		28.0		7.5	5	0	5
			4		4.0		19.3		23.0		7.5	41	0	41
	4		1		4.8		19.3		27.0		7.6	19	0	19
			2		6.6		19.3		27.0		7.7	1	0	1
			3		5.7		19.3		25.0		7.9	169	0	169
			4		4.6		19.4		26.0		7.9	98	0	98
	6		1		7.1		18.9		27.0		7.9	123	0	123
			2		6.3		19.2		24.0		8.0	269	0	269
			3		6.4		19.3		27.0		7.7	25	0	25
			4		6.5		19.4		27.0		7.9	86	0	86
	8		1		6.8		19.0		27.0		7.8	101	0	101
			2		6.9		19.1		25.0		7.8	296	0	296
			3		6.1		19.2		25.0		8.0	287	0	287
			4		6.6		19.2		27.0		7.7	136	0	136
	12		1		6.8		19.2		27.0		7.4	303	0	303
			2		6.5		19.4		28.0		8.0	295	0	295
			3		6.1		19.1		27.0		7.7	397	0	397
			4		2.2		19.2		27.0		7.4	161	0	161

**Table E1. Acute *Neanthes arenaceodentata*, Test 1 (17 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
Control	1	0.5	0.4				0.0		0.5	<b>0.3</b>	100	100	100	<b>100</b>
	2	0.5		0.9				0.0	1.3	<b>0.7</b>	100	100	100	<b>100</b>
	3	0.6			0.2				0.9	<b>0.6</b>	100	100	100	<b>100</b>
	4	0.5				0.5			0.6	<b>0.5</b>	100	100	100	<b>100</b>
2.5	1	6.7	2.9				0.0		3.7	<b>3.3</b>	100	100	100	<b>100</b>
	2	14.5		2.5				0.0	14.6	<b>7.9</b>	100	100	100	<b>100</b>
	3	5.6			5.1				9.6	<b>6.7</b>	100	100	100	<b>100</b>
	4	14.5				9.6			15.0	<b>13.0</b>	100	100	100	<b>100</b>
5	1	31.2	23.0				14.3		32.3	<b>25.2</b>	100	100	100	<b>100</b>
	2	0.0		10.6				4.8	3.8	<b>4.8</b>	100	100	100	<b>80</b>
	3	13.7			6.4				10.9	<b>10.3</b>	100	100	100	<b>100</b>
	4	27.3				11.4			22.4	<b>20.3</b>	100	100	100	<b>100</b>
10	1	62.3	49.3				30.7		59.0	<b>50.3</b>	100	100	100	<b>100</b>
	2	47.5		34.4				7.6	42.6	<b>33.0</b>	100	100	100	<b>100</b>
	3	49.9			27.7				34.2	<b>37.3</b>	100	100	100	<b>100</b>
	4	41.9				23.6			29.1	<b>31.5</b>	100	100	100	<b>100</b>
25	1	97.3	86.8				0.0*		176	<b>120</b>	100	100	100	<b>100</b>
	2	32.1		166				1.5*	51.1	<b>83.2</b>	100	100	100	<b>90</b>
	3	90.5			70.6				130	<b>97.0</b>	100	100	100	<b>100</b>
	4	87.8				70.6			136	<b>98.3</b>	100	100	100	<b>100</b>
50	1	0.0*	70.9				51.5		93.3	<b>71.9</b>	100	100	100	<b>100</b>
	2	154		123				17.2*	164	<b>147</b>	100	100	100	<b>80</b>
	3	163			85.8				135	<b>128</b>	100	100	100	<b>90</b>
	4	135				143			162	<b>147</b>	100	100	100	<b>100</b>

\* Sulfide concentrations decreased during test



Acute Fish Test-96 Hr Survival				
Start Date:	2/17/2006	Test ID:	P060103.18	Sample ID:
End Date:	2/21/2006	Lab ID:	WESTON	Sample Type:
Sample Date:		Protocol:	EPA/600/R	Test Species:
Comments:	Average			
Conc-ug/L	1	2	3	4
Control	1.0000	1.0000	1.0000	1.0000
7.7	1.0000	1.0000	1.0000	1.0000
15.2	1.0000	0.8000	1.0000	1.0000
38	1.0000	1.0000	1.0000	1.0000
99.6	1.0000	0.9000	1.0000	1.0000
123.5	1.0000	0.8000	0.9000	0.0000

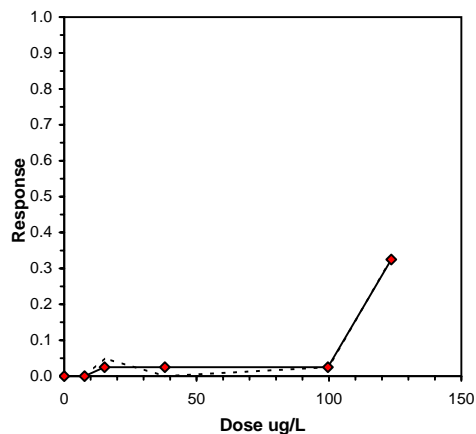
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	Rank Sum	1-Tailed Critical	Isotonic	
			Mean	Min	Max	CV%				Mean	N-Mean
Control	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4			1.0000	1.0000
7.7	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4	18.00	10.00	1.0000	1.0000
15.2	0.9500	0.9500	1.3358	1.1071	1.4120	11.411	4	16.00	10.00	0.9750	0.9750
38	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4	18.00	10.00	0.9750	0.9750
99.6	0.9750	0.9750	1.3713	1.2490	1.4120	5.942	4	16.00	10.00	0.9750	0.9750
123.5	0.6750	0.6750	0.9817	0.1588	1.4120	57.307	4	12.00	10.00	0.6750	0.6750

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.6690895	0.884	-2.315677	10.432854
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	123.5	>123.5		

Treatments vs Control

Point	ug/L	SD	Linear Interpolation (200 Resamples)	
			95% CL(Exp)	Skew
IC05	101.59			
IC10	105.58			
IC15	109.56			
IC20	113.54			
IC25	117.53			
IC40	>123.5			
IC50	>123.5			





## 4-Day Acute Toxicity Test

**Table E2. Acute *Neanthes arenaceodentata*, Test 1 (17 Feb 2006)**

Weston Test ID: P060103.18	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
2.5	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
5	1		10	10	10	10			
	2		10	10	10	8			
	3		10	10	10	10			
	4		10	10	10	10			
10	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
25	1		10	10	10	10			
	2		10	10	10	9			
	3		10	10	10	10			
	4		10	10	10	10			
50	1		10	10	10	10			
	2		10	10	10	8			
	3		10	10	10	9			
	4		10	10	10	10			
Date			2/18/06	2/19/06	2/20/06	2/21/06			
Time			1600	1025	1045	1629			
Initials			EB	EB	EB	JM/TS			



## 4-Day Acute Toxicity Test

**Table E3. Acute *Neanthes arenaceodentata*, Test 1 (17 Feb 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen sulfide	Date Test Started:	17-Feb-06
Client Sample ID:	N/A	Date Test Ended:	21-Feb-06
Weston Test ID:	P060103.18	Study Director:	Brian Hester
Species:	<i>Neanthes arenaceodentata</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/17/06  Time: 1247  Technician: TS	Control		1		7.7		18.7		30		7.67	4	0	4
			2		7.6		19.2		30		7.70	4	0	4
			3		7.5		19.4		30		7.73	5	0	5
			4		7.5		19.4		30		7.69	4	0	4
	2.5		1		7.3		19.7		27		7.75	59	0	59
			2		7.1		19.6		27		7.81	144	0	144
			3		7.4		19.6		27		7.80	54	0	54
			4		7.2		19.6		27		7.83	150	0	150
	5		1		6.8		19.6		27		7.85	337	0	337
			2		6.9		19.4		29		9.84	39	0	39
			3		7.4		19.7		29		7.79	132	0	132
			4		7.0		19.7		27		7.80	265	0	265
	10		1		6.7		19.8		27		7.91	763	0	763
			2		7.3		19.5		27		7.86	523	0	523
			3		7.5		19.7		29		7.78	470	0	470
			4		7.5		19.8		28		7.84	445	0	445
	25		1		6.8		19.5		27		8.05	322	1/5	1610
			2		7.3		19.7		29		7.87	73	1/5	365
			3		6.6		19.7		27		8.06	306	1/5	1530
			4		6.9		19.5		27		8.09	317	1/5	1585
	50		1		5.6		19.0		26		11.50	205	1/5	1025
			2		6.6		19.5		27		8.24	771	1/5	3855
			3		7.6		19.4		29		7.84	347	1/5	1735
			4		7.0		19.5		27		8.25	694	1/5	3470
<b>Day 1</b> <b>AM</b> Date: 2/18/06 Time: 1025 Technician: EB/TS	Control				7.0		18.8		30		7.83	4	0	4
	2.5				7.4		19.6		30		7.92	37	0	37
	5				6.4		19.4		27		7.93	294	0	294
	10				6.4		19.3		27		7.98	701	0	701
	25				6.6		19.3		27		8.21	408	1/5	2040
	50				7.6		19.6		30		8.08	254	1/5	1270
<b>Day 1</b> <b>PM</b> Date: 2/18/06 Time: 1513 Technician: TS/EB	Control				7.5		18.9		30		7.77	8	0	8
	2.5				7.1		19.1		26		7.86	27	0	27
	5				7.7		19.3		30		7.91	132	0	132
	10				6.7		19.3		28		7.96	471	0	471
	25				6.8		19.2		30		7.97	470	1/5	2350
	50				6.1		19.2		27		8.39	864	1/5	4320



## 4-Day Acute Toxicity Test

**Table E3. Acute *Neanthes arenaceodentata*, Test 1 (17 Feb 2006)**

Weston Test ID:	P060103.18	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 2/19/06 Time: 0930 Technician: TS/EB Feed Time:	Control		3		7.1		19.0		30		7.81	2	0	2
	2.5				7.0		19.1		28		7.85	55	0	55
	5				6.9		19.4		29		7.86	71	0	71
	10				6.8		19.4		28		7.89	327	0	327
	25				5.2		19.2		27		8.02	219	1/5	1095
	50				7.1		19.4		30		8.04	282	1/5	1410
<b>Day 2</b> <b>PM</b> Date: 2/19/06 Time: 1417 Technician: TS	Control		4		7.1		18.8		31		7.70	4	0	4
	2.5				7.2		19.1		27		7.87	108	0	108
	5				6.4		19.4		28		7.84	121	0	121
	10				7.3		19.2		29		7.86	262	0	262
	25				6.4		18.9		27		8.02	219	1/5	1095
	50				6.4		19.1		28		8.09	520	1/5	2600
<b>Day 3</b> <b>AM</b> Date: 2/20/06 Time: 0925 Technician: GZ/EB	Control		1		7.2		19.2		31		7.83	0	0	0
	2.5				7.7		19.6		28		7.91	0	0	0
	5				7.0		19.6		27		7.89	168	0	168
	10				6.9		19.4		27		7.96	419	0	419
	25				7.0		19.6		30		7.82	0	1/5	0
	50				7.5		19.8		29		7.98	148	1/5	740
<b>Day 3</b> <b>PM</b> Date: 2/20/06 Time: 1445 Technician: EB	Control		2		7.0		19.2		30		7.89	0	0	0
	2.5				8.1		19.3		29		7.95	9	0	9
	5				9.5		19.5		29		7.98	69	0	69
	10				8.0		19.3		28		7.88	88	0	88
	25				9.4		19.4		30		8.04	5	1/5	25
	50				9.1		19.3		27		8.39	121	1/5	605
<b>Day 4</b>  Date: 2/21/06  Time: 0940  Technician: TS/EB	Control		1		9.1		19.0		30		7.75	4	0	4
			2		9.4		18.7		30		7.79	13	0	13
			3		8.8		19.4		30		7.75	8	0	8
			4		9.0		19.5		30		7.76	5	0	5
	2.5		1		9.4		19.7		28		7.86	41	0	41
			2		8.4		19.5		26		7.84	153	0	153
			3		8.7		19.6		27		7.83	99	0	99
			4		8.5		19.5		26		7.85	161	0	161
	5		1		8.1		19.6		26		7.84	339	0	339
			2		9.7		19.7		30		7.85	42	0	42
			3		8.7		19.8		30		7.76	99	0	99
			4		8.6		19.7		27		7.81	222	0	222
	10		1		8.0		19.5		26		7.90	704	0	704
			2		8.1		19.5		27		7.85	460	0	460
			3		8.5		19.7		27		7.84	361	0	361
			4		8.7		19.7		28		7.87	329	0	329
	25		1		7.3		19.6		26		7.92	438	1/5	2190
			2		9.2		19.8		29		7.87	116	1/5	580
			3		7.5		19.6		27		7.99	378	1/5	1890
			4		7.6		19.5		26		8.01	412	1/5	2060
	50		1		9.1		19.8		30		7.93	242	1/5	1210
			2		6.7		19.6		27		8.26	863	1/5	4315
			3		8.6		19.9		28		7.97	378	1/5	1890
			4		7.7		19.6		27		8.23	795	1/5	3975

**Table E4. Acute *Neanthes arenaceodentata* , Test 2 (17 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	1.1	0.8				0.0		0.6	<b>0.6</b>	100	100	100	<b>100</b>
	2	0.8		0.4				0.4	0.8	<b>0.6</b>	100	100	100	<b>100</b>
	3	0.6			0.0				1.4	<b>0.7</b>	100	100	100	<b>100</b>
	4	0.6				0.2			0.6	<b>0.5</b>	100	100	100	<b>100</b>
<b>2.5</b>	1	3.1	2.0				0.0		3.0	<b>2.0</b>	100	100	100	<b>100</b>
	2	9.6		3.4				1.4	5.6	<b>5.0</b>	100	100	100	<b>90</b>
	3	6.1			3.0				3.7	<b>4.3</b>	100	100	100	<b>90</b>
	4	8.6				4.5			4.0	<b>5.7</b>	100	100	100	<b>100</b>
<b>5</b>	1	18.3	18.9				5.7		10.3	<b>13.3</b>	100	100	100	<b>100</b>
	2	3.8		6.3				3.6	1.8	<b>3.9</b>	100	100	100	<b>100</b>
	3	7.5			4.9				6.4	<b>6.2</b>	100	100	100	<b>100</b>
	4	13.0				6.8			6.6	<b>8.8</b>	100	100	100	<b>100</b>
<b>10</b>	1	56.6	36.4				12.8		21.0	<b>31.7</b>	100	100	100	<b>100</b>
	2	40.9		29.4				19.0	20.2	<b>27.4</b>	100	100	100	<b>90</b>
	3	29.6			14.2				16.3	<b>20.0</b>	100	100	100	<b>100</b>
	4	31.3				7.8			10.2	<b>16.4</b>	100	100	100	<b>100</b>
<b>25</b>	1	140	125				0.0*		86.1	<b>117.1</b>	100	100	100	<b>90</b>
	2	44.0		121				1.5*	38.2	<b>67.8</b>	100	100	100	<b>90</b>
	3	130			30.3				32.9	<b>64.4</b>	100	100	100	<b>100</b>
	4	130				33.4			63.7	<b>75.8</b>	100	100	100	<b>100</b>
<b>50</b>	1	90.8	63.0				38.9		45.1	<b>59.5</b>	100	100	100	<b>100</b>
	2	169		54.0				35.5	92.5	<b>87.7</b>	100	100	100	<b>90</b>
	3	82.0			70.9				96.5	<b>83.1</b>	100	100	100	<b>100</b>
	4	145				83.8			120	<b>116</b>	100	100	100	<b>100</b>

\* Sulfide concentrations decreased during test

Acute Fish Test-96 Hr Survival				
Start Date:	2/17/2006	Test ID:	P060103.17	Sample ID:
End Date:	2/21/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPA/600/R	Test Species:
Comments:	Average			
	Neanthes arenaceodentata			

Conc-ug/L	1	2	3	4
Control	1.0000	1.0000	1.0000	1.0000
4.3	1.0000	0.9000	0.9000	1.0000
8.1	1.0000	1.0000	1.0000	1.0000
23.9	1.0000	0.9000	1.0000	1.0000
81.3	0.9000	0.9000	1.0000	1.0000
86.6	1.0000	0.9000	1.0000	1.0000

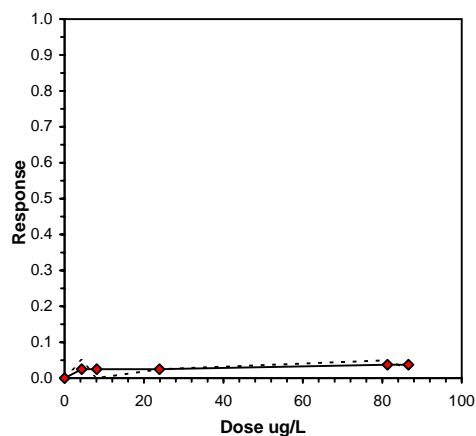
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical	Isotonic	
			Mean	Min	Max	CV%	N			Mean	N-Mean
Control	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4			1.0000	1.0000
4.3	0.9500	0.9500	1.3305	1.2490	1.4120	7.072	4	14.00	10.00	0.9750	0.9750
8.1	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	4	18.00	10.00	0.9750	0.9750
23.9	0.9750	0.9750	1.3713	1.2490	1.4120	5.942	4	16.00	10.00	0.9750	0.9750
81.3	0.9500	0.9500	1.3305	1.2490	1.4120	7.072	4	14.00	10.00	0.9625	0.9625
86.6	0.9750	0.9750	1.3713	1.2490	1.4120	5.942	4	16.00	10.00	0.9625	0.9625

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.8813365	0.884	-0.599255	-0.61569
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	86.6	>86.6		

Treatments vs Control

Point	ug/L	SD	Linear Interpolation (200 Resamples)	
			95% CL(Exp)	Skew
IC05	>86.6			
IC10	>86.6			
IC15	>86.6			
IC20	>86.6			
IC25	>86.6			
IC40	>86.6			
IC50	>86.6			





## 4-Day Acute Toxicity Test

**Table E5. Acute *Neanthes arenaceodentata*, Test 2 (17 Feb 2006)**

Weston Test ID: P060103.17	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
2.5	1		10	10	10	10			
	2		10	10	10	9			
	3		10	10	10	9			
	4		10	10	10	10			
5	1		10	10	10	10			
	2		10	10	10	10			
	3		10	10	10	10			
	4		10	10	10	10			
10	1		10	10	10	10			
	2		10	10	10	9			
	3		10	10	10	10			
	4		10	10	10	10			
25	1		10	10	10	9			
	2		10	10	10	9			
	3		10	10	10	10			
	4		10	10	10	10			
50	1		10	10	10	10			
	2		10	10	10	9			
	3		10	10	10	10			
	4		10	10	10	10			
Date			2/18/06	2/19/06	2/20/06	2/21/06			
Time			1600	1025	1045	1629			
Initials			EB	EB	EB	JM/TS			



## 4-Day Acute Toxicity Test

**Table E6. Acute *Neanthes arenaceodentata*, Test 2 (17 Feb 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen sulfide	Date Test Started:	17-Feb-06
Client Sample ID:	N/A	Date Test Ended:	21-Feb-06
Weston Test ID:	P060103.17	Study Director:	Brian Hester
Species:	<i>Neanthes</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/17/06  Time: 1842  Technician: JM/TS/EB/JW	Control		1		7.5		19.3		30		7.67	8	0	8
			2		7.6		19.2		30		7.69	6	0	6
			3		7.4		19.4		30		7.70	5	0	5
			4		7.4		19.5		30		7.70	5	0	5
	2.5		1		7.5		19.6		27		7.75	27	0	27
			2		7.0		19.3		27		7.75	84	0	84
			3		7.3		19.4		27		7.70	48	0	48
			4		7.3		19.2		27		7.74	74	0	74
	5		1		6.9		19.4		27		7.75	160	0	160
			2		7.6		19.6		29		7.73	32	0	32
			3		7.5		19.5		29		7.75	66	0	66
			4		7.3		19.6		27		7.78	121	0	121
	10		1		6.9		19.4		27		7.85	611	0	611
			2		7.1		19.3		29		7.72	340	0	340
			3		7.2		19.3		28		7.78	277	0	277
			4		7.2		19.5		27		7.80	304	0	304
	25		1		6.9		19.4		29		7.94	369	1/5	1845
			2		7.6		19.6		27		7.78	82	1/5	410
			3		7.2		19.5		27		7.97	362	1/5	1810
			4		6.9		19.3		27		7.98	371	1/5	1855
	50		1		7.6		19.5		26		7.92	226	1/5	1130
			2		7.3		19.3		27		8.23	830	1/5	4150
			3		7.5		19.6		29		8.06	280	1/5	1400
			4		7.3		19.4		27		8.23	711	1/5	3555
<b>Day 1 AM</b> Date: 2/18/06 Time: 1025 Technician: EB/TS	Control				7.4		18.5		30		7.74	7	0	7
	2.5				7.3		19.4		30		7.90	24	0	24
	5				6.6		19.3		27		7.93	241	0	241
	10				6.4		19.3		27		7.98	518	0	518
	25				5.8		19.4		27		7.98	356	1/5	1780
	50				7.5		19.4		30		8.10	236	1/5	1180
<b>Day 1 PM</b> Date: 2/18/06 Time: 1513 Technician: TS/EB	Control				7.4		19.1		30		7.77	4	0	4
	2.5				6.7		19.2		26		7.86	37	0	37
	5				7.8		19.3		30		7.92	80	0	80
	10				6.8		19.2		28		7.83	306	0	306
	25				7.6		19.2		30		7.91	301	1/5	1505
	50				6.5		19.3		27		8.34	339	1/5	1695





## 4-Day Acute Toxicity Test

**Table E6. Acute *Neanthes arenaceodentata*, Test 2 (17 Feb 2006)**

Weston Test ID:	P060103.17	Client:	Marine Research Specialists	Client Sample ID:	
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 2/19/06 Time: 0930 Technician: TS/EB Feed Time:	Control				7.3		18.7		30		7.77	0	0	0
	2.5				7.1		19.1		28		7.84	32	0	32
	5				7.3		19.2		29		7.86	54	0	54
	10				6.5		19.3		28		7.86	157	0	157
	25				4.3		19.2		27		7.94	79	1/5	395
	50				6.8		19.3		30		8.02	223	1/5	1115
			3											
<b>Day 2</b> <b>PM</b> Date: 2/19/06 Time: 1417 Technician: TS	Control				7.0		19.0		31		7.80	2	0	2
	2.5				7.1		19.1		27		7.87	51	0	51
	5				6.9		19.2		28		7.78	64	0	64
	10				7.0		19.2		29		7.87	89	0	89
	25				5.0		19.1		27		7.95	89	1/5	445
	50				4.1		19.2		28		8.10	311	1/5	1555
			4											
<b>Day 3</b> <b>AM</b> Date: 2/20/06 Time: 0925 Technician: GZ/EB	Control				7.1		19.1		31		7.85	0	0	0
	2.5				7.7		19.6		28		7.90	0	0	0
	5				6.6		19.5		27		7.87	64	0	64
	10				6.1		19.5		27		7.92	160	0	160
	25				6.9		19.5		30		7.83	0	1/5	0
	50				6.9		19.8		29		7.96	107	1/5	535
			1											
<b>Day 3</b> <b>PM</b> Date: 2/20/06 Time: 1445 Technician: EB	Control				7.3		19.3		30		7.90	5	0	5
	2.5				7.8		19.2		29		7.84	15	0	15
	5				9.4		19.5		29		7.95	49	0	49
	10				7.3		19.3		28		7.77	174	0	174
	25				9.1		19.3		30		7.95	4	1/5	20
	50				4.2		19.2		27		8.04	115	1/5	575
			2											
<b>Day 4</b>  Date: 2/21/06  Time: 0940  Technician: TS/EB	Control		1		9.0		18.9		30		7.72	5	0	5
			2		9.1		19.2		30		7.77	7	0	7
			3		8.6		19.4		30		7.75	12	0	12
			4		8.8		19.5		30		7.77	6	0	6
	2.5		1		9.2		19.6		28		7.85	33	0	33
			2		8.3		19.4		26		7.84	59	0	59
			3		8.3		19.5		27		7.77	34	0	34
			4		8.4		19.5		26		7.83	41	0	41
	5		1		7.7		19.4		26		7.82	104	0	104
			2		9.6		19.6		30		7.83	19	0	19
			3		8.8		19.7		30		7.81	64	0	64
			4		8.3		19.6		27		7.81	65	0	65
	10		1		7.1		19.4		26		7.86	230	0	230
			2		7.4		19.4		27		7.82	205	0	205
			3		8.0		19.6		27		7.79	155	0	155
			4		8.1		19.6		28		7.82	104	0	104
	25		1		4.1		19.5		26		7.60	111	1/5	555
			2		8.8		19.7		29		7.86	85	1/5	425
			3		5.8		19.6		27		7.93	84	1/5	420
			4		5.8		19.4		26		7.93	162	1/5	810
	50		1		8.0		19.6		30		7.87	103	1/5	515
			2		4.3		19.5		27		8.17	398	1/5	1990
			3		6.5		19.9		28		7.93	248	1/5	1240
			4		5.7		19.6		27		8.18	527	1/5	2635

**Table F1. Acute *Ampelisca abdita*, Test 1 (23 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
<b>Control</b>	1	0.4	0.6				0.0		0.1	<b>0.3</b>	100**	100**	ND	<b>100</b>
	2	0.3		0.2				0.1		<b>0.2</b>	100	100	ND	<b>100</b>
	3	0.4			0.0					<b>0.2</b>	100	100	ND	<b>100</b>
	4	0.3				0.2				<b>0.2</b>	100	100	ND	<b>80</b>
<b>2.5</b>	1	1.5	1.5				0.2		4.5	<b>1.9</b>	100	100	ND	<b>90</b>
	2	1.7		1.0				0.9		<b>1.2</b>	100	80	ND	<b>70</b>
	3	2.2			0.2					<b>1.2</b>	100	100	ND	<b>90</b>
	4	2.0				0.3				<b>1.1</b>	100	100	ND	<b>90</b>
<b>5</b>	1	11.5	13.1				5.8		14.8	<b>11.3</b>	100	100	ND	<b>100</b>
	2	10.8		10.4				6.6		<b>9.3</b>	100	80	ND	<b>80</b>
	3	11.3			6.3					<b>8.6</b>	90	90	ND	<b>80</b>
	4	11.0				6.1				<b>8.5</b>	100	100	ND	<b>90</b>
<b>10</b>	1	27.8	36.0				11.3		33.7	<b>27.2</b>	100**	100	ND	<b>70</b>
	2	27.7		19.3				14.5		<b>20.5</b>	100	90	ND	<b>40</b>
	3	28.6			12.2					<b>20.4</b>	100	100	ND	<b>80</b>
	4	29.5				11.9				<b>20.7</b>	100	100	ND	<b>70</b>
<b>15</b>	1	50.5	42.9				20.5		69.7	<b>45.9</b>	100	100	ND	<b>60</b>
	2	58.2		36.1				41.0		<b>45.1</b>	100	90	ND	<b>60</b>
	3	58.9			34.0					<b>46.5</b>	100	100	ND	<b>40</b>
	4	60.3				27.3				<b>43.8</b>	100	90	ND	<b>30</b>
<b>20</b>	1	81.4	74.7				34.0		115	<b>76.2</b>	100**	100	ND	<b>40</b>
	2	76.0		57.6				75.9		<b>69.9</b>	90	80	ND	<b>40</b>
	3	80.0			52.1					<b>66.0</b>	100	80	ND	<b>0</b>
	4	80.2				27.1				<b>53.6</b>	100	90	ND	<b>0</b>

ND = No data

\*\* Survival miscounted

Acute Fish Test-96 Hr Survival				
Start Date:	2/23/2006	Test ID:	P060103.19	Sample ID:
End Date:	2/27/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPA/600/R	Test Species:
Comments:	Average			
	AP-Ampelisca abdita			

Conc-ug/L	1	2	3	4
Control	1.0000	1.0000	1.0000	0.8000
1.4	0.9000	0.7000	0.9000	0.9000
9.4	1.0000	0.8000	0.8000	0.9000
22.2	0.7000	0.4000	0.8000	0.7000
45.3	0.6000	0.6000	0.4000	0.3000
66.4	0.4000	0.4000	0.0000	0.0000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%	N					
Control	0.9500	1.0000	1.3358	1.1071	1.4120	11.411	4				2	40
1.4	0.8500	0.8947	1.1846	0.9912	1.2490	10.885	4	1.145	2.410	0.3182	6	40
9.4	0.8750	0.9211	1.2188	1.1071	1.4120	11.906	4	0.886	2.410	0.3182	5	40
*22.2	0.6500	0.6842	0.9435	0.6847	1.1071	19.184	4	2.971	2.410	0.3182	14	40
*45.3	0.4750	0.5000	0.7591	0.5796	0.8861	20.120	4	4.367	2.410	0.3182	21	40
*66.4	0.2000	0.2105	0.4217	0.1588	0.6847	71.998	4	6.922	2.410	0.3182	32	40

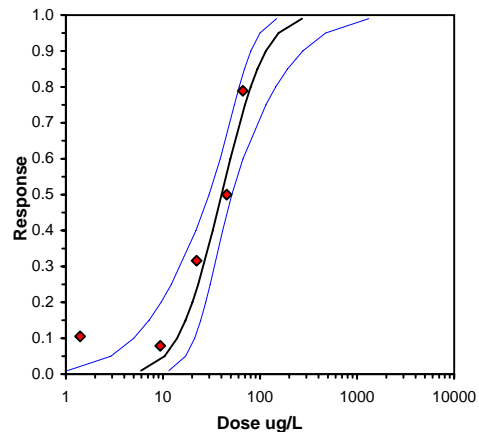
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.9184331	0.884	-0.28886	-1.01471
Bartlett's Test indicates equal variances (p = 0.69)	3.0345705	15.086272		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	9.4	22.2	14.445761		0.2218584	0.2345758	0.4697599	0.0348697	1.5E-05	5, 18

Treatments vs Control

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	2.8041933	0.6593889	1.511791	4.0965956	0.05	2.8585206	7.8147278	0.41	1.6038021	0.3566088	6
Intercept	0.502629	1.0721792	-1.598842	2.6041003							
TSCR	0.0993326	0.0317507	0.0371012	0.161564							
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	5.9457404	1.0381767	11.528925							
EC05	3.355	10.404777	2.9001447	17.091599							
EC10	3.718	14.02134	4.9953273	21.165799							
EC15	3.964	17.1475	7.1875892	24.523791							
EC20	4.158	20.122065	9.5701051	27.648439							
EC25	4.326	23.081982	12.195478	30.742424							
EC40	4.747	32.617921	21.880863	41.233251							
EC50	5.000	40.160775	29.931068	51.122715							
EC60	5.253	49.447906	38.91324	66.690331							
EC75	5.674	69.876486	54.383802	114.83274							
EC80	5.842	80.155186	60.903809	145.2916							
EC85	6.036	94.059654	69.028649	192.4297							
EC90	6.282	115.03092	80.305722	275.75729							
EC95	6.645	155.01414	99.781266	473.39184							
EC99	7.326	271.26785	148.33987	1318.726							

Point	Probits	ug/L	Response
EC01	2.674	5.9457404	0.10
EC05	3.355	10.404777	0.08
EC10	3.718	14.02134	0.32
EC15	3.964	17.1475	0.50
EC20	4.158	20.122065	0.78





## 4-Day Acute Toxicity Test

**Table F2. Acute *Ampelisca abdita*, Test 1 (23 Feb 2006)**

Weston Test ID: P060103.19	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10**	10**	ND	10			
	2		10	10	ND	10			
	3		10	10	ND	10			
	4		10	10	ND	8			
2.5	1		10	10	ND	9			
	2		10	8	ND	7			
	3		10	10	ND	9			
	4		10	10*	ND	9			
5	1		10	10	ND	10			
	2		10	8	ND	8			
	3		9	9	ND	8			
	4		10	10	ND	9			
10	1		10**	10	ND	7			
	2		10	9	ND	4			
	3		10	10	ND	8			
	4		10	10	ND	7			
15	1		10	10	ND	6			
	2		10	9	ND	6			
	3		10	10	ND	4			
	4		10	9	ND	3			
20	1		10**	10	ND	4			
	2		9	8	ND	4			
	3		10	8	ND	0			
	4		10	9	ND	0			
Date			2/24/06	2/25/06		2/27/06			
Time			1210	1530		1640			
Initials			GZ	BH/JW		TS/RE			

\* Note on datasheet that animal accidentally collected w/ sulfides

\*\* Survival miscounted

ND = No data



## 4-Day Acute Toxicity Test

**Table F3. Acute *Ampelisca abdita*, Test 1 (23 Feb 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen sulfide	Date Test Started:	26-Jan-06
Client Sample ID:	N/A	Date Test Ended:	30-Jan-06
Weston Test ID:	P060103.19	Study Director:	Brian Hester
Species:	<i>Ampelisca abdita</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/23/06  Time: 0915  Technician: JM/GZ	Control		1		8.1		19.2		30		7.86	4	0	4
			2		8.0		19.4		29		7.85	3	0	3
			3		8.0		19.6		30		7.87	4	0	4
			4		8.0		19.5		30		7.87	3	0	3
	2.5		1		7.9		19.6		28		7.91	19	0	19
			2		8.0		19.4		28		7.90	21	0	21
			3		7.9		19.7		27		7.89	26	0	26
			4		7.9		19.6		28		7.90	24	0	24
	5		1		8.0		19.6		27		7.93	147	0	147
			2		7.9		19.5		27		7.93	138	0	138
			3		8.0		19.4		27		7.94	147	0	147
			4		7.9		19.5		28		7.93	141	0	141
	10		1		7.9		19.5		27		7.98	396	0	396
			2		7.9		19.6		27		7.97	385	0	385
			3		8.0		19.6		27		7.96	390	0	390
			4		7.9		19.8		27		7.98	420	0	420
	15		1		7.9		19.7		28		8.01	154	1/5	770
			2		7.9		19.5		27		8.00	173	1/5	865
			3		7.9		19.6		27		8.00	175	1/5	875
			4		7.9		19.6		27		8.00	179	1/5	895
	20		1		7.9		19.5		27		8.03	258	1/5	1290
			2		8.0		19.6		27		8.03	241	1/5	1205
			3		7.9		19.7		27		8.04	259	1/5	1295
			4		7.9		19.6		27		8.03	254	1/5	1270
<b>Day 1</b> <b>AM</b> Date: 2/24/06 Time: 0916 Technician: JM	Control				7.9		17.8		30		7.83	6	0	6
	2.5				7.8		17.7		27		7.84	16	0	16
	5				7.7		17.8		28		7.89	155	0	155
	10				7.6		17.6		27		7.92	450	0	450
	15				7.8		17.6		28		7.95	115	1/5	575
	20				7.7		17.5		28		8.01	228	1/5	1140
<b>Day 1</b> <b>PM</b> Date: 2/24/06 Time: 1530 Technician: JM/GZ	Control				7.8		18.7		29		7.94	2	0	2
	2.5				7.9		18.3		27		8.00	15	0	15
	5				7.7		18.8		27		8.04	168	0	168
	10				7.7		18.8		27		8.07	334	0	334
	15				7.8		18.8		27		8.11	136	1/5	680
	20				7.7		18.7		27		8.13	227	1/5	1135



## 4-Day Acute Toxicity Test

**Table F3. Acute *Ampelisca abdita*, Test 1 (23 Feb 2006)**

Weston Test ID:	P060103.19	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 2</b> <b>AM</b> Date: 2/25/06 Time: 0959 Technician: JW Feed Time:	Control		3		7.7		18.9		30		7.87	0		0
	2.5				7.50		18.9		27		7.91	3	0	3
	5				7.4		18.9		27		7.95	84	0	84
	10				7.3		19.0		27		7.97	170	0	170
	15				7.1		19.0		27		8.00	101	1/5	505
	20				7.2		18.9		27		8.03	165	1/5	825
<b>Day 2</b> <b>PM</b> Date: 2/25/06 Time: 1720 Technician: BH	Control		4		7.7		18.0		30		7.90	2	0	2
	2.5				7.7		18.4		28		7.94	4	0	4
	5				7.7		18.3		28		8.01	93	0	93
	10				7.5		18.4		28		8.00	178	0	178
	15				7.5		18.3		28		8.02	85	1/5	425
	20						18.5		28		8.07	94	1/5	470
<b>Day 3</b> <b>AM</b> Date: 2/26/06 Time: 1035 Technician: JW	Control		1		7.5		18.5		30		7.82	0	0	0
	2.5				7.5		18.7		28		7.92	2	0	2
	5				7.5		18.8		28		7.93	74	0	74
	10				7.0		18.6		28		7.95	152	0	152
	15				4.1		18.6		28		7.97	287	0	287
	20				6.9		18.6		28		8.02	106	1/5	530
<b>Day 3</b> <b>PM</b> Date: 2/26/06 Time: 1630 Technician: JW	Control		2		7.6		18.5		30		7.79	1	0	1
	2.5				7.5		18.6		28		7.93	11	0	11
	5				7.5		18.7		27		7.97	92	0	92
	10				7.4		18.5		27		7.96	197	0	197
	15				7.2		18.6		27		8.03	130	1/5	650
	20				7.0		18.5		27		8.18	334	1/5	1670
<b>Day 4</b>  Date: 2/27/06  Time: 0910  Technician: GZ	Control		1		6.9		19.3		31		7.76	4	0	4
			2		7.0		19.3		31		7.78	3	0	3
			3		6.7		19.4		31		7.77	3	0	3
			4		7.0		19.1		31		7.79	6	0	6
	2.5		1		7.2		19.2		28		7.82	38	0	38
			2		6.9		19.1		28		7.85	29	0	29
			3		7.1		19.3		28		7.84	46	0	46
			4		6.9		19.2		28		7.82	42	0	42
	5		1		7.0		19.3		28		7.83	120	0	120
			2		6.8		19.3		28		7.83	125	0	125
			3		7.0		19.0		28		7.85	135	0	135
			4		6.8		19.2		28		7.82	126	0	126
	10		1		6.2		19.2		28		7.84	331	0	331
			2		7.0		19.1		28		7.91	355	0	355
			3		6.8		19.2		28		7.89	360	0	360
			4		6.8		19.2		28		7.88	290	0	290
	15		1		6.7		19.2		28		7.96	195	1/5	975
			2		6.4		19.2		28		7.96	215	1/5	1075
			3		6.5		19.1		28		7.98	216	1/5	1080
			4		6.5		19.0		28		8.00	208	1/5	1040
	20		1		5.8		19.1		28		8.10	409	1/5	2045
			2		5.6		19.1		28		8.08	382	1/5	1910
			3		5.7		19.2		28		8.07	330	1/5	1650
			4		6.1		19.3		28		8.09	384	1/5	1920

**Table F4. Acute *Ampelisca abdita*, Test 2 (23 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4	Mean	Percentage Survival			
		PM	AM	PM	AM	PM	AM	PM	AM		Day 1	Day 2	Day 3	Day 4
Control	1	0.5	0.5				0.0		0.0	<b>0.2</b>	100	100	ND	<b>100</b>
	2	0.5		0.4				0.5		<b>0.5</b>	100	100	ND	<b>80</b>
	3	0.3			0.0					<b>0.1</b>	100	100	ND	<b>80</b>
	4	0.5				0.1				<b>0.3</b>	100	90	ND	<b>50</b>
2.5	1	1.5	0.9				0.2		2.1	<b>1.2</b>	100	100	ND	<b>100</b>
	2	0.9		0.5				0.2		<b>0.5</b>	90	90	ND	<b>90</b>
	3	1.2			0.4					<b>0.8</b>	100**	100	ND	<b>100</b>
	4	1.6				0.2				<b>0.9</b>	90	80	ND	<b>80</b>
5	1	9.0	10.7				0.5*		6.1	<b>8.6</b>	100	100	ND	<b>100</b>
	2	8.4		5.4				0.6*		<b>6.9</b>	100**	100	ND	<b>100</b>
	3	10.2			1.1					<b>5.7</b>	100	100	ND	<b>100</b>
	4	9.0				1.8				<b>5.4</b>	100	90	ND	<b>50</b>
10	1	24.7	24.7				1.9*		8.6	<b>19.3</b>	100**	100**	ND	<b>100</b>
	2	23.6		14.4				1.7*		<b>19.0</b>	100	100	ND	<b>70</b>
	3	26.5			5.0					<b>15.8</b>	100	100	ND	<b>100</b>
	4	27.7				5.7				<b>16.7</b>	100	90	ND	<b>90</b>
15	1	35.1	31.2				4.7*		37.4	<b>34.6</b>	100**	100	ND	<b>100</b>
	2	59.2		35.8				4.8*		<b>47.5</b>	60	70	ND	<b>60</b>
	3	57.1			16.9					<b>37.0</b>	100	100	ND	<b>100</b>
	4	52.5				14.8				<b>33.6</b>	100	80	ND	<b>70</b>
20	1	78.3	63.3				13.6*		54.9	<b>65.5</b>	80	60	ND	<b>60</b>
	2	74.5		50.8				11.7*		<b>62.6</b>	90	90	ND	<b>80</b>
	3	78.8			26.7					<b>52.7</b>	100	100	ND	<b>100</b>
	4	70.5				13.5				<b>42.0</b>	100	90	ND	<b>80</b>

ND = No data

\* Sulfide concentrations decreased during test

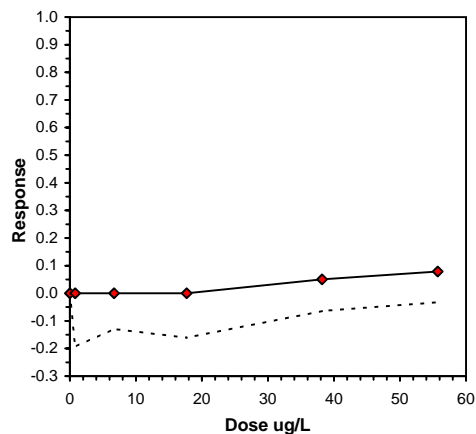
\*\* Survival miscounted

Acute Fish Test-96 Hr Survival				
Start Date:	2/23/2006	Test ID:	P060103.20	Sample ID:
End Date:	2/27/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPA/600/R	Test Species:
Comments:	Average			
Conc-ug/L	1	2	3	4
Control	1.0000	0.8000	0.8000	0.5000
0.8	1.0000	0.9000	1.0000	0.8000
6.7	1.0000	1.0000	1.0000	0.5000
17.7	1.0000	0.7000	1.0000	0.9000
38.2	1.0000	0.6000	1.0000	0.7000
55.7	0.6000	0.8000	1.0000	0.8000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%					Mean	N-Mean
Control	0.7750	1.0000	1.1029	0.7854	1.4120	23.198	4				0.8688	1.0000
0.8	0.9250	1.1935	1.2951	1.1071	1.4120	11.347	4	-1.128	2.410	0.4104	0.8688	1.0000
6.7	0.8750	1.1290	1.2554	0.7854	1.4120	24.958	4	-0.895	2.410	0.4104	0.8688	1.0000
17.7	0.9000	1.1613	1.2661	0.9912	1.4120	15.696	4	-0.958	2.410	0.4104	0.8688	1.0000
38.2	0.8250	1.0645	1.1753	0.8861	1.4120	23.539	4	-0.425	2.410	0.4104	0.8250	0.9496
55.7	0.8000	1.0323	1.1281	0.8861	1.4120	19.154	4	-0.148	2.410	0.4104	0.8000	0.9209

Auxiliary Tests					Statistic		Critical		Skew		Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.9401154		0.884		-0.530185		-0.625577
Bartlett's Test indicates equal variances (p = 0.88)					1.7626121		15.086272				
Hypothesis Test (1-tail, 0.05)		NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test		55.7	>55.7			0.3889244	0.4882226	0.0252639	0.0579903	0.8178229	5, 18
Treatments vs Control											

Point	ug/L	SD	Linear Interpolation (200 Resamples)	
			95% CL(Exp)	Skew
IC05	38.054			
IC10	>55.7			
IC15	>55.7			
IC20	>55.7			
IC25	>55.7			
IC40	>55.7			
IC50	>55.7			







## 4-Day Acute Toxicity Test

**Table F5. Acute *Ampelisca abdita*, Test 2 (23 Feb 2006)**

Weston Test ID:	P060103.20	Client:	Marine Research Specialists	Client Sample ID:	N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	ND	10			
	2		10	10	ND	8			
	3		10	10	ND	8			
	4		10	9	ND	5			
2.5	1		10	10	ND	10			
	2		9	9	ND	9			
	3		10**	10	ND	10			
	4		9	8	ND	8			
5	1		10	10	ND	10			
	2		10**	10	ND	10			
	3		10	10	ND	10			
	4		10	9	ND	5			
10	1		10**	10**	ND	10			
	2		10	10	ND	7			
	3		10	10	ND	10			
	4		10	9	ND	9			
15	1		10**	10	ND	10			
	2		6	7	ND	6			
	3		10	10	ND	10			
	4		10	8	ND	7			
20	1		8	6	ND	6			
	2		9	9	ND	8			
	3		10	10	ND	10			
	4		10	9	ND	8			
Date			2/24/06	2/25/06		2/27/06			
Time			1210	1530		1640			
Initials			GZ	BH/JW		TS/RE			

\*\* Survival miscounted

ND = No data



## 4-Day Acute Toxicity Test

**Table F6. Acute *Ampelisca abdita*, Test 2 (23 Feb 2006)**

Client	Marine Research Specialists	Date Received:	N/A
Project:	Hydrogen sulfide	Date Test Started:	23-Feb-06
Client Sample ID:	N/A	Date Test Ended:	27-Feb-06
Weston Test ID:	P060103.20	Study Director:	Brian Hester
Species:	<i>Ampelisca abdita</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	D.O. (mg/L)	Temp (°C)	Sal. (ppt)	pH	Total Sulfide (µg/L)		
								Value	Dil	Corr value
<b>Day 0</b>  Date: 2/23/06  Time: 0915  Technician: JM/GZ	Control		1	8.0	19.2	30	7.70	4	0	4
			2	7.9	19.6	29	7.82	5	0	5
			3	7.9	19.7	30	7.85	3	0	3
			4	7.9	19.6	30	7.85	5	0	5
	2.5		1	7.9	19.6	28	7.86	17	0	17
			2	7.9	19.5	28	7.88	10	0	10
			3	7.8	19.7	27	7.87	13	0	13
			4	7.9	19.6	28	7.87	18	0	18
	5		1	7.8	19.6	27	7.90	108	0	108
			2	7.8	19.4	27	7.91	103	0	103
			3	7.8	19.5	27	7.91	125	0	125
			4	7.8	19.6	28	7.90	109	0	109
	10		1	7.8	19.6	27	7.95	330	0	330
			2	7.8	19.6	27	7.94	308	0	308
			3	7.8	19.6	27	7.93	339	0	339
			4	7.7	19.8	27	7.94	362	0	362
	15		1	7.7	19.7	28	7.95	94	1/5	470
			2	7.8	19.6	27	7.95	158	1/5	790
			3	7.8	19.6	27	7.94	149	1/5	745
			4	7.7	19.5	27	7.94	137	1/5	685
	20		1	7.7	19.5	27	7.97	218	1/5	1090
			2	7.9	19.7	27	7.98	212	1/5	1060
			3	7.8	19.6	27	8.00	234	1/5	1170
			4	7.8	19.6	27	7.99	205	1/5	1025
<b>Day 1</b> <b>AM</b>  Date: 2/24/06  Time: 0916  Technician: JM	Control		1	7.8	17.8	30	7.70	4	0	4
	2.5			7.7	17.8	27	7.81	9	0	9
	5			7.7	17.9	28	7.88	124	0	124
	10			7.6	17.6	27	7.91	302	0	302
	15			7.6	17.7	28	7.94	82	1/5	410
	20			7.6	17.6	28	7.99	185	1/5	925
<b>Day 1</b> <b>PM</b>  Date: 2/24/06  Time: 1530  Technician: JM/GZ	0		2	7.6	18.8	29	7.73	3	0	3
	2.5			7.6	18.7	27	7.93	6	0	6
	5			7.5	18.9	27	7.99	79	0	79
	10			7.5	18.9	27	8.01	218	0	218
	15			7.5	18.8	27	8.04	116	1/5	580
	20			7.2	18.8	27	8.05	168	1/5	840
<b>Day 2</b> <b>AM</b>  Date: 2/25/06  Time: 0959  Technician: JW Feed Time:	0		3	7.5	19.1	30	7.87	0	0	0
	2.5			7.3	19.1	27	7.91	5	0	5
	5			7.2	19.0	27	7.94	15	0	15
	10			7.0	19.1	27	7.94	65	0	65
	15			6.7	19.0	27	7.95	45	1/5	225
	20			6.4	19.0	27	8.01	81	1/5	405



## 4-Day Acute Toxicity Test

**Table F6. Acute *Ampelisca abdita*, Test 2 (23 Feb 2006)**

Weston Test ID:	P060103.20	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	D.O. (mg/L)	Temp (°C)	Sal. (ppt)	pH	Total Sulfide (µg/L)		
								Value	Dil	Corr value
<b>Day 2</b> <b>PM</b> Date: 2/25/06 Time: 1720 Technician: BH	Control		4	7.5	18.3	30	7.90	1	0	1
	2.5			7.7	18.5	28	7.92	2	0	2
	5			7.5	18.5	28	7.96	24	0	24
	10			7.3	18.5	28	7.95	77	0	77
	15			7.1	18.3	28	8.02	46	1/5	230
	20			6.9	18.6	28	8.03	43	1/5	215
<b>Day 3</b> <b>AM</b> Date: 2/26/06 Time: 1035 Technician: JW	0		1	7.3	18.8	30	7.91	0	0	0
	2.5			7.5	18.8	28	7.93	2	0	2
	5			7.2	18.8	28	7.90	6	0	6
	10			6.6	18.7	28	7.91	24	0	24
	15			6.4	18.8	28	7.93	61	1/5	61
	20			6.2	18.7	28	7.98	39	1/5	195
<b>Day 3</b> <b>PM</b> Date: 2/26/06 Time: 1630 Technician: JW	0		2	7.4	18.5	30	7.87	6	0	6
	2.5			7.4	18.6	28	7.93	3	0	3
	5			7.4	18.5	27	7.93	8	0	8
	10			7.1	18.7	27	7.92	21	0	21
	15			6.5	18.5	27	7.96	13	1/5	65
	20			6.3	18.7	27	8.03	37	1/5	185
<b>Day 4</b> Date: 2/27/06 Time: 0910 Technician: GZ	Control		1	6.9	19.3	31	7.68	4	0	4
			2	7.0	19.4	31	7.78	4	0	4
			3	6.7	19.5	31	7.75	2	0	2
			4	7.1	19.1	31	7.81	3	0	3
	2.5		1	7.0	19.4	28	7.84	13	0	13
			2	6.8	19.1	28	7.82	10	0	10
			3	7.0	19.4	28	7.83	24	0	24
			4	6.8	19.3	28	7.81	10	0	10
	5		1	6.7	19.4	28	7.83	36	0	36
			2	6.5	19.4	28	7.80	44	0	44
			3	6.8	19.1	28	7.84	10	0	10
			4	6.6	19.3	28	7.81	16	0	16
	10		1	5.6	19.2	28	7.80	87	0	87
			2	6.4	19.3	28	7.88	72	0	72
			3	6.1	19.5	28	7.90	46	0	46
			4	6.7	19.3	28	7.88	63	0	63
	15		1	5.5	19.3	28	7.92	42	1/5	210
			2	5.6	19.3	28	7.92	35	1/5	175
			3	5.3	19.2	28	7.93	49	1/5	245
			4	5.4	19.1	28	7.95	44	1/5	220
	20		1	3.4	19.2	28	8.03	92	1/5	460
			2	3.3	19.2	28	8.00	101	1/5	505
			3	3.2	19.2	28	8.00	47	1/5	235
			4	3.9		28	8.03	57	1/5	285
<b>Day 4</b> <b>PM</b> Date: 2/27/06 Time: 1515 Technician: GZ/TS retest	Control		2	7.4	18.9	30	7.81	0	0	0
	2.5			7.3	19.2	28	7.89	25	0	25
	5			7.0	19.3	28	7.84	65	0	65
	10			6.4	18.9	28	7.84	91	0	91
	15			6.0	18.8	28	7.98	107	1/5	535
	20			6.4	18.8	28	8.08	195	1/5	975

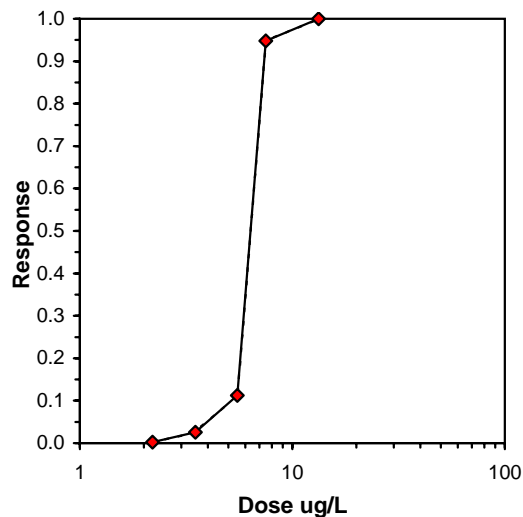
Bivalve Larval Survival and Development Test-Proportion Normal				
Start Date:	Test ID:	Biv T1	Sample ID:	
End Date:	Lab ID:	WESTON - Port Gamble	Sample Type:	
Sample Date:	Protocol:	ASTM 87	Test Species:	M. galloprovincialis
Comments:	Average			

Conc-ug/L	1	2	3	4
Control	1.0000	1.0000	1.0000	1.0000
2.2	0.9895	1.0000	1.0000	1.0000
3.5	0.9615	0.9714	0.9833	0.9810
5.5	0.9059	0.7753	0.9074	0.9515
7.5	0.0851	0.0612	0.0230	0.0337
13.3	0.0000	0.0000	0.0000	0.0000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%	N				
Control	1.0000	1.0000	1.5275	1.5261	1.5310	0.156	4			0	537
*2.2	0.9974	0.9974	1.5069	1.4680	1.5249	1.737	4	10.00	10.00	1	389
*3.5	0.9743	0.9743	1.4120	1.3734	1.4413	2.197	4	10.00	10.00	11	434
*5.5	0.8850	0.8850	1.2365	1.0769	1.3486	9.242	4	10.00	10.00	43	385
*7.5	0.0508	0.0508	0.2207	0.1522	0.2960	29.275	4	10.00	10.00	349	368
*13.3	0.0000	0.0000	0.0507	0.0486	0.0559	6.969	4	10.00	10.00	394	394

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.863901	0.884	-1.0164	4.647608
Bartlett's Test indicates unequal variances (p = 2.22E-06)	34.15617	15.08627		
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	<2.2	2.2		
Treatments vs Control				

Trimmed Spearman-Kärber			
Trim Level	EC50	95% CL	
0.0%			
5.0%	6.2866	6.2004	6.3741
10.0%	6.3489	6.2082	6.4927
20.0%	6.3515	6.3040	6.3994
Auto-0.3%	6.2252	6.1174	6.3350



**Table G.1 Water Quality Observations for Toxicity Test with *Mytilus* sp.**

Time	Test	Sample ID	Temperature (°C)	Salinity (‰)	pH
2/21/06 15:31	1	0	15.2	33	7.72
	1	2	14.8	33	7.76
	1	4	14.7	33	7.73
	1	6	14.7	33	7.73
	1	8	14.8	33	7.73
	1	15	14.7	33	7.74
2/21/06 16:20	1	0	15.2	33	7.72
	1	2	14.8	33	7.76
	1	4	14.7	33	7.73
	1	6	14.7	33	7.73
	1	8	14.8	33	7.73
	1	15	14.7	33	7.74
2/21/06 18:00	1	2	14.8	33	7.76
	1	4	14.7	33	7.73
	1	6	14.7	33	7.73
	1	8	14.8	33	7.73
	1	15	14.7	33	7.74
2/21/06 18:30	1	2	14.8	33	7.76
	1	4	14.7	33	7.73
	1	6	14.7	33	7.73
	1	8	14.8	33	7.73
	1	15	14.7	33	7.74
2/21/06 20:00	1	2	14.8	33	7.64
	1	4	14.7	33	7.64
	1	6	14.7	33	7.64
	1	8	14.8	33	7.66
	1	15	14.7	33	7.69
2/21/06 21:00	1	2	14.8	33	7.64
	1	4	14.7	33	7.64
	1	6	14.7	33	7.64
	1	8	14.8	33	7.66
	1	15	14.7	33	7.69
2/21/06 22:30	1	2	14.8	33	7.7
	1	4	14.7	33	7.71
	1	6	14.7	33	7.72
	1	8	14.8	33	7.74
	1	15	14.7	33	7.76
2/22/06 0:00	1	2	14.8	33	7.7
	1	4	14.7	33	7.71
	1	6	14.7	33	7.72
	1	8	14.8	33	7.74
	1	15	14.7	33	7.76
	1	2	14.8	33	7.7
2/22/06 1:30	1	4	14.7	33	7.71
	1	6	14.7	33	7.72
	1	8	14.8	33	7.74
	1	15	14.7	33	7.76
2/22/06 3:00	1	2	14.8	33	7.58
	1	4	14.7	33	7.64
	1	6	14.7	33	7.66
	1	8	14.8	33	7.67
	1	15	14.7	33	7.69
2/22/06 4:30	1	2	14.8	33	7.58
	1	4	14.7	33	7.64
	1	6	14.7	33	7.66
	1	8	14.8	33	7.67
	1	15	14.7	33	7.69

2/22/06 5:10	1	2	14.8	33	7.58
	1	4	14.7	33	7.64
	1	6	14.7	33	7.66
	1	8	14.8	33	7.67
	1	15	14.7	33	7.69
2/22/06 6:00	1	2	14.8	33	7.58
	1	4	14.7	33	7.64
	1	6	14.7	33	7.66
	1	8	14.8	33	7.67
	1	15	14.7	33	7.69
2/22/06 6:45	1	2	14.8	33	7.58
	1	4	14.7	33	7.64
	1	6	14.7	33	7.66
	1	8	14.8	33	7.67
	1	15	14.7	33	7.69
2/22/06 7:30	1	2	14.8	33	7.95
	1	4	14.7	33	7.96
	1	6	14.7	33	7.96
	1	8	14.8	33	7.97
	1	15	14.7	33	7.98
2/22/06 9:00	1	2	14.8	33	7.87
	1	4	14.7	33	7.94
	1	6	14.7	33	7.93
	1	8	14.8	33	7.94
	1	15	14.7	33	7.96
2/22/06 10:30	1	2	14.8	33	7.87
	1	4	14.7	33	7.94
	1	6	14.7	33	7.93
	1	8	14.8	33	7.94
	1	15	14.7	33	7.96
2/22/06 12:00	1	2	14.8	33	7.87
	1	4	14.7	33	7.94
	1	6	14.7	33	7.93
	1	8	14.8	33	7.94
	1	15	14.7	33	7.96
2/22/06 13:30	1	2	14.8	33	7.91
	1	4	14.7	33	7.92
	1	6	14.7	33	7.93
	1	8	14.8	33	7.92
	1	15	14.7	33	7.94
2/22/06 14:30	1	2	14.8	33	7.91
	1	4	14.7	33	7.92
	1	6	14.7	33	7.93
	1	8	14.8	33	7.92
	1	15	14.7	33	7.94
2/22/06 16:00	1	2	14.8	33	7.91
	1	4	14.7	33	7.92
	1	6	14.7	33	7.93
	1	8	14.8	33	7.92
	1	15	14.7	33	7.94
2/22/06 17:30	1	2	14.8	33	7.91
	1	4	14.7	33	7.92
	1	6	14.7	33	7.93
	1	8	14.8	33	7.92
	1	15	14.7	33	7.94
2/22/06 19:00	1	2	14.8	33	7.91
	1	4	14.7	33	7.92
	1	6	14.7	33	7.93
	1	8	14.8	33	7.92
	1	15	14.7	33	7.94

2/22/06 20:00	1	2	14.8	33	7.84
	1	4	14.7	33	7.84
	1	6	14.7	33	7.87
	1	8	14.8	33	7.9
	1	15	14.7	33	7.93
2/22/06 21:30	1	2	14.8	33	7.82
	1	4	14.7	33	7.85
	1	6	14.7	33	7.88
	1	8	14.8	33	7.9
	1	15	14.7	33	7.93
2/22/06 23:00	1	2	14.8	33	7.86
	1	4	14.7	33	7.88
	1	6	14.7	33	7.9
	1	8	14.8	33	7.91
	1	15	14.7	33	7.89
2/23/06 0:30	1	2	14.8	33	7.86
	1	4	14.7	33	7.9
	1	6	14.7	33	7.92
	1	8	14.8	33	7.94
	1	15	14.7	33	7.9
2/23/06 2:00	1	2	14.8	33	7.81
	1	4	14.7	33	7.87
	1	6	14.7	33	7.88
	1	8	14.8	33	7.9
	1	15	14.7	33	7.94
2/23/06 3:30	1	2	14.8	33	7.87
	1	4	14.8	33	7.87
	1	6	14.9	33	7.9
	1	8	14.9	33	7.91
	1	15	14.9	33	7.95
2/23/06 5:00	1	2	14.8	33	7.83
	1	4	14.8	33	7.9
	1	6	14.9	33	7.93
	1	8	14.9	33	7.96
	1	15	14.9	33	8
2/23/06 6:30	1	2	14.8	33	7.85
	1	4	14.8	33	7.91
	1	6	14.9	33	7.95
	1	8	14.9	33	7.98
	1	15	14.9	33	7.99
2/23/06 8:00	1	2	14.8	33	7.83
	1	4	14.8	33	7.88
	1	6	14.9	33	7.91
	1	8	14.9	33	7.94
	1	15	14.9	33	7.98
2/23/06 10:15	1	2	14.8	33	7.55
	1	4	14.8	33	7.58
	1	6	14.9	33	7.61
	1	8	14.9	33	7.65
	1	15	14.9	33	7.7
2/23/06 12:45	1	2	14.8	33	7.59
	1	4	14.8	33	7.61
	1	6	14.9	33	7.6
	1	8	14.9	33	7.67
	1	15	14.9	33	7.68

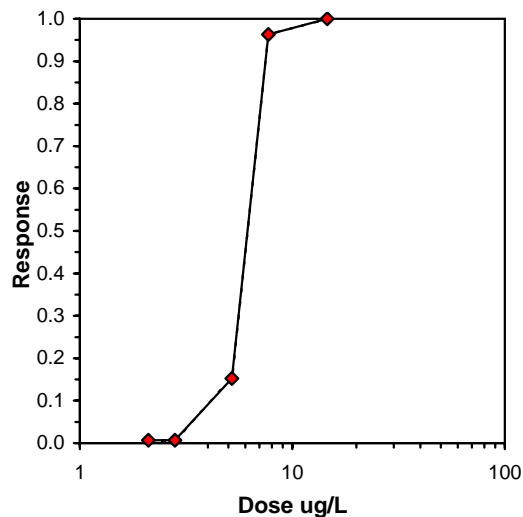
Bivalve Larval Survival and Development Test-Proportion Normal				
Start Date:	Test ID:	Biv T2	Sample ID:	
End Date:	Lab ID:	WESTON - Port Gamble	Sample Type:	
Sample Date:	Protocol:	ASTM 87	Test Species:	M. galloprovincialis
Comments:	Average			

Conc-ug/L	1	2	3	4
Control	1.0000	0.9915	1.0000	0.9910
2.1	0.9894	0.9701	1.0000	1.0000
2.8	0.9789	0.9855	1.0000	1.0000
5.2	0.8091	0.8425	0.8571	0.8795
7.7	0.0556	0.0505	0.0250	0.0185
14.6	0.0000	0.0000	0.0000	0.0000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%						
Control	0.9956	1.0000	1.5009	1.4757	1.5264	1.827	4				2	464
2.1	0.9899	0.9942	1.4640	1.3972	1.5186	3.425	4	1.348	2.410	0.0660	3	279
2.8	0.9911	0.9955	1.4706	1.4252	1.5062	2.679	4	1.107	2.410	0.0660	3	275
*5.2	0.8471	0.8508	1.1702	1.1186	1.2163	3.493	4	12.082	2.410	0.0660	63	404
*7.7	0.0374	0.0376	0.1900	0.1365	0.2379	26.283	4	47.900	2.410	0.0660	363	377
*14.6	0.0000	0.0000	0.0507	0.0481	0.0539	4.944	4	52.988	2.410	0.0660	391	391

Auxiliary Tests					Statistic	Critical	Skew	Kurt			
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.96754	0.884	-0.2648	-0.76906			
Bartlett's Test indicates equal variances (p = 0.02)					13.25373	15.08627					
Hypothesis Test (1-tail, 0.05)		NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test		2.8	5.2	3.815757		0.01347	0.013537	1.815865	0.001498	4.3E-22	5, 18
Treatments vs Control											

Trimmed Spearman-Kärber			
Trim Level	EC50	95% CL	
0.0%			
5.0%	6.0207	5.8918	6.1524
10.0%	6.1145	5.9425	6.2914
20.0%	6.1539	6.0885	6.2200
Auto-0.6%	5.9568	5.8307	6.0857





**Table G.1 Water Quality Observations for Toxicity Test with *Mytilus* sp.**

Time	Test	Sample ID	Temperature (°C)	Salinity (‰)	pH
2/21/06 15:31	2	0	14.9	33	7.7
	2	2	14.7	33	7.73
	2	4	14.6	33	7.72
	2	6	14.5	33	7.73
	2	8	14.5	33	7.74
	2	15	14.5	33	7.74
2/21/06 16:20	2	0	14.9	33	7.7
	2	2	14.7	33	7.73
	2	4	14.6	33	7.72
	2	6	14.5	33	7.73
	2	8	14.5	33	7.74
	2	15	14.5	33	7.74
2/21/06 18:00	2	2	14.7	33	7.73
	2	4	14.6	33	7.72
	2	6	14.5	33	7.73
	2	8	14.5	33	7.74
	2	15	14.5	33	7.74
2/21/06 20:00	2	2	14.7	33	7.65
	2	4	14.6	33	7.65
	2	6	14.5	33	7.66
	2	8	14.5	33	7.67
	2	15	14.5	33	7.69
2/21/06 21:00	2	2	14.7	33	7.65
	2	4	14.6	33	7.65
	2	6	14.5	33	7.66
	2	8	14.5	33	7.67
	2	15	14.5	33	7.69
2/21/06 22:30	2	2	14.7	33	7.7
	2	4	14.6	33	7.7
	2	6	14.5	33	7.72
	2	8	14.5	33	7.7
	2	15	14.5	33	7.74
2/22/06 0:00	2	2	14.7	33	7.7
	2	4	14.6	33	7.7
	2	6	14.5	33	7.72
	2	8	14.5	33	7.7
	2	15	14.5	33	7.74
2/22/06 1:30	2	2	14.7	33	7.7
	2	4	14.6	33	7.7
	2	6	14.5	33	7.72
	2	8	14.5	33	7.7
	2	15	14.5	33	7.74
2/22/06 3:00	2	2	14.7	33	7.63
	2	4	14.6	33	7.65
	2	6	14.5	33	7.65
	2	8	14.5	33	7.66
	2	15	14.5	33	7.68
2/22/06 4:30	2	2	14.7	33	7.63
	2	4	14.6	33	7.65
	2	6	14.5	33	7.65
	2	8	14.5	33	7.66
	2	15	14.5	33	7.68
2/22/06 5:10	2	2	14.7	33	7.63
	2	4	14.6	33	7.65
	2	6	14.5	33	7.65

	2	8	14.5	33	7.66
	2	15	14.5	33	7.68
2/22/06 6:00	2	2	14.7	33	7.63
	2	4	14.6	33	7.65
	2	6	14.5	33	7.65
	2	8	14.5	33	7.66
	2	15	14.5	33	7.68
2/22/06 6:45	2	2	14.7	33	7.63
	2	4	14.6	33	7.65
	2	6	14.5	33	7.65
	2	8	14.5	33	7.66
	2	15	14.5	33	7.68
2/22/06 7:30	2	2	14.7	33	7.95
	2	4	14.6	33	7.96
	2	6	14.5	33	7.96
	2	8	14.5	33	7.97
	2	15	14.5	33	7.98
2/22/06 9:00	2	2	14.7	33	7.91
	2	4	14.6	33	7.9
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.96
2/22/06 10:30	2	2	14.7	33	7.91
	2	4	14.6	33	7.9
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.96
2/22/06 12:00	2	2	14.7	33	7.91
	2	4	14.6	33	7.9
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.96
2/22/06 13:30	2	2	14.7	33	7.91
	2	4	14.6	33	7.92
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.95
2/22/06 14:30	2	2	14.7	33	7.91
	2	4	14.6	33	7.92
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.95
2/22/06 16:00	2	2	14.7	33	7.91
	2	4	14.6	33	7.92
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.95
2/22/06 17:30	2	2	14.7	33	7.91
	2	4	14.6	33	7.92
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.95
2/22/06 19:00	2	2	14.7	33	7.91
	2	4	14.6	33	7.92
	2	6	14.5	33	7.92
	2	8	14.5	33	7.93
	2	15	14.5	33	7.95

2/22/06 20:00	2	2	14.7	33	7.89
	2	4	14.6	33	7.89
	2	6	14.5	33	7.91
	2	8	14.5	33	7.93
	2	15	14.5	33	7.97
2/22/06 21:30	2	2	14.7	33	7.87
	2	4	14.6	33	7.85
	2	6	14.5	33	7.88
	2	8	14.5	33	7.91
	2	15	14.5	33	7.94
2/22/06 23:00	2	2	14.7	33	7.86
	2	4	14.6	33	7.86
	2	6	14.5	33	7.88
	2	8	14.5	33	7.91
	2	15	14.5	33	7.94
2/23/06 0:30	2	2	14.7	33	7.86
	2	4	14.6	33	7.85
	2	6	14.5	33	7.87
	2	8	14.5	33	7.9
	2	15	14.5	33	7.94
2/23/06 2:00	2	2	14.7	33	7.84
	2	4	14.6	33	7.88
	2	6	14.5	33	7.89
	2	8	14.5	33	7.91
	2	15	14.5	33	7.94
2/23/06 3:30	2	2	15	33	7.88
	2	4	15	33	7.88
	2	6	14.9	33	7.91
	2	8	14.9	33	7.96
	2	15	14.9	33	7.99
2/23/06 5:00	2	2	15	33	7.89
	2	4	15	33	7.9
	2	6	14.9	33	7.93
	2	8	14.9	33	7.96
	2	15	14.9	33	8
2/23/06 6:30	2	2	15	33	7.86
	2	4	15	33	7.89
	2	6	14.9	33	7.92
	2	8	14.9	33	7.96
	2	15	14.9	33	7.98
2/23/06 8:00	2	2	15	33	7.83
	2	4	15	33	7.86
	2	6	14.9	33	7.9
	2	8	14.9	33	7.92
	2	15	14.9	33	7.98
2/23/06 10:15	2	2	15	33	7.6
	2	4	15	33	7.58
	2	6	14.9	33	7.64
	2	8	14.9	33	7.67
	2	15	14.9	33	7.73
2/23/06 12:45	2	2	15	33	7.6
	2	4	15	33	7.59
	2	6	14.9	33	7.63
	2	8	14.9	33	7.67
	2	15	14.9	33	7.69

Bivalve Larval Survival and Development Test-Proportion Normal			
Start Date:	Test ID: 0 Hour	Sample ID:	
End Date:	Lab ID: PGL- Port Gamble Laboratory	Sample Type:	
Sample Date:	Protocol: ASTM 87	Test Species:	CG-Crassostrea gigas
Comments:			

Conc-ug/L	1	2	3
Control	0.7900	0.8400	0.7800
1.68	0.7900	0.7800	0.7600
1.92	0.7700	0.8000	0.8100
4.07	0.8100	0.7800	0.7500
6.78	0.4500	0.2800	0.4700
7.7	0.2700	0.2700	0.2600
10.3	0.0000	0.0000	0.0100
16.9	0.0000	0.0000	0.0000
18.9	0.0000	0.0000	0.0000

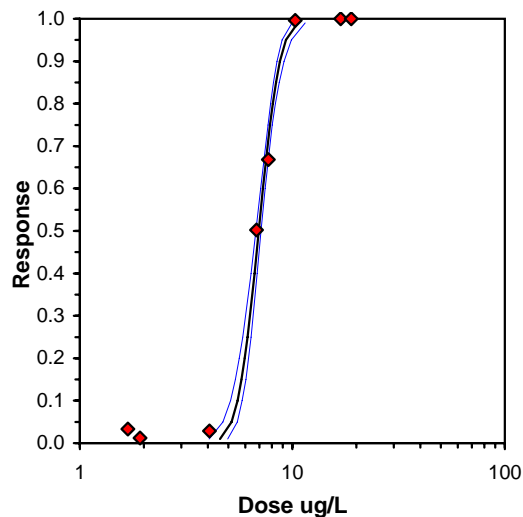
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%	N					
Control	0.8033	1.0000	1.1122	1.0826	1.1593	3.706	3				59	300
1.68	0.7767	0.9668	1.0787	1.0588	1.0948	1.694	3	0.951	2.580	0.0908	67	300
1.92	0.7933	0.9876	1.0992	1.0706	1.1198	2.322	3	0.370	2.580	0.0908	62	300
4.07	0.7800	0.9710	1.0832	1.0472	1.1198	3.350	3	0.824	2.580	0.0908	66	300
*6.78	0.4000	0.4979	0.6828	0.5576	0.7554	15.944	3	12.198	2.580	0.0908	180	300
*7.7	0.2667	0.3320	0.5426	0.5351	0.5464	1.205	3	16.179	2.580	0.0908	220	300
*10.3	0.0033	0.0041	0.0667	0.0500	0.1002	43.383	3	29.696	2.580	0.0908	299	300
*16.9	0.0000	0.0000	0.0500	0.0500	0.0500	0.000	3	30.171	2.580	0.0908	300	300
*18.9	0.0000	0.0000	0.0500	0.0500	0.0500	0.000	3	30.171	2.580	0.0908	300	300

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.01)	0.87461	0.894	-1.21221	5.178625
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	4.07	6.78	5.253056		0.076717	0.095415	0.697054	0.001859	2.1E-18	8, 18
Treatments vs Control										

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	12.79722	1.234702	10.3772	15.21723	0.196667	6.877827	12.59159	0.33	0.841796	0.078142	7
Intercept	-5.77265	1.078852	-7.8872	-3.6581							
TSCR	0.212251	0.011851	0.189023	0.235478							

Point	Probits	ug/L	95% Fiducial Limits	
EC01	2.674	4.570997	4.064858	4.956049
EC05	3.355	5.167307	4.724412	5.499076
EC10	3.718	5.51637	5.117386	5.813978
EC15	3.964	5.765108	5.399857	6.037652
EC20	4.158	5.970771	5.634543	6.222527
EC25	4.326	6.15305	5.843059	6.386661
EC40	4.747	6.637421	6.396053	6.827487
EC50	5.000	6.946985	6.743637	7.117727
EC60	5.253	7.270988	7.094882	7.43623
EC75	5.674	7.843364	7.665391	8.054201
EC80	5.842	8.082809	7.886126	8.332623
EC85	6.036	8.371154	8.142611	8.678745
EC90	6.282	8.748616	8.468199	9.144465
EC95	6.645	9.339605	8.963982	9.8931
EC99	7.326	10.558	9.956252	11.48667



Bivalve Larval Survival and Development Test-Proportion Normal			
Start Date:	Test ID: 6 Hour	Sample ID:	
End Date:	Lab ID: PGL- Port Gamble Laboratory	Sample Type:	
Sample Date:	Protocol: ASTM 87	Test Species:	CG-Crassostrea gigas
Comments:			

Conc-ug/L	1	2	3
Control	0.8200	0.7900	0.7700
6.45	0.7700	0.8100	0.8200
7.81	0.6500	0.7300	0.8200
9.09	0.6300	0.6800	0.7600
12.12	0.6100	0.4600	0.5100
12.81	0.4900	0.4600	0.5200
14.11	0.4900	0.4000	0.3400
17.73	0.4800	0.4200	0.4600
18.4	0.4100	0.4900	0.3800

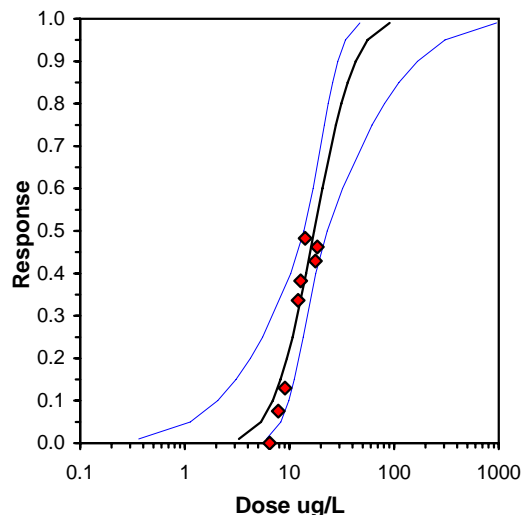
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%	N					
Control	0.7933	1.0000	1.0993	1.0706	1.1326	2.844	3				62	300
6.45	0.8000	1.0084	1.1077	1.0706	1.1326	2.955	3	-0.167	2.580	0.1289	60	300
7.81	0.7333	0.9244	1.0316	0.9377	1.1326	9.466	3	1.356	2.580	0.1289	80	300
9.09	0.6900	0.8697	0.9818	0.9169	1.0588	7.308	3	2.354	2.580	0.1289	93	300
*12.12	0.5267	0.6639	0.8124	0.7454	0.8963	9.465	3	5.744	2.580	0.1289	142	300
*12.81	0.4900	0.6176	0.7754	0.7454	0.8054	3.872	3	6.484	2.580	0.1289	153	300
*14.11	0.4100	0.5168	0.6942	0.6225	0.7754	11.073	3	8.109	2.580	0.1289	177	300
*17.73	0.4533	0.5714	0.7386	0.7051	0.7654	4.161	3	7.221	2.580	0.1289	164	300
*18.4	0.4267	0.5378	0.7115	0.6642	0.7754	8.070	3	7.763	2.580	0.1289	172	300

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.968586	0.894	0.299597	-0.46632
Bartlett's Test indicates equal variances ( $p = 0.68$ )	5.66291	20.09023		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	9.09	12.12	10.49623		0.112867	0.142201	0.086845	0.003744	5.7E-08	8, 18
Treatments vs Control										

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	3.232935	0.762337	1.367564	5.098307	0.206667	27.04121	12.59159	1.4E-04	1.236193	0.309316	12
Intercept	1.003469	0.870977	-1.12773	3.134673							
TSCR	0.181131	0.045459	0.069897	0.292364							
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	3.285602	0.361834	6.037444							
EC05	3.355	5.338415	1.128891	8.292933							
EC10	3.718	6.914928	2.06186	9.863169							
EC15	3.964	8.233922	3.084588	11.12741							
EC20	4.158	9.459438	4.232008	12.29441							
EC25	4.326	10.65521	5.524378	13.45748							
EC40	4.747	14.38229	10.27706	17.78007							
EC50	5.000	17.22632	13.70425	22.90307							
EC60	5.253	20.63275	16.78053	32.12847							
EC75	5.674	27.84988	21.4652	61.73282							
EC80	5.842	31.37039	23.39342	80.93757							
EC85	6.036	36.03948	25.76879	111.3815							
EC90	6.282	42.91385	29.0078	166.9965							
EC95	6.645	55.58694	34.44032	305.5419							
EC99	7.326	90.31716	47.23997	954.6112							

Significant heterogeneity detected ( $p = 1.42E-04$ )



Bivalve Larval Survival and Development Test-Proportion Normal			
Start Date:	Test ID: 12 Hour	Sample ID:	
End Date:	Lab ID: PGL- Port Gamble Laboratory	Sample Type:	
Sample Date:	Protocol: ASTM 87	Test Species:	CG-Crassostrea gigas
Comments:			

Conc-ug/L	1	2	3
Control	0.7300	0.7600	0.7200
0.55	0.6200	0.5800	0.6400
3.06	0.6200	0.7100	0.7000
3.79	0.5900	0.6300	0.7200
8.89	0.5800	0.6800	0.7500
10.71	0.7000	0.6739	0.6200
12.81	0.7400	0.6900	0.7200
17.84	0.6400	0.7800	0.6600
23.56	0.7100	0.7600	0.7000

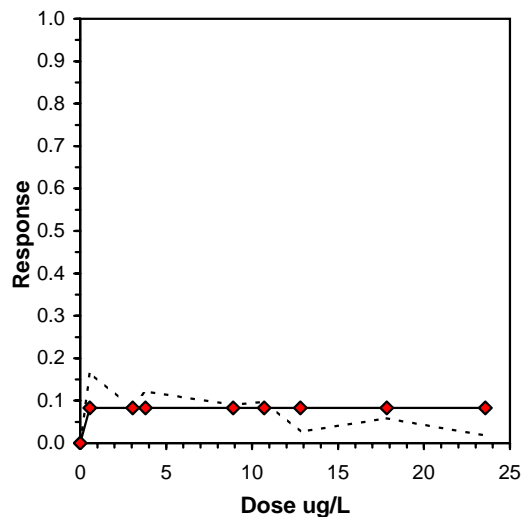
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
Control	0.7367	1.0000	1.0321	1.0132	1.0588	2.304	3				0.7367	1.0000
*0.55	0.6133	0.8326	0.8999	0.8657	0.9273	3.480	3	2.874	2.580	0.1187	0.6755	0.9170
3.06	0.6767	0.9186	0.9666	0.9066	1.0021	5.409	3	1.424	2.580	0.1187	0.6755	0.9170
3.79	0.6467	0.8778	0.9353	0.8759	1.0132	7.536	3	2.104	2.580	0.1187	0.6755	0.9170
8.89	0.6700	0.9095	0.9608	0.8657	1.0472	9.475	3	1.550	2.580	0.1187	0.6755	0.9170
10.71	0.6646	0.9022	0.9536	0.9066	0.9912	4.517	3	1.707	2.580	0.1187	0.6755	0.9170
12.81	0.7167	0.9729	1.0097	0.9803	1.0357	2.761	3	0.487	2.580	0.1187	0.6755	0.9170
17.84	0.6933	0.9412	0.9860	0.9273	1.0826	8.545	3	1.002	2.580	0.1187	0.6755	0.9170
23.56	0.7233	0.9819	1.0174	0.9912	1.0588	3.570	3	0.321	2.580	0.1187	0.6755	0.9170

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.978277	0.894	0.206268	-0.20663
Bartlett's Test indicates equal variances ( $p = 0.63$ )	6.184668	20.09023		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	23.56	>23.56			0.110225	0.149589	0.005368	0.003176	0.168945	8, 18
Treatments vs Control										

Point	ug/L	SD	Linear Interpolation (200 Resamples)	
			95% CL(Exp)	Skew
IC05*	0.3315			
IC10	>23.56			
IC15	>23.56			
IC20	>23.56			
IC25	>23.56			
IC40	>23.56			
IC50	>23.56			

\* indicates IC estimate less than the lowest concentration



Bivalve Larval Survival and Development Test-Proportion Normal			
Start Date:	Test ID: 24 Hour	Sample ID:	
End Date:	Lab ID: PGL- Port Gamble Laboratory	Sample Type:	
Sample Date:	Protocol: ASTM 87	Test Species:	CG-Crassostrea gigas
Comments:			

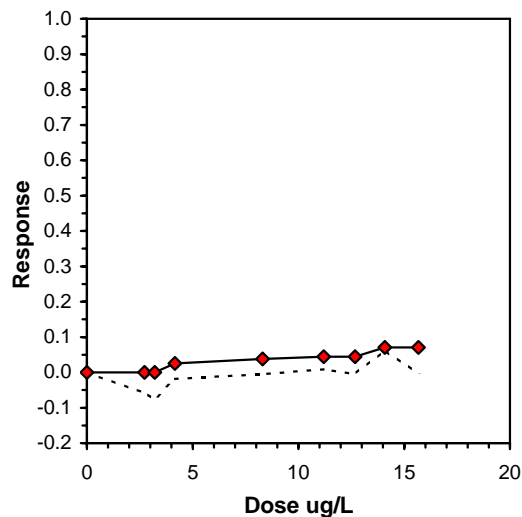
Conc-ug/L	1	2	3
Control	0.7600	0.7400	0.7500
2.73	0.7900	0.7800	0.8100
3.2	0.7700	0.8200	0.8300
4.16	0.7700	0.8300	0.6900
8.3	0.7300	0.7200	0.8100
11.2	0.7300	0.7700	0.7300
12.68	0.7700	0.7900	0.7000
14.08	0.6400	0.6900	0.7800
15.68	0.7000	0.7500	0.8100

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
Control	0.7500	1.0000	1.0472	1.0357	1.0588	1.103	3				0.7833	1.0000
2.73	0.7933	1.0578	1.0990	1.0826	1.1198	1.725	3	-1.170	2.580	0.1142	0.7833	1.0000
3.2	0.8067	1.0756	1.1164	1.0706	1.1458	3.597	3	-1.561	2.580	0.1142	0.7833	1.0000
4.16	0.7633	1.0178	1.0656	0.9803	1.1458	7.777	3	-0.414	2.580	0.1142	0.7633	0.9745
8.3	0.7533	1.0044	1.0525	1.0132	1.1198	5.565	3	-0.118	2.580	0.1142	0.7533	0.9617
11.2	0.7433	0.9911	1.0398	1.0244	1.0706	2.566	3	0.168	2.580	0.1142	0.7483	0.9553
12.68	0.7533	1.0044	1.0522	0.9912	1.0948	5.152	3	-0.111	2.580	0.1142	0.7483	0.9553
14.08	0.7033	0.9378	0.9967	0.9273	1.0826	7.920	3	1.142	2.580	0.1142	0.7283	0.9298
15.68	0.7533	1.0044	1.0527	0.9912	1.1198	6.125	3	-0.123	2.580	0.1142	0.7283	0.9298

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.974191	0.894	0.134843	-0.40634
Bartlett's Test indicates equal variances ( $p = 0.38$ )	8.619522	20.09023		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	15.68	>15.68			0.104516	0.139346	0.00354	0.002938	0.349997	8, 18
Treatments vs Control										

Point	ug/L	SD	Linear Interpolation (200 Resamples)	
			95% CL(Exp)	Skew
IC05	12.972			
IC10	>15.68			
IC15	>15.68			
IC20	>15.68			
IC25	>15.68			
IC40	>15.68			
IC50	>15.68			



Bivalve Larval Survival and Development Test-Proportion Normal			
Start Date:	Test ID: 24 Hour	Sample ID:	
End Date:	Lab ID: PGL- Port Gamble Laboratory	Sample Type:	
Sample Date:	Protocol: ASTM 87	Test Species:	CG-Crassostrea gigas
Comments:			

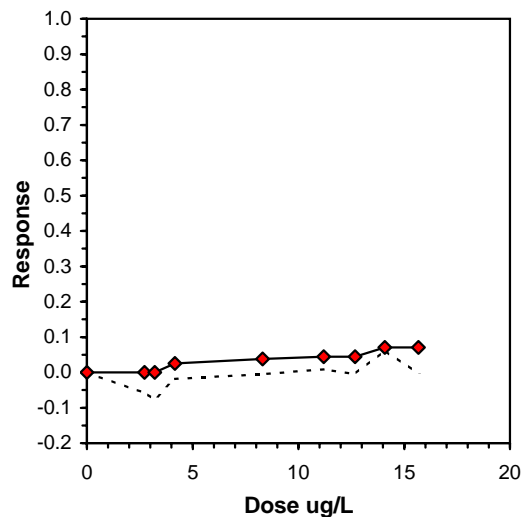
Conc-ug/L	1	2	3
Control	0.7600	0.7400	0.7500
2.73	0.7900	0.7800	0.8100
3.2	0.7700	0.8200	0.8300
4.16	0.7700	0.8300	0.6900
8.3	0.7300	0.7200	0.8100
11.2	0.7300	0.7700	0.7300
12.68	0.7700	0.7900	0.7000
14.08	0.6400	0.6900	0.7800
15.68	0.7000	0.7500	0.8100

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
Control	0.7500	1.0000	1.0472	1.0357	1.0588	1.103	3				0.7833	1.0000
2.73	0.7933	1.0578	1.0990	1.0826	1.1198	1.725	3	-1.170	2.580	0.1142	0.7833	1.0000
3.2	0.8067	1.0756	1.1164	1.0706	1.1458	3.597	3	-1.561	2.580	0.1142	0.7833	1.0000
4.16	0.7633	1.0178	1.0656	0.9803	1.1458	7.777	3	-0.414	2.580	0.1142	0.7633	0.9745
8.3	0.7533	1.0044	1.0525	1.0132	1.1198	5.565	3	-0.118	2.580	0.1142	0.7533	0.9617
11.2	0.7433	0.9911	1.0398	1.0244	1.0706	2.566	3	0.168	2.580	0.1142	0.7483	0.9553
12.68	0.7533	1.0044	1.0522	0.9912	1.0948	5.152	3	-0.111	2.580	0.1142	0.7483	0.9553
14.08	0.7033	0.9378	0.9967	0.9273	1.0826	7.920	3	1.142	2.580	0.1142	0.7283	0.9298
15.68	0.7533	1.0044	1.0527	0.9912	1.1198	6.125	3	-0.123	2.580	0.1142	0.7283	0.9298

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.974191	0.894	0.134843	-0.40634
Bartlett's Test indicates equal variances ( $p = 0.38$ )	8.619522	20.09023		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	15.68	>15.68			0.104516	0.139346	0.00354	0.002938	0.349997	8, 18
Treatments vs Control										

Point	ug/L	SD	Linear Interpolation (200 Resamples)	
			95% CL(Exp)	Skew
IC05	12.972			
IC10	>15.68			
IC15	>15.68			
IC20	>15.68			
IC25	>15.68			
IC40	>15.68			
IC50	>15.68			





Bivalve Larval Survival and Development Test-Proportion Normal			
Start Date:	Test ID: 36 Hour	Sample ID:	
End Date:	Lab ID: PGL- Port Gamble Laboratory	Sample Type:	
Sample Date:	Protocol: ASTM 87	Test Species:	CG-Crassostrea gigas
Comments:			

Conc-ug/L	1	2	3
Control	0.8300	0.8500	0.8100
3.09	0.7800	0.7900	0.7300
4.85	0.7800	0.8300	0.7600
7.65	0.7800	0.7500	0.8100
10.78	0.6900	0.7700	0.7200
12.12	0.7500	0.7900	0.7900
17.09	0.8000	0.8100	0.7700
22.72	0.7900	0.7400	0.8300
34.96	0.6900	0.7800	0.7800

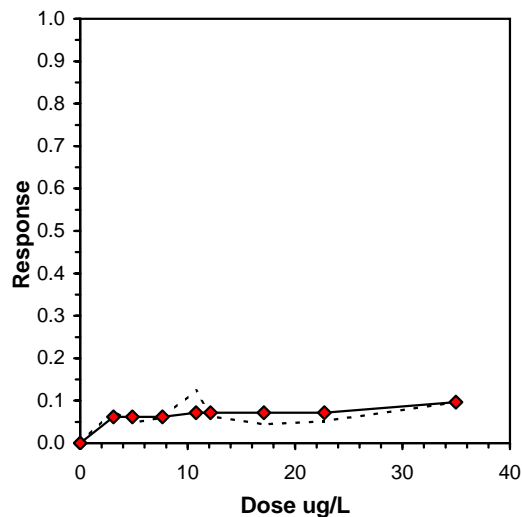
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
Control	0.8300	1.0000	1.1462	1.1198	1.1731	2.326	3				0.8300	1.0000
3.09	0.7667	0.9237	1.0672	1.0244	1.0948	3.524	3	2.332	2.580	0.0874	0.7789	0.9384
4.85	0.7900	0.9518	1.0957	1.0588	1.1458	4.103	3	1.491	2.580	0.0874	0.7789	0.9384
7.65	0.7800	0.9398	1.0832	1.0472	1.1198	3.350	3	1.861	2.580	0.0874	0.7789	0.9384
*10.78	0.7267	0.8755	1.0214	0.9803	1.0706	4.476	3	3.687	2.580	0.0874	0.7708	0.9287
12.12	0.7767	0.9357	1.0789	1.0472	1.0948	2.545	3	1.988	2.580	0.0874	0.7708	0.9287
17.09	0.7933	0.9558	1.0992	1.0706	1.1198	2.322	3	1.389	2.580	0.0874	0.7708	0.9287
22.72	0.7867	0.9478	1.0921	1.0357	1.1458	5.044	3	1.598	2.580	0.0874	0.7708	0.9287
*34.96	0.7500	0.9036	1.0485	0.9803	1.0826	5.633	3	2.886	2.580	0.0874	0.7500	0.9036

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.957752	0.894	-0.23101	-0.98389
Bartlett's Test indicates equal variances ( $p = 0.96$ )	2.55022	20.09023		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	22.72	34.96	28.18317		0.070287	0.084651	0.00365	0.00172	0.088202	8, 18
Treatments vs Control										

Linear Interpolation (200 Resamples)				
Point	ug/L	SD	95% CL(Exp)	Skew
IC05*	2.5089			
IC10	>34.96			
IC15	>34.96			
IC20	>34.96			
IC25	>34.96			
IC40	>34.96			
IC50	>34.96			

\* indicates IC estimate less than the lowest concentration



**Table G.1 Water Quality Observations for Toxicity Test with *Mytilus* sp.**

Time	Sample ID	Temperature (°C)	Salinity (‰)	pH
3.23.06 01:00	4	15.6	31	7.92
	6	15.6	31	7.92
	8	15.6	31	7.92
	10	15.6	31	7.92
	12	15.6	31	8.02
	14	15.6	31	8.00
	16	15.6	31	8.04
	20	15.6	31	8.06
3.23.06 07:30	4	16	32	7.93
	6	16.1	32	7.93
	8	16.1	32	7.92
	10	16.1	32	7.91
	12	16.1	32	7.91
	14	16	32	7.91
	16	16	32	7.91
	20	16	32	7.91
3.23.06 1900	4	15	31	8.02
	6	15	31	8.02
	8	14.9	31	8.02
	10	14.9	31	8.01
	12	14.9	31	8.00
	14	15	31	8.00
	16	15	31	8.01
	20	14.9	31	8.02
3.23.06 1900	4	15.2	31	7.58
	6	15.3	31	7.71
	8	15.3	31	7.75
	10	15.3	31	7.76
	12	15.3	31	7.77
	14	15.3	31	7.77
	16	15.3	31	7.77
	20	15.2	31	7.79
3.23.06 07:30	4	15.1	31	7.85
	6	15	31	7.85
	8	15	31	7.86
	10	15.1	31	7.86
	12	15	31	7.85
	14	15.1	31	7.86
	16	15.2	31	7.87
	20	15.1	31	7.89

**Table G.1 Water Quality Observations for Toxicity Test with *Haliotis* sp.**

Time	Sample ID	Temperature (°C)	Salinity (‰)	pH
4.4.06 15:00	4	15.7	31	7.77
	6	15.7	31	7.78
	8	15.8	31	7.8
	10	15.8	31	7.8
	12	15.7	31	7.82
	14	15.7	31	7.82
	16	15.6	31	7.84
	20	15.7	31	7.84
4.4.06 21:00	4	15.8	31	7.78
	6	15.5	31	7.81
	8	15.4	31	7.82
	10	15.5	31	7.82
	12	15.6	31	7.93
	14	15.6	31	7.84
	16	15.5	31	7.84
	20	15.5	31	7.852
4.5.06 03:0	4	15	32	7.8
	6	15	32	7.85
	8	15.2	32	7.88
	10	15.1	32	7.9
	12	15	32	7.9
	14	15	32	7.91
	16	14.9	32	7.88
	20	15	32	7.9
4.5.06 15:00	4	14.7	32	7.84
	6	14.9	32	7.88
	8	15.2	32	7.57
	10	15.4	32	7.83
	12	15.3	32	7.81
	14	15.3	32	7.8
	16	15.2	32	7.8
	20	15.1	32	7.79
4.6.06 03:00	4	15.3	31	7.89
	6	15.4	31	7.88
	8	15.4	31	7.88
	10	15.4	31	7.87
	12	15.4	31	7.87
	14	15.4	31	7.86
	16	15.3	31	7.86
	20	15.3	31	7.86

# **Abalone Larval Development Test-Proportion Normal**

Start Date:	Test ID: AB Hour 0	Sample ID:
End Date:	Lab ID: WESTON	Sample Type:
Sample Date:	Protocol: MBP 96-Anderson et al.	Test Species: HR-Haliotis rufescens
Comments:		

Conc-ug/L	1	2	3
5.11	0.9000	0.9000	0.9200
6.38	0.9000	0.9200	0.8800
7.7	0.8900	0.8800	0.8400
13.7	0.8700	0.8300	0.7600
16	0.7600	0.7500	0.7500
20.5	0.5900	0.6200	0.5500
20.6	0.6500	0.7200	0.6400
29.3	0.3100	0.3900	0.4300

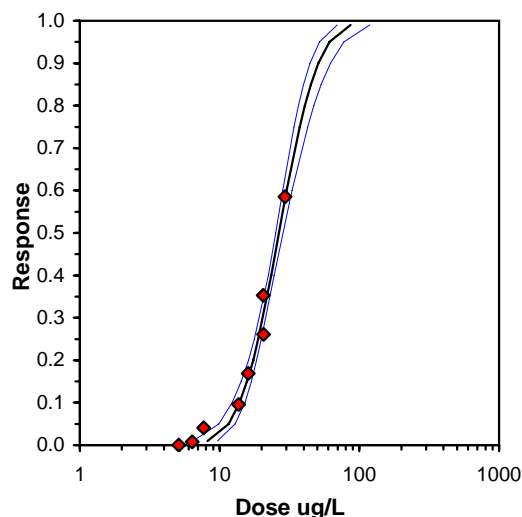
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%	N					
5.11	0.9067	1.0000	1.2607	1.2490	1.2840	1.603	3				28	300
6.38	0.9000	0.9926	1.2500	1.2171	1.2840	2.680	3	0.294	2.560	0.0928	30	300
7.7	0.8700	0.9596	1.2030	1.1593	1.2327	3.216	3	1.591	2.560	0.0928	39	300
*13.7	0.8200	0.9044	1.1355	1.0588	1.2019	6.350	3	3.452	2.560	0.0928	54	300
*16	0.7533	0.8309	1.0511	1.0472	1.0588	0.639	3	5.781	2.560	0.0928	74	300
*20.5	0.5867	0.6471	0.8727	0.8355	0.9066	4.086	3	10.701	2.560	0.0928	124	300
*20.6	0.6700	0.7390	0.9594	0.9273	1.0132	4.885	3	8.308	2.560	0.0928	99	300
*29.3	0.3767	0.4154	0.6601	0.5905	0.7152	9.632	3	16.563	2.560	0.0928	187	300

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.980081	0.884	-0.24692	-0.22814
Bartlett's Test indicates equal variances ( $p = 0.32$ )	8.151669	18.47531		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	7.7	13.7	10.27083		0.06064	0.066866	0.131511	0.001973	1.2E-10	7, 16
Treatments vs 5.11										

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	4.547485	0.433998	3.696849	5.39812	0.093333	6.791379	12.59159	0.34	1.424924	0.219902	5
Intercept	-1.47982	0.577904	-2.61251	-0.34713							
TSCR	0.100888	0.0093	0.08266	0.119116							

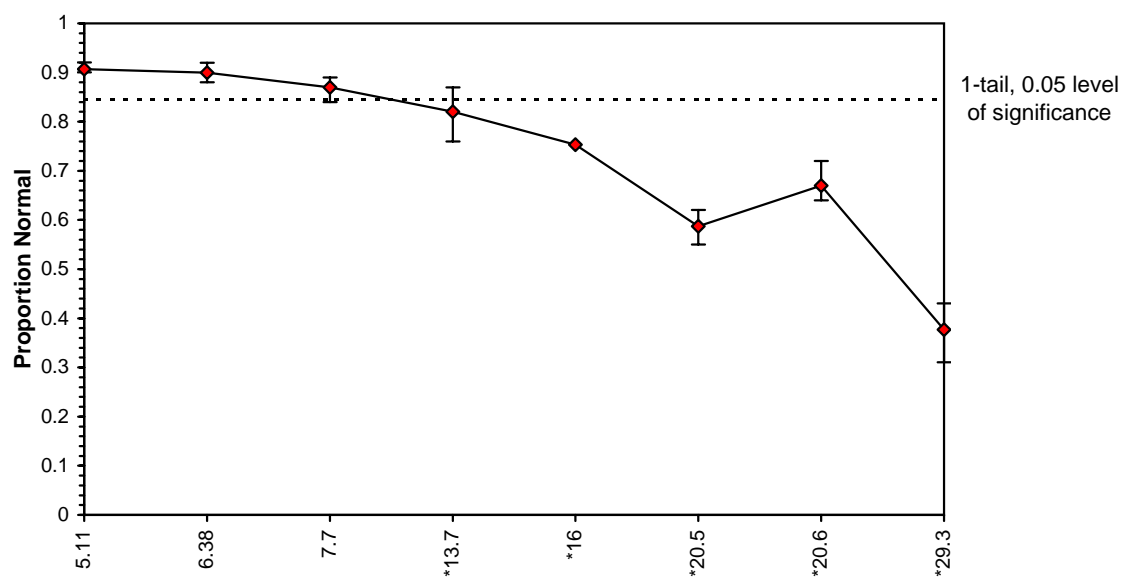
Point	Probits	ug/L	95% Fiducial Limits	
EC01	2.674	8.191366	6.467041	9.649375
EC05	3.355	11.56693	9.856472	12.94418
EC10	3.718	13.90301	12.31963	15.16259
EC15	3.964	15.74022	14.29912	16.89563
EC20	4.158	17.37202	16.06935	18.44468
EC25	4.326	18.90614	17.72355	19.92916
EC40	4.747	23.39984	22.2941	24.64822
EC50	5.000	26.60262	25.21267	28.4328
EC60	5.253	30.24377	28.31236	33.03129
EC75	5.674	37.43225	34.10101	42.66485
EC80	5.842	40.73788	36.67461	47.27617
EC85	6.036	44.96121	39.90457	53.30534
EC90	6.282	50.90259	44.35756	62.02095
EC95	6.645	61.18298	51.86035	77.66862
EC99	7.326	86.39578	69.46141	118.5575



### Abalone Larval Development Test-Proportion Normal

Start Date:	Test ID: AB Hour 0	Sample ID:
End Date:	Lab ID: WESTON	Sample Type:
Sample Date:	Protocol: MBP 96-Anderson et al.	Test Species: HR-Haliotis rufescens
Comments:		

Dose-Response Plot



### Abalone Larval Development Test-Proportion Normal

Start Date: Test ID: AB Hour 6 Sample ID:  
 End Date: Lab ID: WESTON Sample Type:  
 Sample Date: Protocol: MBP 96-Anderson et al. Test Species: HR-Haliotis rufescens  
 Comments:

Conc-ug/L	1	2	3
2.57	0.8500	0.8600	0.8900
6.61	0.8500	0.8700	0.8800
7.92	0.8800	0.8300	0.8300
9.15	0.8800	0.8200	0.8100
10.9	0.8900	0.8900	0.8600
11.5	0.8600	0.8200	0.7900
12.6	0.8500	0.8700	0.8100
14.8	0.8200	0.8200	0.8800

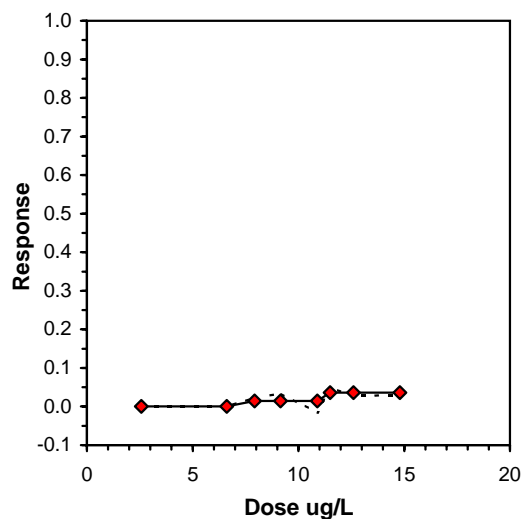
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
2.57	0.8667	1.0000	1.1977	1.1731	1.2327	2.601	3				0.8667	1.0000
6.61	0.8667	1.0000	1.1974	1.1731	1.2171	1.865	3	0.011	2.560	0.0840	0.8667	1.0000
7.92	0.8467	0.9769	1.1696	1.1458	1.2171	3.517	3	0.858	2.560	0.0840	0.8544	0.9859
9.15	0.8367	0.9654	1.1565	1.1198	1.2171	4.569	3	1.256	2.560	0.0840	0.8544	0.9859
10.9	0.8800	1.0154	1.2176	1.1873	1.2327	2.154	3	-0.606	2.560	0.0840	0.8544	0.9859
11.5	0.8233	0.9500	1.1382	1.0948	1.1873	4.087	3	1.813	2.560	0.0840	0.8356	0.9641
12.6	0.8433	0.9731	1.1649	1.1198	1.2019	3.578	3	0.999	2.560	0.0840	0.8356	0.9641
14.8	0.8400	0.9692	1.1608	1.1326	1.2171	4.198	3	1.125	2.560	0.0840	0.8356	0.9641

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.91105	0.884	0.462908	-1.13795
Bartlett's Test indicates equal variances (p = 0.96)	2.005853	18.47531		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	14.8	>14.8			0.061919	0.071406	0.002081	0.001615	0.316574	7, 16
Treatments vs 2.57										

### Linear Interpolation (200 Resamples)

Point	ug/L	SD	95% CL(Exp)	Skew
IC05	>14.8			
IC10	>14.8			
IC15	>14.8			
IC20	>14.8			
IC25	>14.8			
IC40	>14.8			
IC50	>14.8			



# **Abalone Larval Development Test-Proportion Normal**

Start Date:	Test ID: AB Hour 12	Sample ID:
End Date:	Lab ID: WESTON	Sample Type:
Sample Date:	Protocol: MBP 96-Anderson et al.	Test Species: HR-Haliotis rufescens
Comments:		

Conc-ug/L	1	2	3
2.38	0.8200	0.8800	0.8900
4.3	0.8800	0.8500	0.8000
7.01	0.8800	0.8600	0.8500
8.44	0.8900	0.8300	0.8800
8.53	0.8600	0.8800	0.8800
9.43	0.8700	0.8100	0.8800
10	0.8600	0.8700	0.8300
13.8	0.8400	0.9300	0.9300

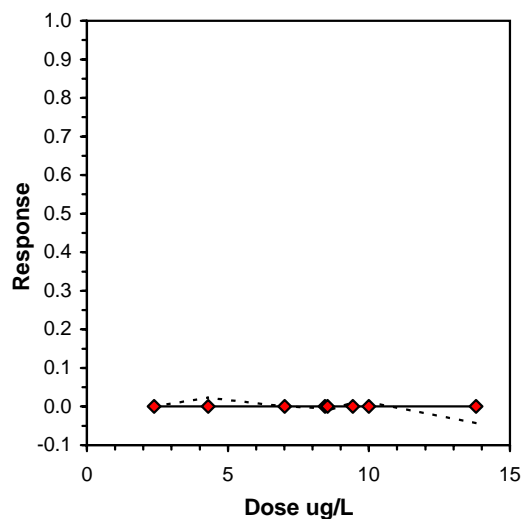
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
2.38	0.8633	1.0000	1.1941	1.1326	1.2327	4.508	3				0.8646	1.0000
4.3	0.8433	0.9768	1.1658	1.1071	1.2171	4.745	3	0.706	2.560	0.1029	0.8646	1.0000
7.01	0.8633	1.0000	1.1925	1.1731	1.2171	1.881	3	0.041	2.560	0.1029	0.8646	1.0000
8.44	0.8667	1.0039	1.1985	1.1458	1.2327	3.865	3	-0.109	2.560	0.1029	0.8646	1.0000
8.53	0.8733	1.0116	1.2071	1.1873	1.2171	1.423	3	-0.323	2.560	0.1029	0.8646	1.0000
9.43	0.8533	0.9884	1.1796	1.1198	1.2171	4.438	3	0.362	2.560	0.1029	0.8646	1.0000
10	0.8533	0.9884	1.1783	1.1458	1.2019	2.471	3	0.393	2.560	0.1029	0.8646	1.0000
13.8	0.9000	1.0425	1.2551	1.1593	1.3030	6.613	3	-1.517	2.560	0.1029	0.8646	1.0000

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.910256	0.884	-0.79458	-0.31774
Bartlett's Test indicates equal variances (p = 0.60)	5.501075	18.47531		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	13.8	>13.8			0.077571	0.089706	0.002201	0.002422	0.524252	7, 16
Treatments vs 2.38										

## **Linear Interpolation (200 Resamples)**

Point	ug/L	SD	95% CL(Exp)	Skew
IC05	>13.8			
IC10	>13.8			
IC15	>13.8			
IC20	>13.8			
IC25	>13.8			
IC40	>13.8			
IC50	>13.8			



# **Abalone Larval Development Test-Proportion Normal**

Start Date:	Test ID: AB Hour 24	Sample ID:
End Date:	Lab ID: WESTON	Sample Type:
Sample Date:	Protocol: MBP 96-Anderson et al.	Test Species: HR-Haliotis rufescens
Comments:		

Conc-ug/L	1	2	3
5.68	0.7900	0.7800	0.7700
7.85	0.8400	0.7400	0.8100
15.2	0.7500	0.7700	0.7700
17.2	0.7800	0.7800	0.7900
20	0.8000	0.7600	0.8600
20.2	0.7700	0.7000	0.7700
21.7	0.7400	0.7300	0.7700
26.6	0.7500	0.6900	0.7200

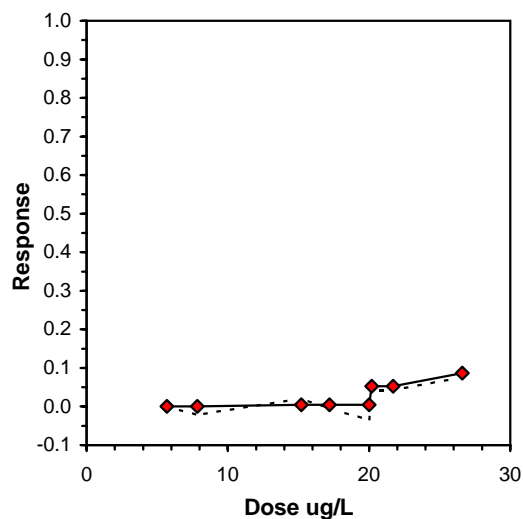
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
5.68	0.7800	1.0000	1.0827	1.0706	1.0948	1.115	3				0.7883	1.0000
7.85	0.7967	1.0214	1.1049	1.0357	1.1593	5.711	3	-0.693	2.560	0.0822	0.7883	1.0000
15.2	0.7633	0.9786	1.0628	1.0472	1.0706	1.272	3	0.618	2.560	0.0822	0.7844	0.9951
17.2	0.7833	1.0043	1.0866	1.0826	1.0948	0.647	3	-0.124	2.560	0.0822	0.7844	0.9951
20	0.8067	1.0342	1.1178	1.0588	1.1873	5.806	3	-1.093	2.560	0.0822	0.7844	0.9951
20.2	0.7467	0.9573	1.0441	0.9912	1.0706	4.394	3	1.200	2.560	0.0822	0.7467	0.9471
21.7	0.7467	0.9573	1.0436	1.0244	1.0706	2.308	3	1.217	2.560	0.0822	0.7467	0.9471
26.6	0.7200	0.9231	1.0136	0.9803	1.0472	3.300	3	2.152	2.560	0.0822	0.7200	0.9133

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.970257	0.884	-0.18481	0.433158
Bartlett's Test indicates equal variances (p = 0.11)	11.77428	18.47531		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	26.6	>26.6			0.071573	0.091754	0.00366	0.001547	0.072867	7, 16
Treatments vs 5.68										

## **Linear Interpolation (200 Resamples)**

Point	ug/L	SD	95% CL(Exp)	Skew
IC05	20.188			
IC10	>26.6			
IC15	>26.6			
IC20	>26.6			
IC25	>26.6			
IC40	>26.6			
IC50	>26.6			





### Abalone Larval Development Test-Proportion Normal

Start Date: Test ID: AB Hour 36 Sample ID:  
 End Date: Lab ID: WESTON Sample Type:  
 Sample Date: Protocol: MBP 96-Anderson et al. Test Species: HR-Haliotis rufescens  
 Comments:

Conc-ug/L	1	2	3
3.07	0.5800	0.5600	0.5500
4.74	0.4000	0.4900	0.5300
6.74	0.5500	0.4700	0.5600
10.1	0.6200	0.5300	0.6800
10.2	0.5300	0.5800	0.4900
12.8	0.5500	0.5400	0.4800
13.7	0.5800	0.5300	0.5800
18.3	0.6200	0.5000	0.4000

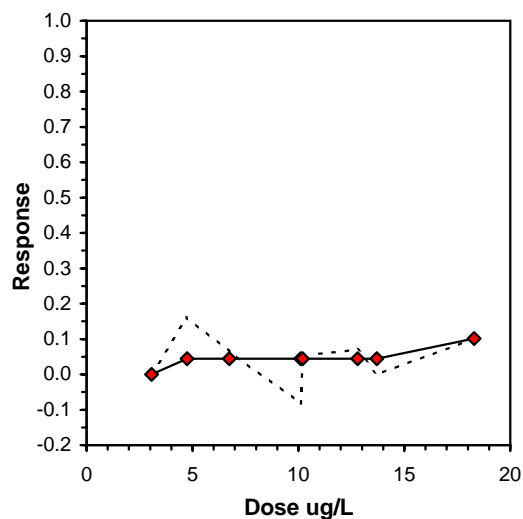
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
3.07	0.5633	1.0000	0.8489	0.8355	0.8657	1.815	3				0.5633	1.0000
4.74	0.4733	0.8402	0.7585	0.6847	0.8154	8.828	3	1.813	2.560	0.1277	0.5383	0.9556
6.74	0.5267	0.9349	0.8121	0.7554	0.8455	6.084	3	0.738	2.560	0.1277	0.5383	0.9556
10.1	0.6100	1.0828	0.8972	0.8154	0.9695	8.637	3	-0.968	2.560	0.1277	0.5383	0.9556
10.2	0.5333	0.9467	0.8189	0.7754	0.8657	5.529	3	0.603	2.560	0.1277	0.5383	0.9556
12.8	0.5233	0.9290	0.8088	0.7654	0.8355	4.686	3	0.805	2.560	0.1277	0.5383	0.9556
13.7	0.5633	1.0000	0.8490	0.8154	0.8657	3.423	3	-0.001	2.560	0.1277	0.5383	0.9556
18.3	0.5067	0.8994	0.7922	0.6847	0.9066	14.022	3	1.137	2.560	0.1277	0.5067	0.8994

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.97533	0.884	-0.11604	0.279685
Bartlett's Test indicates equal variances ( $p = 0.39$ )	7.342123	18.47531		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	18.3	>18.3			0.127308	0.225982	0.005268	0.00373	0.267052	7, 16
Treatments vs 3.07										

Linear Interpolation (200 Resamples)				
Point	ug/L	SD	95% CL(Exp)	Skew

IC05	14.160			
IC10	18.252			
IC15	>18.3			
IC20	>18.3			
IC25	>18.3			
IC40	>18.3			
IC50	>18.3			



**Table I1. Chronic *Menidia beryllina* (13 Mar 2006)**

Treatment	Rep	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Mean
		PM	AM	AM	AM	AM	AM	AM	AM	
<b>Control</b>	1	0.0	0.0				0.0		0.4	<b>0.1</b>
	2	0.1		0.0				0.3	0.2	<b>0.2</b>
	3	0.0			0.1				0.3	<b>0.2</b>
	4	0.3				0.0			0.5	<b>0.3</b>
<b>4</b>	1	2.3	0.7				0.1*		0.1	<b>1.1</b>
	2	2.1		2.1				1.4	1.4	<b>1.7</b>
	3	4.2			1.1				1.0	<b>2.1</b>
	4	2.0				0.0*			1.2	<b>1.6</b>
<b>8</b>	1	8.7	8.8				2.0*		7.1	<b>8.2</b>
	2	6.8		8.2				8.7	6.7	<b>7.6</b>
	3	9.7			7.2				7.3	<b>8.0</b>
	4	7.8				4.6			5.1	<b>5.8</b>
<b>16</b>	1	15.2	13.4				14.4		7.8	<b>12.7</b>
	2	13.8		13.6				14.0	9.6	<b>12.8</b>
	3	14.6			6.9				10.2	<b>10.6</b>
	4	12.5				5.6			9.2	<b>9.1</b>
<b>24</b>	1	25.5	24.9				26.4		12.9	<b>22.4</b>
	2	24.6		22.6				22.5	13.1	<b>20.7</b>
	3	25.7			12.1				10.7	<b>16.2</b>
	4	24.6				11.0			13.5	<b>16.4</b>
<b>32</b>	1	38.3	33.8				41.4		20.0	<b>33.3</b>
	2	36.1		30.7				30.3	22.7	<b>30.0</b>
	3	35.0			19.3				19.4	<b>24.5</b>
	4	33.9				14.6			13.9	<b>20.8</b>
<b>48</b>	1	56.5	50.3				53.2		33.2	<b>48.3</b>
	2	59.0		61.6				44.7	34.8	<b>50.0</b>
	3	52.1			36.3				32.5	<b>40.3</b>
	4	53.7				28.8			29.5	<b>37.3</b>

\* Sulfide concentrations decreased during test

**Table I1. Chronic *Menidia beryllina* (13 Mar 2006)**

Treatment	Percentage Survival							Individual Biomass (mg/ind.)	Combined Endpoint
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7		
<b>Control</b>	100	100	100	90	90	90	<b>90</b>	<b>0.73</b>	<b>0.66</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.80</b>	<b>0.80</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.85</b>	<b>0.85</b>
	100	100	90	90	90	90	<b>90</b>	<b>0.75</b>	<b>0.68</b>
<b>4</b>	100	100	100	100	100	100	<b>100</b>	<b>0.71</b>	<b>0.71</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.83</b>	<b>0.83</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.74</b>	<b>0.74</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.75</b>	<b>0.75</b>
<b>8</b>	90	90	90	90	90	90	<b>90</b>	<b>0.83</b>	<b>0.75</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.79</b>	<b>0.79</b>
	100	90	90	90	90	90	<b>80</b>	<b>1.12</b>	<b>0.90</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.59</b>	<b>0.59</b>
<b>16</b>	100	100	100	100	100	100	<b>100</b>	<b>0.76</b>	<b>0.76</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.81</b>	<b>0.81</b>
	90	90	90	90	90	90	<b>90</b>	<b>0.72</b>	<b>0.64</b>
	100	100	100	100	100	100	<b>100</b>	<b>0.79</b>	<b>0.79</b>
<b>24</b>	90	90	80	80	80	80	<b>80</b>	<b>0.61</b>	<b>0.49</b>
	100	90	90	90	90	90	<b>90</b>	<b>0.85</b>	<b>0.77</b>
	100	90	90	90	90	90	<b>90</b>	<b>1.06</b>	<b>0.95</b>
	100	90	80	80	80	80	<b>80</b>	<b>0.92</b>	<b>0.74</b>
<b>32</b>	90	70	70	70	70	70	<b>70</b>	<b>0.75</b>	<b>0.52</b>
	100	100**	100	100	100	100	<b>100</b>	<b>0.76</b>	<b>0.76</b>
	100	80	80	80	80	80	<b>80</b>	<b>0.79</b>	<b>0.63</b>
	100	70	70	70	70	70	<b>70</b>	<b>0.93</b>	<b>0.65</b>
<b>48</b>	100	30	20	20	20	20	<b>20</b>	<b>1.34</b>	<b>0.27</b>
	80	10	10	10	10	10	<b>0</b>		
	100	50	50	40	30	20	<b>10</b>	<b>0.91</b>	<b>0.09</b>
	80	40	40	40	40	40	<b>40</b>	<b>1.16</b>	<b>0.46</b>

\*\* Survival miscounted

### Larval Fish Growth and Survival Test-7 Day Survival

Start Date:	3/13/2006	Test ID:	P060103.26	Sample ID:	
End Date:	3/20/2006	Lab ID:	WESTON - Port Gamble	Sample Type:	
Sample Date:		Protocol:	EPAM 94-EPA/600/4-91/003	Test Species:	MB-Menidia beryllina
Comments:	Average				

Conc-ug/L	1	2	3	4
Control	0.9000	1.0000	1.0000	0.9000
1.6	1.0000	1.0000	1.0000	1.0000
7.4	0.9000	1.0000	0.8000	1.0000
11.3	1.0000	1.0000	0.9000	1.0000
18.9	0.8000	0.9000	0.9000	0.8000
27.2	0.7000	1.0000	0.8000	0.7000
44	0.2000	0.0000	0.1000	0.4000

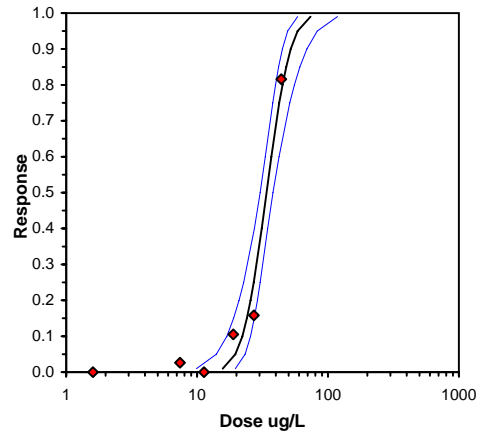
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%	N				
Control	0.9500	1.0000	1.3305	1.2490	1.4120	7.072	4			2	40
1.6	1.0000	1.0526	1.4120	1.4120	1.4120	0.000	4	22.00	10.00	0	40
7.4	0.9250	0.9737	1.2951	1.1071	1.4120	11.347	4	17.00	10.00	3	40
11.3	0.9750	1.0263	1.3713	1.2490	1.4120	5.942	4	20.00	10.00	1	40
18.9	0.8500	0.8947	1.1781	1.1071	1.2490	6.954	4	12.00	10.00	6	40
27.2	0.8000	0.8421	1.1254	0.9912	1.4120	17.662	4	13.00	10.00	8	40
*44	0.1750	0.1842	0.4072	0.1588	0.6847	54.767	4	10.00	10.00	33	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.9623308	0.896	0.424825	0.7295437
Equality of variance cannot be confirmed				

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	27.2	44	34.594797	

Treatments vs Control

Maximum Likelihood-Probit											
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	6.9279808	1.294047	4.3916485	9.464313	0.05	7.3390115	9.4877291	0.12	1.5304083	0.1443422	9
Intercept	-5.602639	1.9716715	-9.467116	-1.738163							
TSCR	0.043823	0.0159894	0.0124838	0.0751621							
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	15.653741	9.8817434	19.577664							
EC05	3.355	19.632994	14.022914	23.277831							
EC10	3.718	22.152684	16.854664	25.596044							
EC15	3.964	24.03296	19.044192	27.342879							
EC20	4.158	25.640522	20.948067	28.867315							
EC25	4.326	27.105099	22.691029	30.297858							
EC40	4.747	31.177382	27.419637	34.641655							
EC50	5.000	33.916289	30.348465	38.017246							
EC60	5.253	36.895807	33.22371	42.18192							
EC75	5.674	42.439051	37.88068	51.115297							
EC80	5.842	44.863153	39.731778	55.40468							
EC85	6.036	47.864047	41.923971	60.976938							
EC90	6.282	51.926647	44.764321	68.930237							
EC95	6.645	58.590892	49.201774	82.884479							
EC99	7.326	73.484975	58.478643	117.66388							



### Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	Test ID: Menidia T1	Sample ID:	
End Date:	Lab ID: WESTON - Port Gamble	Sample Type:	
Sample Date:	Protocol: EPAM 94-EPA/600/4-91/003	Test Species:	MB-Menidia beryllina
Comments: Average			

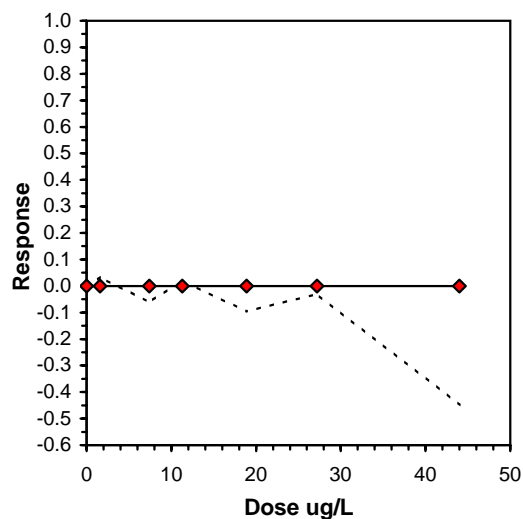
Conc-ug/L	1	2	3	4
Control	0.7300	0.8040	0.8500	0.7533
1.6	0.7130	0.8250	0.7390	0.7510
7.4	0.8344	0.7870	1.1238	0.5880
11.3	0.7580	0.8130	0.7156	0.7880
18.9	0.6075	0.8511	1.0589	0.9212
27.2	0.7457	0.7620	0.7888	0.9329
44	1.3400	0.9100	1.1600	

Conc-ug/L	Mean	N-Mean	Transform: Untransformed					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
Control	0.7843	1.0000	0.7843	0.7300	0.8500	6.831	4				0.8496	1.0000
1.6	0.7570	0.9652	0.7570	0.7130	0.8250	6.344	4	0.277	2.613	0.2579	0.8496	1.0000
7.4	0.8333	1.0624	0.8333	0.5880	1.1238	26.535	4	-0.496	2.613	0.2579	0.8496	1.0000
11.3	0.7686	0.9800	0.7686	0.7156	0.8130	5.455	4	0.159	2.613	0.2579	0.8496	1.0000
18.9	0.8597	1.0961	0.8597	0.6075	1.0589	21.983	4	-0.763	2.613	0.2579	0.8496	1.0000
27.2	0.8073	1.0293	0.8073	0.7457	0.9329	10.596	4	-0.233	2.613	0.2579	0.8496	1.0000
44	1.1367	1.4492	1.1367	0.9100	1.3400	18.998	3	-3.304	2.613	0.2786	0.8496	1.0000

Auxiliary Tests					Statistic	Critical	Skew	Kurt		
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.919634	0.894	0.009685	1.159082		
Bartlett's Test indicates equal variances (p = 0.03)					14.0707	16.81189				
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Bonferroni t Test	44	>44			0.278587	0.35519	0.055054	0.019492	0.037073	6, 20
Treatments vs Control										

Linear Interpolation (200 Resamples)				
Point	ug/L	SD	95% CL(Exp)	Skew

IC05	>44			
IC10	>44			
IC15	>44			
IC20	>44			
IC25	>44			
IC40	>44			
IC50	>44			





## 7-Day Chronic Toxicity Test

Table I2. Chronic *Menidia beryllina* (13 Mar 2006)

Weston Test ID: P060103.26	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	9	9	9	9
	2		10	10	10	10	10	10	10
	3		10	10	10	10	10	10	10
	4		10	10	9	9	9	9	9
4	1		10	10	10	10	10	10	10
	2		10	10	10	10	10	10	10
	3		10	10	10	10	10	10	10
	4		10	10	10	10	10	10	10
8	1		9	9	9	9	9	9	9
	2		10	10	10	10	10	10	10
	3		10	9	9	9	9	9	8
	4		10	10	10	10	10	10	10
16	1		10	10	10	10	10	10	10
	2		10	10	10	10	10	10	10
	3		9	9	9	9	9	9	9
	4		10	10	10	10	10	10	10
24	1		9	9	8	8	8	8	8
	2		10	9	9	9	9	9	9
	3		10	9	9	9	9	9	9
	4		10	9	8	8	8	8	8
32	1		9	7	7	7	7	7	7
	2		10	10*	10	10	10	10	10
	3		10	8	8	8	8	8	8
	4		10	7	7	7	7	7	7
48	1		10	3	2	2	2	2	2
	2		8	1	1	1	1	1	0
	3		10	5	5	4	3	2	1
	4		8	4	4	4	4	4	4
Date			3/14/06	3/15/06	3/16/06	3/17/06	3/18/06	3/19/06	3/20/06
Time			950	943	915	925	1000	915	1050
Initials			GZ	JM	TS	GZ		TS	JM/BG

\* Survival miscounted



## 7-Day Chronic Toxicity Test

**Table I3. Chronic *Menidia beryllina* (13 Mar 2006)**

Weston Test ID:	P060103.26	Client: Marine Research Specialists	Client Sample ID:	N/A
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Concentration	Rep	Boat Number	Weight Empty Boat (mg)	Weight Boat & Animals (mg)	Total Biomass	Number of Survivors	Biomass (mg/ind.)	Combined Endpoint
Control	1	1	83.51	90.08	6.57	9	0.73	0.66
	2	2	70.04	78.08	8.04	10	0.80	0.80
	3	3	77.08	85.58	8.5	10	0.85	0.85
	4	4	74.72	81.50	6.78	9	0.75	0.68
4	1	5	71.51	78.64	7.13	10	0.71	0.71
	2	6	77.13	85.38	8.25	10	0.83	0.83
	3	7	77.59	84.98	7.39	10	0.74	0.74
	4	8	75.51	83.02	7.51	10	0.75	0.75
8	1	9	78.48	85.99	7.51	9	0.83	0.75
	2	10	99.62	107.49	7.87	10	0.79	0.79
	3	11	78.60	87.59	8.99	8	1.12	0.90
	4	12	98.09	103.97	5.88	10	0.59	0.59
16	1	13	98.91	106.49	7.58	10	0.76	0.76
	2	14	81.31	89.44	8.13	10	0.81	0.81
	3	15	91.06	97.50	6.44	9	0.72	0.64
	4	16	86.84	94.72	7.88	10	0.79	0.79
24	1	17	83.82	88.68	4.86	8	0.61	0.49
	2	18	95.50	103.16	7.66	9	0.85	0.77
	3	19	104.48	114.01	9.53	9	1.06	0.95
	4	20	101.45	108.82	7.37	8	0.92	0.74
32	1	21	101.74	106.96	5.22	7	0.75	0.52
	2	22	78.99	86.61	7.62	10	0.76	0.76
	3	23	83.10	89.41	6.31	8	0.79	0.63
	4	24	89.05	95.58	6.53	7	0.93	0.65
48	1	1a	82.89	85.57	2.68	2	1.34	0.27
	2	ND	ND	ND	ND	0	ND	ND
	3	2a	74.90	75.81	0.91	1	0.91	0.09
	4	3a	98.52	103.16	4.64	4	1.16	0.46

ND = No data



## 7-Day Chronic Toxicity Test

**Table I4. Chronic *Menidia beryllina* (13 Mar 2006)**

Client	Marine Research Specialists	Date Received:	NA
Project:	Hydrogen Sulfide	Date Test Started:	13-Mar-06
Client Sample ID:	NA	Date Test Ended:	20-Mar-06
Weston Test ID:	P060103.26	Study Director:	Brian Hester
Species:	<i>Menidia beryllina</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 3/13/06  Time: 1703  Technician: TS/GZ/JM	Control		1		8.4		19.0		31		7.70	0	0	0
			2		8.2		19.2		31		7.71	1	0	1
			3		8.1		19.5		31		7.77	0	0	0
			4		8.1		19.7		31		7.79	3	0	3
	4		1		8.1		19.2		30		7.83	24	0	24
			2		8.1		19.0		30		7.83	21	0	21
			3		8.1		19.1		30		7.75	36	0	36
			4		8.0		19.5		30		7.82	20	0	20
	8		1		8.1		19.4		30		7.84	91	0	91
			2		8.0		19.4		30		7.84	71	0	71
			3		8.0		19.3		30		7.81	95	0	95
			4		8.0		19.7		30		7.84	83	0	83
	16		1		8.1		19.5		30		7.85	82	1:2	164
			2		8.0		19.5		30		7.86	76	1:2	152
			3		8.1		19.3		30		7.79	69	1:2	138
			4		8.0		19.6		30		7.86	69	1:2	138
	24		1		8.1		19.5		30		7.87	143	1:2	286
			2		8.0		19.5		30		7.87	138	1:2	276
			3		8.0		19.4		30		7.86	141	1:2	282
			4		7.9		19.7		30		7.87	139	1:2	278
	32		1		8.0		19.5		30		7.90	229	1:2	458
			2		8.0		19.5		30		7.90	216	1:2	432
			3		8.0		19.4		30		7.88	200	1:2	400
			4		7.9		19.7		30		7.89	200	1:2	400
	48		1		8.0		19.4		30		7.93	359	1:2	718
			2		8.0		19.4		30		7.94	383	1:2	766
			3		8.0		19.5		30		7.92	325	1:2	650
			4		7.9		19.7		30		7.94	352	1:2	704
<b>Day 1</b> <b>AM</b> Date: 3/14/06 Time: 0902 Technician: TS/GZ	Control				8.0		19.2		31		7.76	0	0	0
	4				7.4		19.6		30		7.79	7	0	7
	8				7.1		19.8		30		7.82	90	0	90
	16				7.0		20.0		30		7.82	69	1:2	138
	24				7.1		19.9		30		7.85	136	1:2	272
	32				7.0		19.9		30		7.87	192	1:2	384
	48				7.1		19.7		30		7.92	316	1:2	632
<b>Day 1</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND





## 7-Day Chronic Toxicity Test

**Table 14. Chronic *Menidia beryllina* (13 Mar 2006)**

Weston Test ID:	P060103.26	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>Day 2</b> <b>AM</b> Date: 3/15/06 Time: 0832 Technician: GZ/JM	Control				6.9		18.9		32		7.68	0	0	0
	4				6.6		19.5		30		7.71	17	0	17
	8				7.4		19.8		30		7.74	71	0	71
	16				7.4		19.8		30		7.75	60	1:2	120
	24				7.5		19.9		30		7.74	98	1:2	196
	32				7.3		19.9		30		7.75	136	1:2	272
	48				7.4		19.7		30		7.81	307	1:2	614
<b>Day 2</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND
<b>Day 3</b> <b>AM</b> Date: 3/16/06 Time: 1021 Technician: JM	Control				7.0		18.8		31		7.65	1	0	1
	4				7.6		19.3		30		7.77	10	0	10
	8				7.5		19.6		30		7.78	67	0	67
	16				7.3		19.8		30		7.79	66	0	66
	24				7.4		19.7		30		7.76	109	0	109
	32				7.4		19.7		30		7.80	188	0	188
	48				7.2		19.8		30		7.78	341	0	341
<b>Day 3</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND
<b>Day 4</b> <b>AM</b> Date: 3/17/06 Time: 0845 Technician: GZ/TS	Control				6.1		19.7		31		7.88	0	0	0
	4				7.5		19.8		30		7.92	0	0	0
	8				7.4		19.9		30		7.94	60	0	60
	16				7.4		19.9		30		7.96	77	0	77
	24				7.4		19.8		30		7.98	157	0	157
	32				7.4		19.8		30		7.94	192	0	192
	48				7.4		19.8		30		7.94	379	0	379
<b>Day 4</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND



## 7-Day Chronic Toxicity Test

**Table 14. Chronic *Menidia beryllina* (13 Mar 2006)**

Weston Test ID:	P060103.26	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>Day 5</b> <b>AM</b> Date: 3/18/06 Time: 0914 Technician: TS/DM	Control				7.4		19.4		30		7.66	0	0	0
	4				7.6		19.7		30		7.77	1	0	1
	8				7.5		19.8		30		7.78	19	0	19
	16				7.6		19.5		30		7.73	121	0	121
	24				7.3		19.9		30		7.76	239	0	239
	32				7.2		20.0		30		7.77	383	0	383
	48				7.1		19.8		30		7.84	566	0	566
<b>Day 5</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND
<b>Day 6</b> <b>AM</b> Date: 3/19/06 Time: 0754 Technician: TS	Control				7.2		19.2		30		7.64	2	0	2
	4				7.5		19.8		30		7.79	13	0	13
	8				7.3		19.5		30		7.78	81	0	81
	16				7.3		19.9		30		7.79	135	0	135
	24				7.3		19.9		30		7.81	226	0	226
	32				7.2		19.9		30		7.83	317	0	317
	48				7.0		19.6		30		7.86	493	0	493
<b>Day 6</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND
<b>Day 7</b> Date: 3/20/06 Time: 0935 Technician: JM/BCG	Control		1		7.3		19.7		31		7.72	3	0	3
			2		7.0		19.6		31		7.75	2	0	2
			3		7.1		19.6		31		7.76	3	0	3
			4		7.0		19.5		31		7.73	4	0	4
	4		1		7.5		19.6		32		7.78	1	0	1
			2		7.4		19.7		30		7.81	14	0	14
			3		7.3		19.6		30		7.81	10	0	10
			4		7.4		19.6		30		7.81	12	0	12
	8		1		7.4		19.8		30		7.83	74	0	74
			2		7.3		19.9		30		7.83	70	0	70
			3		7.1		19.6		30		7.82	74	0	74
			4		7.1		19.6		30		7.82	52	0	52
	16		1		7.2		19.9		30		7.83	81	0	81
			2		7.2		19.9		30		7.82	98	0	98
			3		7.1		19.8		30		7.78	96	0	96
			4		7.2		19.8		30		7.79	88	0	88
	24		1		7.1		19.9		30		7.81	129	0	129
			2		7.0		19.9		30		7.83	137	0	137
			3		7.0		19.6		30		7.84	113	0	113
			4		7.1		19.6		30		7.84	143	0	143
	32		1		7.0		19.8		30		7.85	217	0	217
			2		7.1		19.8		30		7.86	252	0	252
			3		7.1		19.6		30		7.85	209	0	209
			4		7.1		19.6		30		7.86	153	0	153
	48		1		6.9		19.5		30		7.89	389	0	389
			2		6.8		19.5		30		7.89	407	0	407
			3		6.9		19.7		30		7.88	375	0	375
			4		6.9		19.6		30		7.88	339	0	339

**Table I5. Chronic *Menidia beryllina* (14 Mar 2006)**

Treatment	Rep	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Mean
		PM	AM	AM	AM	AM	AM	AM	AM	
Control	1		0.0					0.2	0.6*	0.1
	2	0.0			0.0				0.4*	0.0
	3			1.0		0.0			0.1*	0.5
	4						0.0		0.5*	0.0
4	1		2.2					2.0	0.8*	2.1
	2	2.1			0.0*				1.0*	2.1
	3			2.0		0.5*			1.2*	2.0
	4						1.5		1.1*	1.5
8	1		10.7					7.7	2.9*	9.2
	2	8.2			4.1				2.3*	6.2
	3			9.6		3.1*			3.9*	9.6
	4						9.7		5.1*	9.7
16	1		17.2					11.3	2.6*	14.2
	2	14.7			6.1				3.9*	10.4
	3			13.8		7.7			5.6*	10.7
	4						14.9		5.3*	14.9
24	1		21.2					13.4	7.2*	17.3
	2	16.2			6.8*				6.9*	16.2
	3			16.0		13.2			6.9*	14.6
	4						20.3		7.4*	20.3
32	1		30.1					12.8	8.2*	21.4
	2	19.3			11.9				8.2*	15.6
	3			23.3		16.9			8.5*	20.1
	4						28.0		10.3*	28.0
48	1		34.3					23.6	15.2*	28.9
	2	31.2			14.4				14.5*	22.8
	3			25.3		20.7			14.3*	23.0
	4						25.8		16.7*	25.8

\* Sulfide concentrations decreased during test

**Table I5. Chronic *Menidia beryllina* (14 Mar 2006)**

Treatment	Rep	Percentage Survival							Individual Biomass (mg/ind.)	Combined Endpoint
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7		
<b>Control</b>	1	90	90	80	80	80	80	<b>80</b>	<b>0.88</b>	<b>0.71</b>
	2	90	90	90	90**	90	90	<b>90</b>	<b>0.86</b>	<b>0.77</b>
	3	90	90	80	80	80	80	<b>80</b>	<b>0.85</b>	<b>0.68</b>
	4	100	100	100	100	100	100	<b>100</b>	<b>0.66</b>	<b>0.66</b>
<b>4</b>	1	100	100	100	100	100	100	<b>100</b>	<b>0.90</b>	<b>0.90</b>
	2	90	80	90	90	90	90	<b>90</b>	<b>0.84</b>	<b>0.75</b>
	3	100**	100**	100	100**	100**	100	<b>100</b>	<b>0.84</b>	<b>0.84</b>
	4	100	100	100	100	100	100	<b>100</b>	<b>0.73</b>	<b>0.73</b>
<b>8</b>	1	100	100	100	100	100	100	<b>100</b>	<b>0.81</b>	<b>0.81</b>
	2	90	90	80	80	80	80	<b>80</b>	<b>0.88</b>	<b>0.70</b>
	3	90	90	90**	90**	90	90	<b>90</b>	<b>0.79</b>	<b>0.71</b>
	4	100	100	100	100	100	100	<b>100</b>	<b>0.89</b>	<b>0.89</b>
<b>16</b>	1	90	90	90	90	90	90	<b>90</b>	<b>0.82</b>	<b>0.74</b>
	2	100	90	90	90	90	90	<b>90</b>	<b>0.77</b>	<b>0.69</b>
	3	80	70	70	70	70	70	<b>70</b>	<b>0.72</b>	<b>0.51</b>
	4	100	100	90	80	80	80	<b>80</b>	<b>1.03</b>	<b>0.82</b>
<b>24</b>	1	80	80	80**	80	80	80	<b>80</b>	<b>0.87</b>	<b>0.69</b>
	2	100	90	60	60	60	60	<b>60</b>	<b>0.96</b>	<b>0.58</b>
	3	100	100	100	100	100	100	<b>100</b>	<b>0.86</b>	<b>0.86</b>
	4	90	70	60	60	60	60	<b>60</b>	<b>0.71</b>	<b>0.43</b>
<b>32</b>	1	70	50	40	40	40	40	<b>40</b>	<b>0.90</b>	<b>0.36</b>
	2	90	80	80	60	60	60	<b>60</b>	<b>0.93</b>	<b>0.56</b>
	3	70	50	50	30	30	30	<b>30</b>	<b>0.83</b>	<b>0.25</b>
	4	80	60	60	50	50	50	<b>50</b>	<b>0.79</b>	<b>0.40</b>
<b>48</b>	1	50	40	30**	30	30	30	<b>30</b>	<b>0.89</b>	<b>0.27</b>
	2	70	60	50	50	50	50	<b>50</b>	<b>0.86</b>	<b>0.43</b>
	3	70	70	50	40	40	30	<b>30</b>	<b>0.95</b>	<b>0.28</b>
	4	90	70	50	30	30	30	<b>30</b>	<b>1.20</b>	<b>0.36</b>

\*\* Survival miscounted

Larval Fish Growth and Survival Test-7 Day Survival				
Start Date:	3/14/2006	Test ID:	P060103.27	Sample ID:
End Date:	3/21/2006	Lab ID:	WESTON - Port Gamble	Sample Type:
Sample Date:		Protocol:	EPAM 94-EPA/600/4-91/003	Test Species:
Comments:	Average			
	MB-Menidia beryllina			

Conc-ug/L	1	2	3	4
Control	0.8000	0.9000	0.8000	1.0000
1.9	1.0000	0.9000	1.0000	1.0000
8.7	1.0000	0.8000	0.9000	0.1000
12.6	0.9000	0.9000	0.7000	0.8000
17.1	0.8000	0.6000	1.0000	0.6000
21.3	0.4000	0.6000	0.3000	0.5000
25.1	0.3000	0.5000	0.3000	0.3000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%	N					
Control	0.8750	1.0000	1.2188	1.1071	1.4120	11.906	4				5	40
1.9	0.9750	1.1143	1.3713	1.2490	1.4120	5.942	4	-0.942	2.451	0.3969	1	40
8.7	0.7000	0.8000	1.0225	0.3218	1.4120	47.285	4	1.213	2.451	0.3969	12	40
12.6	0.8250	0.9429	1.1491	0.9912	1.2490	10.856	4	0.431	2.451	0.3969	7	40
17.1	0.7500	0.8571	1.0728	0.8861	1.4120	23.208	4	0.902	2.451	0.3969	10	40
*21.3	0.4500	0.5143	0.7340	0.5796	0.8861	17.942	4	2.995	2.451	0.3969	22	40
*25.1	0.3500	0.4000	0.6311	0.5796	0.7854	16.302	4	3.631	2.451	0.3969	26	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.90109	0.896	-1.176279	4.6228683
Bartlett's Test indicates equal variances ( $p = 0.03$ )	13.937789	16.811893		

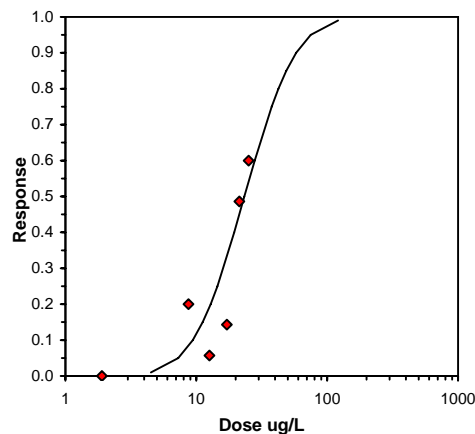
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	17.1	21.3	19.084811		0.3446191	0.3910981	0.2766441	0.052419	0.0018697	6, 21

Treatments vs Control

Treatments vs Control				Maximum Likelihood-Probit						
Parameter	Value	SE	95% Fiducial Limits	Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	3.2483004	1.6529761	-1.341097 7.8376978	0.125	13.575388	9.4877291	8.8E-03	1.3682626	0.3078533	12
Intercept	0.5554719	2.0998742	-5.274714 6.3856575							
TSCR	0.0903592	0.0587039	-0.072629 0.2533474							
Point	Probits	ug/L	95% Fiducial Limits							
EC01	2.674	4.4883711								
EC05	3.355	7.2759384								
EC10	3.718	9.4131018								
EC15	3.964	11.19936								
EC20	4.158	12.857803								
EC25	4.326	14.475005								
EC40	4.747	19.510522								
EC50	5.000	23.348696								
EC60	5.253	27.941929								
EC75	5.674	37.662272								
EC80	5.842	42.399282								
EC85	6.036	48.677929								
EC90	6.282	57.915186								
EC95	6.645	74.926631								
EC99	7.326	121.46093								

Point	Probits	ug/L	Response (approx)
EC01	2.674	4.4883711	0.0
EC05	3.355	7.2759384	0.2
EC10	3.718	9.4131018	0.05
EC15	3.964	11.19936	0.15
EC20	4.158	12.857803	0.48
EC25	4.326	14.475005	0.6
EC40	4.747	19.510522	0.58

Significant heterogeneity detected ( $p = 8.78E-03$ )



### Larval Fish Growth and Survival Test-7 Day Growth

Start Date:	Test ID: Menidia T2	Sample ID:
End Date:	Lab ID: WESTON - Port Gamble	Sample Type:
Sample Date:	Protocol: EPAM 94-EPA/600/4-91/003	Test Species: MB-Menidia beryllina
Comments: Average		

Conc-ug/L	1	2	3	4
Control	0.8825	0.8567	0.8487	0.6640
1.9	0.8970	0.8356	0.8360	0.7320
8.7	0.8130	0.8750	0.7933	8.8600
12.6	0.8211	0.7656	0.7214	1.0300
17.1	0.8675	0.9583	0.8620	0.7133
21.3	0.8950	0.9267	0.8300	0.7920
25.1	0.8933	0.8580	0.9467	1.2033

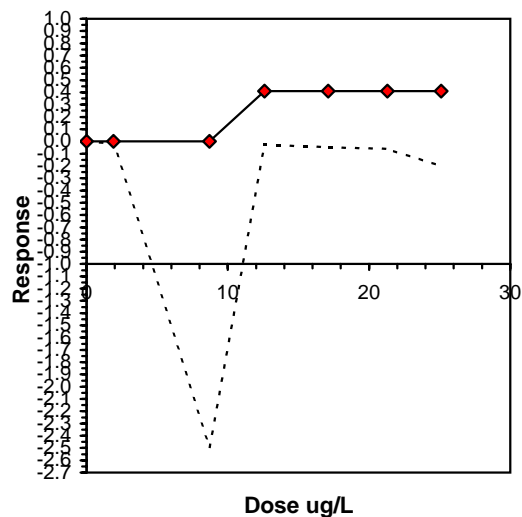
Conc-ug/L	Mean	N-Mean	Transform: Untransformed					Rank Sum	1-Tailed Critical	Isotonic	
			Mean	Min	Max	CV%	N			Mean	N-Mean
Control	0.8130	1.0000	0.8130	0.6640	0.8825	12.345	4			1.4912	1.0000
1.9	0.8251	1.0150	0.8251	0.7320	0.8970	8.298	4	17.00	10.00	1.4912	1.0000
8.7	2.8353	3.4876	2.8353	0.7933	8.8600	141.662	4	19.00	10.00	1.4912	1.0000
12.6	0.8345	1.0265	0.8345	0.7214	1.0300	16.363	4	17.00	10.00	0.8803	0.5903
17.1	0.8503	1.0459	0.8503	0.7133	0.9583	11.929	4	21.00	10.00	0.8803	0.5903
21.3	0.8609	1.0590	0.8609	0.7920	0.9267	7.094	4	20.00	10.00	0.8803	0.5903
25.1	0.9753	1.1997	0.9753	0.8580	1.2033	16.026	4	25.00	10.00	0.8803	0.5903

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution ( $p \leq 0.01$ )	0.493403	0.896	3.20876	16.13507
Bartlett's Test indicates unequal variances ( $p = 1.19E-15$ )	82.30203	16.81189		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	25.1	>25.1		
Treatments vs Control				

Linear Interpolation (200 Resamples)				
Point	ug/L	SD	95% CL(Exp)	Skew

IC05	9.176			
IC10	9.652			
IC15	10.128			
IC20	10.604			
IC25	11.080			
IC40	12.508			
IC50	>25.1			





## 7-Day Chronic Toxicity Test

**Table I6. Chronic *Menidia beryllina* (14 Mar 2006)**

Weston Test ID: P060103.27	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		9	9	8	8	8	8	8
	2		9	9	9	9*	9	9	9
	3		9	9	8	8	8	8	8
	4		10	10	10	10	10	10	10
4	1		10	10	10	10	10	10	10
	2		9	9*	9	9	9	9	9
	3		10*	10*	10	10*	10*	10	10
	4		10	10	10	10	10	10	10
8	1		10	10	10	10	10	10	10
	2		9	9	8	8	8	8	8
	3		9	9	9*	9*	9	9	9
	4		10	10	10	10	10	10	10
16	1		9	9	9	9	9	9	9
	2		10	9	9	9	9	9	9
	3		8	7	7	7	7	7	7
	4		10	10	9	8	8	8	8
24	1		8	8	8*	8	8	8	8
	2		10	9	6	6	6	6	6
	3		10	10	10	10	10	10	10
	4		9	7	6	6	6	6	6
32	1		7	5	4	4	4	4	4
	2		9	8	8	6	6	6	6
	3		7	5	5	3	3	3	3
	4		8	6	6	5	5	5	5
48	1		5	4	3*	3	3	3	3
	2		7	6	5	5	5	5	5
	3		7	7	5	4	4	3	3
	4		9	7	5	3	3	3	3
Date			3/15/06	3/16/06	3/17/06	3/18/06	3/19/06	3/20/06	3/21/06
Time			930	1000	851	905	945	1205	1550
Initials			DM	DM	TS	DM	TS	BG	JE

\* Survival miscounted



# 7-Day Chronic Toxicity Test

**Table I7. Chronic *Menidia beryllina* (14 Mar 2006)**

Weston Test ID:	P060103.27	Client:	Marine Research Specialists	Client Sample ID	N/A
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Concentration	Rep	Boat Number	Weight Empty Boat (mg)	Weight Boat & Animals (mg)	Total Biomass	Number of Survivors	Biomass (mg/ind.)	Combined Endpoint
Control	1	1	150.12	157.18	7.06	8	0.88	0.71
	2	2	142.83	150.54	7.71	9	0.86	0.77
	3	3	150.10	156.89	6.79	8	0.85	0.68
	4	4	89.38	96.02	6.64	10	0.66	0.66
4	1	5	98.31	107.28	8.97	10	0.90	0.90
	2	6	84.14	91.66	7.52	9	0.84	0.75
	3	7	89.66	98.02	8.36	10	0.84	0.84
	4	8	88.17	95.49	7.32	10	0.73	0.73
8	1	9	127.14	135.27	8.13	10	0.81	0.81
	2	10	98.34	105.34	7	8	0.88	0.70
	3	11	106.21	113.35	7.14	9	0.79	0.71
	4	12	111.54	120.40	8.86	10	0.89	0.89
16	1	13	96.82	104.21	7.39	9	0.82	0.74
	2	14	98.68	105.57	6.89	9	0.77	0.69
	3	15	99.87	104.92	5.05	7	0.72	0.51
	4	16	87.64	95.88	8.24	8	1.03	0.82
24	1	17	157.92	164.86	6.94	8	0.87	0.69
	2	18	146.67	152.42	5.75	6	0.96	0.58
	3	19	155.23	163.85	8.62	10	0.86	0.86
	4	20	102.85	107.13	4.28	6	0.71	0.43
32	1	21	108.00	111.58	3.58	4	0.90	0.36
	2	22	115.97	121.53	5.56	6	0.93	0.56
	3	23	112.89	115.38	2.49	3	0.83	0.25
	4	24	69.25	73.21	3.96	5	0.79	0.40
48	1	1a	138.02	140.70	2.68	3	0.89	0.27
	2		132.79	137.08	4.29	5	0.86	0.43
	3	2a	156.56	159.40	2.84	3	0.95	0.28
	4	3a	140.46	144.07	3.61	3	1.20	0.36





## 7-Day Chronic Toxicity Test

**Table I8. Chronic *Menidia beryllina* (14 Mar 2006)**

Client	Marine Research Specialists	Date Received:	NA
Project:	Hydrogen Sulfide	Date Test Started:	14-Mar-06
Client Sample ID:	NA	Date Test Ended:	21-Mar-06
Weston Test ID:	P060103.27	Study Director:	Brian Hester
Species:	<i>Menidia beryllina</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
												Value	Dil	Corr value	H <sub>2</sub> S
<b>Day 0</b>  Date: 3/14/06  Time: 1808  Technician: TS	Control		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		8.1		19.0		31		7.80	0	0	0	0.00
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	4		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		8.0		19.4		30		7.88	24	0	24	2.10
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	8		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		8.0		19.4		30		7.89	96	0	96	8.23
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	16		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.5		30		7.91	179	0	179	14.65
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	24		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.6		30		7.93	207	0	207	16.18
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	32		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.6		30		7.93	247	0	247	19.31
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
	48		1		ND		ND		ND		ND	ND	ND	ND	ND
			2		7.9		19.7		30		7.87	352	0	352	31.15
			3		ND		ND		ND		ND	ND	ND	ND	ND
			4		ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 1</b> <b>AM</b> Date: 3/15/06 Time: 0815 Technician: DM	Control				7.8		19.4		30		7.79	0	0	0	0.00
	4				7.6		19.4		30		7.75	19	0	19	2.18
	8				7.8		19.4		30		7.77	97	0	97	10.68
	16				7.7		18.9		30		7.79	160	0	160	17.18
	24				7.7		19.3		30		7.84	222	0	222	21.20
	32				7.9		18.5		30		7.81	288	0	288	30.07
	48				7.5		19.3		30		7.81	337	0	337	34.27
<b>Day 1</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND



Weston Test ID:	P060103.27	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
Day 2	Control		3		7.2		19.6		30		7.65	5	0	5	0.98
AM	4				7.2		19.8		30		7.70	23	0	23	2.01
Date: 3/16/06	8				7.2		19.9		30		7.70	96	0	96	9.63
Time: 1100	16				7.2		19.9		30		7.75	101	0	101	13.78
Technician: DM/JM	24				7.2		20.0		30		7.73	149	0	149	15.95
	32				7.1		20.0		30		7.75	176	0	176	23.30
	48				6.9		20.0		30		7.71	280	0	280	25.29
Day 2	Control				ND		ND		ND		ND	ND	ND	ND	ND
PM	4				ND		ND		ND		ND	ND	ND	ND	ND
Date:	8				ND		ND		ND		ND	ND	ND	ND	ND
Time:	16				ND		ND		ND		ND	ND	ND	ND	ND
Technician:	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
Day 3	Control		2		7.4		19.3		31		7.92	0	0	0	0.00
AM	4				7.3		19.8		30		7.98	0	0	0	0.00
Date: 3/17/06	8				7.2		19.7		30		7.97	58	0	58	4.15
Time: 0900	16				7.0		19.8		30		7.99	90	0	90	6.15
Technician: GZ/TS	24				7.3		19.8		30		7.99	99	0	99	6.76
	32				7.0		19.9		30		8.01	182	0	182	11.86
	48				7.2		19.9		30		8.00	216	0	216	14.39
Day 3	Control				ND		ND		ND		ND	ND	ND	ND	ND
PM	4				ND		ND		ND		ND	ND	ND	ND	ND
Date:	8				ND		ND		ND		ND	ND	ND	ND	ND
Time:	16				ND		ND		ND		ND	ND	ND	ND	ND
Technician:	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
Day 4	Control		3		7.3		19.7		30		7.70	0	0	0	0.00
AM	4				7.6		19.5		30		7.70	4	0	4	0.51
Date: 3/18/06	8				7.5		19.9		30		7.78	29	0	29	3.08
Time: 0850	16				7.2		20.0		30		7.79	74	0	74	7.67
Technician: DM	24				7.3		20.0		30		7.75	117	0	117	13.17
	32				7.2		20.0		30		7.77	156	0	156	16.85
	48				7.3		19.9		30		7.78	195	0	195	20.70
Day 4	Control				ND		ND		ND		ND	ND	ND	ND	ND
PM	4				ND		ND		ND		ND	ND	ND	ND	ND
Date:	8				ND		ND		ND		ND	ND	ND	ND	ND
Time:	16				ND		ND		ND		ND	ND	ND	ND	ND
Technician:	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND



## 7-Day Chronic Toxicity Test

**Table I8. Chronic *Menidia beryllina* (14 Mar 2006)**

Weston Test ID:	P060103.27	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)			
<b>Day 5</b> <b>AM</b> Date: 3/19/06 Time: 0805 Technician: TS	Control				7.3		19.2		31		7.73	0	0	0	0.00
	4				7.5		19.5		30		7.8	15	0	15	1.55
	8				7.5		19.6		30		7.82	98	0	98	9.66
	16				7.3		19.8		30		7.83	155	0	155	14.87
	24				7.2		19.9		30		7.83	212	0	212	20.27
	32				7.2		19.9		30		7.85	305	0	305	27.97
	48				7.0		19.9		30		7.84	276	0	276	25.85
<b>Day 5</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 6</b> <b>AM</b> Date: 3/20/06 Time: 1143 Technician: JM/BG	Control				7.0		19.7		30		7.56	1	0	1	0.17
	4				7.4		19.9		29		7.78	19	0	19	1.98
	8				7.2		20.0		29		7.82	79	0	79	7.72
	16				7.3		20.0		29		7.84	120	0	120	11.25
	24				7.1		20.0		29		7.85	146	0	146	13.40
	32				7.1		20.1		29		7.85	140	0	140	12.81
	48				7.2		20.0		29		7.87	268	0	268	23.59
<b>Day 6</b> <b>PM</b> Date: Time: Technician:	Control				ND		ND		ND		ND	ND	ND	ND	ND
	4				ND		ND		ND		ND	ND	ND	ND	ND
	8				ND		ND		ND		ND	ND	ND	ND	ND
	16				ND		ND		ND		ND	ND	ND	ND	ND
	24				ND		ND		ND		ND	ND	ND	ND	ND
	32				ND		ND		ND		ND	ND	ND	ND	ND
	48				ND		ND		ND		ND	ND	ND	ND	ND
<b>Day 7</b>  Date: 3/21/06  Time: 1155  Technician: JE/JW	Control		1		6.9		20.8		31		7.73	5	0	5	0.57
			2		6.9		21.2		30		7.84	4	0	4	0.36
			3		6.9		21.2		31		7.82	1	0	1	0.09
			4		6.9		21.1		31		7.83	5	0	5	0.46
	4		1		7.1		21.1		29		7.87	9	0	9	0.76
			2		6.9		21.2		30		7.86	12	0	12	1.03
			3		7.1		21.1		30		7.88	15	0	15	1.24
			4		7.1		21.1		29		7.89	13	0	13	1.06
	8		1		7.1		21.2		29		7.9	36	0	36	2.86
			2		6.8		21.1		30		7.89	28	0	28	2.27
			3		7.2		21.1		29		7.92	51	0	51	3.89
			4		7.2		21.2		30		7.91	66	0	66	5.11
	16		1		7.0		21.0		29		7.92	34	0	34	2.60
			2		7.0		21.1		30		7.92	51	0	51	3.88
			3		7.0		21.1		29		7.92	74	0	74	5.65
			4		6.9		21.1		29		7.92	70	0	70	5.34
	24		1		7.0		21.1		29		7.92	94	0	94	7.18
			2		6.9		21.1		30		7.92	91	0	91	6.92
			3		7.1		21.0		30		7.93	93	0	93	6.94
			4		7.0		21.1		29		7.94	101	0	101	7.39
	32		1		6.9		21.1		30		7.93	110	0	110	8.18
			2		7.0		21.0		30		7.93	110	0	110	8.21
			3		7.0		21.0		30		7.94	116	0	116	8.47
			4		7.1		21.0		29		7.94	141	0	141	10.35
	48		1		7.0		21.0		30		7.95	213	0	213	15.23
			2		6.9		21.0		30		7.95	203	0	203	14.51
			3		7.0		21.0		29		7.95	199	0	199	14.29
			4		6.9		21		29		7.96	237	0	237	16.66

**Table J1. Chronic *Cyprinodon variegatus* Test 1 (15 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4		Day 5		Day 6		Day 7	Mean
		PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	
Control	1	0.2	1.4				0.1				0.2				0.3	0.4
	2	0.1		1.5				0.8				0.4			0.4	0.6
	3	0.2			0.6				0.0				1.1		0.2	0.4
	4	0.2				0.2				0.0				0.0	0.3	0.1
15	1	26.0	47.2				19.6				39.3				22.1	30.8
	2	29.1		52.2				32.9				33.8			20.0	33.6
	3	34.0			54.4				29.7				67.0		21.1	41.2
	4	34.1				32.7				9.8				38.7	19.1	26.9
25	1	24.0	75.3				12.0*				62.9				36.4	49.7
	2	28.3		67.7				70.6				54.1			38.7	51.9
	3	25.3			87.9				47.3				97.8		32.5	58.2
	4	29.0				37.7				35.2				80.4	33.6	43.2
50	1	31.5	132				39.9				123				58.4	77.0
	2	38.0		128				90.6				95.5			64.1	83.2
	3	42.9			122				49.8				108		56.2	75.8
	4	40.9				62.3				43.3				138	58.9	68.7
75	1	84.5	205				33.4*				136				50.0	119
	2	74.7		216				147				156			50.4	129
	3	57.2			239				39.7*				182		68.9	137
	4	97.9				111				34.5*				141	81.3	107.8
100	1	80.3	214				50.7				160				105	122
	2	115		217				102				170			142	149
	3	52.5			231				49.4				288		133	151
	4	55.2				115				64.5				208	106	110

\* Sulfide concentrations decreased during test

**Table J1. Chronic *Cyprinodon variegatus* Test 1 (15 Feb 2006)**

Treatment	Rep	Percentage Survival							Individual Biomass (mg/ind.)	Combined Endpoint
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7		
<b>Control</b>	1	100	100	80	70	60	60	<b>60</b>	<b>0.53</b>	<b>0.32</b>
	2	100	90	90	90	90	70	<b>70</b>	<b>0.60</b>	<b>0.42</b>
	3	100	100	100**	100**	100**	100	<b>100</b>	<b>0.61</b>	<b>0.61</b>
	4	100	100	70	70	60	60	<b>60</b>	<b>0.80</b>	<b>0.48</b>
<b>15</b>	1	100	90	70	70	70	70	<b>70</b>	<b>0.51</b>	<b>0.35</b>
	2	100	100	90	80	80	80	<b>80</b>	<b>0.70</b>	<b>0.56</b>
	3	100	100	100	90	90	90	<b>90</b>	<b>0.60</b>	<b>0.54</b>
	4	100	90	80	70	50	50	<b>50</b>	<b>0.58</b>	<b>0.29</b>
<b>25</b>	1	100	80	80	80	80	80	<b>80</b>	<b>0.72</b>	<b>0.57</b>
	2	100	100	80	80	80	90	<b>80</b>	<b>0.66</b>	<b>0.53</b>
	3	100	100	80	80	80	80	<b>80</b>	<b>0.65</b>	<b>0.52</b>
	4	100	90	90	90	90	90	<b>90</b>	<b>0.64</b>	<b>0.58</b>
<b>50</b>	1	70	70	70	70	70	70	<b>70</b>	<b>0.51</b>	<b>0.36</b>
	2	90	90	80	80	80	80	<b>60</b>	<b>0.53</b>	<b>0.32</b>
	3	80	80	80**	80**	80	80	<b>80</b>	<b>0.74</b>	<b>0.59</b>
	4	100	100	100	90	90	70	<b>70</b>	<b>0.74</b>	<b>0.52</b>
<b>75</b>	1	100	80	50**	50**	50	50	<b>50</b>	<b>0.59</b>	<b>0.29</b>
	2	100	100	80	70	70	70	<b>60</b>	<b>0.44</b>	<b>0.26</b>
	3	100	100	100	90	90	80	<b>80</b>	<b>0.41</b>	<b>0.33</b>
	4	100	100	90	60	60	60	<b>50</b>	<b>0.42</b>	<b>0.21</b>
<b>100</b>	1	100	90	90	30	30	0	<b>T</b>	<b>NC</b>	<b>NC</b>
	2	100	100	100	80	80	90	<b>70</b>	<b>0.40</b>	<b>0.28</b>
	3	100	100	80	50	50	40	<b>40</b>	<b>0.53</b>	<b>0.21</b>
	4	100	100	90	80	80	60	<b>50</b>	<b>0.56</b>	<b>0.28</b>

T = Test terminated due to 0% survival

NC = Not calculable

\*\* Survival miscounted

Larval Fish Growth and Survival Test-7 Day Survival					
Start Date:	2/15/2006	Test ID:	P060103.16	Sample ID:	
End Date:	2/22/2006	Lab ID:	WESTON - Port Gamble	Sample Type:	
Sample Date:		Protocol:	EPAM 94-EPA/600/4-91/003	Test Species:	CV-Cyprinodon variegatus
Comments:	Average				

Conc-ug/L	1	2	3	4
Control	0.6000	0.7000	1.0000	0.6000
33.1	0.7000	0.8000	0.9000	0.5000
50.7	0.8000	0.8000	0.8000	0.9000
76.2	0.7000	0.6000	0.8000	0.7000
123.1	0.5000	0.6000	0.8000	0.5000
133	0.0000	0.7000	0.4000	0.5000

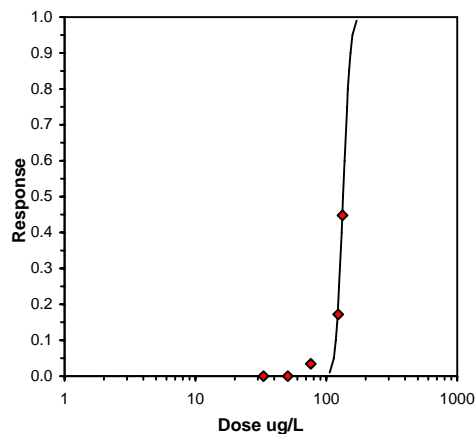
Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root					t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%	N					
Control	0.7250	1.0000	1.0438	0.8861	1.4120	23.989	4				11	40
33.1	0.7250	1.0000	1.0332	0.7854	1.2490	18.969	4	0.072	2.410	0.3568	11	40
50.7	0.8250	1.1379	1.1426	1.1071	1.2490	6.209	4	-0.667	2.410	0.3568	7	40
76.2	0.7000	0.9655	0.9939	0.8861	1.1071	9.086	4	0.337	2.410	0.3568	12	40
123.1	0.6000	0.8276	0.8910	0.7854	1.1071	17.027	4	1.032	2.410	0.3568	16	40
*133	0.4000	0.5517	0.6550	0.1588	0.9912	54.129	4	2.626	2.410	0.3568	24	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)	0.9563594	0.884	-0.267747	1.4656789
Bartlett's Test indicates equal variances (p = 0.13)	8.5282106	15.086272		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	123.1	133	127.95429		0.3448129	0.4615478	0.1157283	0.0438376	0.0584785	5, 18

Treatments vs Control

Parameter	Value	SE	95% Fiducial Limits	Maximum Likelihood-Probit							
				Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter	
Slope	22.949353	13.912075	-4.318316 50.217021	0.275	1.861037	7.8147278	0.6	2.1279882	0.0435742	5	
Intercept	-43.83595	29.397211	-101.4545 13.782584								
TSCR	0.25625	0.0345132	0.1886041 0.3238959								
Point	Probits	ug/L	95% Fiducial Limits								
EC01	2.674	106.32108									
EC05	3.355	113.84527									
EC10	3.718	118.07163									
EC15	3.964	121.01143									
EC20	4.158	123.40001									
EC25	4.326	125.48673									
EC40	4.747	130.90275									
EC50	5.000	134.27284									
EC60	5.253	137.72969									
EC75	5.674	143.67412									
EC80	5.842	146.10368									
EC85	6.036	148.98754									
EC90	6.282	152.69709									
EC95	6.645	158.36579									
EC99	7.326	169.57311									



# Larval Fish Growth and Survival Test-7 Day Growth

Start Date: Test ID: Cyp T1 Sample ID:  
End Date: Lab ID: WESTON - Port Gamble Sample Type:  
Sample Date: Protocol: EPAM 94-EPA/600/4-91/003 Test Species: CV-Cyprinodon variegatus  
Comments: Average

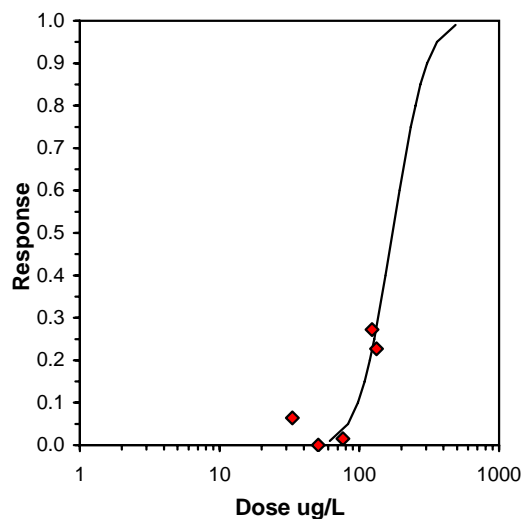
Conc-ug/L	1	2	3	4
Control	0.5350	0.5957	0.6120	0.8050
33.1	0.5057	0.6988	0.6033	0.5760
50.7	0.7150	0.6625	0.6512	0.6400
76.2	0.5071	0.5250	0.7350	0.7414
123.1	0.5880	0.4367	0.4088	0.4220
133	0.3957	0.5250	0.5560	

Conc-ug/L	Mean	N-Mean	Transform: Untransformed					t-Stat	1-Tailed		Mean	N-Mean
			Mean	Min	Max	CV%	N		Critical	MSD		
Control	0.6369	1.0000	0.6369	0.5350	0.8050	18.345	4				0.6369	0.0000
33.1	0.5959	0.9357	0.5959	0.5057	0.6988	13.411	4	0.620	2.567	0.1696	0.5959	0.0643
50.7	0.6672	1.0475	0.6672	0.6400	0.7150	4.972	4	-0.458	2.567	0.1696	0.6672	-0.0475
76.2	0.6271	0.9846	0.6271	0.5071	0.7414	20.488	4	0.148	2.567	0.1696	0.6271	0.0154
*123.1	0.4639	0.7283	0.4639	0.4088	0.5880	18.011	4	2.620	2.567	0.1696	0.4639	0.2717
133	0.4922	0.7728	0.4922	0.3957	0.5560	17.272	3	2.028	2.567	0.1831	0.4922	0.2272

Auxiliary Tests					Statistic	Critical	Skew	Kurt			
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.943793	0.881	0.4526	-0.73715			
Bartlett's Test indicates equal variances (p = 0.50)					4.3304	15.08627					
Hypothesis Test (1-tail, 0.05)		NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Bonferroni t Test		76.2	123.1	96.85154		0.18314	0.287537	0.02598	0.008726	0.04139	5, 17
Treatments vs Control											

Maximum Likelihood-Probit												
Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter	
Slope	5.16847	5.13776	-4.90154	15.23848	0	1.309854	7.814728	0.73	2.237712	0.193481	33	
Intercept	-6.56555	10.78958	-27.7131	14.58203								

Point	Probits	ug/L	95% Fiducial Limits
EC01	2.674	61.32084	
EC05	3.355	83.07382	
EC10	3.718	97.66888	
EC15	3.964	108.9386	
EC20	4.158	118.8158	
EC25	4.326	128.0003	
EC40	4.747	154.4166	
EC50	5.000	172.8669	
EC60	5.253	193.5217	
EC75	5.674	233.4602	
EC80	5.842	251.5067	
EC85	6.036	274.3102	
EC90	6.282	305.9621	
EC95	6.645	359.7159	
EC99	7.326	487.3217	





## 7-Day Chronic Toxicity Test

**Table J2. Chronic *Cyprinodon variegatus* Test 1 (15 Feb 2006)**

Weston Test ID: P060103.16	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	8	7	6	6	6
	2		10	9	9	9	9	7	7
	3		10	10	10*	10*	10*	10	10
	4		10	10	7	7	6	6	6
15	1		10	9	7	7	7	7	7
	2		10	10	9	8	8	8	8
	3		10	10	10	9	9	9	9
	4		10	9	8	7	5	5	5
25	1		10	8	8	8	8	8	8
	2		10	10	8	8	8	9	8
	3		10	10	8	8	8	8	8
	4		10	9	9	9	9	9	9
50	1		7	7	7	7	7	7	7
	2		9	9	8	8	8	8	6
	3		8	8	8*	8*	8	8	8
	4		10	10	10	9	9	7	7
75	1		10	8	5*	5*	5	5	5
	2		10	10	8	7	7	7	6
	3		10	10	10	9	9	8	8
	4		10	10	9	6	6	6	5
100	1		10	9	9	3	3	0	0
	2		10	10	10	8	8	9	7
	3		10	10	8	5	5	4	4
	4		10	10	9	8	8	6	5
Date			2/16/06	2/17/06	2/18/06	2/19/06	2/20/06	2/21/06	2/22/06
Time			1057	1710	1315	1110	1100	937	1340
Initials			EB/JM	TS/JM/EB	TS/EB	TS/EB	GZ	JM	GZ/JM





# 7-Day Chronic Toxicity Test

**Table J3. Chronic *Cyprinodon variegatus* Test 1 (15 Feb 2006)**

Weston Test ID:	P060103.16	Client:	Marine Research Specialists	Client Sample ID:	N/A
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Concentration	Rep	Jar #	Boat Number	Weight Empty Boat (mg)	Weight Boat & Animals (mg)	Total Biomass	Number of Survivors	Individual Biomass (mg/ind.)	Combined Endpoint
Control	1		1	141.11	144.32	3.21	6	0.53	0.32
	2		2	149.05	153.22	4.17	7	0.60	0.42
	3		3	153.04	159.16	6.12	10	0.61	0.61
	4		4	153.34	158.17	4.83	6	0.80	0.48
15	1		5	149.25	152.79	3.54	7	0.51	0.35
	2		6	155.84	161.43	5.59	8	0.70	0.56
	3		7	160.25	165.68	5.43	9	0.60	0.54
	4		8	161.02	163.90	2.88	5	0.58	0.29
25	1		9	157.19	162.91	5.72	8	0.72	0.57
	2		10	99.87	105.17	5.3	8	0.66	0.53
	3		11	102.34	107.55	5.21	8	0.65	0.52
	4		12	103.92	109.68	5.76	9	0.64	0.58
50	1		13	102.32	105.87	3.55	7	0.51	0.36
	2		14	103.39	106.54	3.15	6	0.53	0.32
	3		15	107.91	113.79	5.88	8	0.74	0.59
	4		16	99.12	104.31	5.19	7	0.74	0.52
75	1		17	95.50	98.44	2.94	5	0.59	0.29
	2		18	97.04	99.66	2.62	6	0.44	0.26
	3		19	102.42	105.69	3.27	8	0.41	0.33
	4		20	89.81	91.92	2.11	5	0.42	0.211
100	1		21				0		
	2		22	88.78	91.55	2.77	7	0.40	0.277
	3		23	78.27	80.37	2.1	4	0.53	0.21
	4		24	93.63	96.41	2.78	5	0.56	0.278



## 7-Day Chronic Toxicity Test

**Table J4. Chronic *Cyprinodon variegatus* Test 1 (15 Feb 2006)**

Client	Marine Research Specialists	Date Received:	NA
Project:	Hydrogen Sulfide	Date Test Started:	15-Feb-06
Client Sample ID:	NA	Date Test Ended:	22-Feb-06
Weston Test ID:	P060103.16	Study Director:	Brian Hester
Species:	<i>Cyprinodon variegatus</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/15/06  Time: 1030  Technician: GZ/TS	Control		1		8		19.4		29		8.04	3	0	3
			2		7.6		19.5		29		8.08	1	0	1
			3		7.5		19.4		29		8.03	3	0	3
			4		7.5		19.1		29		8.04	4	0	4
	15		1		7.6		18.9		26		8.24	130	1/5	650
			2		7.5		19.5		27		8.22	140	1/5	700
			3		7.3		19.5		25		8.19	151	1/5	755
			4		7.4		19.4		26		8.18	149	1/5	745
	25		1		7.4		19.4		26		8.24	120	1/5	600
			2		7.3		19.5		26		8.26	148	1/5	740
			3		7.3		19.4		26		8.28	138	1/5	690
			4		7.5		19.2		26		8.31	169	1/5	845
	50		1		7.3		19.4		26		8.42	235	1/5	1175
			2		7.3		19.4		26		8.39	265	1/5	1325
			3		7.4		19.3		26		8.38	293	1/5	1465
			4		7.4		19.3		26		8.40	292	1/5	1460
	75		1		7.2		19.4		25		8.55	168	1/25	4200
			2		7.3		19.2		26		8.54	146	1/25	3650
			3		7.3		19.3		26		8.56	117	1/25	2925
			4		7.3		19.4		26		8.51	179	1/25	4475
	100		1		7.3		19.3		26		8.66	206	1/25	5150
			2		7.3		19.3		26		8.65	289	1/25	7225
			3		7.3		19.1		26		8.62	123	1/25	3075
			4		7.3		19.3		26		8.67	145	1/25	3625
<b>Day 1</b> <b>AM</b> Date: 2/16/06 Time: 0950 Technician: JM/EB	Control				7.1		20.3		29		7.72	12	0	12
	15				6.9		20.4		27		7.95	126	1/5	630
	25				7.0		20.4		27		8.04	244	1/5	1220
	50				6.9		20.3		26		8.21	619	1/5	3095
	75				6.9		20.3		26		8.29	229	1/25	5725
	100				6.9		20.4		26		8.38	292	1/25	7300
<b>Day 1</b> <b>PM</b> Date: 2/16/06 Time: Technician:	Control				7.4		19.1		30		7.82	15	0	15
	15				7.4		19.3		28		7.93	134	1/5	670
	25				7.1		19.8		27		8.06	229	1/5	1145
	50				7.1		19.8		28		8.19	579	1/5	2895
	75				7.0		19.5		28		8.32	261	1/25	6525
	100				6.9		19.8		28		8.39	306	1/25	7650



## 7-Day Chronic Toxicity Test

**Table J4. Chronic *Cyprinodon variegatus* Test 1 (15 Feb 2006)**

Weston Test ID:	P060103.16	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>AM</b> Date: 2/17/06 Time: 1100 Technician: EB/JM	Control		3	A	7.0		20.1		30		7.78	6	0	6
	15				6.8		20.4		28		7.91	134	1/5	670
	25				6.4		20.4		28		7.92	221	1/5	1105
	50				6.6		20.4		29		8.10	456	1/5	2280
	75				6.6		20.4		29		8.26	254	1/25	6350
	100				6.9		20.4		29		8.34	293	1/25	7325
<b>Day 2</b> <b>PM</b> Date: 2/17/06 Time: 1623 Technician: TS	Control		4		7.1		18.7		30		7.78	2	0	2
	15				6.8		18.8		28		7.93	84	1/5	420
	25				6.7		18.9		28		7.95	101	1/5	505
	50				6.5		18.9		28		8.15	258	1/5	1290
	75				6.5		18.8		28		8.28	123	1/25	3075
	100				6.3		18.9		29		8.38	159	1/25	3975
<b>Day 3</b> <b>AM</b> Date: 2/18/06 Time: 1015 Technician: EB/TS	Control		1		7.0		18.5		31		7.97	2	0	2
	15				6.3		18.5		29		8.06	334	0	334
	25				6.3		18.7		28		8.04	39	1/5	195
	50				5.8		18.8		28		8.23	197	1/5	985
	75				5.5		18.5		28		8.23	33	1/25	825
	100				2.0		18.5		29		8.50	92	1/25	2300
<b>Day 3</b> <b>PM</b> Date: 2/18/06 Time: 1530 Technician: TS/EB	Control		2		8.2		18.9		31		7.69	6	0	6
	15				7.7		18.5		29		7.81	66	1/5	330
	25				6.0		18.5		28		7.88	163	1/5	815
	50				8.2		18.0		28		8.10	336	1/5	1680
	75				6.1		18.0		28		8.20	136	1/25	3400
	100				7.9		18.0		29		8.42	154	1/25	3850
<b>Day 4</b> <b>AM</b> Date: 2/19/06 Time: 0925 Technician: EB/TS	Control		3		6.7		18.7		30		7.80	0	0	0
	15				5.0		18.6		28		7.89	70	1/5	350
	25				5.9		18.7		29		8.01	145	1/5	725
	50				0.2		18.8		29		8.07	174	1/5	870
	75				3.2		18.6		28		8.25	41	1/25	1025
	100				0.2		18.6		29		8.37	67	1/25	1675
<b>Day 4</b> <b>PM</b> Date: 2/19/06 Time: 1530 Technician: TS	Control		4		6.6		18.0		31		7.70	0	0	0
	15				5.8		18.6		29		8.12	38	1/5	190
	25				6.0		18.6		29		8.21	167	1/5	835
	50				6.6		18.5		30		8.28	241	1/5	1205
	75				5.3		18.6		29		8.45	56	1/25	1400
	100				6.9		18.6		29		8.56	134	1/25	3350



## 7-Day Chronic Toxicity Test

**Table J4. Chronic *Cyprinodon variegatus* Test 1 (15 Feb 2006)**

Weston Test ID:	P060103.16	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>Day 5</b> <b>AM</b> Date: 2/20/06 Time: 1020 Technician: GZ/EB	Control		1		6.9		18.9		30		7.83	2	0	2
	15				6.5		19.4		28		7.97	110	1/5	550
	25				6.4		19.5		28		8.06	214	1/5	1070
	50				5.2		19.5		28		8.19	555	1/5	2775
	75				4.8		19.3		28		8.33	168	1/25	4200
	100				5.3		19.4		28		8.45	259	1/25	6475
<b>Day 5</b> <b>PM</b> Date: 2/20/06 Time: 1440 Technician: GZ/EB	Control		2		6.6		19.3		30		7.87	5	0	5
	15				6.4		19.5		28		8.01	103	1/5	515
	25				6.1		19.4		28		8.08	192	1/5	960
	50				5.3		19.1		27		8.18	420	1/5	2100
	75				5.0		19.2		28		8.30	180	1/25	4500
	100				5.5		19.2		28		8.51	315	1/25	7875
<b>Day 6</b> <b>AM</b> Date: 2/21/06 Time: 1159 Technician: TS	Control		3		7.7		19.3		30		7.66	8	0	8
	15				7.3		19.3		27		7.74	115	1/5	575
	25				5.5		19.3		27		7.76	175	1/5	875
	50				6.6		19.3		27		7.96	293	1/5	1465
	75				6.8		19.3		27		8.18	160	1/25	4000
	100				6.6		19.2		28		8.16	244	1/25	6100
<b>Day 6</b> <b>PM</b> Date: 2/21/06 Time: 1545 Technician: TS	Control		4		6.6		18.9		30		7.72	0	0	0
	15				6.4		19.5		27		7.87	87	1/5	435
	25				6.3		19.5		27		7.96	219	1/5	1095
	50				5.3		19.5		27		7.98	392	1/5	1960
	75				5.3		19.5		27		8.27	151	1/25	3775
	100				5.2		19.5		27		8.22	200	1/25	5000
<b>Day 7</b>  Date: 2/22/06  Time: 0917  Technician: JM/GZ	Control		1		6.7		18.6		29		8.01	4	0	4
			2		6.6		19.0		29		8.05	6	0	6
			3		6.4		19.1		30		8.05	4	0	4
			4		6.6		19.1		29		8.06	5	0	5
	15		1		6.6		19.0		27		8.13	87	1/5	435
			2		6.3		19.1		27		8.12	77	1/5	385
			3		6.2		19.1		27		8.13	83	1/5	415
			4		6.8		18.9		27		8.14	77	1/5	385
	25		1		6.5		19.0		27		8.23	179	1/5	895
			2		6.2		19.0		27		8.22	186	1/5	930
			3		6.4		19.0		26		8.21	152	1/5	760
			4		6.6		18.9		26		8.21	157	1/5	785
	50		1		5.0		19.0		26		8.25	298	1/5	1490
			2		5.7		18.9		26		8.24	320	1/5	1600
			3		5.9		19.1		26		8.20	257	1/5	1285
			4		5.8		19.0		26		8.23	288	1/5	1440
	75		1		5.3		19.0		26		8.41	73	1/25	1825
			2		5.0		18.9		26		8.43	77	1/25	1925
			3		2.9		19.0		26		8.51	126	1/25	3150
			4		5.3		18.9		26		8.49	142	1/25	3550
	100		1		5.2		18.9		27		8.48	180	1/25	4500
			2		5.0		19.0		27		8.46	234	1/25	5850
			3		4.9		19.0		27		8.39	187	1/25	4675
			4		4.7		18.8		27		8.44	166	1/25	4150

**Table J5. Chronic *Cyprinodon variegatus* Test 2 (15 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4		Day 5		Day 6		Day 7	Mean
		PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	
<b>Control</b>	1	0.2	0.8				0.2				0.0				0.1	<b>0.3</b>
	2	0.2		2.3				0.7				0.4			0.3	<b>0.8</b>
	3	0.3			0.3				0.0				0.0		0.4	<b>0.2</b>
	4	0.3				0.4				0.0				0.2	0.3	<b>0.2</b>
<b>15</b>	1	24.3	47.3				4.3*				2.0*				2.7*	<b>35.8</b>
	2	18.3		40.7				26.5				17.1			3.0*	<b>25.7</b>
	3	32.7			31.0				1.0*				32.8		5.4*	<b>32.2</b>
	4	40.0				15.2				1.5*				1.4*	2.9*	<b>27.6</b>
<b>25</b>	1	30.3	75.1				7.2*				18.7				15.3	<b>34.9</b>
	2	34.5		79.1				47.9				23.4			24.1	<b>41.8</b>
	3	42.7			70.0				2.0*				34.2		16.3	<b>40.8</b>
	4	32.2				19.9				1.8*				19.9	9.1*	<b>24.0</b>
<b>50</b>	1	36.0	131				7.3*				17.7				6.1*	<b>61.7</b>
	2	46.5		104				65.8				27.8			5.8*	<b>61.1</b>
	3	44.1			96.5				41.6				41.8		31.7*	<b>56.0</b>
	4	45.1				55.3				38.4				85.9	15.5*	<b>56.2</b>
<b>75</b>	1	75.4	214				3.8*				19.0				9.8*	<b>102.7</b>
	2	62.3		213				128				47.3			11.0*	<b>112.7</b>
	3	64.0			188				63.4				18.7		16.5*	<b>83.6</b>
	4	66.7				87.9				2.2*				14.6*	19.2*	<b>77.3</b>
<b>100</b>	1	86.2	241				19.3				31.9				3.2*	<b>94.7</b>
	2	58.5		192				74.7				37.3			7.0*	<b>90.6</b>
	3	73.9			207				39.4				96.6		13.6*	<b>104.3</b>
	4	55.9				97.9				35.7				40.7	20.0*	<b>57.6</b>

\* Sulfide concentrations decreased during test

**Table J5. Chronic *Cyprinodon variegatus* Test 2 (15 Feb 2006)**

Treatment	Rep	Percentage Survival							Individual Biomass (mg/ind.)	Combined Endpoint
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7		
Control	1	100	100	100	100	100	100	100	0.50	0.50
	2	100	90	70	70	70	70	70	0.57	0.40
	3	100	100	80	80	80	80	80	0.50	0.40
	4	100	100	100	90	90	90**	90	0.53	0.47
15	1	80	80**	80**	80	80	80	80	0.52	0.42
	2	100	90	80	80	80	80	80	0.54	0.44
	3	100	90	90	90	90	90	90	0.48	0.43
	4	100	90	90	90	90	90	90	0.54	0.49
25	1	100	100	100	90	90	90	90	0.54	0.49
	2	100	90	70	70	70	70	70	0.75	0.53
	3	100	100	100	100	100	100	100	0.42	0.42
	4	100	70	60	50	50	50	50	0.46	0.23
50	1	90	90	80	80	80	80	80	0.44	0.35
	2	90	90	90	90	90	90**	90	0.39	0.35
	3	90	90	90	90	90**	90	90	0.42	0.37
	4	100	80	80	80	80	80	80	0.38	0.30
75	1	80	90	70	60	60	60	60	0.43	0.26
	2	100	90	80	60	60	60	60	0.44	0.26
	3	100	100	60	60	60	60	60	0.39	0.23
	4	90	90	70	60	60	60	50	0.43	0.22
100	1	90	90	30	20	20**	20**	20	0.41	0.08
	2	70	60	60	60	60	60	60	0.34	0.21
	3	100	100	80	80	80	50	50	0.42	0.21
	4	90	70**	70**	70**	70**	70	70	0.45	0.32

\*\* Survival miscounted

### Larval Fish Growth and Survival Test-7 Day Survival

Start Date: 2/15/2006	Test ID: P060103.15	Sample ID:
End Date: 2/22/2006	Lab ID: WESTON - Port Gamble	Sample Type:
Sample Date:	Protocol: EPAM 94-EPA/600/4-91/003	Test Species: CV-Cyprinodon variegatus
Comments: Average		

Conc-ug/L	1	2	3	4
Control	1.0000	0.7000	0.8000	0.9000
30.3	0.8000	0.8000	0.9000	0.9000
35.4	0.9000	0.7000	0.1000	0.5000
58.8	0.8000	0.9000	0.9000	0.8000
86.8	0.2000	0.6000	0.5000	0.7000
94.1	0.6000	0.6000	0.6000	0.5000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				N	t-Stat	1-Tailed Critical	MSD	Number Resp	Total Number
			Mean	Min	Max	CV%						
Control	0.8500	1.0000	1.1898	0.9912	1.4120	15.281	4				6	40
30.3	0.8500	1.0000	1.1781	1.1071	1.2490	6.954	4	0.081	2.410	0.3512	6	40
*35.4	0.5500	0.6471	0.8368	0.3218	1.2490	46.878	4	2.422	2.410	0.3512	18	40
58.8	0.8500	1.0000	1.1781	1.1071	1.2490	6.954	4	0.081	2.410	0.3512	6	40
*86.8	0.5000	0.5882	0.7816	0.4636	0.9912	29.171	4	2.801	2.410	0.3512	20	40
94.1	0.5750	0.6765	0.8609	0.7854	0.8861	5.847	4	2.257	2.410	0.3512	17	40

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )	0.9279178	0.884	-0.64536	2.5106286
Bartlett's Test indicates equal variances ( $p = 0.02$ )	13.999896	15.086272		

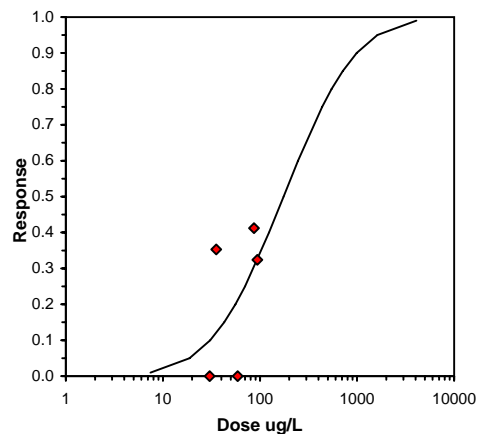
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	30.3	35.4	32.750878		0.3086579	0.3581717	0.1544404	0.0424822	0.0189622	5, 18

Treatments vs Control

Parameter	Value	SE	95% Fiducial Limits	Maximum Likelihood-Probit						
				Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	1.6981185	2.1567899	-5.165749 8.5619864	0.15	16.942727	7.8147278	7.3E-04	2.2421	0.5888871	7
Intercept	1.1926486	3.9742812	-11.45529 13.840585							
TSCR	0.1556361	0.1356302	-0.276 0.5872719							

Point	Probits	ug/L	95% Fiducial Limits
EC01	2.674	7.4497865	
EC05	3.355	18.770117	
EC10	3.718	30.719268	
EC15	3.964	42.83096	
EC20	4.158	55.780211	
EC25	4.326	69.968363	
EC40	4.747	123.85318	
EC50	5.000	174.62242	
EC60	5.253	246.20273	
EC75	5.674	435.8111	
EC80	5.842	546.66324	
EC85	6.036	711.93815	
EC90	6.282	992.63379	
EC95	6.645	1624.5495	
EC99	7.326	4093.1379	

Significant heterogeneity detected ( $p = 7.26E-04$ )



# Larval Fish Growth and Survival Test-7 Day Growth

Start Date: Test ID: Cyp T2 Sample ID:  
End Date: Lab ID: WESTON - Port Gamble Sample Type:  
Sample Date: Protocol: EPAM 94-EPA/600/4-91/003 Test Species: CV-Cyprinodon variegatus  
Comments: Average

Conc-ug/L	1	2	3	4
Control	0.5030	0.5686	0.5013	0.5256
30.3	0.5225	0.5438	0.4800	0.5400
35.4	0.5422	0.7500	4.1800	0.4620
58.8	0.4400	0.3856	0.4156	0.3775
86.8	1.2950	0.4367	0.4660	0.3100
94.1	0.1383	0.3433	0.3500	0.6360

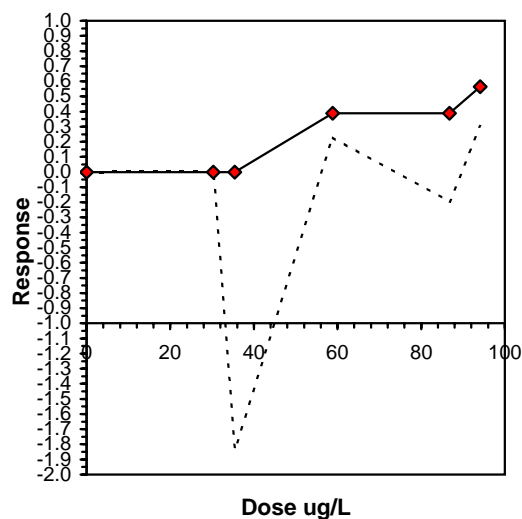
Conc-ug/L	Mean	N-Mean	Transform: Untransformed					Rank Sum	1-Tailed Critical	Isotonic	
			Mean	Min	Max	CV%	N			Mean	N-Mean
Control	0.5246	1.0000	0.5246	0.5013	0.5686	5.974	4			0.8432	1.0000
30.3	0.5216	0.9942	0.5216	0.4800	0.5438	5.601	4	18.00	10.00	0.8432	1.0000
35.4	1.4836	2.8280	1.4836	0.4620	4.1800	121.446	4	21.00	10.00	0.8432	1.0000
*58.8	0.4047	0.7714	0.4047	0.3775	0.4400	7.091	4	10.00	10.00	0.5158	0.6117
86.8	0.6269	1.1951	0.6269	0.3100	1.2950	71.860	4	14.00	10.00	0.5158	0.6117
94.1	0.3669	0.6994	0.3669	0.1383	0.6360	55.743	4	14.00	10.00	0.3669	0.4351

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution ( $p \leq 0.01$ )	0.651774	0.884	2.709599	11.58776
Bartlett's Test indicates unequal variances ( $p = 5.20E-11$ )	56.94041	15.08627		

Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	35.4	58.8	45.62368	
Treatments vs Control				

Linear Interpolation (200 Resamples)				
Point	ug/L	SD	95% CL(Exp)	Skew

IC05	38.413			
IC10	41.426			
IC15	44.439			
IC20	47.452			
IC25	50.465			
IC40	87.283			
IC50	91.418			







## 7-Day Chronic Toxicity Test

**Table J6. Chronic *Cyprinodon variegatus* Test 2 (15 Feb 2006)**

Weston Test ID: P060103.15	Client: Marine Research Specialists	Client Sample ID: N/A
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	10	10	10	10	10	10
	2		10	9	7	7	7	7	7
	3		10	10	8	8	8	8	8
	4		10	10	10	9	9	9*	9
15	1		8	8*	8*	8	8	8	8
	2		10	9	8	8	8	8	8
	3		10	9	9	9	9	9	9
	4		10	9	9	9	9	9	9
25	1		10	10	10	9	9	9	9
	2		10	9	7	7	7	7	7
	3		10	10	10	10	10	10	10
	4		10	7	6	5	5	5	5
50	1		9	9	8	8	8	8	8
	2		9	9	9	9	9	9*	9
	3		9	9	9	9	9*	9	9
	4		10	8	8	8	8	8	8
75	1		8	9	7	6	6	6	6
	2		10	9	8	6	6	6	6
	3		10	10	6	6	6	6	6
	4		9	9	7	6	6	6	5
100	1		9	9	3	2	2*	2*	2
	2		7	6	6	6	6	6	6
	3		10	10	8	8	8	5	5
	4		9	7*	7*	7*	7*	7	7
Date			2/16/06	2/17/06	2/18/06	2/19/06	2/20/06	2/21/06	2/22/06
Time			1057	1710	1315	1110	1100	937	1340
Initials			EB/JM	TS/JM/EB	EB/TS	EB/TS	GZ	JM	GZ

\* Survival miscounted



## 7-Day Chronic Toxicity Test

**Table J7. Chronic *Cyprinodon variegatus* Test 2 (15 Feb 2006)**

Weston Test ID:	P060103.15	Client:	Marine Research Specialists	Client Sample ID:	N/A
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Concentration	Rep	Jar #	Boat Number	Weight Empty Boat (mg)	Weight Boat & Animals (mg)	Total Biomass	Number of Survivors	Individual Biomass (mg/ind.)	Combined Endpoint
Control	1		25	98.08	103.11	5.03	10.00	0.50	0.50
	2		26	95.29	99.27	3.98	7.00	0.57	0.40
	3		27	95.64	99.65	4.01	8.00	0.50	0.40
	4		28	98.94	103.67	4.73	9.00	0.53	0.47
15	1		29	73.15	77.33	4.18	8.00	0.52	0.42
	2		30	76.44	80.79	4.35	8.00	0.54	0.44
	3		31	110.72	115.04	4.32	9.00	0.48	0.43
	4		32	115.29	120.15	4.86	9.00	0.54	0.49
25	1		33	101.62	106.50	4.88	9.00	0.54	0.49
	2		34	104.08	109.33	5.25	7.00	0.75	0.53
	3		35	101.52	105.70	4.18	10.00	0.42	0.42
	4		36	94.71	97.02	2.31	5.00	0.46	0.23
50	1		37	78.41	81.93	3.52	8.00	0.44	0.35
	2		38	74.18	77.65	3.47	9.00	0.39	0.35
	3		39	69.90	73.64	3.74	9.00	0.42	0.37
	4		40	71.92	74.94	3.02	8.00	0.38	0.30
75	1		41	80.40	82.99	2.59	6.00	0.43	0.26
	2		42	86.13	88.75	2.62	6.00	0.44	0.26
	3		43	97.98	100.31	2.33	6.00	0.39	0.23
	4		44	92.91	95.08	2.17	5.00	0.43	0.22
100	1		45	100.86	101.69	0.83	2.00	0.41	0.08
	2		46	79.63	81.69	2.06	6.00	0.34	0.21
	3		47	82.68	84.78	2.10	5.00	0.42	0.21
	4		48	92.57	95.75	3.18	7.00	0.45	0.32



## 7-Day Chronic Toxicity Test

**Table J8. Chronic *Cyprinodon variegatus* Test 2 (15 Feb 2006)**

Client	Marine Research Specialists	Date Received:	NA
Project:	Hydrogen Sulfide	Date Test Started:	15-Feb-06
Client Sample ID:	NA	Date Test Ended:	22-Feb-06
Weston Test ID:	P060103.15	Study Director:	Brian Hester
Species:	<i>Cyprinodon variegatus</i>	# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/15/06  Time: 1030  Technician: GZ,TS	Control		1		7.3		19.7		29		8.05	3	0	3
			2		7.4		19.6		29		8.08	4	0	4
			3		7.6		19.3		29		8.04	5	0	5
			4		7.4		19.4		29		8.08	3	0	5
	15		1		7.9		18.9		26		8.20	111	1:05	555
			2		7.5		19.5		27		8.22	88	1:05	440
			3		7.0		19.5		25		8.21	152	1:05	760
			4		7.2		19.4		26		8.19	179	1:05	895
	25		1		7.1		19.6		26		8.23	148	1:05	740
			2		7.1		19.5		26		8.26	180	1:05	900
			3		7.0		19.4		26		8.22	204	1:05	1,020
			4		7.3		19.2		26		8.32	192	1:05	960
	50		1		6.8		19.5		26		8.22	172	1:05	860
			2		7.0		19.2		26		8.41	339	1:05	1,695
			3		7.2		19.4		26		8.36	288	1:05	1,440
			4		7.4		19.4		26		8.40	322	1:05	1,610
	75		1		6.9		19.3		25		8.55	150	1:25	3,750
			2		6.6		19.4		26		8.42	93	1:25	2,325
			3		6.8		19.4		26		8.51	117	1:25	2,925
			4		6.9		19.4		26		8.51	122	1:25	3,050
	100		1		7.0		19.4		26		8.62	202	1:25	5,050
			2		7.0		19.3		26		8.72	172	1:25	4,300
			3		7.0		19.4		26		8.69	203	1:25	5,075
			4		6.9		19.2		26		8.70	157	1:25	3,925
<b>Day 1</b> <b>AM</b> Date: 2/16/06 Time: 0940 Technician: EB/JM	Control				7.1		20.0		29		7.77	7	0	7
	15				6.8		20.4		27		7.93	121	1:05	605
	25				6.8		20.5		27		8.01	228	1:05	1,140
	50				6.8		20.4		26		8.19	587	1:05	2,935
	75				6.7		20.3		26		8.26	223	1:25	5,575
	100				6.7		20.3		26		8.36	315	1:25	7,875
<b>Day 1</b> <b>PM</b> Date: Time: Technician:	Control				7.2		19.6		30		7.74	20	0	20
	15				7.1		19.8		28		7.97	114	1:05	570
	25				6.9		19.9		27		8.04	256	1:05	1,280
	50				6.7		20.1		28		8.22	503	1:05	2,515
	75				6.1		19.9		28		8.22	206	1:25	5,150
	100				6.4		19.8		28		8.39	271	1:25	6,775



## 7-Day Chronic Toxicity Test

**Table H8. Chronic *Cyprinodon variegatus* Test 2 (15 Feb 2006)**

Weston Test ID:	P060103.15	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>Day 2</b> <b>AM</b> Date: 2/17/06 Time: 1100 Technician: EB/JM	Control		3		7.3		19.2		30		7.76	3	0	3
	15				6.4		20.3		28		7.87	70	1:05	350
	25				6.3		20.4		28		7.92	176	1:05	880
	50				6.2		20.4		29		8.07	337	1:05	1,685
	75				6.4		20.4		29		8.23	187	1:25	4,675
	100				6.6		20.3		29		8.34	263	1:25	6,575
<b>Day 2</b> <b>PM</b> Date: 2/17/06 Time: 1623 Technician: TS	Control		4		7.1		18.8		30		7.81	4	0	4
	15				6.3		18.8		28		7.88	35	1:05	175
	25				6.2		18.9		28		7.91	49	1:05	245
	50				5.5		18.8		28		8.13	219	1:05	1,095
	75				5.5		18.8		28		8.28	97	1:25	2,425
	100				6.2		19.0		29		8.40	142	1:25	3,550
<b>Day 3</b> <b>AM</b> Date: 2/18/06 Time: 1015 Technician: TS/EB	Control		1		7.1		18.5		31		7.83	2	0	2
	15				6.1		18.5		29		8.03	69	0	69
	25				5.7		18.7		28		8.01	22	1:05	110
	50				4.4		18.7		28		8.13	29	1:05	145
	75				2.3		18.5		28		8.36	5	1:25	125
	100				1.9		18.5		29		8.50	35	1:25	875
<b>Day 3</b> <b>PM</b> Date: 2/18/06 Time: 1530 Technician: TS/EB	Control		2		8.6		18.8		31		7.66	5	0	5
	15				8.2		18.5		29		7.80	52	1:05	260
	25				7.7		18.5		28		7.86	106	1:05	530
	50				7.5		18.0		28		8.05	219	1:05	1,095
	75				4.6		18.0		28		8.07	89	1:25	2,225
	100				6.3		17.9		29		8.36	99	1:25	2,475
<b>Day 4</b> <b>AM</b> Date: 2/19/06 Time: 0925 Technician: EB/TS	Control		3		6.8		18.5		30		7.75	0	0	0
	15				5.5		18.6		28		7.83	2	1:05	10
	25				4.8		18.6		29		7.80	4	1:05	20
	50				3.2		18.8		29		8.03	133	1:05	665
	75				3.9		18.5		28		8.24	64	1:25	1,600
	100				1.3		18.6		29		8.34	50	1:25	1,250
<b>Day 4</b> <b>PM</b> Date: 2/19/06 Time: 1530 Technician: TS	Control		4		6.8		18.5		31		7.97	0	0	0
	15				5.5		18.6		29		8.05	5	1:05	25
	25				5.1		18.6		29		8.11	7	1:05	35
	50				5.6		18.5		30		8.27	209	1:05	1,045
	75				4.3		18.5		29		8.38	3	1:25	75
	100				5.2		18.6		29		8.57	76	1:25	1,900



## 7-Day Chronic Toxicity Test

**Table H8. Chronic *Cyprinodon variegatus* Test 2 (15 Feb 2006)**

Weston Test ID:	P060103.15	Client:	Marine Research Specialists	Client Sample ID:	N/A
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>Day 5</b> <b>AM</b> Date: 2/20/06 Time: 1020 Technician: GZ/EB	Control		1		6.5		19.3		30		7.77	0	0	0
	15				5.6		19.5		28		7.92	5	1:05	25
	25				4.7		19.6		28		8.01	57	1:05	285
	50				0.8		19.5		28		8.05	59	1:05	295
	75				1.2		19.3		28		8.28	21	1:25	525
	100				2.1		19.4		28		8.37	43	1:25	1,075
<b>Day 5</b> <b>PM</b> Date: 2/20/06 Time: 1440 Technician: GZ/EB	Control		2		6.5		19.3		30		7.84	4	0	4
	15				5.6		19.5		28		7.92	43	1:05	215
	25				4.8		19.4		28		7.98	67	1:05	335
	50				2.9		19.3		27		8.05	92	1:05	460
	75				4.0		19.3		28		8.16	40	1:25	1,000
	100				4.5		19.3		28		8.51	69	1:25	1,725
<b>Day 6</b> <b>AM</b> Date: 2/21/06 Time: 1159 Technician: TS	Control		3		7.4		19.4		30		7.58	0	0	0
	15				5.5		19.2		27		7.65	47	1:05	235
	25				5.3		19.2		27		7.75	60	1:05	300
	50				4.6		19.3		27		7.86	92	1:05	460
	75				1.8		19.2		27		7.95	10	1:25	250
	100				4.0		19.2		29		8.06	66	1:25	1,650
<b>Day 6</b> <b>PM</b> Date: 2/21/06 Time: 1545 Technician: TS	Control		4		5.6		19.4		30		7.46	1	0	1
	15				6.0		19.4		27		7.64	2	1:05	10
	25				4.6		19.5		27		7.85	43	1:05	215
	50				3.2		19.5		27		7.80	167	1:05	835
	75				2.0		19.5		27		8.11	11	1:25	275
	100				2.1		19.5		27		8.17	35	1:25	875
<b>Day 7</b>  Date: 2/22/06  Time: 0917  Technician: JM/GZ	Control		1		6.2		19.0		29		8.00	2	0	2
			2		6.3		19.0		29		8.02	4	0	4
			3		6.2		19.2		30		7.99	6	0	6
			4		5.7		19.1		29		8.00	5	0	5
	15		1		5.2		19.1		27		8.05	9	1:05	45
			2		5.3		19.0		27		8.09	11	1:05	55
			3		5.6		19.0		27		8.05	18	1:05	90
			4		6.1		19.0		27		7.96	8	1:05	40
	25		1		5.2		19.1		27		8.07	53	1:05	265
			2		4.9		19.0		27		8.11	91	1:05	455
			3		5.1		19.0		26		8.13	64	1:05	320
			4		5.3		19.0		26		8.11	34	1:05	170
	50		1		4.3		18.9		26		8.15	25	1:05	125
			2		3.8		19.0		26		8.19	26	1:05	130
			3		3.6		19.1		26		8.15	130	1:05	650
			4		5.1		18.9		26		8.14	62	1:05	310
	75		1		3.6		18.9		26		8.33	12	1:25	300
			2		0.5		18.9		26		8.24	11	1:25	275
			3		3.2		18.9		26		8.37	22	1:25	550
			4		4.3		18.9		26		8.36	25	1:25	625
	100		1		3.0		18.9		27		8.34	4	1:25	100
			2		1.8		19.0		27		8.30	8	1:25	200
			3		3.7		18.9		27		8.25	14	1:25	350
			4		6.5		18.8		27		8.30	23	1:25	575

**Table K1. Chronic *Americamysis bahia* (1 Feb 2006)**

Treatment	Rep	Day 0	Day 1		Day 2		Day 3		Day 4		Day 5		Day 6		Day 7	Mean
		PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	PM	AM	
Control	1	0.0	0.0				0.2				0.5				0.0	0.1
	2	0.0		0.0				0.6				0.0			0.0	0.1
	3	0.2			0.3				0.3				0.0		0.0	0.2
	4	0.0				0.4				0.2				0.0	0.0	0.1
2.5	1	6.3	6.8				3.2				2.8				1.0*	4.8
	2	5.5		4.7				4.4				2.1			0.5*	4.2
	3	6.1			1.5				2.7				0.8*		0.7*	3.4
	4	5.8				2.9				1.9				1.4*	0.4*	3.5
5	1	16.9	14.9				10.2				7.8				2.7*	12.4
	2	16.0		13.0				12.8				9.1			2.2*	12.7
	3	15.0			2.8*				9.5				3.5*		1.8*	12.2
	4	16.9				7.8				7.0				3.8*	2.6*	10.6
10	1	37.6	35.5													36.6
	2	32.6		31.1												31.8
	3	35.1			15.3											25.2
	4	33.4				16.0										24.7
15	1	64.7	56.2													60.5
	2	55.9		50.6												53.3
	3	58.7														58.7
	4	85.2														85.2
20	1	78.0	72.7													75.4
	2	73.1		65.0												69.0
	3	70.8														70.8
	4	69.4														69.4

\* Sulfide concentrations decreased during test

**Table K1. Chronic *Americamysis bahia* (1 Feb 2006)**

Treatment	Rep	Percentage Survival							Individual Biomass (mg/ind.)	Combined Endpoint
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7		
Control	1	100	80	80	80**	80	80	80	0.26	0.21
	2	80**	80	80	80	80	80	80	0.32	0.25
	3	100	100	100	100	90	90	90	0.24	0.21
	4	100	100	100	100	100	100	100	0.25	0.25
2.5	1	100	100	100	90	90	90	90	0.26	0.24
	2	100	100	90	80	80	80	80	0.20	0.16
	3	100	100	100	100	100	100	100	0.25	0.25
	4	100	100	100	100	100	100	100	0.20	0.20
5	1	100	80	20	20	20	20	20	0.19	0.04
	2	100	100	40	30	20	20	20	0.50	0.10
	3	100	90	80	60	60	50	30	0.20	0.06
	4	100	80	60	30	20	10	0	NC	NC
10	1	90	70	0	T	T	T	T	NC	NC
	2	50	10	0	T	T	T	T	NC	NC
	3	70	10	0	T	T	T	T	NC	NC
	4	100	10	0	T	T	T	T	NC	NC
15	1	90	10	0	T	T	T	T	NC	NC
	2	90	0	T	T	T	T	T	NC	NC
	3	70	0	T	T	T	T	T	NC	NC
	4	70	0	T	T	T	T	T	NC	NC
20	1	80	0	T	T	T	T	T	NC	NC
	2	70	0	T	T	T	T	T	NC	NC
	3	80	0	T	T	T	T	T	NC	NC
	4	50	0	T	T	T	T	T	NC	NC

T = Test terminated due to 0% survival

NC = Not calculable

\*\* Survival miscounted

# Mysid Survival, Growth and Fecundity Test-7 Day Survival

Start Date: 2/1/2006 Test ID: P060103.07 Sample ID:  
End Date: 2/8/2006 Lab ID: WESTON - Port Gamble Sample Type:  
Sample Date: Protocol: EPAM 94-EPA/600/4-91/003 Test Species: MY-Mysidopsis bahia  
Comments: Average

Conc-ug/L	1	2	3	4
Control	0.8000	0.8000	0.9000	1.0000
4	0.9000	0.8000	1.0000	1.0000
12	0.2000	0.2000	0.3000	0.0000
29.6	0.0000	0.0000	0.0000	0.0000
64.4	0.0000	0.0000	0.0000	0.0000
71.2	0.0000	0.0000	0.0000	0.0000

Conc-ug/L	Mean	N-Mean	Transform: Arcsin Square Root				Rank Sum	1-Tailed Critical	Number Resp	Total Number
			Mean	Min	Max	CV%				
Control	0.8750	1.0000	1.2188	1.1071	1.4120	11.906	4		5	40
4	0.9250	1.0571	1.2951	1.1071	1.4120	11.347	4	20.50	3	40
*12	0.1750	0.2000	0.4164	0.1588	0.5796	43.287	4	10.00	33	40
*29.6	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	40	40
*64.4	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	40	40
*71.2	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	4	10.00	40	40

## Auxiliary Tests

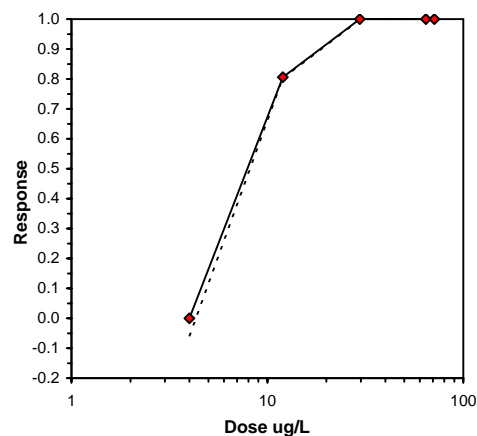
Shapiro-Wilk's Test indicates normal distribution ( $p > 0.01$ )  
Equality of variance cannot be confirmed

## Hypothesis Test (1-tail, 0.05)

Steel's Many-One Rank Test

Treatments vs Control

Trim Level	EC50	95% CL	
0.0%	8.4165	7.4257	9.5394
5.0%	8.2170	7.1771	9.4075
10.0%	8.0565	7.0095	9.2598
20.0%	7.9105	7.1153	8.7946
Auto-0.0%	8.4165	7.4257	9.5394





### Mysid Survival, Growth and Fecundity Test-Growth-Weight

Start Date:	Test ID: Mysid T1	Sample ID:
End Date:	Lab ID: WESTON - Port Gamble	Sample Type:
Sample Date:	Protocol: EPAM 94-EPA/600/4-91/003	Test Species: MY-Mysidopsis bahia
Comments: Average		

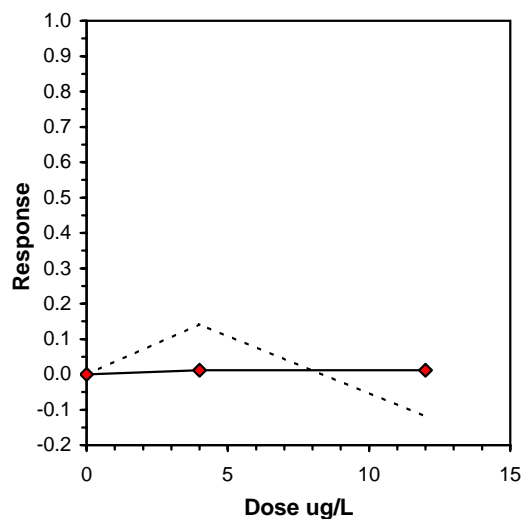
Conc-ug/L	1	2	3	4
Control	0.2625	0.3175	0.2378	0.2470
4	0.2611	0.2000	0.2500	0.2030
12	0.1850	0.5050	0.2033	

Conc-ug/L	Mean	N-Mean	Transform: Untransformed					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
Control	0.2662	1.0000	0.2662	0.2378	0.3175	13.408	4				0.2662	1.0000
4	0.2285	0.8585	0.2285	0.2000	0.2611	13.810	4	0.564	2.306	0.1540	0.2632	0.9886
12	0.2978	1.1186	0.2978	0.1850	0.5050	60.345	3	-0.438	2.306	0.1664	0.2632	0.9886

Auxiliary Tests					Statistic	Critical	Skew	Kurt			
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.875517	0.792	1.363909	3.371033			
Bartlett's Test indicates equal variances (p = 0.02)					8.389171	9.21034					
Hypothesis Test (1-tail, 0.05)		NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Bonferroni t Test		12	>12			0.166376	0.625018	0.004192	0.008924	0.641382	2, 8
Treatments vs Control											

Linear Interpolation (200 Resamples)				
Point	ug/L	SD	95% CL(Exp)	Skew

IC05	>12
IC10	>12
IC15	>12
IC20	>12
IC25	>12
IC40	>12
IC50	>12





## 7-Day Chronic Toxicity Test

**Table K2. Chronic *Americamysis bahia* (1 Feb 2006)**

Weston Test ID: P060103.07	Client: Marine Research Specialists	Client Sample ID:
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### Survival Data

Concentration	Rep	Jar #	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Control	1		10	8	8	8*	8	8	8
	2		8*	8	8	8	8	8	8
	3		10	10	10	10	9	9	9
	4		10	10	10	10	10	10	10
2.5	1		10	10	10	9	9	9	9
	2		10	10	9	8	8	8	8
	3		10	10	10	10	10	10	10
	4		10	10	10	10	10	10	10
5	1		10	8	2	2	2	2	2
	2		10	10	4	3	2	2	2
	3		10	9	8	6	6	5	3
	4		10	8	6	3	2	1	0
10	1		9	7	0	0	0	0	0
	2		5	1	0	0	0	0	0
	3		7	1	0	0	0	0	0
	4		10	1	0	0	0	0	0
15	1		9	1	0	0	0	0	0
	2		9	0	0	0	0	0	0
	3		7	0	0	0	0	0	0
	4		7	0	0	0	0	0	0
20	1		8	0	0	0	0	0	0
	2		7	0	0	0	0	0	0
	3		8	0	0	0	0	0	0
	4		5	0	0	0	0	0	0
Date			2/2/2006	2/3/2006	2/4/2006	2/5/2006	2/6/2006	2/7/2006	2/8/2006
Time			1025	1015	1035	919	1300	1030	1735
Initials			GZ	JM/GZ	JM	JM	GZ	GZ	AM

\* Survival miscounted



# 7-Day Chronic Toxicity Test

**Table K3. Chronic *Americamysis bahia* (1 Feb 2006)**

Weston Test ID:	P060103.07	Client:	Marine Research Specialists	Client Sample ID:	N/A
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Concentration	Rep	Jar #	Weight Empty Boat (mg)	Weight Boat & Animals (mg)	Total Biomass	Number of Survivors	Individual Biomass (mg/ind.)	Combined Endpoint
Control	1	1	85.99	88.09	2.1	8	0.26	0.21
	2	2	82.13	84.67	2.54	8	0.32	0.25
	3	3	78.61	80.75	2.14	9	0.24	0.21
	4	4	60.47	62.94	2.47	10	0.25	0.25
2.5	1	5	79.05	81.40	2.35	9	0.26	0.24
	2	6	73.18	74.78	1.6	8	0.20	0.16
	3	7	74.15	76.65	2.5	10	0.25	0.25
	4	8	76.36	78.39	2.03	10	0.20	0.20
5	1	9	77.16	77.53	0.37	2	0.19	0.04
	2	10	67.18	68.19	1.01	2	0.50	0.10
	3	11	62.97	63.58	0.61	3	0.20	0.06
	4	12	58.26			0		
10	1	13	77.36			0		
	2	14	66.73			0		
	3	15	72.33			0		
	4	16	68.64			0		
15	1	17	68.62			0		
	2	18	71.13			0		
	3	19	84.57			0		
	4	20	82.04			0		
20	1	21	73.47			0		
	2	22	76.90			0		
	3	23	79.44			0		
	4	24	78.84			0		



## 7-Day Chronic Toxicity Test

**Table K4. Chronic *Americamysis bahia* (1 Feb 2006)**

Client	Marine Research Specialists
Project:	Hydrogen Sulfide
Client Sample ID:	N/A
Weston Test ID:	P060103.07
Species:	<i>Americamysis bahia</i>

Date Received:	
Date Test Started:	1-Feb-06
Date Test Ended:	8-Feb-06
Study Director:	Brian Hester
# Organisms/Chamber:	10

	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
												Value	Dil	Corr value
<b>Day 0</b>  Date: 2/1/06  Time: 1704  Technician: JM/GZ	Control		1		8.5		19.4		28		7.76	0	0	0
			2		8.4		18.8		28		7.77	0	0	0
			3		8.4		18.8		29		7.81	2	0	2
			4		8.5		19.5		29		7.54	0	0	0
	2.5		1		8.2		19.6		26		7.83	65	0	65
			2		8.1		19.3		26		7.86	60	0	60
			3		8.2		19.1		27		7.85	66	0	66
			4		8.1		19.3		27		7.84	61	0	61
	5		1		8.2		19.3		27		7.85	182	0	182
			2		8.1		19.5		27		7.87	180	0	180
			3		8.1		19.6		26		7.9	179	0	179
			4		8.1		19.7		26		7.86	185	0	185
	10		1		8.2		19.5		27		7.93	481	0	481
			2		8.1		19.7		27		7.94	425	0	425
			3		8.1		19.9		27		7.93	449	0	449
			4		8.1		19.7		27		7.95	445	0	445
	15		1		8.1		19.9		26		7.92	161	1/5	805
			2		8.1		20.1		27		7.98	159	1/5	795
			3		8.1		20.0		27		7.98	167	1/5	835
			4		8.1		19.8		27		7.84	180	1/5	900
	20		1		8.1		19.7		27		7.98	222	1/5	1110
			2		8.1		19.8		27		8.00	217	1/5	1085
			3		8.1		19.7		27		8.01	215	1/5	1075
			4		8.0		19.8		27		8.03	220	1/5	1100
<b>Day 1</b> <b>AM</b> Date: 2/2/06 Time: 0946 Technician: TS/JM	Control		1		8.0		19.9		28		7.7	0	0	0
	2.5				7.1		20.3		27		7.8	66	0	66
	5				7.7		20.2		27		7.9	178	0	178
	10				7.6		20.3		27		7.9	426	0	426
	15				7.7		20.1		27		8.0	167	1/5	835
	20				7.8		19.9		28		8.0	217	1/5	1085
<b>Day 1</b> <b>PM</b> Date: 2/2/06 Time: 1445 Technician: GZ	Control		2		7.9		19.7		29		7.77	0	0	0
	2.5				7.8		19.8		27		7.84	50	0	50
	5				7.8		20.0		27		7.86	143	0	143
	10				7.8		20.0		27		7.91	381	0	381
	15				7.6		20.1		27		7.95	135	1/5	675
	20				7.7		20.0		27		7.99	189	1/5	945



## 7-Day Chronic Toxicity Test

**Table K4. Chronic *Americamysis bahia* (1 Feb 2006)**

Weston Test ID:	P060103.07	Client:	Marine Research Specialists	Client Sample ID:	
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>Day 2</b> <b>AM</b> Date: 2/3/06 Time: 0844 Technician: JM/TS/GZ	Control		3		8.0		19.6		30		7.90	3	0	3
	2.5				7.8		20.0		27		7.92	19	0	19
	5				7.7		20.0		27		8.07	49	0	49
	10				7.6		19.9		27		8.09	277	0	277
	15				7.5		19.9		27		8.15	110	1/5	550
	20				7.7		19.9		27		8.16	167	1/5	835
<b>Day 2</b> <b>PM</b> Date: 2/3/06 Time: 1400 Technician: TS/JM/GZ	Control		4		8.0		19.4		29		7.87	5	0	5
	2.5				7.7		19.9		28		8.00	43	0	43
	5				7.8		19.9		27		8.05	129	0	129
	10				7.7		19.9		28		8.09	290	0	290
	15				7.7		20.1		28		8.16	132	1/5	660
	20				7.6		20.1		27		8.20	209	1/5	1045
<b>Day 3</b> <b>AM</b> Date: 2/4/06 Time: 0905 Technician: BH/JM	Control		1		7.2		19.3		30		7.77	2	0	2
	2.5				7.4		19.2		28		7.67	24	0	24
	5				7.3		19.5		29		7.68	78	0	78
	10				7.3		19.5		28		7.73	207	0	207
	15				7.2		19.5		29		7.78	93	1/5	465
	20				7.1		19.4		29		7.85	113	1/5	565
<b>Day 3</b> <b>PM</b> Date: 2/4/06 Time: 1715 Technician: BH	Control		2		7.9		19.3		30		7.83	6	0	6
	2.5				7.7		19.6		27		7.88	50	0	50
	5				7.7		19.6		27		7.91	157	0	157
	10				7.4		19.7		27		7.99	379	0	379
	15				7.5		19.6		27		8.01	140	1/5	700
	20				7.3		19.7		27		8.04	168	1/5	840
<b>Day 4</b> <b>AM</b> Date: 2/5/06 Time: 0827 Technician: JM	Control		3		8.1		18.6		29		7.77	3	0	3
	2.5				7.8		19.1		27		7.87	30	0	30
	5				7.7		19.3		27		7.89	111	0	111
	10				7.5		19.4		27		7.90	288	0	288
	15				6.9		19.5		27		7.95	105	1/5	525
	20				6.1		19.5		27		7.92	120	1/5	600
<b>Day 4</b> <b>PM</b> Date: 2/5/06 Time: 1935 Technician: BH	Control		4		7.8		19.3		30		7.94	3	0	3
	2.5				7.5		19.7		27		7.99	27	0	27
	5				7.5		19.7		26		8.03	111	0	111
	10				7.2		19.6		27		8.04	236	0	236
	15				7.3		19.7		27		8.11	119	1/5	595
	20				7.2		19.5		27		8.11	148	1/5	740



## 7-Day Chronic Toxicity Test

**Table K4. Chronic *Americamysis bahia* (1 Feb 2006)**

Weston Test ID:	P060103.07	Client:	Marine Research Specialists	Client Sample ID:	
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	Conc.	Jar #	Rep	Meter #	D.O. (mg/L)	Meter #	Temp (°C)	Meter #	Sal. (ppt)	Meter #	pH	Total Sulfide (µg/L)		
<b>Day 5</b> <b>AM</b> Date: 2/6/06 Time: 0955 Technician: GZ/TS	Control		1		7.5		19.8		28		7.72	4	0	4
	2.5				7.5		19.9		27		7.84	29	0	29
	5				7.6		20.0		27		7.88	90	0	90
	10				7.4		20.1		27		7.91	205	0	205
	15				7.2		20.1		27		7.95	63	1/5	315
	20				7.2		20.1		27		7.95	84	1/5	420
<b>Day 5</b> <b>PM</b> Date: 2/6/06 Time: 1600 Technician: GZ	Control		2		7.7		19.6		29		7.74	0	0	0
	2.5				7.8		19.7		27		7.82	21	0	21
	5				7.8		19.8		27		7.88	104	0	104
	10				7.7		19.9		27		7.92	203	0	203
	15				7.7		19.8		27		7.96	86	1/5	430
	20				7.8		19.7		27		8.00	115	1/5	575
<b>Day 6</b> <b>AM</b> Date: 2/7/06 Time: 0935 Technician: GZ/JW	Control		3		7.4		19.8		29		7.88	0	0	0
	2.5				7.4		19.9		28		7.95	10	0	10
	5				7.5		19.9		27		7.96	47	0	47
	10				7.2		19.9		27		7.97	140	0	140
	15				7.1		20		27		8.00	45	1/5	225
	20				6.6		19.9		4		8.32	290	1/5	1450
<b>Day 6</b> <b>PM</b> Date: 2/7/06 Time: 1408 Technician: TS/JM	Control		4		7.6		19.5		29		7.84	0	0	0
	2.5				7.5		19.8		27		7.90	17	0	17
	5				7.4		19.9		27		7.93	49	0	49
	10				7.6		19.7		27		7.94	126	0	126
	15				7.1		19.9		27		7.94	280	0	280
	20				6.9		20.0		27		8.03	397	0	397
<b>Day 7</b>  Date: 2/8/06  Time: 1615  Technician: AM/JM/TS	Control		1		7.4		19.4		28		7.82	0	0	0
			2		7.4		19.8		28		7.83	0	0	0
			3		7.4		19.9		28		7.82	0	0	0
			4		7.2		19.9		28		7.85	0	0	0
	2.5		1		7.1		19.8		26		7.85	11	0	11
			2		7.4		19.9		26		7.86	5	0	5
			3		7.2		19.8		26		7.85	7	0	7
			4		7.2		19.9		26		7.85	4	0	4
	5		1		7.5		19.8		26		7.92	33	0	33
			2		7.3		19.9		26		7.94	28	0	28
			3		7.5		19.8		26		7.94	24	0	24
			4		7.3		19.7		26		7.87	29	0	29
	10		1		7.2		19.9		26		7.91	114	0	114
			2		7.1		19.9		26		7.93	80	0	80
			3		7.3		19.9		26		7.91	94	0	94
			4		7.1		19.8		26		7.88	113	0	113
	15		1		7.2		19.7		26		7.95	236	0	236
			2		5.2		20.0		26		7.92	222	0	222
			3		6.7		19.8		26		7.96	256	0	256
			4		6.8		19.7		26		7.93	245	0	245
	20		1		6.6		19.9		26		7.91	306	0	306
			2		6.7		19.8		26		7.97	295	0	295
			3		6.3		19.8		26		7.97	311	0	311
			4		6.6		19.8		26		7.98	325	0	325

## **9.0 APPENDIX V: STATISTICAL RESULTS FOR THE PROTECTIVE LEVEL**

### Appendix V1. Primary Data Set Used in PRA Analysis

Phylum	Common Name	Species	Concentration (µg/L H <sub>2</sub> S)	Reference
Mollusca	Bay Mussel	<i>Mytilus galloprovincialis</i>	7	This study
Mollusca	Bay Mussel	<i>Mytilus galloprovincialis</i>	8	This study
Arthropoda	Mysid	<i>Americamysis bahia</i>	8	This study
Arthropoda	Mysid	<i>Americamysis bahia</i>	9	This study
Mollusca	Bay Mussel	<i>Mytilus edulis</i>	9	Knezovich et al., 1996
Mollusca	Bay Mussel	<i>Mytilus</i> sp.	10	Knezovich et al., 1996
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	11	Holland, et al., 1960
Arthropoda	Mysid	<i>Americamysis bahia</i>	12	This study
Echinodermata	Purple Sea Urchin	<i>Strongylocentrotus purpuratus</i>	13	Knezovich et al., 1996
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	15	Holland, et al., 1960
Arthropoda	Mysid	<i>Americamysis bahia</i>	17	This study
Mollusca	Red Abalone	<i>Haliotis rufescens</i>	17	This study
Echinodermata	Purple Sea Urchin	<i>Strongylocentrotus purpuratus</i>	19	Knezovich et al., 1996
Mollusca	Red Abalone	<i>Haliotis rufescens</i>	20	This study
Echinodermata	White Sea Urchin	<i>Lytechinus pictus</i>	21	Thompson et al., 1991
Chordata	Topsmelt	<i>Atherinops affinis</i>	23	This study
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	23	Holland, et al., 1960
Chordata	Inland Silverside	<i>Menidia beryllina</i>	26	This study
Chordata	Inland Silverside	<i>Menidia beryllina</i>	26	This study
Chordata	Inland Silverside	<i>Menidia beryllina</i>	28	This study
Chordata	Inland Silverside	<i>Menidia beryllina</i>	29	This study
Chordata	Inland Silverside	<i>Menidia beryllina</i>	30	This study
Chordata	Inland Silverside	<i>Menidia beryllina</i>	30	This study
Chordata	Northern Anchovy	<i>Engraulis mordax</i>	34	Bagarinao and Vetter, 1989
Echinodermata	Sea Urchin	<i>Paracentrotus lividus</i>	34	Knezovich et al., 1996
Arthropoda	Amphipod	<i>Ampelisca abdita</i>	39	This study
Chordata	Topsmelt	<i>Atherinops affinis</i>	40	This study
Chordata	Inland Silverside	<i>Menidia beryllina</i>	40	This study
Echinodermata	White Sea Urchin	<i>Lytechinus pictus</i>	43	Thompson et al., 1991
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	45	Holland, et al., 1960
Chordata	Inland Silverside	<i>Menidia beryllina</i>	48	This study
Arthropoda	Amphipod	<i>Ampelisca abdita</i>	48	This study
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	60	Holland, et al., 1960
Arthropoda	Penaeid Shrimp	<i>Penaeus indicus</i>	63	Gopakumar and Kuttyamma, 1996
Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	73	This study
Arthropoda	Penaeid Shrimp	<i>Metapenaeus dobsoni</i>	77	Gopakumar and Kuttyamma, 1996
Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	84	This study
Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	91	This study
Echinodermata	Sea Urchin	<i>Paracentrotus lividus</i>	95	Thompson et al., 1991
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	96	Holland, et al., 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	96	Holland, et al., 1960
Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	99	This study
Chordata	Speckled Sanddab	<i>Citharichthys stigmaeus</i>	102	Bagarinao and Vetter, 1989
Bacillariophyta	Diatom	<i>Skeletonema costatum</i>	105	Breteler et al., 1991
Annelida	Polychaete worm	<i>Neanthes arenaceodentata</i>	105	This study
Annelida	Polychaete worm	<i>Neanthes arenaceodentata</i>	105	This study
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	107	Holland, et al., 1960
Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	110	This study
Arthropoda	Penaeid Shrimp	<i>Penaeus indicus</i>	117	Gopakumar and Kuttyamma, 1996
Arthropoda	Penaeid Shrimp	<i>Penaeus indicus</i>	119	Gopakumar and Kuttyamma, 1996
Arthropoda	Penaeid Shrimp	<i>Metapenaeus dobsoni</i>	125	Gopakumar and Kuttyamma, 1996



Phylum	Common Name	Species	Concentration (µg/L H <sub>2</sub> S)	Reference
Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	132	This study
Chordata	Giant Kelpfish	<i>Heterostichus rostratus</i>	136	Bagarinao and Vetter, 1989
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	143	Holland et al, 1960
Arthropoda	Penaeid Shrimp	<i>Penaeus indicus</i>	144	Gopakumar and Kuttyamma, 1996
Arthropoda	Penaeid Shrimp	<i>Metapenaeus dobsoni</i>	147	Gopakumar and Kuttyamma, 1996
Arthropoda	Amphipoda	<i>Rhepoxynius abronius</i>	147	Knezovich et al., 1996
Chordata	Sheepshead Minnow	<i>Cyprinodon variegatus</i>	154	This study
Arthropoda	Amphipoda	<i>Rhepoxynius abronius</i>	160	Caldwell, 1975
Chordata	Black Surf Perch	<i>Embiotoca jacksoni</i>	170	Bagarinao and Vetter, 1989
Arthropoda	Penaeid Shrimp	<i>Penaeus indicus</i>	189	Gopakumar and Kuttyamma, 1996
Arthropoda	Amphipoda	<i>Eohaustorius estuarius</i>	192	Knezovich et al., 1996
Arthropoda	Amphipoda	<i>Anisogammarus confervicola</i>	200	Caldwell, 1975
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	200	Holland, et al., 1960
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	204	Holmer and Bondgaard, 2001
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	204	Holmer and Bondgaard, 2001
Arthropoda	Penaeid Shrimp	<i>Metapenaeus dobsoni</i>	219	Gopakumar and Kuttyamma, 1996
Arthropoda	Penaeid Shrimp	<i>Penaeus indicus</i>	281	Gopakumar and Kuttyamma, 1996
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	287	Holland et al, 1960
Chordata	Chinook Salmon	<i>Oncorhynchus tshawytscha</i>	300	Vismann, 1996
Chordata	Black Sea Turbot	<i>Rhombus maeoticus</i>	310	Ivanov et al., 1973
Arthropoda	Pacific Oyster	<i>Crassostrea gigas</i>	320	Caldwell, 1975
Arthropoda	Amphipoda	<i>Eohaustorius estuarius</i>	332	Knezovich et al., 1996
Arthropoda	Penaeid Shrimp	<i>Metapenaeus dobsoni</i>	340	Gopakumar and Kuttyamma, 1996
Arthropoda	Penaeid Shrimp	<i>Penaeus indicus</i>	342	Gopakumar and Kuttyamma, 1996
Arthropoda	Penaeid Shrimp	<i>Metapenaeus dobsoni</i>	378	Gopakumar and Kuttyamma, 1996
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	408	Vismann, 1996
Magnoliophyta	Eelgrass	<i>Zostera marina</i>	408	Vismann, 1996
Chordata	Long-Jawed Mudsucker	<i>Gillichthys mirabilis</i>	417	Bagarinao and Vetter, 1989
Chordata	Kelp Bass	<i>Paralabrax clathratus</i>	476	Bagarinao and Vetter, 1989
Chordata	Sand Bass	<i>Paralabrax nebulifer</i>	476	Bagarinao and Vetter, 1989
Arthropoda	Red Rock Crab	<i>Cancer magister</i>	500	Caldwell, 1975
Phaeophyta	Rock Weed	<i>Fucus serratus</i>	560	Chapman and Fletcher, 2002
Chordata	Long-Jawed Mudsucker	<i>Gillichthys mirabilis</i>	625	Bagarinao and Vetter, 1989
Chordata	Atlantic Salmon	<i>Salmo salar</i>	670	Kiemer et al., 1995
Arthropoda	copepods	mixed	765	Marcus et al. 1987
Annelida	Nereidae	<i>Neanthes arenaceodentata</i>	780	Dillon et al., 1993
Chordata	Bay Blenny	<i>Hypsoblennius gentilis</i>	782	Bagarinao and Vetter, 1989
Chordata	California Killifish	<i>Fundulus parvipinnis</i>	833	Bagarinao and Vetter, 1989
Arthropoda	Rock Crab	<i>Cancer antennarius</i>	1000	Vetter et al. 1987
Arthropoda	Dungeness Crab	<i>Cancer magister</i>	1000	Caldwell, 1975
Arthropoda	Amphipoda	<i>Corophium salmonis</i>	1000	Caldwell, 1975
Arthropoda	Pacific Oyster	<i>Crassostrea gigas</i>	1000	Caldwell, 1975
Chordata	California Halibut	<i>Paralichthys californicus</i>	1122	Bagarinao and Vetter, 1989
Mollusca	Pacific Oyster	<i>Crassostrea gigas</i>	1400	Caldwell, 1975
Chordata	Striped Mullet	<i>Mugil cephalus</i>	1428	Bagarinao and Vetter, 1989
Annelida	Capitellida	<i>Capitella capitata</i>	1724	Dubilier, 1988
Chordata	California Killifish	<i>Fundulus parvipinnis</i>	1802	Bagarinao and Vetter, 1989
Chordata	Long-Jawed Mudsucker	<i>Gillichthys mirabilis</i>	1802	Bagarinao and Vetter, 1989
Annelida	Nereidae	<i>Neanthes arenaceodentata</i>	2035	Dillon et al., 1993
Annelida	Spionida	<i>Streblospio benedicti</i>	2244	Llanso, 1991
Arthropoda	Isopoda	<i>Gnorimosphaeroma oregonensis</i>	5200	Caldwell, 1975
Mollusca	Bent Nose Clam	<i>Macoma balthica</i>	6000	Caldwell, 1975

Historic Data								
No.	Value	log[conc]	Cum%		No.	Value	log[conc]	Cum%
1	9	0.9542	1%		36	287.0	2.4579	51%
2	10	1.0000	3%		37	300.0	2.4771	53%
3	13.0	1.1139	4%		38	310	2.4914	54%
4	19	1.2788	6%		39	320	2.5051	56%
5	21	1.3222	7%		40	332	2.5211	57%
6	34	1.5315	9%		41	340	2.5315	59%
7	34	1.5315	10%		42	342	2.5340	60%
8	42.5	1.6284	11%		43	378	2.5775	61%
9	60.0	1.7782	13%		44	408	2.6107	63%
10	63	1.7993	14%		45	408	2.6107	64%
11	77	1.8865	16%		46	417	2.6201	66%
12	95	1.9763	17%		47	476	2.6776	67%
13	96	1.9800	19%		48	476	2.6776	69%
14	95.5	1.9800	20%		49	500	2.6990	70%
15	102.0	2.0086	21%		50	560	2.7482	71%
16	105	2.0212	23%		51	625	2.7959	73%
17	107	2.0294	24%		52	670.0	2.8261	74%
18	117.0	2.0682	26%		53	765	2.8837	76%
19	119.0	2.0755	27%		54	780	2.8921	77%
20	125	2.0969	29%		55	782	2.8932	79%
21	136	2.1335	30%		56	833	2.9206	80%
22	143	2.1553	31%		57	1000	3.0000	81%
23	144.0	2.1584	33%		58	1000	3.0000	83%
24	147	2.1673	34%		59	1000.0	3.0000	84%
25	147	2.1673	36%		60	1000.0	3.0000	86%
26	160	2.2041	37%		61	1122	3.0500	87%
27	170	2.2304	39%		62	1400	3.1461	89%
28	189.0	2.2765	40%		63	1428	3.1547	90%
29	192.0	2.2833	41%		64	1724	3.2365	91%
30	200	2.3010	43%		65	1802	3.2558	93%
31	200	2.3010	44%		66	1802	3.2558	94%
32	204	2.3096	46%		67	2035	3.3086	96%
33	204	2.3096	47%		68	2244	3.3510	97%
34	219	2.3404	49%		69	5200	3.7160	99%
35	281.0	2.4487	50%		70	6000	3.7782	100%

N=70

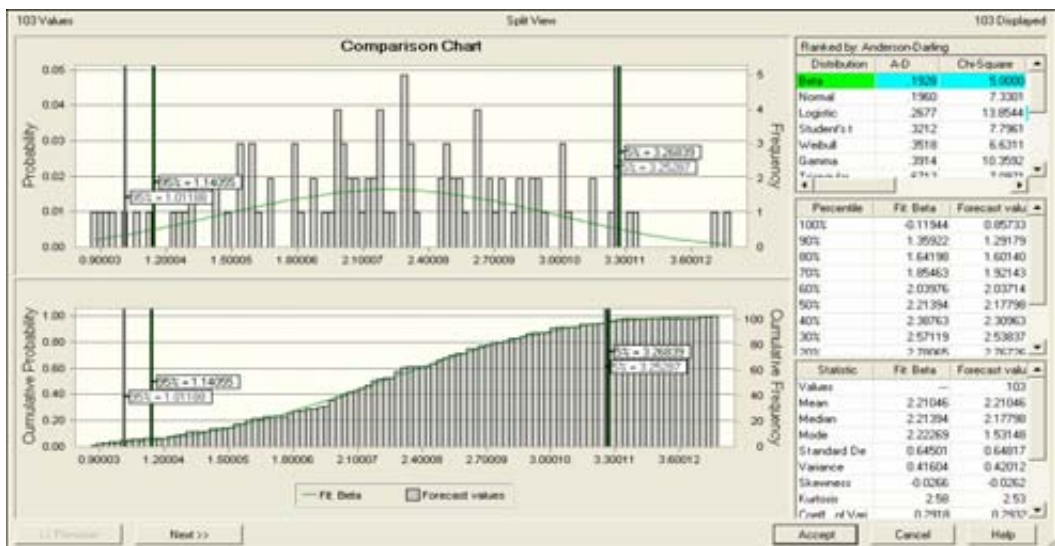
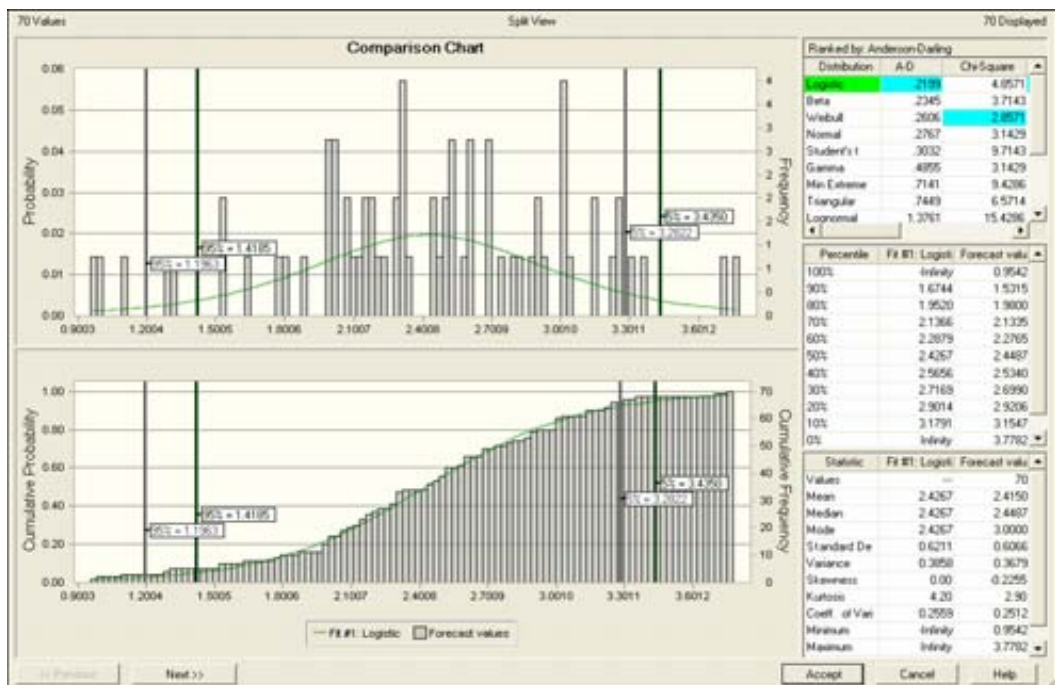
Mean = 616.36

Stdev = 1012.3

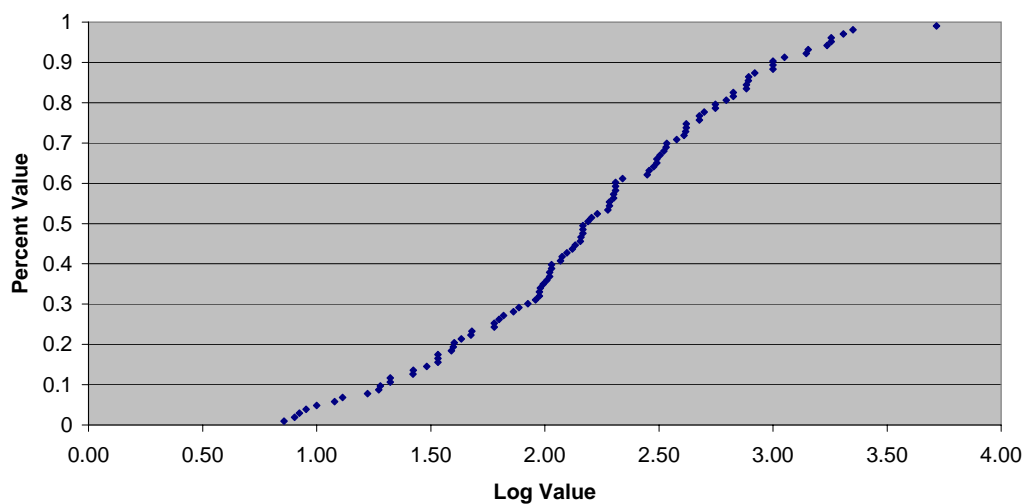
# **All Data**

No.	Value	log[conc]	Cum%	valuesonly	No.	Value	log[conc]	Cum%	valuesonly	No.	Value	log[conc]	Cum%	valuesonly
1	7.2	0.857	1%	0.85733	36	98.5	1.993	35%	1.99344	71	340	2.531	69%	2.53148
2	8	0.903	2%	0.90309	37	102	2.009	36%	2.00860	72	342	2.534	70%	2.53403
3	8.4	0.924	3%	0.92428	38	105	2.021	37%	2.02119	73	378	2.577	71%	2.57749
4	9	0.954	4%	0.95424	39	105.1	2.022	38%	2.02160	74	408	2.611	72%	2.61066
5	10	1.000	5%	1.00000	40	107	2.029	39%	2.02938	75	414	2.617	73%	2.61700
6	12	1.079	6%	1.07918	41	107	2.029	40%	2.02938	76	417	2.620	74%	2.62014
7	13	1.114	7%	1.11394	42	117	2.068	41%	2.06819	77	417	2.620	75%	2.62014
8	16.7	1.223	8%	1.22272	43	119	2.076	42%	2.07555	78	476	2.678	76%	2.67761
9	18.7	1.272	9%	1.27184	44	125	2.097	43%	2.09691	79	476	2.678	77%	2.67761
10	19	1.279	10%	1.27875	45	132.1	2.121	44%	2.12090	80	500	2.699	78%	2.69897
11	21	1.322	11%	1.32222	46	136	2.134	45%	2.13354	81	560	2.748	79%	2.74819
12	21	1.322	12%	1.32222	47	143	2.155	46%	2.15534	82	560	2.748	80%	2.74819
13	26.4	1.422	13%	1.42160	48	144	2.158	47%	2.15836	83	625	2.796	81%	2.79588
14	26.5	1.423	14%	1.42325	49	147	2.167	48%	2.16732	84	670	2.826	82%	2.82607
15	30.4	1.483	15%	1.48287	50	147	2.167	49%	2.16732	85	670	2.826	83%	2.82607
16	34	1.531	16%	1.53148	51	147	2.167	50%	2.16732	86	765	2.884	83%	2.88366
17	34	1.531	17%	1.53148	52	154.4	2.189	50%	2.18865	87	765	2.884	84%	2.88366
18	34	1.531	17%	1.53148	53	160	2.204	51%	2.20412	88	780	2.892	85%	2.89209
19	39	1.591	18%	1.59106	54	170	2.230	52%	2.23045	89	782	2.893	86%	2.89321
20	39.7	1.599	19%	1.59879	55	189	2.276	53%	2.27646	90	833	2.921	87%	2.92065
21	40.1	1.603	20%	1.60314	56	192	2.283	54%	2.28330	91	1000	3.000	88%	3.00000
22	43	1.633	21%	1.63347	57	192	2.283	55%	2.28330	92	1000	3.000	89%	3.00000
23	47.5	1.677	22%	1.67669	58	200	2.301	56%	2.30103	93	1,000	3.000	90%	3.00000
24	47.9	1.680	23%	1.68034	59	200	2.301	57%	2.30103	94	1122	3.050	91%	3.04999
25	60	1.778	24%	1.77815	60	204	2.310	58%	2.30963	95	1400	3.146	92%	3.14613
26	60	1.778	25%	1.77815	61	204	2.310	59%	2.30963	96	1428	3.155	93%	3.15473
27	63	1.799	26%	1.79934	62	204	2.310	60%	2.30963	97	1724	3.237	94%	3.23654
28	66	1.820	27%	1.81954	63	219	2.340	61%	2.34044	98	1802	3.256	95%	3.25575
29	72.9	1.863	28%	1.86273	64	281	2.449	62%	2.44871	99	1802	3.256	96%	3.25575
30	77	1.886	29%	1.88649	65	287	2.458	63%	2.45788	100	2035	3.309	97%	3.30856
31	84.2	1.925	30%	1.92531	66	300	2.477	64%	2.47712	101	2244	3.351	98%	3.35102
32	91	1.959	31%	1.95904	67	310	2.491	65%	2.49136	102	5200	3.716	99%	3.71600
33	94.7	1.976	32%	1.97635	68	310	2.491	66%	2.49136	103	6000	3.778	100%	3.77815
34	94.7	1.976	33%	1.97635	69	320	2.505	67%	2.50515	N = 103				
35	95.5	1.980	34%	1.98000	70	332	2.521	68%	2.52114					

Verification and Check			All Data Means	
	95% Log	95% [Conc]	mean	454.5
AllData	1.14055	13.8	sumlogs	227.6771
All Data2	1.01188	10.3	log geomean	2.210
			geomean	162.4



Logs Values of All Paired Toxicity Data



Species and Date of Test	LOEC				EC50			
	96 h (4 d) Avg	168 h (7 d) Surv Avg	168 h (7 d) Growth Avg	168 h (7 d) Biomass Avg	96 h (4 d) Avg	168 h (7 d) Surv Avg	168 h (7 d) Growth Avg	168 h (7 d) Biomass Avg
<b><i>Americamysis bahia</i></b>								
Acute: 1.13.06								
Acute: 1.19.06								
Acute: 1.25.06	33.1				9.4			
Acute: 2.05.06	9.9				11.1			
Chronic Test 1: 2.01.06		12.0	12.0			8.4	12.0	
Acute Test 1: 2.10.06	7.1				6.5			
MEAN	16.7				9.0	8.4	12.0	
<b><i>Ampelisca</i></b>								
Acute Test 1: 2.23.06	22.2				40.2			
Acute Test 2: 2.23.06	55.7				55.7			
Mean	39.0				47.9			
<b><i>Atherinops affinis</i></b>								
Acute: 3.20.06	11.4				41.6			
Acute: 3.21.06	34.5				37.8			
Mean	23.0				39.7			
<b><i>Cyprinodon variegatus</i></b>								
Acute: 2.10.06	40.1				40.1			
Acute Test 1: 2.15.06	118.4				171.6			
Acute Test 2 : 2.15.06	60.3				83.9			
Chronic Test 1: 2.15.06		133.0	123.1	133.0		134.3	172.9	128.4
Chronic Test 2: 2.15.06		35.4	58.8	86.8		174.6	91.4	90.6
Mean	72.9	84.2	91.0	109.9	98.5	154.4	132.1	109.5
<b><i>Menidia beryllina</i></b>								
Acute: 1.13.06	63.4				34.6			
Acute: 1.19.06	25.5				20.8			
Acute: 1.25.06	53.7				65.0			
Chronic: 2.10.06		14.0	22.1	22.1		22.1	22.1	22.1
Chronic: 3.13.06		44.0	44.0	44.0		33.9	44.0	37.1
Chronic: 3.14.06		21.3	25.1	21.3		23.3	25.1	23.7
Mean	47.5	26.4	30.4	29.1	40.1	26.5	30.4	27.6
<b><i>Neanthes arenaceodentata</i></b>								
4 d Test 1: 2.17.06	123.5				123.5			
4 d Test 2: 2.17.06	86.6				86.6			
Mean	105.1				105.1			
<b><i>M. galloprovincialis</i></b>								
0 Hour Spike	6.8				6.9			
6 Hour Spike	12.8				17.2			
12 Hour Spike	23.6				23.6			
24 Hour Spike	15.7				15.7			
36 Hour Spike	35.0				35.0			
<b><i>Continuous</i></b>	9.0				10.0			
Test 1	7.5				7.6			
Test 2	5.2				6.5			
Mean	7.2				8.0			

red font means > than values